Pedobiologia 58 (2015) 129–135

Contents lists available at ScienceDirect

Pedobiologia - Journal of Soil Ecology

journal homepage: www.elsevier.de/pedobi

The role of seedling nutrient status on development of ectomycorrhizal fungal communities in two soil types following surface mining disturbance

Jacob Gaster, Justine Karst*, Simon M. Landhäusser

[a] University of Alberta, Department of Renewable Resources, 4-42 Earth Sciences Building, Edmonton, Alberta T6G 2E3, Canada

ARTICLE INFO

Article history:
Received 26 November 2014
Received in revised form 3 July 2015
Accepted 14 July 2015

Keywords:
Forest restoration
Populus tremuloides
Nutrition
Root carbon reserves
Soil substrate

ABSTRACT

Severe disturbances, such as those caused by surface mining, sever connections between ectomycorrhizal (ECM) fungi and their hosts. In this study, we examined the importance of internal plant nutritional status and soil type on the abundance and composition of ECM communities in newly established reclamation areas. Ectomycorrhizal fungi were examined on two aspen (*Populus tremuloides*) seedling stock types that differed in root nutrient concentrations at time of planting. Stock types were planted into two salvaged soil types, an upland forest soil and a lowland peat-mineral soil mix, placed on a reclaimed mining site. Two growing seasons following planting, ECM fungal richness and abundance was low with only four operational taxonomic units identified across the reclamation site. Initial seedling nutrient status affected the total amount of ECM fungi on seedling roots; seedlings with initially high root nutrient reserves (N, P and starch) had more root tips colonized by ECM fungi than seedlings with initially lower nutrient tissue concentrations. Soil type did not affect total amount of ECM colonization; however, the relative abundance of an individual species, *Cenococcum geophilum*, was influenced by soil type. Seedling nutrient reserves, independent of soil nutrition, affects the amount of ECM root colonization, while soil type affects the relative abundance of some ECM fungi colonizing roots.

1. Introduction

Restoring functional interactions between vegetation and soils is important to create self-sustaining ecosystems following landscape level disturbances (Macdonald et al., 2012). In the boreal forest, trees develop intimate associations with ectomycorrhizal (ECM) fungi, which affect tree survival and growth through their effects on resource uptake. Ectomycorrhizal hyphae extending from colonized roots are physical linkages that functionally connect tree roots to soils, where hosts supply the fungi with photosynthetically-derived sugars and the fungi provide water and soil-derived nutrients to their hosts. Communities of ECM fungi are highly diverse on micro-spatial scales, with multiple species often within centimeters of each other (Bruns, 1995). Though the relationship between mycorrhizal species diversity and host plant productivity is often context-dependent (Jonsson et al., 2001), high mycorrhizal diversity has been demonstrated to increase nutrient uptake and seedling growth (Baxter and Dighton, 2001; Velmala et al., 2014). During severe soil disturbances such as surface mining, vegetation, soils, and parent geological material are stripped to access resource deposits. Following mining, ECM associations must re-establish with planted seedlings. However, depending on the type and severity of disturbance, the diversity of the ECM fungal community is often much lower than in undisturbed areas (Kipfer et al., 2011; Read, 1991). Restoring ECM fungal communities can be a challenge in heavily disturbed soils, which generally have a low ectomycorrhizal inoculum potential (Bois et al., 2005; Hankin et al., 2015).

Seedling establishment is the first step towards re-vegetation on heavily disturbed sites. On these sites, seedlings are often exposed to stress such as poor nutrient availability, drought, or mineral toxicity. Elevated nutrient (nitrogen, phosphorous, and potassium) and non-structural carbon reserves (sugar and starch) in the tissues of planted seedlings can increase seedling growth and nutrient acquisition in stressful conditions such as those found in disturbed areas (Quoreshi and Timmer 2000a; Landhäusser et al., 2012; Schott, 2013). Although the higher reserves in seedlings are often only temporary, the improved growth

Abbreviations: ECM, ectomycorrhizal; FFM, forest floor material; NSC, non-structural carbohydrate; PMM, peat mineral mix.

* Corresponding author. Fax: +1 780 492 4323.
E-mail addresses: justine@karst.ca (J. Karst), Simon.Landhausser@ualberta.ca (S.M. Landhäusser).

http://dx.doi.org/10.1016/j.pedobi.2015.07.001

performance persists beyond the presence of elevated tissue nutrient levels (Schott, 2013). What underlies this longer-term response is not clear; however, it could be driven by belowground interactions such as those formed between plants and mycorrhizal fungi. The effect of seedling nutrient and carbon reserve status on the establishment of mycorrhizal symbionts in disturbed sites has received little attention. Prior studies have shown that soil nutrient availability influences the outcome between mycorrhizal fungi and their host (Johnson, 1993; Johnson et al., 1992); however, host nutrient and carbon reserve status may also influence the outcome of mycorrhizal interactions (Nylund, 1988). For instance, Quoreshi and Timmer (1998, 2000b) found that nutrient loading (i.e., artificially increasing nutrient reserves) black spruce seedlings (*Picea mariana* [Mill.] BSP.) stimulated mycorrhizal formation during inoculation. Since a balance exists between internal plant carbon and nutrients which governs mycorrhizal symbioses (Johnson, 2010), alterations to the nutrient levels in plant tissues may influence the development of the ECM symbiosis. High amounts of nutrients in the roots, particularly N, can inhibit the development of ECM fungi (Richards and Wilson, 1963). As ECM fungi rely predominantly on their host for carbon, variation in carbon reserves in seedling roots may also influence the abundance of ECM fungi.

In addition to seedling physiology, soil characteristics may also influence the abundance and species composition of ECM fungi occurring at disturbed sites. Soils commonly used in restoration of surface mines in the boreal forests are materials salvaged prior to mining: forest floor material (FFM), which is composed of the litter layer plus a portion of the underlying mineral soil from upland forest sites, and peat, which is salvaged from lowland peatlands and is often mixed with underlying mineral subsoil resulting in a peat-mineral mix (PMM). As can be expected, these two materials differ greatly in soil structure and chemistry, nutrient availability, and their plant and fungal propagule bank, which reflects the plant and fungal communities present at the sites prior to salvage (McMillan et al., 2007; Dimitriu et al., 2010; Schott, 2013). Salvaged soils may differ in the ECM propagules they retain in addition to acting as different habitats suitable for some ECM species, dependent upon fungal physiology, and thus lead to the development of dissimilarly structured ECM fungal communities.

The objective of this study is to characterize the respective influence of host internal plant nutritional status and soil type on the establishment of ECM fungal communities in highly disturbed areas. Specifically, we assessed the influence of *P. tremuloides* seedling nutrient and carbon reserve status on the early development of an ECM fungal community two years after planting and how the early ECM community was influenced by soil type (FFM and PMM). We hypothesized that seedlings with initially lower nutrient reserves would have greater ECM fungal abundance compared to seedlings with higher nutrient reserves because a lower nutrient status ought to lead to a greater necessity to develop and facilitate the establishment of the ECM symbiosis (Johnson et al., 1997). As *P. tremuloides* is an upland tree species, we expected a more abundant ECM community when seedlings were planted in FFM than in PPM.

2. Materials and methods

2.1. Site description

The study is located in the Central Mixedwood subregion of the boreal forest (Natural Regions Committee, 2006). Uplands of this region are typically dominated by white spruce (*Picea glauca* (Moench) Voss) and trembling aspen (*P. tremuloides* Michx.). The low lying areas are wetlands dominated by black spruce (*Picea mariana* (Mill.) Britton, Sterns & Poggenb.). The Central Mixedwood subregion endures long, cold winters and short, warm summers. Average daily temperatures range from −18.8 °C in January to 16.8 °C in July (Environment Canada 2013). Average annual rainfall and snow is 342 mm and 156 mm, respectively. The growing season (May–September) of our sample collection (2012) was near average with 350 mm of precipitation and a 12.7 °C average temperature (Environment Canada, 2013).

2.2. Soil salvage and placement

The field site was a reclaimed overburden dump (>100 ha) 26 km north of Fort McMurray, Alberta, Canada. Over six months beginning August 2008 and prior to building the overburden structure, the surface soils were stripped and stockpiled by soil type (forest floor material and peat-mineral mix). Upland forest floor material (FFM) was comprised of a mixture of the top 30 cm and included the organic L, F, H, and the mineral A and a portion of the B soil horizons of a Gray Luvisol occurring under stands of white birch (*Betula papyrifera* Marsh.), balsam poplar (*Populus balsamifera* L.), and/or trembling aspen (*P. tremuloides* Michx.). Peat-mineral mix (PMM) was salvaged from lowland sites where the surface material was stripped to a depth of roughly 30 cm, which included the transition/peat layer and mineral soil beneath it and was monitored to ensure a mix of 60:40 (volume:volume) peat to mineral soil. In 2009, after the soils had been stripped and stockpiled on-site, the dump was filled with overburden material (sodic and/or saline) and the site was sloped with a height to volume ratio of six. In 2010, the construction of the overburden dump finished and 1 m of subsoil (low-sodic soil salvaged from the C-horizon from a depth of 60 cm to 300 cm) was placed across the surface. Capping soil was placed at a depth of 50 cm; placement commenced in August 2010 and was finished in June 2011. The study site was 1.5 ha in size; the salvaged and stockpiled FFM and PMM soil types were placed in alternating 20 m wide and 65 m long strips. A pair of FFM and PMM strips (40 m × 65 m) was considered an experimental block ($n = 5$).

2.3. Tree seedling production and planting

Aspen seedlings were grown commercially from an open-pollinated seed source collected in the Fort McMurray (Alberta, Canada) region. Seedling growing conditions during nursery production are described in more detail in Schott et al. (2013). Briefly, seeds were sown into Styroblock containers (5-12A, 220 ml, Beaver Plastic, Edmonton, Alberta) and grown for a single growing season at Smoky Lake Nursery (Smoky Lake, Alberta, Canada). Once seedlings had reached an average height of 35 cm, they were assigned to two treatments: *high feed* and standard. Standard seedlings were grown under typical seedling production conditions and fertilized with a mixture of macro and chelated micronutrients (78 ppm N, 77 ppm P, 161 ppm K, 46 ppm S) while the high feed seedlings received double the amount of the same fertilizer (i.e., 156 ppm N, 154 ppm P, 322 ppm K, 92 ppm S, including chelated micronutrients). Fertilization continued at these concentrations until early fall after which all fertilization ceased. Dormancy and hardening was induced by leaving seedlings outside and seedlings were lifted and stored frozen (−3 °C) once day temperatures were below freezing. Roots of seedlings were not examined for ectomycorrhizas prior to planting. Aspen seedlings were planted in early spring of 2011 in alternating rows of high feed and standard seedlings within each capping treatment and block. Seedlings were regular spaced at 1.3 m (5917 stems/ha). At the time of planting, high feed seedlings were of similar size as