Université du Québec en Abitibi-Témiscamingue

LES MÉCANISMES DE TRANSITION ENTRE DEUX ÉCOSYSTÈMES, LES FORÊTS OUVERTES À LICHEN ET LES FORÊTS FERMÉES À MOUSSE, ET LA RELATION ENTRE CES DEUX ÉCOSYSTÈMES ET L'HÉRITAGE DES INCENDIES DANS LA FORÊT BORÉALE

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MECHANISMS FOR THE TRANSITION BETWEEN TWO ECOSYSTEMS, OPEN-CANOPY LICHEN AND CLOSED-CANOPY MOSS FORESTS, AND THE RELATIONSHIP OF THESE TWO ECOSYSTEMS TO THE FIRE LEGACY IN THE BOREAL FOREST

Thesis
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This doctoral thesis is the culmination of nearly three and a half years of research on forest ecology in boreal forest ecosystems, the magnificent landscapes of which fulfilled my childhood imagination. In 2017, my academic career led me to join the Inner Mongolia University in China. My master's thesis focused on characterizing soil carbon sequestration by reforestation. This experience gave me the opportunity to pursue my research into forest ecology, by completing a PhD in forest ecology under the supervision of the Université du Québec in Abitibi-Témiscamingue (UQAT), in Canada.

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DEDICATION

In memory of my grandmother, Lin Yang
She gave me the most important thing in the world, love.

To my mentor, Dr. Yves Bergeron.
His wisdom and encouragement have guided me along the way.

To my mentor, Dr. David Paré.
His patience and guidance became a boost on my road to success of doctorate.

FOREWORD

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Introduction

Chapter 1. Gao, L., Paré, D., Chavardès, Raphaël D., & Bergeron, Y. (2023). Initiating the transition from open-canopy lichen woodland to productive forest by transplanting moss, results from a 10-year experiment. *Plant and Soil,* 1-14. https://doi.org/10.1007/s11104-023-05977-w

Chapter 2. Gao, L., Paré, D., Martineau, C., Yin, X., Rodríguez-Rodríguez, J.C., Gagné, P., & Bergeron, Y. (2023). Response of the soil microbial communities to forest ground cover manipulation in a boreal forest. *Forest Ecology and Management*, 553(C):121615. https://doi.org/10.1016/j.foreco.2023.121615

Chapter 3. Gao, L., Paré, D., Braghiroli, F., Bergeron, Y. (2023) Relationships between charcoal property and ecosystem status in the boreal forest (Manuscript).

General conclusion

The thesis is my own composition and involved collaboration with my supervisors and other collaborators. My supervisors helped me set the research direction, the design and sampling of the fieldwork, the data analysis and the academic writing. **In chapter 1**, Gao Lei: conceptualization, data curation, formal analysis, methodology, software, visualization, writing – original draft. Paré David: conceptualization, formal analysis, , writing – review & editing. Chavardès, Raphaël D.: methodology, writing – review & editing. Bergeron Yves: conceptualization, supervision, writing – review & editing. **In chapter 2**, Gao Lei: conceptualization, data curation, formal analysis, methodology, software, visualization, writing – original draft. Paré David: conceptualization, formal analysis, project administration, supervision, writing – review & editing. Martineau Christine: methodology, resources, software, visualization, writing – review & editing. Yin Xiangbo: methodology, software, visualization, writing – review & editing. Rodríguez-Rodríguez Juanita: methodology, software, visualization, writing – review & editing. Gagné Patrick: data curation, methodology, software, visualization, writing – review & editing. Bergeron Yves: conceptualization, formal analysis, funding acquisition, project administration, supervision, writing – review & editing. **In chapter 3**, Gao Lei: conceptualization, data curation, formal analysis, supervision, writing—original draft. Paré David: conceptualization, writing—original draft. Braghiroli Flavia: conceptualization, methodology. Bergeron Yves: conceptualization, writing—original draft. I have permission from all co-authors to include publications or manuscripts in my thesis.

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RÉSUMÉ

Avec le changement global, le taux de perturbations naturelles devrait augmenter dans de grandes parties de la région boréale, comme la fréquence accrue des incendies et les perturbations combinées (épidémies d'insectes et incendies) qui rendent la forêt plus vulnérable à la transformation écologique. Dans les forêts boréales de l'est du Canada, on a observé le passage de forêts productives à couvert fermé de mousses (*Pleurozium schreberi* (Brid.) Mitt.) à des forêts ouvertes de lichens (*Cladonia* spp.) peu productives. Ces deux écosystèmes forestiers (mousses et lichens) sont considérés comme des états stables alternatifs distincts, les premiers favorisant les conditions biotiques et abiotiques qui maintiennent les forêts productives et les seconds favorisant les conditions qui maintiennent les sites à canopée ouverte à faible productivité. On a émis l'hypothèse qu'une sévérité élevée des incendies serait l'une des causes de cette transition. Les propriétés du charbon de bois étant affectées par la gravité des incendies, nous avons émis l'hypothèse que la quantité et les propriétés du charbon de bois seraient différentes et indiqueraient une plus grande gravité des incendies dans les forêts à canopée ouverte que dans les forêts à canopée fermée. En outre, le charbon de bois issu des perturbations causées par le feu a un grand potentiel pour améliorer la qualité du sol et favoriser la croissance de la végétation. Toutefois, les causes qui contribuent à la transition entre les deux états alternatifs sont mal connues.

Afin de mieux comprendre la transition entre ces deux états alternatifs dans les forêts boréales, nous avons examiné les effets de la transplantation de mousses dans une forêt de lichens ouverte sur la croissance des arbres, les nutriments foliaires, les conditions du sol et les communautés microbiennes du sol, y compris les bactéries et les champignons, après 10 ans, ainsi que les différences dans les quantités et les propriétés du charbon de bois entre ces deux types d'écosystèmes. À notre connaissance, il s'agit de la première expérience utilisant la transplantation de mousse. Notre dispositif expérimental a permis d'étudier le rôle direct de la couverture végétale. Les études précédentes, utilisant des approches observationnelles, ont été limitées dans leur interprétation par la corrélation des facteurs déterminants, y compris les attributs du site et du terrain et la végétation de couverture. Nos résultats ont confirmé que les mousses hypnacées peuvent s'établir, survivre et rester en bonne santé dans un environnement précédemment occupé par le lichen. Le remplacement du lichen par les mousses hypnacées crée des conditions de sol propices à une meilleure croissance des arbres (Chapitre 1). La modification du couvert forestier a un impact significatif sur la diversité, la composition et la fonction des communautés microbiennes du sol. La transplantation de mousse a augmenté de manière significative l'abondance relative du genre fongique Piloderma, responsable de l'élimination de l'azote organique (Chapitre 2). Nous avons également constaté que la quantité moyenne de charbon de bois dans les forêts de mousses était significativement plus élevée que dans les forêts de lichens. La présence de charbon de bois après un incendie peut contribuer au rétablissement et à la régénération de la végétation sous une certaine quantité de charbon de bois, et semble favoriser la survie de la mousse. Cela pourrait démontrer le rôle potentiel du charbon de bois dans l'explication de la cooccurrence de deux états alternatifs stables (Chapitre 3).

En ce qui concerne la productivité de la forêt boréale, le rôle des mousses hypnacées a été démontré à l'aide d'un dispositif expérimental (Chapitre 1-2). Compte tenu du rôle des mousses et des lichens en tant que boucles de rétroaction potentielles sur les conditions du sol, les communautés microbiennes du sol et la croissance des arbres, il est possible de maintenir des conditions forestières productives en favorisant des conditions favorables aux mousses hypnacées plutôt qu'aux lichens. Cette étude démontre également que deux écosystèmes forestiers, la mousse et le lichen, ont une différence significative sur la quantité de charbon de bois, mais ne semblent pas avoir une différence significative sur les propriétés chimiques et physiques du charbon de bois, bien qu'il y ait une plus grande variabilité des propriétés du charbon de bois pour les peuplements à canopée ouverte, donc ces résultats ne nous permettent pas de valider ou de rejeter complètement notre hypothèse sur le rôle de la sévérité du feu dans le changement entre les peuplements à canopée ouverte et fermée. Cependant, ils suggèrent que la variabilité des conditions d'incendie ainsi que la quantité de charbon de bois produite sont différentes entre les types d'écosystèmes. En outre, étant donné le rôle potentiel du charbon de bois dans la dynamique de la forêt boréale, d'autres études seront nécessaires pour évaluer la relation entre le charbon de bois et le sol forestier et la nature du feu, et pour développer des stratégies de gestion forestière appropriées.

Mots clés: Mousses hypnacées; Lichen; Conditions du sol; Microbes du sol; Croissance des arbres; Charbon de bois; Productivité des forêts; L'héritage du feu; Écosystème; Forêt boréale

ABSTRACT

With global change, the rate of natural disturbances is predicted to increase over large portions of the boreal region, such as increased fire disturbance and compound disturbances (insect outbreaks and fires) which are making the forest more vulnerable to ecological transformation. Shifts from productive closed-canopy feather moss (*Pleurozium schreberi* (Brid.) Mitt.) forests to low-productivity open lichen (*Cladonia* spp.) woodlands in boreal forests of eastern Canada have been observed. These two forest ecosystems (moss vs lichen) are considered as distinct alternative stable states, with the former favoring biotic and abiotic conditions maintaining productive woodlands and the latter favoring condition that maintain in low-productivity open-canopy sites. It has been hypothesized that a high severity of fires would be the one cause of this transition. Because charcoal properties are affected by fire severity, we have put forward the hypothesis that the amount and properties of charcoal would be different and indicative of a greater fire severity for open-canopy forests compared to closed canopy ones. Additionally, charcoal from fire disturbances may have a great potential to improve soil quality and promote vegetation growth. However, the causes that contribute to the transition between the two alternative states are poorly known.

To better understand the transition between these two alternative states in boreal forests, we examined the effects of moss transplantation in an open lichen woodland on tree growth, foliar nutrients, soil conditions and soil microbial communities, including bacteria and fungi, after 10 years, as well as the differences in charcoal amounts and properties between these two ecosystems. To our knowledge, this is the first experiment using moss transplantation. Our experimental set-up made possible to investigate the direct role of ground cover vegetation. Previous studies, using observational approaches were limited in their interpretation by the correlation of driving factors including site and terrain attributes and ground cover vegetation. Our results confirmed that feather moss can establish, survive, and remain healthy in an environment previously occupied by lichen. The replacement of lichen by feather moss establishes soil conditions that were conducive to better tree growth (Chapter 1). Changing the forest ground cover also has a significant impact on the diversity, composition and function of soil microbial communities. Moss transplantation significantly increased the relative abundance of the organic nitrogen-scavenging fungal genus, *Piloderma* (Chapter 2). We also found that the amount of charcoal in moss forests was significantly higher than that in lichen woodlands. The presence of charcoal after a fire may contribute to the recovery and regeneration of vegetation under an amount of charcoal and seems to favor the survival of moss. This may demonstrate that the potential role of charcoal in explaining the co-occurrence of two stable alternative states (Chapter 3).

In terms of boreal forest productivity, the role of feather moss was demonstrated with an experimental set-up (Chapter 1-2). Considering the role of moss and lichen as potential feedback loops on soil conditions, soil microbial communities and tree growth, productive forest conditions can be maintained by promoting conditions that are favorable to feather moss rather than lichen. This study also demonstrates that two forest ecosystems, moss and lichen, had a significant difference on the amount of charcoal, but did not seem to have a significant difference on charcoal chemical and physical properties albeit a greater variability of charcoal properties for open canopy stands. Thus, these results do not allow us to fully validate or reject our hypothesis on the role of fire severity in the shift between open and closed canopy stands. However, they suggested that the variability in fire conditions as well as the amount of charcoal produced are different between ecosystem types. Furthermore, given the potential role of charcoal in boreal forest dynamics, further studies will be needed to assess the relationship between charcoal and forest soil and the nature of fire, and to develop the appropriate forest management strategies.

Keywords: Feather moss; Lichen; Soil conditions; Soil microbes; Tree growth; Charcoal; Forest productivity; Fire legacy; Ecosystem; Boreal forest

INTRODUCTION

Background. Global climate change is having increasing impacts on terrestrial ecosystems, such as leading to warmer and drier conditions, thus increasing fire activity, especially in the boreal forests of North America (Boulanger et al. 2014; Ellis et al. 2022; Davis et al. 2023). In the boreal forest, fire is one of the dominant natural disturbances that are making the forest more vulnerable to ecological transformation (Baltzer et al. 2021; Davis et al. 2023). Shifts from productive closed-canopy feather moss forests to low-productivity open lichen woodlands in boreal forests of eastern Canada have been reported (Girard et al. 2008; Pacé et al. 2020a), and may be more frequent due to increasing fire severity and frequency (Veraverbeke et al. 2017; Baltzer et al. 2021). Moreover, the possibility that they are an alternative stable state and not simply a successional stage that culminates in a closed-moss forest (Girard et al. 2008). An alternative stable state is an ecosystem that can persist (i.e., pass through one or several turnovers) under the same environmental and climatic conditions as a different ecosystem type (Connell and Sousa 1983; Girard et al. 2008). A useful heuristic device that we use to represent shifting between alternative stable states is the ball-in-cup analogy outlined in Figure 0.1. This is often presented as an uneven surface with balls, because of the disturbance, moving to low areas where it is difficult to get out from (a stable state with positive feedback loops) (Fig.0.1). However, the causes that contribute to the transition between the two alternative states are poorly known.

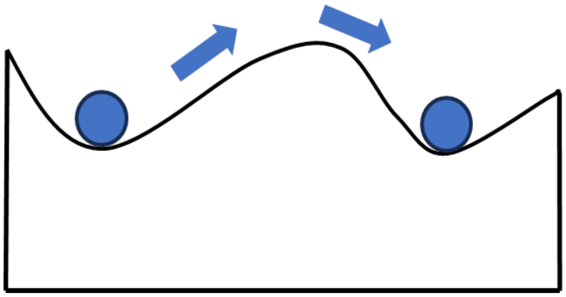

Figure 0.1 Ball-in-cup heuristic of system stability. Valleys represent stability domains, balls represent the system, and arrows represent disturbances (Beisner et al. 2003).

In eastern Canada, the structure of lichen woodlands is simple with two main strata: sparse trees (mainly black spruce and/or jack pine) several meters apart (< 25% canopy cover) and large expanses of terrestrial fruticose lichens (*Cladonia* spp.) (Payette et al. 2000; Ouimet et al. 2018). These structures can be detrimental to the forest productivity in boreal forests, which, combined with the arid conditions created by well-drained lichen woodlands (Haughian and Burton 2015), can contribute to more frequent fire disturbances in the region occupied by lichen (Kang et al. 2006). Lichens also have different roles in forest ecosystems. For example, lichens can increase seedling biomass accumulation, increase needle nitrogen uptake, serve as fodder for reindeer and caribou (Stark et al. 2007; Kytöviita and Stark 2009). However, compared to moss ground cover, reports of lichen causing tree regeneration failure through allelopathic effect and nutrient limitation deserve more attention (Hawkes and Menges 2003; Mallik and Kayes 2018; Pacé et al. 2020b). Feather moss cover tends to favor biotic and abiotic conditions maintaining productive woodlands, while lichen cover favors conditions that maintain in low-productivity open-canopy sites (Fig.0.2). However, a previous study by Pacé et al. (2020a) suggested that the difference in the edaphic characteristics associated to these two ecosystem states are not much contrasted, and suggested that the history of disturbance may be a major contributing factor to these two different ecosystem states. Fire frequency, fire severity, or compounding disturbances with insect outbreaks and thus forest regeneration failure or poor tree growth may be drivers of the transition between these two forest ecosystems (Fig.0.2). To our knowledge, there is no research on the potential for forest moss to invade lichen woodlands and to potentially break the resilience of stable open-canopy woodlands that could lead to more productive forests.

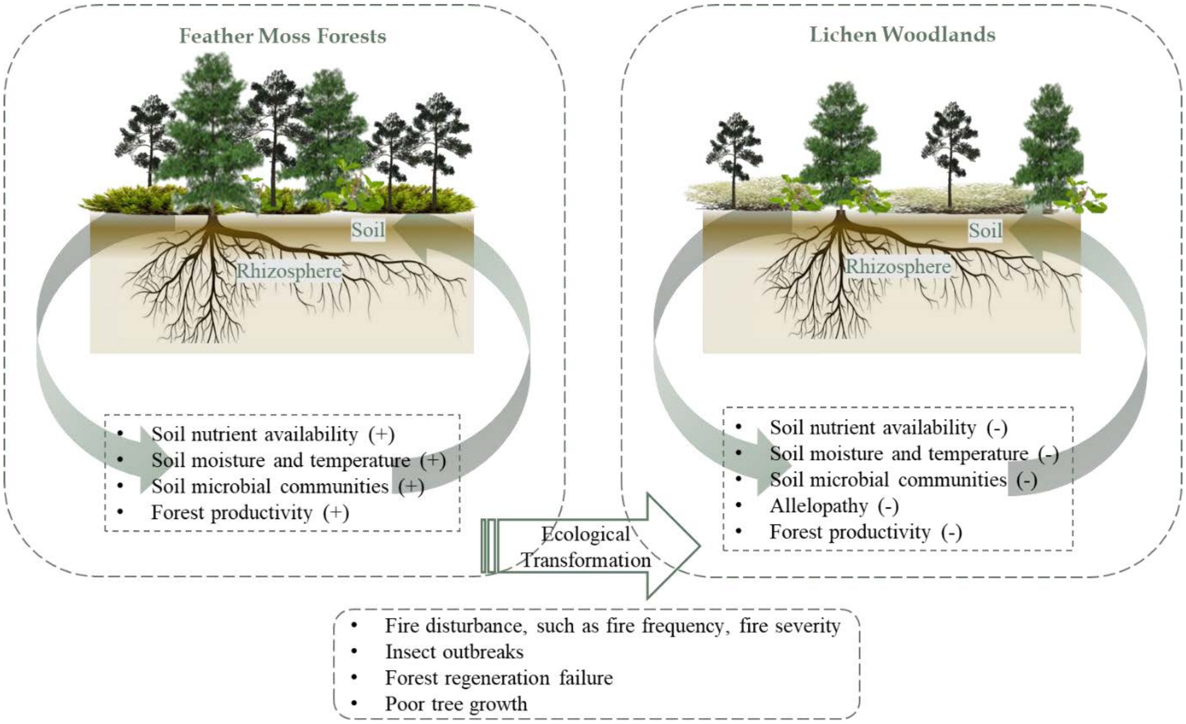

Figure 0.2 Drivers for maintaining two stable forest ecosystems, moss forests and lichen woodlands (Payette 1992; Sedia and Ehrenfeld 2006; Mallik and Kayes 2018; Pacé et al. 2020a) and the potential causes of transition between these two forest ecosystems (Girard et al. 2008; Brandt et al. 2013; Baltzer et al. 2021) in the North American boreal forests. (+) indicates the positive effects of moss on the ecosystem, (-) indicates the negative effects of lichen on the ecosystem.

Fire disturbance not only affects ecological transformation, but also produces fire-related pyrogenic substances, such as charcoal. In the forest fire, the charcoal formed by burning biomass resulting in the deposition of charcoal particles, most of which remain on site or in the vicinity for hundreds to thousands of years due to their stable aromatic structure (Forbes et al. 2006; Preston and Schmidt 2006; DeLuca and Aplet 2008). Charcoal accumulation was found to be highly variable across stands and plots likely due to heterogeneity of fire severity (Brimmer 2006; Preston et al. 2017). Ohlson et al. (2009) estimated that a total of approximately 1 Pg of carbon is contained in the soil charcoal of the world's boreal forests, an amount equivalent to about 15% of annual anthropogenic emissions from fossil fuel burning. Charcoal is characterized by its highly porous structure and high adsorption capacity (Brimmer 2006; Preston and Schmidt 2006). Additionally, charcoal may have an essential contribution to the re-establishment and growth of post-fire vegetation in the early stages after a forest fire (Zackrisson et al. 1996; Wardle et al. 1998; Robertson et al. 2012; Licht and Smith 2020). However, the potential role of charcoal in shaping both ecosystems, open-lichen woodlands and closed-moss forests, is still unclear.

Based on this information, we believe that assessing the potential for forest ground moss transplantation in a lichen woodland in the boreal forest and the relationship of post-fire charcoal to moss and lichen forests is necessary for future management and restoration of forest ecosystems. Our project implemented moss transplantation in an open lichen woodland in the boreal forest of eastern Canada. Treatments included: 1) removal of the lichen cover, 2) removal of the lichen cover followed by transplantation of a feather moss cover, 3) a control with the lichen cover kept in place (i.e., lichen control), and 4) a natural forest site with feather moss cover (i.e., moss control). We assessed the effects of moss transplantation on soil conditions, tree growth, foliar nutrients and soil microbial communities. Furthermore, considering that poor forest productivity may be possibly linked to fire severity (Girard et al. 2008; Brandt et al. 2013; Baltzer et al. 2021) and the indicative nature of charcoal for the recording of fire history (Pingree et al. 2012; Soucémarianadin et al. 2015; Santín et al. 2016), we analyzed the amount and properties of charcoal in lichen woodlands and moss forests, respectively. We predicted that moss transplantation would alter soil conditions, soil microbial communities, and improve tree growth. In addition, the amount of charcoal and its physic-chemical properties would differ between moss forests and lichen woodlands.

Disturbance dynamics in boreal forests. Boreal forest is one of the largest forest ecosystems in the world. Boreal ecosystems experience dynamic changes driven by a complex interplay of natural factors, including climate, fire, insects, and diseases, and their intricate interactions (Girard et al. 2008; Brandt et al. 2013). These disturbances have historically played crucial roles in rejuvenating ecosystems, regulating biogeochemical cycles, and shaping landscape diversity in terms of species composition, size, and succession. Boreal ecosystems are inherently dynamic, influenced by diverse intrinsic and extrinsic factors across various spatial and temporal scales.

Changes in climate, atmospheric carbon dioxide concentration and fire regimes have been occurring for decades in the global boreal forest, with future climate change likely to increase fire frequency—the primary disturbance agent in most boreal forests (Bond-Lamberty et al. 2007). In the Canadian boreal forest, most fires are stand-replacing, burn rates (percentage area burned annually) determine forest age-class distribution, as well as structure and composition at site and landscape levels, and drive the regional carbon balance in boreal forests (Payette 1992; Bond-Lamberty et al. 2007). Studies suggest this boreal biome will experience rapid temperature increases during the 21st century, with a potential 30–500% increase in burn rates (Balshi et al. 2009b; Bergeron et al. 2010; Boulanger et al. 2014). Both fire size and the frequency of large fire years are expected to increase (Kasischke and Turetsky 2006), with a cascading effect on ecosystem dynamics (Girard et al. 2008; Boiffin and Munson 2013) and carbon storage (Balshi et al. 2009a). Intense fire disturbances in certain boreal forest ecosystems challenge their resilience, particularly under high burn rates. These large and severe fires can disrupt seed sources and seedbeds for tree recruitment, potentially prompting transitions to new forest ecosystems (Arseneault 2001; Johnstone et al. 2010b). Moreover, shortened fire-free intervals might accelerate young, sexually immature trees to the maturation of tree populations, favoring faster- maturing species (Johnstone and Chapin 2006; Brown and Johnstone 2012; Buma et al. 2013). Notably, the cumulative forest carbon stocks face depletion due to the impacts of substantial and severe fires as well as shortened fire-free intervals (Brown and Johnstone 2011; Turetsky et al. 2011).

Under future climate and fire, the relative dominance of deciduous broadleaf trees nearly doubles by 2100, with commensurate declines in contributions from evergreen conifer trees and herbaceous plants in the Alaska boreal ecosystem (Mekonnen et al. 2019). Currently, the North American boreal landscape is characterized by evergreen conifers, particularly black spruce, which has demonstrated remarkable resilience over the Holocene. Through repeated self-replacement from aerial seedbanks, black spruce stands have withstood stand-replacing fires, underscoring the species resilience (Ilisson and Chen 2009; Johnstone et al. 2010a). In this context, fires occur more frequently than the life span of postfire tree cohorts (typically <100 to 150 years), thereby initial patterns of tree regeneration and relative growth rates determine the trajectory of future forest composition until the next fire event (Johnstone et al. 2010a; Héon et al. 2014). As a result, the initial years following a fire hold the key to seedling recruitment, which critically determines the composition of mature boreal forest stands for boreal tree species (Greene et al. 2004; Ilisson and Chen 2009; Johnstone et al. 2010a). Recent studies have indicated that fires of unusually high severity and/or short return intervals can disrupt the successful regeneration strategy of black spruce, stimulating shifts to alternative states dominated by deciduous broadleaf trees or jack pine (Jasinski and Payette 2005; Girard et al. 2008; Baltzer et al. 2021) or even non-forest (Splawinski et al. 2018; Whitman et al. 2019), changes indicative of reduced black spruce resilience (Baltzer et al. 2021). Thus, fire disturbances may result in forest regeneration failures, by inadequate seed supply due to fire burning (Davis et al. 2019) and severe fires that may result in loss of nutrients through volatilization from complete combustion of forest ground organic matter (e.g., nitrogen) (Brais et al. 2000; Certini 2005; Mehdi et al. 2012). Over the past few decades, the area occupied by closed-crown moss forests has decreased dramatically, and has shifted to open lichen woodlands in the boreal forests of eastern Canada (Girard et al. 2008). Therefore, there is a need to understand the causes for this shift and the role of abiotic and biotic factors in maintaining these two stable states.

Effects of lichen and moss on boreal ecosystems. The lichen woodland is one of the most widespread forest ecosystems in North America. The combined effects of insect infestation and fire may lead to a drastic reduction in forest tree regeneration after fire, thus inducing the conversion of spruce moss forests to lichen woodlands (Payette et al. 2000; Jasinski and Payette 2005). Lichens, symbiotic organisms consisting of mycobionts (fungi) and photobionts (algae and/or cyanobacteria), could serve as fodder for reindeer and caribou (Stark et al. 2007; Kytöviita and Stark 2009), and are a source of energy for soil microorganisms (Stark and Hyvärinen 2003). The lichen woodland is a typical nutrient-poor forest surviving on acidic, well-drained, moraine-derived soils and granitic outcrops composed of post-fire seral communities tightly tuned to a recurrent pattern of fire activity (Payette 1992; Pacé et al. 2020a). A previous study observed that lichen grazing positively affects Scots pine growth (Macias Fauria et al. 2008). Pacé et al. (2016) showed that lichen removal in Canadian boreal forest locally increased pine fine root biomass and that jack pine roots tended to be thinner under lichens. Lichen removal significantly increased soil potassium and base cations including calcium, sodium and magnesium, which confirmed that lichens also had short-term effects on soil chemical properties. Other studies have shown that lichen appears to be detrimental to the growth of trees (Hawkes and Menges 2003; Pacé et al. 2019). Lichen could reduce the availability of soil nutrients (Wheeler et al. 2011; Bastianelli et al. 2017), inhibit microbial communities (Sedia and Ehrenfeld 2003), and maintain lower soil moisture (Mallik and Kayes 2018) and allelopathy (Pacé et al. 2020b).

Contrary to lichen cover that is light dependent, feather moss cover tends to survive in shaded conditions of closed-canopy forests (Bonan and Shugart 1989; Haughian and Burton 2015). It influences forest ecosystem dynamics through its control of soil moisture and temperature (Zackrisson et al. 1997; Gornall et al. 2011; Mallik and Kayes 2018), regulation of soil nutrient availability (Wheeler et al. 2011; Bastianelli et al. 2017; Ouimet et al. 2018), modulation of soil microbial community activity (Sedia and Ehrenfeld 2003), and incidentally, the accumulation and degradation of organic matter (Sedia and Ehrenfeld 2005). As a result, ground cover composition plays a crucial role in shaping soil carbon and nitrogen cycles (Turetsky 2003; Smith et al. 2017). Moreover, the research by DeLuca et al. (2022) indicate that feather moss could act as a nitrogen source for forest ecosystems, potentially contributing to the nitrogen supply in boreal forest ecosystems.

Ecological roles of charcoal. Charcoal, is usually present and recalcitrant at the organic-mineral soil interface (Hart and Luckai 2013), and it is a C-rich, nitrogen (N)-poor thermogenic material with a highly aromatic molecular structure (DeLuca and Aplet 2008). Charcoal, due to its aromatic structure, exhibits high recalcitrance to decomposition in soil, leading to its long-lasting presence in the soil ecosystem for hundreds to thousands of years (DeLuca and Aplet 2008). Ohlson et al. (2009) estimated that a total of approximately 1 Pg of carbon is contained in the soil charcoal of the world's boreal forests, an amount equivalent to about 15% of annual anthropogenic emissions from fossil fuel burning. Along similar lines, Jones et al. (2019) suggested that cumulative pyrogenic charcoal production is 60 Pg since 1750 which could be equivalent to as much as 40% of the global anthropogenic carbon lost through land use change in this period. Charcoal accumulation was found to be highly variable across stands and plots likely due to heterogeneity of fire severity at the time of burning (Brimmer 2006; Preston et al. 2017) and stand biomass spatial variation. Some studies showed that charcoal formation seems to decrease with increasing fire severity (Pingree et al. 2012; Buma et al. 2014). Furthermore, fire severity is related to the charcoal physical properties, such as porosity (Shetty et al. 2021), and the chemical properties, such as carbon content and pH (Lehmann et al. 2011). It was shown that the maximum temperatures reached during fire and the associated prolonged heating times were associated with greater C enrichment, increased thermal recalcitrance and degree of charcoal aromaticity (Soucémarianadin et al. 2015; Santín et al. 2016). Therefore, the soil charcoal pool could offer an invaluable record of site-specific fire history.

Charcoal is characterized by its highly porous structure and high adsorption capacity (Brimmer 2006; Preston and Schmidt 2006). It contains residual ash, including calcium and magnesium, on its surfaces (Makoto and Koike 2021). The porous structure of charcoal increases its surface area, facilitating the adsorption of substances (Hart and Luckai 2013). For example, charcoal can adsorb organic compounds that may have an inhibitory effect on plants or microorganisms, which is important in phenol-rich ecosystems such as boreal forests (DeLuca and Aplet 2008). The porous nature of charcoal can provide many potentially beneficial physical functions to the soil, including increased soil water holding capacity and reduced soil bulk density (Gundale and DeLuca 2006; Lehmann et al. 2006), and increased nutrient retention (Hart and Luckai 2013). A study in the Canadian boreal forest, is the first effort to quantify charcoal produced from forest fire, and to examine the relationship between charcoal quantity and forest soil and plant growth comprehensively (Gale and Thomas 2021). They showed that under certain/or optimum dosage levels, charcoal can increase soil pH, soil nutrients (P, K, Mg), reduce soil bulk density and promote plant growth. Also, they suggested that charcoal is very useful for the growth of plants that regenerate after fire disturbance and is more important in more acidic and nutrient-poor boreal soils. Another study in the Russian Far East, showed that the presence of charcoal was correlated with fine root vitality in forest soils after surface fires, i.e., there was a positive correlation between charcoal and fine root vitality for overstory vegetation (Bryanin and Makoto 2017). However, there is less information on the links between charcoal and the two forest ecosystems, e.g., lichen woodlands and moss forests.

Research objectives and hypotheses. The general objectives of this doctoral thesis are to understand and characterize whether moss transplantation could successfully survive in open lichen woodlands, and to determine the effects of the changes in forest ground cover on soil conditions, tree growth, leaf nutrients and soil microbes; as well as to understand the relationship between charcoal (fire legacy) and lichen woodlands and moss forests in boreal forests.

Effects of ground cover manipulation on soil conditions and tree growth

Objectives:

1) Assess the 10-year effects of ground cover manipulation (lichen, lichen removal, lichen removal with transplantation of feather moss) on soil nutrients, and soil moisture and temperature.

2) Evaluate the effects of moss transplantation on tree growth as well as on leaf nutrient concentrations.

Hypotheses:

1) We hypothesize that the moss transplantation will change the soil moisture, soil temperature and nutrient availability to make it more conducive to the growth of trees.

2) We hypothesize that feather moss transplantation would result in better tree growth and foliar nutrition than lichen removal, and lichen, in that order.

Responses of soil microbes

Objectives:

1) Evaluate the effect of changes in forest ground cover on the composition and the diversity of soil bacterial and fungal communities for period of 10 years.

2) Determine the relationships between soil conditions and the soil microbial communities.

Hypotheses:

1) Different forest ground covers lead to distinct soil microbial communities.

2) Differences in microbial communities are linked to differences in soil conditions (nutrients, soil temperature and moisture) associated to different ground covers.

3) Forest ground covers leading to higher soil nutrient content and tree growth (moss transplantation, control moss) harbor higher proportions of copiotrophic/beneficial taxa.

Relationships between charcoal amount and properties and the two forest ecosystems (lichen woodlands versus moss forests)

Objectives:

1) Assess the difference in charcoal amount between lichen woodlands and moss forests.

2) Evaluate the relationship of charcoal amount to the above vegetation status.

3) Determine the difference in charcoal properties (C, N, H, O, S, ash content and surface area) between lichen and moss forests.

Hypotheses:

1) Charcoal amount is lower in lichen woodlands than in moss woodlands due to the previous occurrence of more severe fire in lichen woodlands compared to moss forests.

2) Charcoal amount will be associated with site productivity status, i.e., site productivity will be greater with increasing charcoal amount.

3) Because high fire severity (high pyrolysis temperature) could decreases the N, H, O content of charcoal, and increase ash, C and specific surface area (Weber and Quicker 2018; El-Naggar et al. 2019), we hypothesize that these charcoal properties be indicative of higher fire severity in lichen than in moss forests indicating that fire severity was greater in open-lichen woodland and suggesting that charcoal amount is related to the transition of closed to open canopy forests.

Study sites.

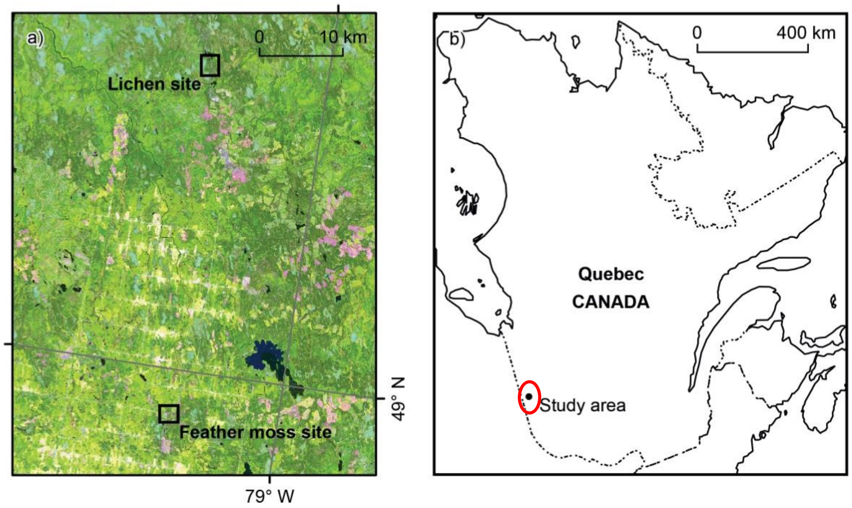

Figure 0.3 Study site of a) the open lichen woodland and b) the location in Quebec, Canada.

The experimental site (Fig. 0.3) is located in Western Quebec, Canada (49° 19’ 59” N; 79° 11’ 51” W), where is dominated by spruce-feather moss bioclimatic (Saucier et al. 2011). The average temperature and precipitation in the study area near La Sarre are 0 ± 2.9 °C and 909 mm, respectively (Pacé et al. 2020a). The age of jack pine stand in our designated fields was about 40 years, and the experimental site was selected in September 2011. The stand lies over sandy to coarse-grained fluvioglacial and glaciolacustrine deposits (MFFP. 2022). Ground cover is mainly composed of terricolous lichens including *Cladonia* stellaris (Opiz) Pouzar & Veda, C. *rangiferina* (L.) F.H. Wigg. and C. *mitis* Sandst (Pacé et al. 2020a). A sawfly outbreak was present near the study area during 2012–2014 (MFFP 2012). In June 2021, we selected a nearby naturally productive jack pine site, which had a continuous feather moss cover composed of *Pleurozium schreberi.* All soils are Humo-ferric Podzols or Dystric Brunisoils (Soil Classification Working Group 1998).

For the charcoal sites, see Pacé et al. (2020a) for more information, twenty-seven pure jack pine stands with lichen and feather moss ground cover were selected and sampled in our study. As several sites where the last major disturbance was harvesting, we do not include these sites. We only selected that the last major disturbance is fire. Ten were located near the locality of La Sarre (48° 48′N; 79°12′W), whereas the other seventeen were located near the locality of Chibougamau (49° 53′N; 74° 20′W). The last major disturbance in all sites was a forest wildfire. Average annual temperatures are 0.2 ± 3.7 °C and average annual precipitations are 995.8 mm in the near Chibougamau sites (Environment Canada 2018).

Thesis outline. This thesis is structured into five chapters, as shown in the figure (Figure 0.4). The main content of these five chapters is as follows:

Introduction (this chapter) presents the research background, objectives, methodology and thesis outline.

Chapter 1 presents the ten-year effects of an experimental ground cover manipulation experiment that included the transplantation of feather moss in an open lichen woodland. The effect of treatments on soil nutrients, soil moisture, soil temperature and on tree growth and needle nutrient concentrations were investigated. Thus, this study assesses the potential restoration of lichen woodlands to closed-canopy forests by moss transplantation.

Chapter 2 presents the effects of ground cover manipulation on the diversity and composition of soil microbial communities in the boreal forest and assesses soil environmental factors that may drive changes in soil microbial communities (indices of bacterial and fungal richness and diversity, as well as the relative abundance of the main genera). It then assesses the possible effects of this manipulation on soil function, specifically analyzing the functional community of soil fungi and the response of possible nitrogen-fixing organisms to this. Further, we explore the key role of soil microbes in this manipulation process and the implications for boreal forest dynamics.

Chapter 3 presents differences in charcoal amounts between lichen woodlands and moss forests, and the relationship between charcoal amounts and forest site productivity indices (degree of canopy opening, tree density, and site productivity indices) and ground cover biomass. As well, it investigates the differences in charcoal properties (C, N, H, O, S, ash content, specific surface area) between lichen woodlands and moss forests.

Conclusion briefly summarizes the results obtained in the thesis, discusses their significance, identifies research limitations and proposes future directions for research.

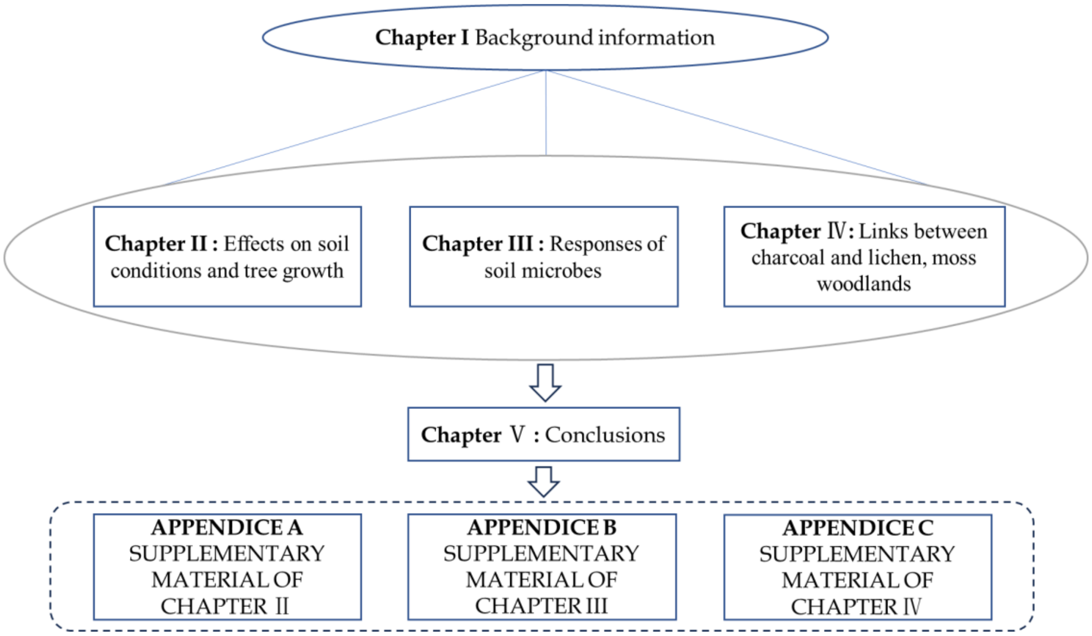

Figure 0.4 The thesis outline.

1. INITIER LA TRANSITION D'UNE FORÊT DE LICHENS À CANOPÉE OUVERTE À UNE FORÊT PRODUCTIVE EN TRANSPLANTANT DES MOUSSES, RÉSULTATS D'UNE EXPÉRIENCE DE 10 ANS

INITIATING THE TRANSITION FROM OPEN-CANOPY LICHEN WOODLAND TO PRODUCTIVE FOREST BY TRANSPLANTING MOSS, RESULTS FROM A 10-YEAR EXPERIMENT

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1.1 Résumé

Contexte et objectifs

Les forêts de lichens ouverts (*Cladonia* spp.) à faible productivité se sont rapidement étendues dans la forêt boréale à couvert fermé (*Pleurozium schreberi* (Brid.) Mitt.) de l'est du Canada. Bien que les zones de forêts ouvertes progressent, on dispose de peu d'informations sur la capacité des forêts de lichens ouverts à se reconstituer en forêts à canopée fermée.

Méthodes utilisées

Un dispositif expérimental utilisant la transplantation de mousses a été installé sur un peuplement pauvre de pin gris (*Pinus banksiana* Lamb.) avec une couverture végétale de lichens en 2011. Les traitements comprenaient: 1) l'élimination du lichen, 2) l'élimination du lichen et la transplantation d'une couverture de mousse, 3) le contrôle du lichen, et 4) un site naturel de pin gris avec une couverture de mousse (contrôle de la mousse). Nous avons extrait des carottes d'incréments de troncs d'arbres et recueilli des aiguilles et des échantillons de sol pour l'analyse des nutriments.

Résultats

Le traitement à base de mousse transplantée peut contrecarrer les effets négatifs du lichen sur la croissance du pin gris. Ce traitement a amélioré la nutrition foliaire et les nutriments du sol, en particulier l'ammonium (N-NH4+) et le nitrate (N-NO3-). Avec ce traitement, les conditions du sol (par exemple, les nutriments du sol, l'humidité du sol) et la nutrition foliaire étaient plus proches de celles du contrôle de la mousse. Il est surprenant de constater que l'élimination des lichens n'a pas amélioré la croissance et a entraîné une croissance plus faible du pin gris et des conditions de sol plus difficiles.

Conclusion

Les mousses hypnacées peuvent s'établir, survivre et rester en bonne santé dans un environnement précédemment occupé par le lichen. Le remplacement du lichen par les mousses hypnacées crée des conditions de sol qui semblent propices à une meilleure croissance des arbres et qui ont le potentiel de restaurer la productivité des forêts boréales dans les forêts à lichen à canopée ouverte.

Mots-clés: Mousses hypnacées; Lichen; Restauration; Capacité de récupération; Pin gris; Forêts boréales

1.2 Abstract

Background

Low productivity open lichen (*Cladonia* spp.) woodlands have been rapidly expanding in the closed-crown feather moss (*Pleurozium schreberi* (Brid.) Mitt.) boreal forest of eastern Canada. While open-woodland areas are progressing, there is little information on the recoverability of open lichen woodlands back to closed-canopy forests.

Methods

An experimental set-up using moss transplantation was installed on a poor jack pine (*Pinus banksiana* Lamb.) stand with lichen ground cover in 2011. Treatments included: 1) lichen cover removed, 2) lichen cover removed and transplantation of a feather moss cover, 3) lichen control, and 4) a natural jack pine site with feather moss cover (moss control). We extracted tree stem increment cores and collected needles and soil samples for nutrient analysis.

Results

The transplanted-moss treatment can counteract the adverse effects of lichen on jack pine growth. This treatment enhanced foliar nutrition and soil nutrients, especially ammonium ($N\text{-}NH_4^+$) and nitrate ($N\text{-}NO_3^-$). With this treatment, the soil conditions (e.g., soil nutrients, soil moisture) and foliar nutrition were closer to that of moss control. Surprisingly, lichen removal treatment did not improve growth and resulted in poorer jack pine growth and harsher soil conditions.

Conclusion

Feather moss can establish, survive, and remain healthy in an environment previously occupied by lichen. The replacement of lichen by feather moss establishes soil conditions that appear conducive to better tree growth and have the potential of restoring the productivity of boreal forests in open-canopy lichen woodlands.

Keywords: Feather moss; Lichen; Restoration; Recoverability; Jack pine; Boreal forests

1.3 Introduction

With global change, the rate of natural disturbances is predicted to increase over large portions of the boreal region (Boulanger and Pascual Puigdevall 2021). Open lichen (*Cladonia* spp.) woodlands, through an increased frequency of fires and through compound disturbances (insect outbreaks and fires), have been rapidly expanding in closed-canopy boreal forests of eastern Canada (Girard et al. 2008; Pacé et al. 2020a). Open lichen woodlands were described as an alternative stable state for closed-canopy feather moss (*Pleurozium schreberi* (Brid.) Mitt.) forests (Payette et al. 2000; Jasinski and Payette 2005). In eastern Canada, the structure of lichen woodlands is simple with two main strata: sparse trees (mainly black spruce and/or jack pine) several meters apart (generally 10–40% cover) and large expanses of fruticose lichens of the genus *Cladonia* (Payette et al. 2000). Feather moss tends to survive in shaded conditions of closed-canopy forests, while lichen tends to dominate in well-drained and high light conditions (Bonan and Shugart 1989; Sedia and Ehrenfeld 2003; Haughian and Burton 2015). Furthermore, lichen and feather moss, as two types of common ground cover in boreal forests, could affect forest growth and regeneration by influencing the physical and biochemical condition of the soil (Sedia and Ehrenfeld 2006; DeLuca et al. 2013; Mallik and Kayes 2018; Pacé et al. 2020b). Therefore, a better understanding of the factors that contribute to the transition between the two alternative states is needed to better promote sustainable management of boreal forests.

Feather moss is a common ground cover in boreal forests. It affects forest ecosystem processes by controlling soil moisture and temperature (Zackrisson et al. 1997; Gornall et al. 2011; Mallik and Kayes 2018), regulating soil nutrient availability (Wheeler et al. 2011;

Bastianelli et al. 2017; Ouimet et al. 2018), and influencing the activity of soil microbial communities (Sedia and Ehrenfeld 2003) and incidentally the accumulation and mineralization of organic matter (Sedia and Ehrenfeld 2005). Ground cover composition can therefore exert an important control over soil carbon and nitrogen cycles (Turetsky 2003; Smith et al. 2017). Furthermore, DeLuca et al. (2022) showed that N_2 fixated in feather moss mats is retained in moss tissue for extended periods and then slowly transferred to the Organic (O) layer of the forest soil as the moss tissue decomposes. These observations suggest that feather mosses are a source of nitrogen for forest ecosystems and likely contribute to the nitrogen supply of boreal forest ecosystems (DeLuca et al. 2002; Haughian and Burton 2015). Lichens also have different roles in forest ecosystems. For example, lichens can increase seedling biomass accumulation, increase needle nitrogen uptake, serve as fodder for reindeer and caribou (Stark et al. 2007; Kytöviita and Stark 2009), and are a source of energy for soil microorganisms (Stark and Hyvärinen 2003). In contrast, other previous studies have shown that lichen appears to be detrimental to the growth of trees (Hawkes and Menges 2003; Pacé et al. 2019). Lichen could reduce the availability of soil nutrients (Wheeler et al. 2011; Pacé et al. 2016; Bastianelli et al. 2017), inhibit microbial communities (Sedia and Ehrenfeld 2003), and maintain lower soil moisture (Mallik and Kayes 2018) and allelopathy (Pacé et al. 2020b). These studies give solid scientific evidence for an important role of lichen in maintaining open woodland conditions. However, to our knowledge, there is no research on the potential for forest mosses to invade lichen woodlands and to potentially break the resilience of stable open-canopy woodlands that could lead to more productive forests. Moreover, processes of natural succession, from lichen to mosses, as well as the success of man-made transplantation of forest mosses have not been evaluated.

Our interest in investigating the potential conversion of lichen woodlands to closed-canopy moss forests was linked to the observation of an increase in the area covered by open-canopy lichen forests at the northern limit of the commercial boreal forest over the past decades (Girard et al. 2008). Additionally, ecosystem services provided by boreal forests, including biodiversity conservation and timber supply, could be negatively impacted by the expansion of lichen woodlands. The main goal of this research was to test the impact of changing the ground cover to enhance tree growth and improve soil conditions in slow-growing open-canopy lichen woodlands. The objectives of this study were (i) to determine the 10-year effects of ground cover manipulation (lichen, lichen removal, lichen removal with transplantation of feather moss) on the growth and foliar nutrient status of mature jack pine in boreal forests, (ii) to observe the response of soil properties to the manipulation of ground cover, and (iii) to gain information on the potential for a transition of lichen woodlands to more productive closed-canopy forests through the manipulation of ground cover. Based on previous studies, we hypothesized that feather moss transplantation would improve soil properties (soil nutrients and soil moisture) in open lichen woodlands. We also hypothesized that feather moss transplantation would result in better tree growth and foliar nutrition than lichen removal, and lichen, in that order.

1.4 Method

1.4.1 Study area

The study area location (49° 19’ 59” N; 79° 11’ 51” W) is in the spruce-feather moss bioclimatic domain of western Quebec, Canada (Saucier et al. 2011). The mean annual temperature and precipitation in the study area are 0 ± 2.9 °C and 909 mm, respectively (Pacé et al. 2020a). Our experiment was implemented in a 40-year-old jack pine (*Pinus banksiana* Lamb.) stand of 15 ha planted in approximately 1980 (MFFP. 2022). The stand lies over sandy to coarse-grained fluvioglacial and glaciolacustrine deposits (MFFP. 2022). Ground cover is mainly composed of terricolous lichens including *Cladonia stellaris* (Opiz) Pouzar & Veda, *C. rangiferina* (L.) F.H. Wigg. and *C. mitis* Sandst (Pacé et al. 2020a). A sawfly outbreak was present near the study area during 2012–2014 (MFFP 2012). In June 2021, we selected a nearby naturally productive jack pine site, which had a continuous feather moss cover composed of *Pleurozium schrberi.* All soils are Humo-ferric Podzols or Dystric Brunisoils (Soil Classification Working Group 1998).

1.4.2 Experimental design and field sampling

The site that was selected in September 2011 for this study had homogeneous stand and site conditions. Thirty focal trees were selected at the center of plots. The selected trees were all the same age, and the spacing of the trees was regular with trees at least 15 m apart from each other. Each focal tree represented the center of a 160 m^2 circular plot (experimental units). We randomly applied treatments to each tree (10 replicated focal trees times 3 treatments). Three ground layer treatments (Fig. 1.1) were randomly and equally assigned to these plots: 1) complete lichen cover removal by hand (n = 10 plots), 2) complete lichen cover removal and feather moss (*Pleurozium schreberi*) transplantation (n = 10 plots), and 3) lichen control (no treatment, n = 10 plots). We obtained the moss transplants from a mature productive forest located less than 2 km away. Large sections of moss were cut out and transported in a trailer. Each experimental unit received several sections of intact mosses that were placed side by side with no spacing for infill. In addition, in June 2021, we selected 10 plots in a nearby productive jack pine site, which had a continuous feather moss cover composed mostly of *Pleurozium schrberi*, to serve as a control benchmark for this forest type and to compare with the transplanted moss treatment. For control moss sites, we considered a nearby closed-canopy jack pine stand with feather moss ground cover, flat topography, and sandy to coarse-grained fluvioglacial and glaciolacustrine deposits. We selected 10 plots (center trees), and like the lichen site, a distance of at least 15 m between selected trees was ensured. In total, there were 40 focal trees, 40 plots and 4 treatments: lichen removal (No Lichen); transplanted moss (Moss Transplanted); control-lichen (No Treatment), and control-productive-moss (Moss Control). In all treatments, the understory was sparse and composed of only a few common species, including *Epigaea repens* (L.), *Vaccinium angustifolium* (Ait.), and *Kalmia* *angustifolia* (L.). We observed in the field that *Pleurozium* survived in all plots and expansion was not obvious. Additionally, composition of similar stands and vegetation were reported for the same region (Boudreault et al. 2002).

Figure 1.1 Appearance of forest and ground cover for the different treatments in 2013 and 2021.

In 2021, tree stem increment cores were collected at breast height (1.3 m above ground). Needles were collected from each focal tree. All cores were prepared following standard dendrochronological procedures (Stokes 1996), and then scanned at 1,200 dots per inch resolution to measure ring-width series using the program CooRecorder version 9.6 (Larsson 2020). We clipped branches from the crown of each focal tree to collect current and older year needles. Needle samples were oven-dried at 60°C for 24 h and then ground for chemical analysis.

We extracted soil samples from each plot as follows. Within each plot, three locations were randomly sampled and pooled per layer. Both the forest floor (complete O layer; the depth of this layer varied from 2 to 8 cm) and the top 20 cm of the mineral soil were sampled at each of these locations. Moist samples were air-dried and sieved using 6-mm (forest floor) or 2-mm (mineral soil) meshes. Considering the low productivity of the sites, *in situ* available N was expected to be extremely low. Thus, we incubated the soil samples in the field prior to extraction to generate higher concentrations and obtain more reliable values. Specifically, as a relative index of N availability, we measured soluble N following an 8-week period using *in situ* buried bags for the forest floor and mineral soil separately (Hart et al. 1994; Kranabetter et al. 2021). We retrieved forest floors without decayed wood and placed them into polyethylene bags. Mineral soils were extracted to 20 cm and gently poured back into a polyethylene bag lining the sample hole. Forest floor samples were placed on top of the mineral soil bags and covered with lichen, moss or leaf litter. After 8 weeks, the bags were retrieved, and each sample ran through a 6-mm (forest floor) and 2-mm (mineral soil) sieve. One subsample was taken for moisture content, while a second subsample was kept frozen until extracted for NH_4^+-N, NO_3^--N, total dissolved nitrogen (TDN), and dissolved organic nitrogen (DON) determinations (Kranabetter et al. 2021). Soil temperature was recorded each hour from June to September 2021 using temperature data loggers (Spectrum®1000 Series) that were buried at a depth of 10 cm and at an approximate distance of 30 cm from focal trees. In each plot, soil moisture was measured with a portable TDR probe (Spectrum® TDR300) six times during the growing season (June-August) at two-week intervals.

1.4.3 Chemical analyses

Needle total carbon and nitrogen concentrations were measured by dry combustion using a Leco TruMac (Leco Corp., St-Joseph, MI, USA). Major and minor nutrients (P, K, Ca, Mg, Mn, Cu, Zn, Al, Fe, Mn, B, Sr, Na) were analyzed by inductively coupled plasma (ICP) using an optical emission spectrometer (Optima 7300 DV, PerkinElmer, Waltham, MA, USA) after ashing at 500°C for 2 hours and recovery in 1M HCl following Kalra (1997).

Soil pH was measured both in $CaCl_2$ and demineralized water solutions with a glass electrode and a pH meter (Orion 2 Star) (Carter and Gregorich 2007). Phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), manganese (Mn), aluminum (Al), iron (Fe), sodium (Na) and strontium (Sr) were extracted with a Mehlich III extraction solution (Carter and Gregorich 2007) and analyzed by inductively coupled plasma (ICP) using an optical emission spectrometer (Optima 7300 DV, PerkinElmer, Waltham, MA, USA). The effective cation exchange capacity (CEC) was computed as the sum of exchangeable base cations (K, Ca, Mg, Mn, Al, Fe, Na). Total carbon (TC) and total nitrogen (TN) concentrations were measured by dry combustion using a Leco TruMac CNS analyzer (Leco Corp., St-Joseph, MI, USA). Soil available N (NH_4^+, NO_3^- and TDN) was first extracted from incubated samples with a 1.0 M KCl solution (Carter and Gregorich 2007) and then analyzed by the FIA on a Lachat QuikChem® 8500 Series 2. Dissolved organic nitrogen (DON) was obtained by subtracting mineral nitrogen from total dissolved nitrogen after persulfate oxidation (Cabrera and Beare 1993).

1.4.4 Statistical analyses

The resulting ring-width series were statistically crossdated using the programs CDendro version 9.6 (Larsson 2020) and COFECHA (Holmes 1983). For each series, we estimated the distance to pith (Duncan 1989) by calculating basal area increments (BAI) for one tree using the R package “dplr” (Bunn 2008). We visually compared mean BAI values for series in each treatment and the control as follows. We focused our comparison of mean BAI on the nine years before (2002-2010) and nine years after (2012-2020) the treatment year, i.e., 2011. In one plot of the lichen cover with the transplantation of feather moss treatment, the branch of a focal tree was partially broken. We did not include the series for this focal tree in the analysis of mean BAI.

Differences in foliar nutrients and soil nutrients between ground cover treatments were analyzed by one-way ANOVA with Turkey post-hoc tests ($\alpha = 0.05$). Because TN, K, Ca, Mg and Mn concentrations in mineral soil were extremely low, the statistical results, even if significant, were not practically meaningful. Therefore, we did not include such nutrient information. In our data, extremely low values are those near or equal to the detection limit I (Mengel and Kirkby 2001). Control moss was not randomized as the other treatments; however, the stands were nearby and covered a similar area, thus we still considered this treatment. Data were transformed to meet the assumptions of normality and homogeneity of variance as necessary. All statistical analyses of ANOVA were performed in SPSS 26.0 (SPSS Inc., Chicago, USA). Principal component analysis (PCA) was conducted to visualize the soil properties and foliar nutrients of the four forest-ground treatments using the R software, version 4.2.2 (R Development Core Team, 2022).

1.5 Results

Ten-years after transplantation, the moss had survived. However, we did not observe in the field an expansion of the planted moss outside of the area where it was transplanted, nor a colonization of lichen on the transplanted moss.

1.5.1 Effects of ground cover on soil properties

Ten years after ground cover treatments, there were clear differences in soil properties between ground cover treatments (Table 1.1; Fig. 1.2; Fig. 1.3). Soil nutrients were mainly concentrated in the forest floor, and forest floor nutrients responded more strongly to ground cover treatments than the mineral soil (Table 1.1). Overall, soil properties of the transplanted-moss treatment were significantly different from those of the control lichen and lichen removal, but more similar to those of the control moss (Table 1.1; Fig. 1.3). On the forest floor, the transplanted-moss treatment and control moss had significantly higher TC and TN, followed by control lichen, while the lowest concentrations were found in the lichen removal treatment. No such effect was found for the C/N ratio. Moreover, the transplanted moss treatment had significantly higher available P and exchangeable Sr, K, Ca, Mg, Mn, Al, Fe, and CEC than control lichen and lichen removal treatments, while values were more similar to those of control moss. Both transplanted-moss treatment and control moss showed significantly higher TDN, N-NH_4^+, N-NO_3^- and DON concentrations than control lichen, whereas the lichen removal treatment showed the lowest values. In the mineral soil, except for exchangeable Sr, Fe, DON and soil pH, the element concentrations did not differ significantly between ground cover treatments. Mineral soil pH ($CaCl_2$) of control moss was more acidic and lower by 0.4 units compared to the lichen treatments, while that of transplanted moss was intermediate and not statistically different from other treatments.

Compared to other treatments, the lichen removal treatment showed significantly higher soil temperature and lower moisture content (Fig. 1.2). The transplanted moss treatment did not bring significant changes to these properties compared to the lichen control (Fig.1.2). However, PCA indicated that both moss treatments showed conditions tending to be wetter and cooler than lichen and lichen removal treatments (Fig. 1.3).

Table 1.1 Effects of ground cover treatment on soil nutrients (forest floor and mineral soil). Significant differences between ground-cover treatments are represented by different letters. Control-L is control lichen; Lichen- is lichen removal; Moss+ is moss transplantation; Control-M is control moss. Significant P values are shown in bold.

		Control-L (*n* = 10)	**Lichen–** (*n* = 10)	**Moss+** (*n* = 10)	**Control-M** (*n* = 10)	*df*	***P*-value**
Forest floor							
TC	%	5.79(0.85)b	1.35(0.12)c	13.07(1.30)a	13.82(1.32)a	4	**<0.001**
TN	%	0.11(0.02)b	0.03(0.00)c	0.30(0.03)a	0.27(0.02)a	4	**<0.001**
C/N		51.72(0.66)	56.11(6.56)	43.80(1.05)	52.14(1.61)	4	0.099
pH	H_2O	3.74(0.09)b	4.15(0.09)a	3.91(0.04)ab	3.78(0.04)b	4	**0.001**
pH	$CaCl_2$	3.00(0.09)b	3.59(0.10)a	3.22(0.05)b	3.07(0.04)b	4	**<0.001**
P	mg kg^{-1}	16.54(2.05)b	12.68(2.10)b	39.27(3.15)a	29.66(2.83)a	4	**<0.001**
Sr	mg kg^{-1}	1.61(0.40)b	0.68(0.09)b	4.17(0.33)a	3.82(0.34)a	4	**<0.001**
K	*cmol*(+) kg^{-1}	0.22(0.03)b	0.08(0.01)b	0.56(0.05)a	0.56(0.06)a	4	**<0.001**
Ca	*cmol*(+) kg^{-1}	0.48(0.11)c	0.17(0.03)c	4.45(0.41)a	2.36(0.36)b	4	**<0.001**
Mg	*cmol*(+) kg^{-1}	0.16(0.03)b	0.06(0.01)b	0.80(0.09)a	0.95(0.09)a	4	**<0.001**
Mn	*cmol*(+) kg^{-1}	0.03(0.01)b	0.01(0.00)b	0.26(0.04)a	0.05(0.01)b	4	**<0.001**
Al	*cmol*(+) kg^{-1}	9.35(0.53)b	12.09(2.10)ab	14.12(0.84)a	10.17(0.84)ab	4	**0.045**
Fe	*cmol*(+) kg^{-1}	1.88(0.12)ab	1.53(0.12)b	2.07(0.10)a	1.95(0.08)a	4	**0.008**
Na	*cmol*(+) kg^{-1}	0.02(0.00)bc	0.01(0.00)c	0.05(0.00)b	0.15(0.02)a	4	**<0.001**
CEC	*cmol*(+) kg^{-1}	12.14(0.65)b	13.94(2.03)b	22.30(1.18)a	16.18(1.20)b	4	**<0.001**
TDN	mg kg^{-1}	13.99(1.54)b	7.12(1.00)b	45.95(6.69)a	36.22(5.42)a	4	**<0.001**
N-NH_4^+	mg kg^{-1}	4.31(1.07)b	2.16(0.82)b	25.14(4.54)a	21.04(4.06)a	4	**<0.001**
DON	mg kg^{-1}	9.79(1.26)bc	5.16(0.38)c	20.79(2.62)a	15.17(2.30)ab	4	**<0.001**
N-NO_3^-	mg kg^{-1}	0.25(0.01)b	0.25(0.00)b	0.29(0.01)a	0.27(0.01)ab	4	**0.005**
Mineral layer							

Table 1.1 continued

TC	%	0.46(0.03)	0.35(0.04)	0.51(0.06)	0.43(0.05)	4	0.118
pH	H_2O	5.1(0.01)a	5.1(0.02)a	5.0(0.02)a	4.9(0.05)b	4	**0.006**
pH	$CaCl_2$	5.0(0.02)a	5.0(0.05)a	4.8(0.05)ab	4.6(0.1)b	4	**0.003**
P	mg kg^{-1}	14.60(1.51)	16.35(2.54)	16.80(2.07)	15.26(2.38)	4	0.844
Sr	mg kg^{-1}	0.10(0.00)b	0.11(0.01)b	0.19(0.01)a	0.17(0.02)a	4	**<0.001**
Al	*cmol*(+) kg^{-1}	28.46(0.33)	28.12(0.34)	28.50(0.48)	26.92(0.82)	4	0.140
Fe	*cmol*(+) kg^{-1}	0.30(0.02)b	0.27(0.03)b	0.39(0.02)b	0.68(0.09)a	4	**<0.001**
CEC	*cmol*(+) kg^{-1}	28.81(0.34)	28.44(0.33)	28.98(0.49)	27.69(0.79)	4	0.317
TDN	mg kg^{-1}	1.32(0.14)	1.23(0.51)	1.71(0.30)	3.48(1.13)	4	0.063
N-NO_3^-	mg kg^{-1}	0.17(0.01)	0.21(0.03)	0.17(0.00)	0.20(0.01)	4	0.178
DON	mg kg^{-1}	1.14(0.23)ab	0.50(0.21)b	1.57(0.25)ab	2.45(0.80)a	4	**0.031**
N-NH_4^+	mg kg^{-1}	0.15(0.05)	0.72(0.40)	0.39(0.12)	1.29(0.48)	4	0.086

Note: Significant differences between ground-cover treatments are represented by different letters. Standard error values are in parentheses.

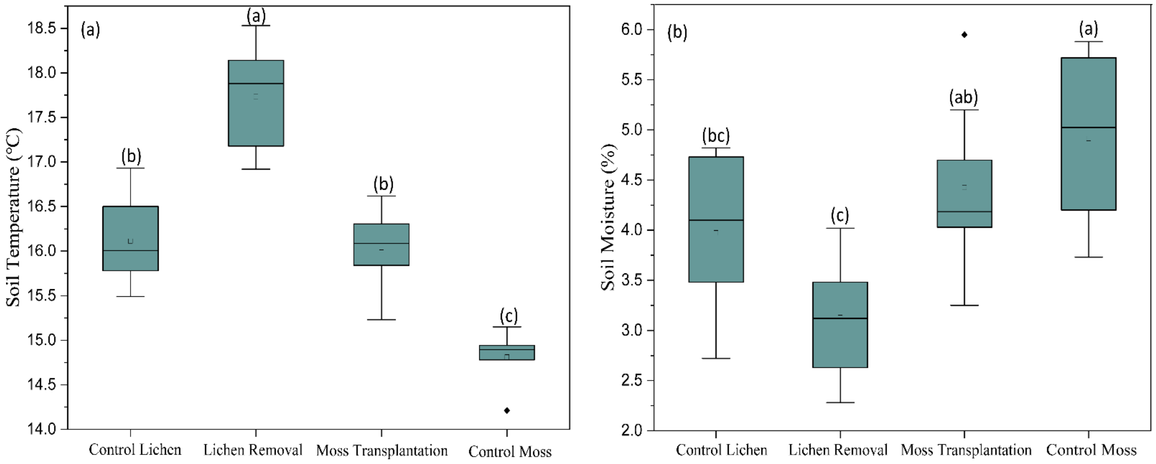

Figure 1.2 The response of soil temperature (a) and soil moisture (b) to ground cover treatments. Sampling took place between June and September 2021. The soil temperature and moisture data for each treatment is an average of the period. Significant differences between ground-cover treatments are represented by different letters.

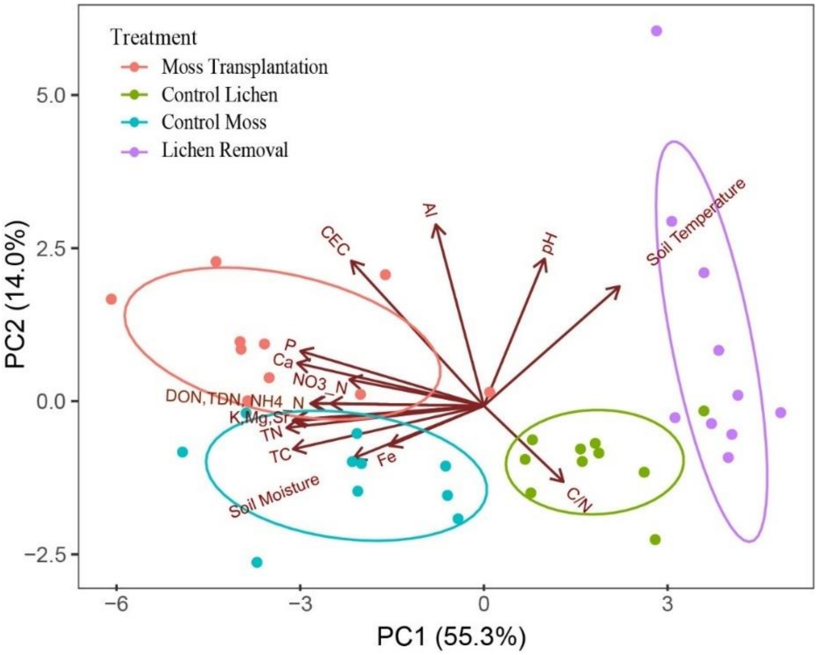

Figure 1.3 Principal component analysis (PCA) biplot of individuals (i.e., treatment plots, n = 40) and explanatory variables (i.e., forest floor soil properties, n = 18). The biplot shows PCA scores of explanatory variables as vectors (dark-red arrows) and individuals of each forest-ground treatment (circles), of the first (x-axis) and second (y-axis) principal components (PCs). Individuals on the same side as a given explanatory variable should be interpreted as having a high contribution on it.

1.5.2 Foliar nutrients

The foliar nutrient composition of jack pine trees showed significant differences between ground cover treatments (Table 1.2). The current-year foliage and the one-year-old foliage showed similar variability in composition between ground cover treatments (Table 1.2; Fig. 1.4; Fig. S1.1). For current-year as well as one-year-old foliage, there were no significant differences between ground cover treatments in foliar N, P, C/N and C/P concentrations. However, the transplanted-moss treatment had significantly higher foliar Ca, Mg, Mn, Zn and Na concentrations than control lichen and lichen removal treatments. The transplanted-moss treatment showed foliar concentration values that were comparable to those of the control moss treatment, particularly for Ca, Mg, Zn and Na (Table 1.2; Fig. 1.4). These values were significantly higher than those of the control lichen and lichen removal treatments ($P<0.001$). The foliar K concentration in the control moss treatment was significantly higher than with other treatments ($P<0.05$), whereas foliar nutrient concentrations of lichen removal were similar to values found in control lichen, with low foliar nutrient concentrations.

Table 1.2 Effects of ground cover treatment on jack pine foliar nutrients (current year and old years). Significant differences between ground-cover treatments are represented by different letters. Control-L is control lichen; Lichen- is lichen removal; Moss+ is moss transplantation; Control-M is control moss. The significant P values are shown in bold.

		Control-L (*n* = 10)	**Lichen-** (*n* = 10)	**Moss+** (*n* = 10)	**Control-M** (*n* = 10)	***df***	***P*-value**
Foliage (old years)							
N	%	1.00(0.03)	1.01(0.02)	0.98(0.03)	0.95(0.02)	4	0.432
P	g kg^{-1}	0.88(0.02)	0.92(0.04)	0.90(0.02)	0.85(0.02)	4	0.349
C/N		54(1.19)	54(1.03)	55(1.82)	56(1.23)	4	0.726
N/P		11.35(0.88)	11.11(0.98)	10.92(1.12)	11.18(0.74)	4	0.781
K	g kg^{-1}	2.50(0.17)ab	2.33(0.15)b	2.34(0.10)b	2.91(0.11)a	4	**0.014**
Ca	g kg^{-1}	2.44(0.20)b	2.47(0.16)b	4.62(0.27)a	4.44(0.34)a	4	**<0.001**
Mg	g kg^{-1}	0.37(0.03)c	0.44(0.04)c	0.62(0.03)b	0.80(0.07)a	4	**<0.001**
Mn	g kg^{-1}	0.25(0.02)b	0.37(0.04)b	0.52(0.05)a	0.29(0.02)b	4	**<0.001**
Zn	mg kg^{-1}	25.57(1.99)c	43.25(4.11)b	68.82(4.33)a	70.85(5.18)a	4	**<0.001**
Al	g kg^{-1}	0.38(0.03)ab	0.49(0.03)a	0.33(0.03)b	0.35(0.03)b	4	**0.003**
Fe	mg kg^{-1}	67.62(5.85)a	65.41(5.02)ab	74.46(5.88)a	47.58(3.18)b	4	**0.005**
B	mg kg^{-1}	9.52(0.46)	9.29(0.57)	8.32(0.84)	8.96(0.83)	4	0.642
Sr	mg kg^{-1}	5.77(0.65)b	8.98(1.28)a	7.42(0.54)ab	5.46(0.56)b	4	**0.015**
Na	mg kg^{-1}	6.37(1.14)b	11.13(1.51)b	13.07(1.36)ab	21.90(4.05)a	4	**<0.001**
Foliage (current)							
N	%	0.92(0.02)	1.00(0.04)	0.95(0.03)	1.00(0.02)	4	0.189
P	g kg^{-1}	0.94(0.02)	0.96(0.04)	0.99(0.02)	0.96(0.02)	4	0.534
C/N		57.08(1.35)	53.08(1.80)	54.77(1.56)	52.24(1.23)	4	0.127
N/P		9.85(0.58)ab	10.40(0.51)a	9.61(0.65)b	10.36(0.61)a	4	**0.011**

Table 1.2 continued

K	g kg^{-1}	2.64(0.19)b	2.57(0.16)b	2.80(0.06)b	3.63(0.16)a	4	**<0.001**
Ca	g kg^{-1}	1.37(0.11)c	1.47(0.11)bc	2.34(0.16)a	1.95(0.12)ab	4	**<0.001**
Mg	g kg^{-1}	0.49(0.03)c	0.59(0.03)bc	0.68(0.03)ab	0.75(0.03)a	4	**<0.001**
Mn	g kg^{-1}	0.17(0.02)b	0.25(0.03)ab	0.31(0.03)a	0.17(0.01)b	4	**<0.001**
Zn	mg kg^{-1}	27.80(1.16)c	34.96(2.24)c	42.93(2.01)b	52.02(2.03)a	4	**<0.001**
Al	g kg^{-1}	0.24(0.02)b	0.32(0.02)a	0.21(0.02)b	0.25(0.01)ab	4	**0.003**
Fe	mg kg^{-1}	33.32(2.40)a	27.29(0.67)ab	29.81(2.26)ab	25.31(1.57)b	4	**0.026**
B	mg kg^{-1}	9.85(0.48)	9.49(0.45)	8.82(0.77)	9.82(0.96)	4	0.705
Sr	mg kg^{-1}	3.16(0.37)ab	4.75(0.65)a	3.46(0.31)ab	2.55(0.25)b	4	**0.007**
Na	mg kg^{-1}	12.95(1.92)b	12.77(1.58)b	17.70(3.29)b	28.50(3.40)a	4	**<0.001**

Note: Standard error values are in parentheses.

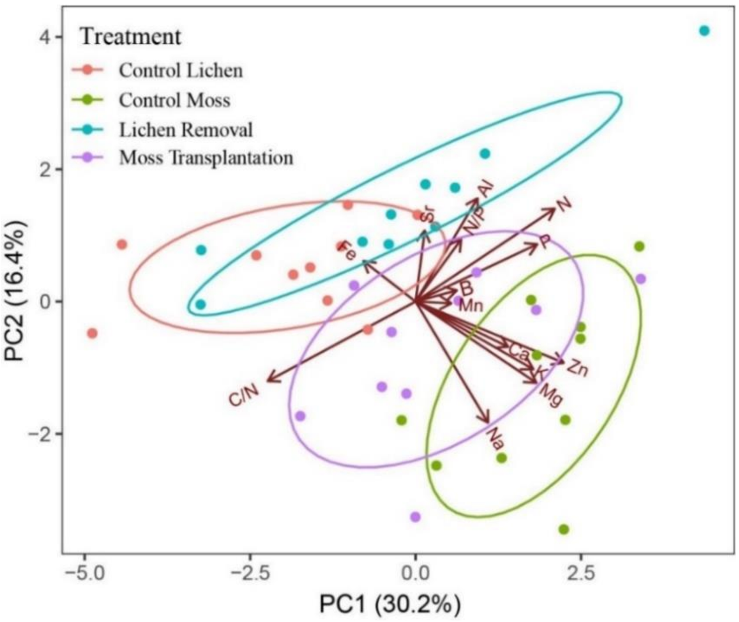

Figure 1.4 Principal component analysis (PCA) biplot of individuals (i.e., treatment plots, n = 40) and explanatory variables (i.e., current foliar nutrients, n = 14). The biplot shows the PCA scores of the explanatory variables as vectors (dark-red arrows) and individuals of each forest-ground treatment (circles), of the first (x-axis) and second (y-axis) principal components (PCs). Individuals on the same side as a given explanatory variable should be interpreted as having a high contribution on it. For the PCA analysis of the nutrition of older-year foliar nutrients, see Supplementary Material Figure S1.1.

1.5.3 Tree growth

We measured radial growth of jack pine over 2002-2020. Overall, there was a similar trend in jack pine growth (basal area increment, BAI) between ground cover treatments until 2011, the treatment year. After 2011, jack pine growth showed distinct trends (Fig. 1.5). The growth of jack pine with a lichen ground cover (control lichen) declined over time but revealed sharper decline between 2012 and 2014. The lichen removal treatment showed sharp decline after the disturbance in 2011 and then remained with a low growth rate. In comparison, the transplanted-moss treatment initially declined after disturbance, and then recovered from 2014 and maintained higher growth until the end of the observation period. Overall, all three treatments showed a similar declining trend in the first three years after 2011 (the treatment year).

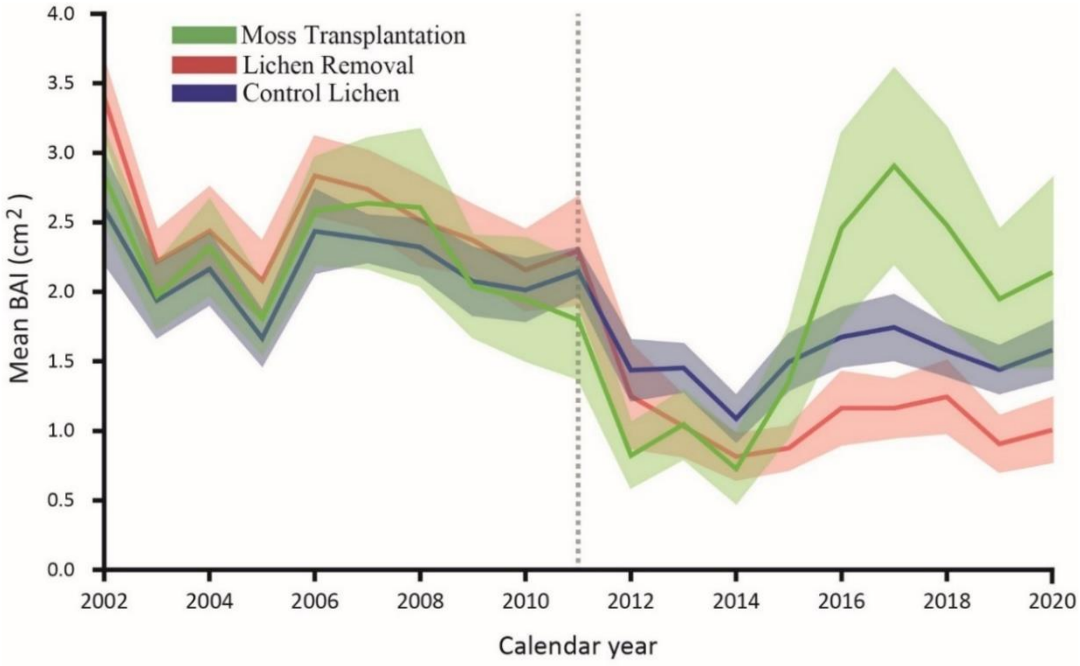

Figure 1.5 Jack pine mean basal area increment (BAI) during 2002-2020 (Control Lichen/Lichen Removal, n = 10; Moss Transplantation, n = 9). The horizontal dotted line is the beginning of the treatments. The legend is as follows: Green line, transplanted moss; Blue line: no treatment, control lichen; Red line: lichen removal. Shaded areas represent the standard error of the mean (for a visualization including the control moss group, see Supplementary Material Fig. S1.2).

1.6 Discussion

To our knowledge, this study represents the first experimental study reporting on the effects of transplanting moss in an open-canopy lichen woodland. The treatments had effects on the soil, tree growth, and foliar nutrition.

1.6.1 Ground cover and soil conditions

The transplanted moss treatment generated soil conditions that were similar to those of moss control and enhanced soil nutrients with respect to lichen control. These effects were mostly observed on the forest floor only. Exchangeable cations, extractable P, and all N availability indices (total N, total dissolved N, nitrate, ammonium, and DON) were significantly higher in the forest floor of the transplanted moss treatment compared to the lichen control. Previous research has shown that feather moss mats have a high potential for nitrogen fixation thanks to associated cyanobacteria (DeLuca et al. 2002; DeLuca et al. 2008; Bay et al. 2013; Rousk et al. 2013; Jean et al. 2021; Renaudin et al. 2022). We also observed that the transplanted-moss soil had a slightly higher nitrogen concentration than the control moss in the forest floor, even though this difference was not statistically significant. A possible explanation is that the transplanted-moss treatment was in open lichen woodlands where sunlight is more available and conditions are warmer than forests with greater canopy closure (Gundale et al. 2012). However, direct measurement of N fixation activity would be needed to evaluate the source of available soil N and distinguish N provided by imports in the transplanted moss versus the amount that was fixed since the onset of the transplantation if this were the case.

Our results showed that the soil temperatures of the transplanted-moss treatment were significantly higher than that of control moss, and control moss and transplanted-moss were associated with higher soil moisture than the other treatments. Some studies have shown that soil moisture availability is an important factor regulating soil mineral weathering rates (Gordon 2005; Egli et al. 2006; Brady et al. 2008). Similarly, the leaching of organic acid can favor mineral weathering. While we did not assess the flux of dissolved organic carbon, it is presumably higher in moss treatments that contain much more organic carbon. Pacé et al. (2019) showed that feather mosses host a greater diversity of ectomycorrhizal fungi than lichens. In summary, more favorable physical, chemical, and biological conditions of the moss layer may explain our results; namely, a higher availability of base cations, and some trace elements, in the soil and foliage under moss control and transplanted moss treatment.

Soil nutrient concentrations were somewhat lower in both control lichen and lichen removal, and soil nutrients were slightly lower overall in the lichen removal than in the control lichen. A similar finding by Sedia and Ehrenfeld (2005, 2006) indicates that lichen creates low nutrient microhabitats, possibly due to the slower decomposition of litter under lichen than under moss. Additionally, Pacé et al. (2020b) indicated a potential allelopathic effect of local lichens on jack pine seedlings.

1.6.2 Foliar nutrients and soil properties

The effect of ground cover on jack pine was reflected in foliar nutrient concentrations, and the variation in foliar nutrient concentrations between ground covers were similar in older and current year needles. In Canadian boreal forests, nitrogen and phosphorus are the most common limiting nutrients (Paquin et al. 1998; Maynard et al. 2014). Our study showed no significant differences in foliar N, P, C/N and N/P concentrations among the ground covers, except for foliar N/P in the current year. The foliar C/N ratio ranged from 52.24 (±1.23) to 57.08 (±1.35) and the foliar N/P ratio ranged from 9.61 (±0.65) to 11.35 (±0.88). Our N/P ratio results were similar to Vallicrosa et al. (2022), who reported a value of 12.35 (*SD* = 1.73) in boreal forests. However, we found that transplanted moss treatment significantly increased foliar Ca, Mg, Mn, Zn, Na in comparison to lichen control and yielded foliar concentrations that were similar to control moss. Foliar concentrations were generally lower and much more similar between control lichen and lichen removal.

Results from the soil and foliage were not fully coherent. While all cations as well as available P and N in the forest floor were higher in the moss transplant treatment than under other treatments, only the cations showed a positive foliar response. This was surprising because jack pine stands in particular have shown an almost ubiquitous positive response to N and to P fertilization in Canadian boreal forests (Maynard et al. 2014). The absence of a significant foliar N and P difference may be due to dilution (Imo and Timmer 1998), namely more N and P were taken up by the trees that produce more abundant foliage without modifying their foliar nutrient concentrations. Our result showed that the specific needle weight (dry mass current year needle par 100 needles) was not different between treatments (Supplementary Material, Table S1.1), indicating that if such an effect occurred following the onset of the treatments, it is not present today and perhaps the trees are producing a more extended canopy with stable N and P concentrations. The enhanced cation concentration in the foliage could indicate that trees are less water limited. Mass flow is the main process by which plants take up Ca and Mg (McGonigle and Grant 2015), so that a greater uptake may reflect a greater water flow through tree stems. These latter results were coherent with the greater soil water content observed in treatments with a moss cover. The drier conditions in the lichen removal treatment may have contributed to a lower tree nutrient uptake (Houle et al. 2016).

1.6.3 Treatment effect on tree growth

Compared to control lichen and lichen removal, the transplanted-moss treatment had a different effect on jack pine growth after 2011 than before this date. In the first three years after treatment installation, growth of jack pine decreased in all treatments. After this period, growth recovered and declined again until 2014. After 2014, the growth of trees in the transplanted moss treatment increased and recovered to the pre-experimental period, while that of the lichen and the lichen removal treatments remained lower than that of the transplanted moss treatment. The delay in a positive response in the transplanted moss treatment is probably due to the direct effects of the disturbance, potentially including root damage induced by the treatment, as well as to the slow acclimatation of the root system. Another potential explanation is that the supply of N fixed by cyanobacteria living in feather moss mats is preserved in the moss tissue for a long time before being transferred to the forest floor during the decomposition of the moss tissue (DeLuca et al. 2022). A potential explanation for the decline in growth for all treatments between 2011 and 2014 is that all trees may have been influenced by a combination of harsher climate (such as drought) and jack pine sawfly damage (*Neodiprion swainei* Middleton). A sawfly outbreak was present near the study area during this period, but we lack confirmation of an occurrence in our plots (MFFP 2012).

Enhanced tree growth in the transplanted moss treatment for the latest portion of the experiment (post 2011, and more specifically post 2014) was consistent with findings by Wheeler et al. (2011) and Pacé et al. (2020b) who showed that feather moss facilitates the establishment and growth of tree seedlings. Several factors can be responsible for enhanced tree growth in the transplanted moss treatment. A greater soil water availability was measured in the transplanted moss treatment. This is not surprising because the moss layer has a strong capacity to retain water and this may greatly change the amount of soil available water following a rainfall event (Ilek et al. 2015). In the coarse sandy soils of our study area, water availability may be critical even if the climate is not considered arid. Another factor that may influence growth is nutrient availability. Jack pine is responsive to nitrogen and phosphorus fertilization (Newton and Amponsah 2006; Maynard et al. 2014). Here, we found enhanced N and P availability in the soil but not in foliar nutrient concentrations. As discussed previously, a potential explanation is a dilution effect with homeostatic nutrient concentration in the foliage but a greater overall foliage mass, although this would need to be validated. Lastly, another potential cause for enhanced growth with mosses is a chemical inhibition of the plants or mycorrhizae from lichen. Mallik and Kayes (2018) showed that lichen seedbeds inhibit black spruce seedling regeneration, potentially through the presence of usnic acid, a common germination inhibiting allelochemical.

Our study also showed that lichen removal not only does not improve growth but appears to result in poorer jack pine growth. This is somewhat surprising because removal of lichen probably would reduce a source of allelochemicals, such as the usnic acid. Pacé et al. (2016) show that lichen removal increases fine root biomass of pine trees, whereas Fauria et al. (2008) indicate that lichen removal by grazing can enhance adult pine growth. Our results may be due to reduced accumulation of organic matter on the ground following lichen removal and direct exposure of the mineral soil, resulting in lower soil moisture and greater diurnal temperature fluctuation (Hawkes and Menges 2003; Lavoie et al. 2006; Houle et al. 2016) (Also, see our results Fig. 1.2). In addition, understory vegetation plays an important role in soil nutrient availability, tree production, and soil-plant interrelationships (Landuyt et al. 2019; Zhou et al. 2022). Therefore, the removal of the understory may adversely affect soil nutrients, soil water content, and microbial activity (Zhang et al. 2022).

Our findings suggested that feather moss has a key role in promoting and maintaining mature jack pine growth. Moreover, our results also suggested that the role of feather mosses in water regulation and litter decomposition rates may be a mechanism to promote tree growth in poor lichen woodlands. To our knowledge, our study is the first to suggest that controlling the ground layer could initiate a transition from open-lichen woodland conditions to those of a more productive closed-canopy moss forest. Although we recognize that the lichen woodland is a unique habitat playing an important role for the preservation of biodiversity, maintaining a mosaic of ecosystems of various composition and productivity contributes to ecological function diversity and thus enhances resilience to disturbance and environmental changes (Thompson et al. 2009).

1.7 Conclusion

Feather mosses are an important component in boreal forests, contributing to boreal forest growth and improving soil properties. Mansuy et al. (2013) suggested that afforestation of open lichen woodlands in boreal forests can be a means of increasing forest productivity. However, without appropriate soil conditions, productive closed-canopy forest conditions may not be achieved. Furthermore, to our knowledge, there are no studies documenting the recoverability of lichen woodlands to closed-canopy forests in Canada. Our 10-year results indicated that it is possible to replace a lichen cover with a feather moss cover and that feather moss can establish, survive, and remain healthy in an environment previously occupied by lichen. The replacement of lichen by feather moss establishes soil conditions that appear conducive to better tree growth and has the potential of restoring the productivity of boreal forests in open-canopy lichen woodlands. The mechanisms involved are not fully elucidated and could be related to a greater availability of water and nutrients thanks to inputs from cyanobacteria associated with *Pleurozium* moss mats and to a greater level of weathering of soil minerals. However, the importance of such mechanisms needs to be evaluated. Also, our results of no moss expansion after 10 years suggested that the effect may be local, at least in the short term. Therefore, the feasibility of transplanting moss over large areas and the long-term survival of moss need to be evaluated. Removing lichens as an alternative to increase productivity does not seem to be a good approach, as lichen removal conversely had an adverse effect on tree growth in our experiments. Moreover, considering the sensitivity of feather moss to high light conditions, as well as the potential damage to the source forest caused by the transplantation process, transplantation may not be needed. Instead, productive forest conditions can be maintained by promoting low light conditions in the understory that are favorable to feather moss rather than to lichen. Dense plantations or restocking natural stands could be possible solutions. Finally, forest managers could give preference to sites already dominated by feather moss that may be beneficial for tree growth when reforestation occurs.

1.8 Acknowledgements

We would like to thank Danielle Charron of the Université du Québec in Montréal (UQAM) for the preparation and organization of field work. We thank Mathieu Lamarche for field work and lab work assistance. This research was supported by the MITACS Accelerate grant with Greenfirst, industry partner in La Sarre, QC, Canada.

2. RÉPONSE DES COMMUNAUTÉS MICROBIENNES DU SOL À LA MANIPULATION DU COUVERT FORESTIER DANS UNE FORÊT BORÉALE

RESPONSE OF THE SOIL MICROBIAL COMMUNITIES TO FOREST GROUND COVER MANIPULATION IN A BOREAL FOREST

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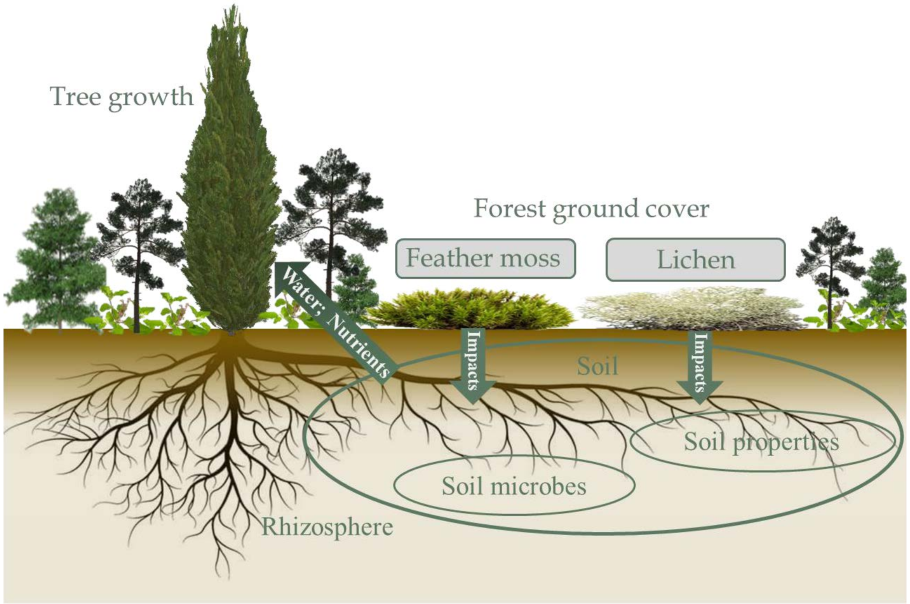
2.1 Résumé

Dans l'est du Canada, les forêts boréales subissent localement un changement entre deux états stables alternatifs, des forêts productives de mousses hypnacées (*Pleurozium schreberi* (Brid.) Mitt.) à couvert fermé et des boisés de lichens ouverts (*Cladonia* spp.) à faible productivité. Bien que ce changement ait des conséquences importantes sur la structure et la productivité de l'écosystème, on sait peu de choses sur les modifications de la diversité et de la composition de la communauté microbienne du sol qui peuvent être induites par ce processus. Nous avons évalué les effets de la transplantation de mousses pendant 10 ans sur les communautés microbiennes du sol dans une forêt de lichens ouverte. Les traitements comprenaient 1) l'élimination de la couverture de lichen, 2) l'élimination de la couverture de lichen suivie de la transplantation d'une couverture de mousse, 3) un contrôle avec la couverture de lichen maintenue en place (*i.e.* contrôle de lichen), et 4) un site de forêt naturelle avec une couverture de mousse (*i.e.* contrôle de mousse). Nous avons constaté que la modification du couvert forestier a un impact significatif sur la diversité, la composition et la fonction des communautés microbiennes du sol. La diversité alpha fongique était plus sensible aux changements dans la couverture de lichens et de mousses que la diversité bactérienne. La composition des communautés microbiennes du sol a montré des différences significatives entre toutes les couvertures forestières, mais avec de plus grandes similitudes entre les traitements de transplantation de mousse et de mousse de contrôle. Plus important encore, les changements de la couverture forestière ont affecté de manière significative la structure des communautés microbiennes et des groupes fonctionnels fongiques. La transplantation de mousse a augmenté de manière significative l'abondance relative du genre fongique *Piloderma*, responsable de l'élimination de l'azote organique. En outre, la transplantation de mousse a augmenté de manière significative l'abondance relative globale des champignons ectomycorhiziens et a diminué la proportion de champignons mycorhiziens éricoïdes. L'humidité et la température du sol sont les principales variables environnementales associées à la modification de la composition de la communauté microbienne. Notre étude montre que la transplantation de mousses dans les forêts de lichens à canopée ouverte contribue à réguler et à modifier la composition, la structure et la fonction des communautés microbiennes du sol, avec des implications potentielles pour expliquer les changements dans les processus de l'écosystème associés à ces deux types de forêts.

Mots-clés: Mousses hypnacées; Lichen; Diversité microbienne du sol; Composition microbienne; Fonction microbienne; Rhizosphère; Forêt boréale

2.2 Abstract

In eastern Canada, boreal forests are locally experiencing a shift between two alternative stable states, productive closed-canopy feather moss (*Pleurozium schreberi* (Brid.) Mitt.) forests to low-productivity open lichen (*Cladonia* spp.) woodlands. While this shift has important consequences for ecosystem structure and productivity, little is known about the changes occurring in the diversity and composition of the soil microbial community which may be driven by this process. We evaluated the effects of 10-year moss transplantation on soil microbial communities in an open-lichen woodland. Treatments included: 1) removal of the lichen cover, 2) removal of the lichen cover followed by transplantation of a feather moss cover, 3) a control with the lichen cover kept in place (lichen control), and 4) a natural forest site with a feather moss cover (moss control). We found that changing the forest ground cover has a significant impact on the diversity, composition and function of soil microbial communities. Fungal alpha diversity was more sensitive to changes in lichen and moss cover, compared to bacterial diversity. Soil microbial community composition showed significant differences among all forest ground covers, but with greater similarities between the moss transplantation and control moss treatments. More importantly, changes of forest ground cover significantly affected the structure of microbial communities and fungal functional groups. Moss transplantation significantly increased the relative abundance of the organic nitrogen-scavenging fungal genus, *Piloderma*. Furthermore, moss transplantation significantly increased the overall relative abundance of ectomycorrhizal fungi and decreased the proportion of ericoid mycorrhizal fungi. Soil moisture and temperature were the main environmental variables associated to the shift in microbial community composition. Our study points out that moss transplantation in open-canopy lichen woodlands contributes to regulate and modify the composition, structure, and function of the soil microbial communities with potential implications for explaining the changes in ecosystem processes associated with these two forest types.



Graphical abstract

Keywords: Feather moss; Lichen; Soil microbial diversity; Microbial composition; Microbial function; Rhizosphere; Boreal forest

2.3 Introduction

Boreal forests face challenges associated with climate change, such as increased natural disturbances and forest shifts (Boulanger and Puigdevall 2021; Berner and Goetz 2022). Notably, shifts from productive closed-canopy feather moss forests to low-productivity open lichen woodlands in boreal forests of eastern Canada have been reported (Girard et al. 2008; Pacé et al. 2020a), and may be more frequent due to increasing fire severity and frequency (Veraverbeke et al. 2017; Baltzer et al. 2021). Lichen (*Cladonia* spp.) and feather moss (*Pleurozium schreberi* (Brid.) Mitt.) are two types of common ground covers in boreal forests whose presence is linked to forest canopy openness (Pacé et al. 2020a). A feather moss ground cover is typically found under shaded conditions of closed-canopy forests, usually associated with fair quality soil conditions and a productive forest; while a lichen ground cover is typically found on well-drained nutrient poor soil supporting a sparse, low productivity forest (Haughian and Burton 2015; Pacé et al. 2019; Gao et al. 2023). Meanwhile, lichen have different roles in forest ecosystems, e.g., to promote vegetation growth and as fodder for reindeer and caribou (Stark et al. 2007; Kytöviita and Stark 2009). However, frequent or severe fire disturbances can limit forest regeneration leading to open canopy conditions with a lichen ground covers on sites previously supporting productive forests (Baltzer et al. 2021).

Constant feedback between aboveground-belowground communities shape species composition and related ecological function(Augusto et al. 2015). Soil microbial communities, including bacteria and fungi, play important roles in plant growth, nutrient availability, and soil health (Clemmensen et al. 2013; Philippot et al. 2013). The composition of soil bacterial and fungal communities is mainly driven by variations in forest composition in boreal forests, even more than changes in ground vegetation(Rodríguez-Rodríguez et al. 2023). Nevertheless, other research in northern ecosystems also indicate a close association between soil microbial communities and ground vegetation, with differences in diversity and composition of microbial communities depending on ground vegetation (Xiao et al. 2022; Ortiz-Rivero et al. 2023). Similarly, rhizosphere and bulk soil is also strongly altered in community structure and ecological functions by shifts in vegetation (Bahram et al. 2020; Thacker and Quideau 2021; Mundra et al. 2022). Understory vegetation dominated by lichens or mosses have different effects on soil microbial activity, which may be related to the microhabitats they create (Sedia and Ehrenfeld 2005; Merilä et al. 2010). Pacé et al. (2019) indicated that lichens were associated with reduced abundance and modified composition of the root ectomycorrhizal community. In boreal forests, soil microorganisms are particularly critical as important regulators of plant productivity, *i.e.*, obtaining limiting nutrients through plant symbioses (Van Der Heijden et al. 2008). For instance, mycorrhizal fungi and nitrogen-fixing bacteria are responsible for most of the nitrogen (N) and phosphorus (P) that plants acquire annually (Van Der Heijden et al. 2008; Tedersoo and Bahram 2019). Also, soil fungal communities drive soil function by regulating carbon storage (Clemmensen et al. 2013), and in particular ectomycorrhizal fungi are important contributors to soil organic matter turnover (Phillips et al. 2014). Therefore, belowground microbial communities have a pivotal role in boreal ecosystems that are crucial for nutrient cycling, and forest productivity.

An increase in the area covered by open-canopy lichen woodlands has been observed over the past few decades in the boreal forests of eastern Canada (Girard et al. 2008). This may affect boreal forest ecosystem services such as climate change mitigation, carbon sequestration, biodiversity conservation and timber supply. A 10-year manipulative study conducted in a slow growing open lichen woodland, which included lichen, lichen removal and moss transplantation treatments, revealed that the forest ground cover has a direct influence on soil properties and tree growth (Gao et al. 2023). This experimental design offers the opportunity to test for the effect of ground cover on soil microbial communities without the confounding effects that are often present in observational studies where site and soil conditions are often correlated with vegetation. More importantly, this experimental set-up gives us the opportunity to explore the role of the soil microbial communities in the restoration of ecosystem processes that follows moss encroachment in open lichen woodlands.

Using this experimental design, our main objective was to evaluate the effect of changes in forest ground cover on the diversity, community structure, and composition of soil bacterial and fungal communities 10 years after the treatment and to determine the relationships between microbial communities and soil properties. In addition, we compared these results with those from a productive closed-canopy forest with a moss ground cover used as a control. Previous results have shown that the ground cover plays an important role in maintaining alternate ecosystem stable states in boreal forests (Pacé et al. 2020a; Gao et al. 2023). However, the role played by the soil microbial communities in this dynamic is poorly known. Based on these analyses and previous studies, we hypothesized that: 1) Different forest ground covers lead to distinct soil microbial communities; 2) Differences in microbial communities are linked to differences in soil conditions (*e.g.*, nutrients, soil temperature and moisture) associated to different ground covers; 3) Forest ground covers leading to higher soil nutrient content and tree growth (*i.e.*, moss transplantation, control moss) harbor higher proportions of copiotrophic/beneficial taxa. We refer to beneficial taxa as those microorganisms that play a positive role in ecosystem functioning, plant health, soil function or other ecological processes.

2.4. Method

2.4.1 Site description and soil sampling

The experimental site is located in Western Quebec, Canada (49° 19’ 59” N; 79° 11’ 51” W), within the spruce-feather moss bioclimatic domain (Saucier et al. 2011). The average temperature and precipitation in the study area are 0 ± 2.9 °C and 909 mm, respectively (Pacé et al. 2020a). The age of the jack pine (*Pinus banksiana* Lamb.) stand in our site was about 40 years. The stand lies over sandy to coarse-grained fluvioglacial and glaciolacustrine deposits (MFFP. 2022). Ground cover is mainly composed of terricolous lichens including *Cladonia stellaris* (Opiz) Pouzar & Veda, *Cladonia rangiferina* (L.) F.H. Wigg. and *Cladonia mitis* Sandst (Pacé et al. 2020a). The study site and experimental design were the same as in Gao et al. (2023). Specifically, in September 2011, 30 circular plots with experimental units 160 m^2, were selected. Three ground layer treatments were randomly assigned to 10 plots of the 30 focal tree plots: 1) complete lichen cover removal by hand (n = 10 plots, lichen removal, RL), 2) complete lichen cover removal and feather moss (*P. schreberi*) transplantation (n = 10 plots, moss transplanted, MT) and 3) no treatment control (n = 10 plots, control lichen, CL). In addition, in June 2021, we selected a nearby naturally productive jack pine site with closed canopy and continuous feather moss cover, consisting mostly of *P. schreberi*, as a natural benchmark (n = 10 plots, control moss, CM) for comparison with artificially converted ground cover. In total, there were 40 plots and 4 treatments. All soils are Humo-ferric Podzols or Dystric Brunisoils (Soil Classification Working Group 1998). In all treatments, the understory was sparse and composed of a few common species, including *Epigaea repens* L., *Vaccinium angustifolium* Ait., and *Kalmia angustifolia* L. (Gao et al. 2023).

Soil samples were collected with a small shovel during the same week in June 2021. For bulk soil, both the forest floor (complete organic layer) and the top 20 cm of the mineral soil were sampled at each of plot and brought back to the laboratory. Within each plot, three points were randomly sampled and pooled to generate a representative composite sample that was thoroughly mixed and sieved through a 2 mm mesh. A subsample of each composite sample was transferred to a 100 mL sterile plastic tube and stored at -20°C prior to DNA extraction. Rhizosphere soil was collected by following lateral roots out from the trunk until the fine roots were reached, ensuring that samples belonged to the focal tree. Then roots were placed in a 50 mL sterile plastic tube and stored in a cooler with ice until samples were brought to the laboratory and stored at -20°C prior to further processing.

To recover the rhizosphere soil, roots were transferred into 50ml tubes with 25ml of a PBS

1X + Tween 20 (0.1%) solution, and the tubes were vortexed for 5 minutes at maximum speed. The roots were then removed from the tubes and the rhizosphere soil was recovered by centrifuging the tubes for 20 minutes at 4500g and 4°C. The supernatant was removed, and the rhizosphere soil was transferred to an absorbent paper to remove residual PBS prior to DNA extraction.

2.4.2 Soil abiotic factors

Soil abiotic factors mainly included Mehlich III extractable phosphorus (P), potassium (K), calcium (Ca), magnesium (Cornelissen et al.), manganese (Mn), aluminum (Al), iron (Fe), sodium (Na) and strontium (Sr), total carbon (TC), as well as total nitrogen (TN), soil pH (H_2O), effective cation exchange capacity (CEC), soil available N (NH_4^+, NO_3^- and TDN), dissolved organic nitrogen (DON), soil temperature and moisture in the forest floor (organic layer) and mineral soil. Here, given the very low concentrations of soluble N in boreal forest soils, we incubated the soils for 8 weeks *in situ* before determining soluble N, which we used as an indicator of plant available N. The experimental methods and instrumentation are detailed in a previous study (Gao et al. 2023).

2.4.3 DNA extraction and high-throughput sequencing

Soil DNA was extracted from 0.1 g of organic bulk soil, or 0.25 g of mineral bulk soil and rhizosphere soil, using the DNeasy Powersoil Pro kit with the QIAcube system following the manufacturer's instructions (QIAGEN, Valencia, CA, USA). For each batch of 23 samples, a negative control (*i.e.*, kit reagents only, no soil added) was processed and sequenced together with the samples. The concentration of all DNA extracts was measured with the Qubit™ dsDNA HS Assay Kit and the Qubit™ 3.0 fluorometer device (Thermo Fisher). Library preparation for Illumina sequencing was performed according to the manufacturer’s instructions for user-defined primers (Illumina 2013). Bacterial communities were amplified using primers 515F-Y and 926R targeting the V4–V5 regions of the 16S rRNA gene of bacteria and archaea (Parada et al. 2016); The ITS2 region of the fungal DNA was amplified using the primer set ITS9F and ITS4R (White et al. 1990; Menkis et al. 2012). The 16S rRNA and ITS amplicon pools were sequenced on an Illumina MiSeq instrument using MiSeq Reagent Kit v3 (2x300 bp) at the Next Generation Sequencing Platform of the CHU de Québec-Université Laval Research Centre.

2.4.4 Processing of 16S and ITS gene data

Bioinformatic analyses were conducted in the QIIME2 software (Bolyen et al. 2019) for 16S rRNA gene and ITS2 region. We used the DADA2 pipeline as established in QIIME2 (v2022.2.0) to process the Illumina-sequenced paired-end fastq files and to generate a table of amplicon sequence variants (ASVs), which are higher-resolution analogs of the traditional OTUs (Callahan et al. 2016). First, the primers were removed using Qiime2 implementation of CutAdapt (Martin 2011) for ITS2 and by position trimming in DADA2 for 16S. Resulting forward and reverse reads were then merged, low quality sequences, chimeras and rare ASV (frequency < 0.05% of mean ASV frequency) were removed. The taxonomy was assigned using the Silva 138 database (Quast et al. 2012) for bacteria and the Unite 8.0 database for fungi (Abarenkov et al. 2010; Nilsson et al. 2019). ASVs with non-target taxonomy (non fungi for ITS2, eukaryotes, chloroplast, and mitochondria for 16S) were filtered out with the Qiime2 taxa filter-table command. The filtered ASVs were then rarefied to 8427 (bacteria), 2849 (fungi) feature count and the resulting file was used to build the ASV table. After the processing, 14,547 and 2,911 ASVs remained for bacteria and fungi, respectively.

FUNGuild was used to analyze the functional groups of fungi in the soil (Nguyen et al. 2016). In order to avoid over-interpreting the fungal functional groups, the assigned functional categories were only considered if the confidence was highly probable and probable. The communities that could not be identified or identified as multiple complex nutrition modes were defined as "others".

2.4.5 Statistical analyses

Statistical analyses and their visualization were performed with the R statistical program (R-Core-Team, 2022) and the RStudio 4.2.2 interface. The phyloseq R package was used to analyze the taxonomically annotated ASV table (McMurdie and Holmes 2013). We analyzed differences in the bacterial and fungal diversity, composition, relative abundance, and fungal functional groups between forest cover treatment (CL, RL, MT and CM). The number of observed ASV (richness) as well as the Shannon and InvSimpson indices were calculated using the phyloseq package (estimate_richness function, McMurdie and Holmes 2013) computed from rarefied ASV relative abundance data. Differences in these alpha diversity indices among forest cover treatments were tested using a Generalized linear model (GLM) with Poisson distribution for the Observed ASV and Gamma distribution for the Shannon index and InvSimpson index. The Shannon diversity index is a commonly used formula to measure biodiversity. It is calculated as (Morris et al. 2014):

$$H = -\Sigma (pi * \ln(pi))$$

The InvSimpson Index is another biodiversity index used to measure the diversity of species within a community. It is calculated as (Morris et al. 2014):

$$InvSimpson = 1 / \Sigma (pi^2)$$

Where:

H represents the Shannon diversity index.

Σ signifies a sum over all species or categories.

pi is the proportion of individuals of a particular species (or category) relative to the total number of individuals in the community.

ln(pi) is the natural logarithm of pi.

Then significant *p*-values were calculated by ANOVA with Tukey's honestly significant difference test ($\alpha = 0.05$). The soil microbial Beta diversity patterns were presented for bacteria and fungi by non-metric multidimensional scaling (NMDS) ordinations based on Bray-Curtis dissimilarities computed from rarefied ASV relative abundance data using the vegan package in R. The ordination was presented two times, adding *a posteriori*, the microbial phyla and the abiotic factors with the envfit function (P <0.05, vegan package). Significant differences in Beta diversity between treatments were determined by permutational multivariate analysis of variance (PERMANOVA) on Bray-Curtis dissimilarities with 999 permutations using the adonis2 function in R (Oksanen et al. 2013). We also used post hoc pairwise PERMANOVAs (999 permutations, “pairwise.adonis” function, Hervé (2014)) with the multiple comparison correction method suggested by Benjamini and Hochberg (1995) to compare differences between ground cover treatments.

Microbiome relative abundances are compositional data that range between 0 and 1 and are also generally zero-inflated (Peng et al. 2016), therefore differential relative abundance analysis was conducted using the glmmTMB package (Brooks et al. 2017). It is a valuable tool for comparing relative abundance between groups due to its ability to fit zero-inflated mixed models (Brooks et al. 2017).

2.5 Results

2.5.1 Differences of soil microbial alpha diversity

Forest ground cover treatments affected soil alpha diversity of both bacterial and fungal communities (Fig.2.1). However, a significant effect on the alpha bacterial diversity was only found in the mineral soil ($p<0.05$). Specifically, control moss had the highest Observed diversity, while lichen removal treatment had the lowest. In contrast to soil bacteria, the alpha diversity of the soil fungal community responded more significantly to forest ground covers, with significant effects detected for most of the measured alpha diversity indices (Fig. 2.1, right panels). Interestingly, trends observed in the organic layer and the rhizosphere were similar, with an overall lower alpha diversity in the control moss treatment than in the other forest cover treatments. In contrast, in the mineral soil, the alpha diversity indices were generally higher for the control moss treatment than for the control lichen and lichen removal treatments, while the moss transplantation treatment led to intermediate values.

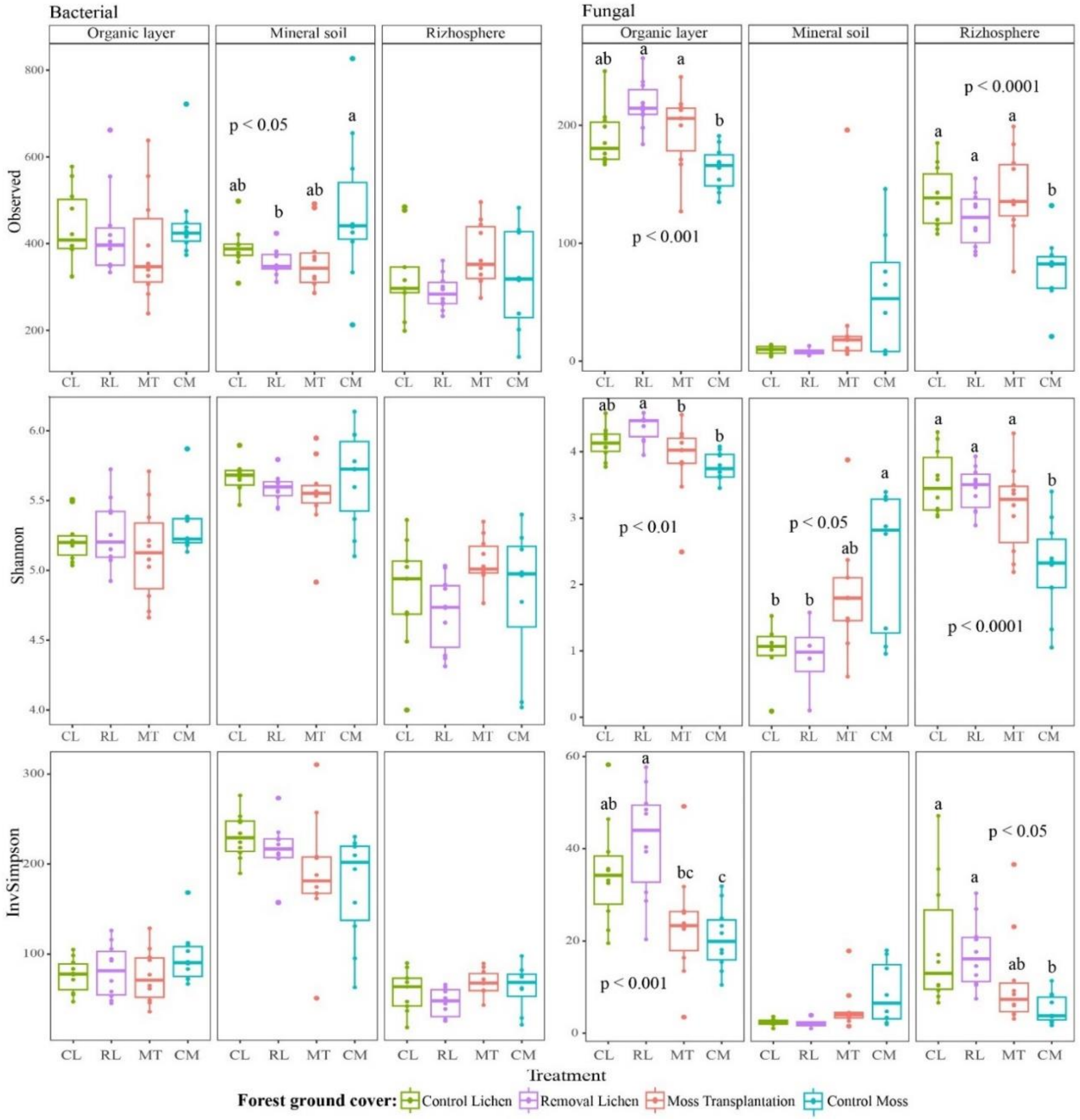


Figure 2.1 Bacterial and Fungal alpha diversity among forest ground cover treatments in forest soil. Forest ground cover treatments dominated by control lichen (CL), lichen removal (RL), moss transplantation (MT) and control moss (CM). Significant differences between ground-cover treatments (p-values) were evaluated by ANOVA using Tukey's honestly significant difference test (α = 0.05). The horizontal line in each box is the median value. Different lowercase letters indicate significantly different means across treatments. For more details on diversity, see Table S2.1.

2.5.2 Effects of forest ground covers on soil microbial community composition

The composition of the soil bacterial (Fig.2.2) and fungal (Fig.2.3) communities were significantly different among treatments in the organic layer, mineral soil and rhizosphere (PERMANOVA, Table S2.2). In most cases, the soil microbial communities under different treatments were clearly discriminated in the ordinations. Effect of treatments on bacterial community composition were stronger in the organic layer and the mineral soil than in the rhizosphere, with communities of the moss transplantation treatment closer to those of the control moss treatment, especially on Axis1. The lichen removal treatment, which created contrasting soil conditions, was also characterized by a distinct bacterial community in the organic layer. The soil bacterial composition was significantly correlated with soil temperature, soil moisture, NH_4^+-N, TDN and other nutrients, with higher soil nutrients, higher soil moisture, and lower soil temperature associated with the moss transplantation treatment and control moss, while higher soil temperature and lower soil nutrients were associated with the lichen removal treatment (Fig.2.2). Several bacterial phyla were significantly correlated to specific forest ground cover treatments. Of note, the *WPS_2*, *Chloroflexi*, *Firmicutes*, and *Actinobacteriota* phyla were related to the lichen removal treatment, while the *Proteobacteria* phylum was mainly related to moss transplantation treatment and control moss.

Fungal community composition also showed significant differences among treatments, especially in the organic layer and rhizosphere (Fig.2.3). Fungal communities of the organic layer showed a significant gradient along the first axis of the ordination, with, from left to right, samples exposed to the extreme conditions from the lichen removal treatment, followed by the control lichen treatment, and finally the moss transplantation treatment and control moss, which were more similar. Fungal communities of the moss control were also clearly distinct from that of the other treatments in the rhizosphere, but not in the mineral soil. Several environmental variables were significantly correlated to the fungal community composition in the organic layer, with communities of the lichen removal treatment associated with conditions of higher soil temperature, while moss transplantation treatment and control moss soils were related to higher soil nutrients and moisture. In addition, fungal phyla such as the *Basidiomycota* was associated with the control moss and moss transplantation treatments, while the *Ascomycota* was associated with the control lichen and lichen removal treatments.

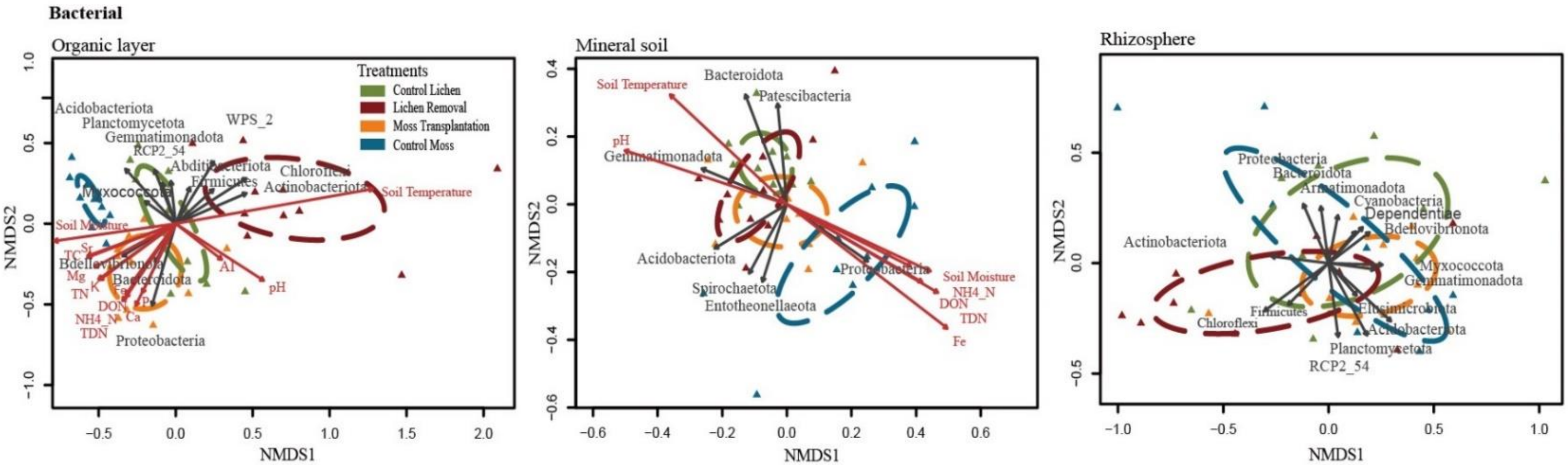


Figure 2.2 Two-dimensional non-metric multidimensional scaling (NMDS) of the soil bacterial community from the different forest cover treatments (stress value: organic layer 0.15, mineral soil 0.25, rhizosphere 0.14). Ellipses correspond to standard deviation of ordination scores for samples according to the Treatment. Black and red arrows indicate the correlation between sample level and ordination axes scores for bacterial phyla and environmental variables, respectively, added a posteriori in the ordination (displayed phyla of maximum estimated P < 0.05). Ordination based on Bray-Curtis dissimilarities of the rarefied ASV relative abundances.

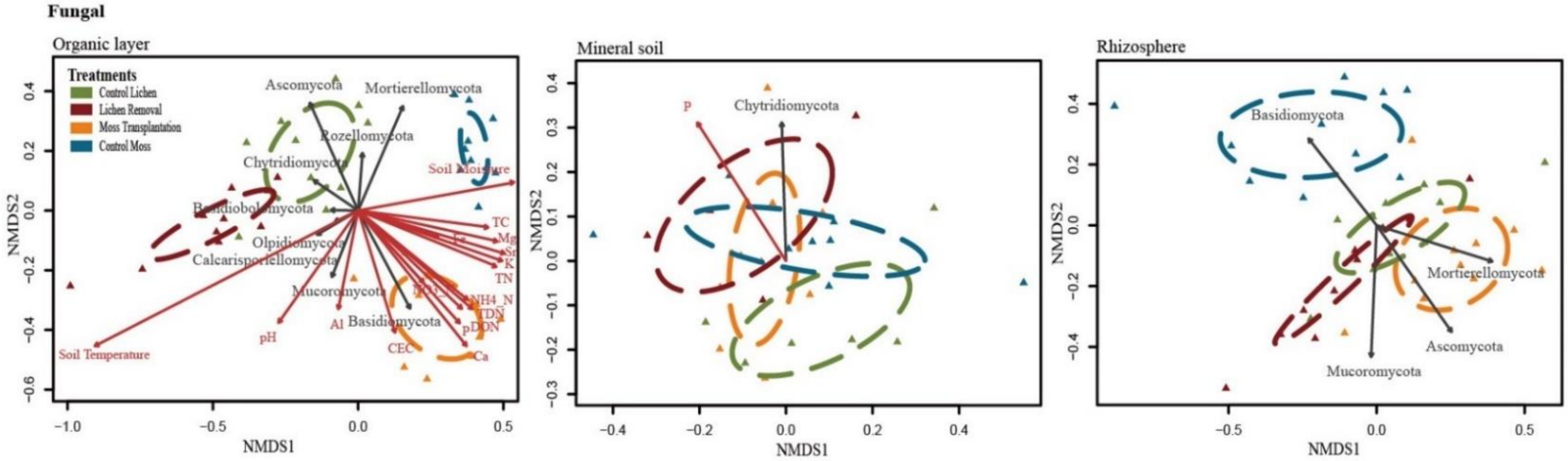


Figure 2.3 Two-dimensional non-metric multidimensional scaling (NMDS) of the soil fungal community from the different forest cover treatments (stress value: organic layer 0.14, mineral soil 0.17, rhizosphere 0.18). Ellipses correspond to standard deviation of ordination scores for samples according to the Treatment. Black and red arrows indicate the correlation between sample level and ordination axes scores for fungal phyla and environmental variables, respectively, added a posteriori in the ordination (displayed phyla of maximum estimated $P < 0.05$). Ordination based on Bray-Curtis dissimilarities of the rarefied ASV relative abundances.

2.5.3 Effects of forest ground covers on the relative abundance of the main soil microbial genera

Forest ground cover treatments significantly influenced soil bacterial (Fig.2.4) and fungal (Fig.2.5) genera abundances in the organic layer, mineral soil and rhizosphere. The soil bacterial genera were dominated by *Bradyrhizobium*, *Mycobacterium*, *Acidipila*, and ASVs assigned to the uncultured lineage *WD260* of the *Gammaproteobacteria* and *Subgroup_2* of the *Acidobacteria*. Significant differences in the abundance of all main genera were detected (Fig.2.4, Table S2.3). Specifically, lichen removal increased the relative abundances of *Mycobacterium*, *Tundrisphaera*, *Bryobacter*, *Conexibacter*, *WPS_2* and Candidatus_*Xiphinematobacter* in the organic layer, while the relative abundances of *Aquisphaera* and *Roseiarcus* were higher in moss transplantation and control moss. In the mineral soil, *Bradyrhizobium* and *RCP2_54* were significantly higher in control moss than in other treatments, and *Mucilaginibacter* and *AD3* were higher in control lichen. In the rhizosphere, the abundance of *Mycobacterium*, *Tundrisphaera* and *Candidatus_Xiphinematobacter* were higher, but *Roseiarcus* was lower in lichen removal than in other treatments.

For fungi, moss transplantation treatment and control moss had significantly higher relative abundances of *Piloderma* and *Cenococcum*, but lower abundances of *Pezoloma* and *Meliniomyces* compared to control lichen and lichen removal treatments in the organic layer (Fig.2.5). In the mineral soil, most dominant fungal taxa were mainly enriched in the moss transplantation treatment and control moss. In the rhizosphere, moss transplantation treatment had a significantly higher relative abundance of *Piloderma* than other treatments. Furthermore, both moss transplantation treatment and control moss had higher relative abundance of *Hydnellum* than control lichen and lichen removal treatments, while the relative abundance of *Pezoloma* was higher in lichen removal than in other treatments.

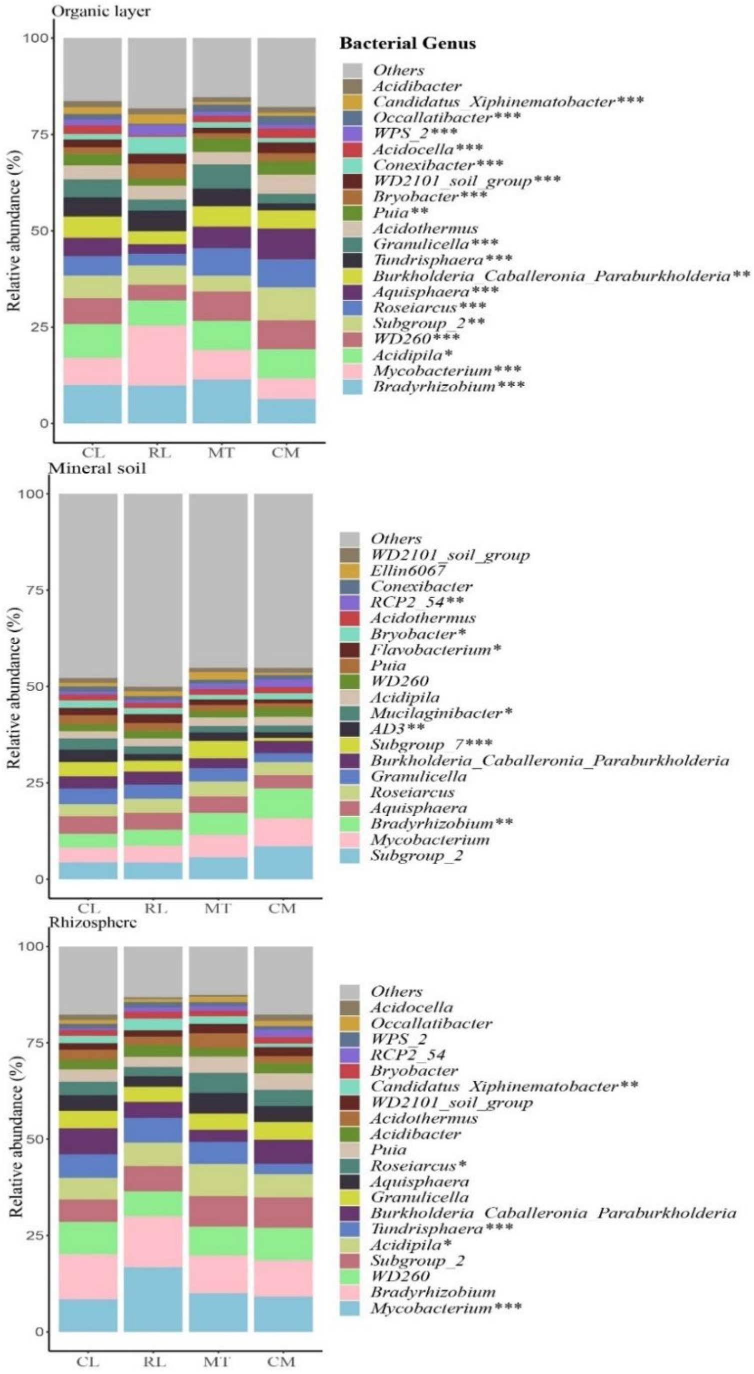
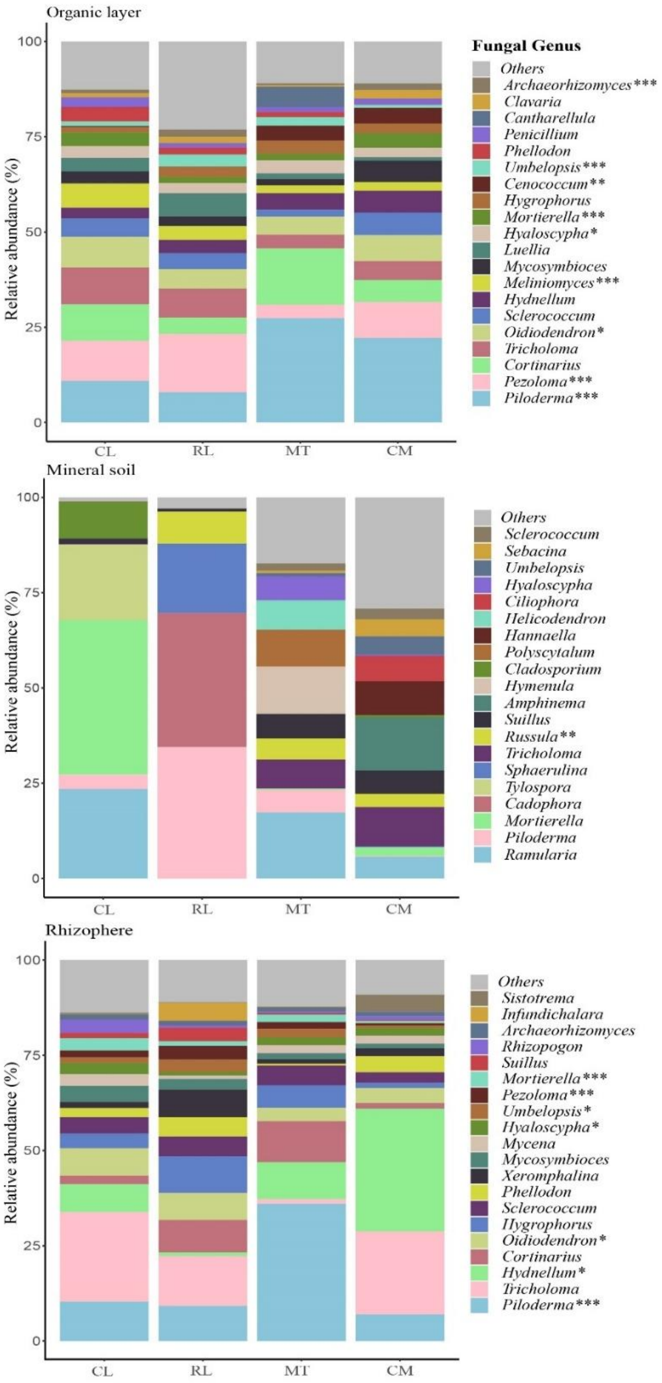


Figure 2.4 Differences in relative abundance (%) of ASVs assigned to Top 20 most relatively abundant bacterial genera for each soil layer among the forest cover treatments: control lichen (CL), lichen removal (RL), moss transplantation (MT) and control moss (CM). Stars indicate significant differences in the relative abundance of a genus among forest cover treatments ($p < 0.001$*, $p < 0.01$**, $p < 0.05$*). Results of multiple comparisons between treatments are shown in Table S2.3.**



Organic layer

Mineral soil

Rhizophere

Figure 2.5 Differences in relative abundance (%) of ASVs assigned to Top 20 most relatively abundant fungal genera for each soil layer among the forest cover treatments: control lichen (CL), lichen removal (RL), moss transplantation (MT) and control moss (CM). Stars indicate significant differences in the relative abundance of a genus among forest cover treatments ($p < 0.001$*, $p < 0.01$**, $p < 0.05$*). Results of multiple comparisons between treatments are shown in Table S2.3.**

2.5.4 Effects of forest ground cover on fungal functional groups

To explore whether forest ground cover treatments affected microbial functions, we used the FUNGuild tool to assign trophic modes and functional guilds to fungal ASVs based on their taxonomy. The relative abundances of the main trophic modes and functional guilds changed significantly with forest ground cover treatments (Fig.S2.3, Fig.2.6 and Table S2.4). For fungal trophic mode (Fig.S2.3), we found that moss transplantation treatment and control moss had higher relative abundances of symbiotroph than control lichen and lichen removal in both organic layer and rhizosphere. In the mineral soil, lichen removal had higher relative abundances of symbiotroph, but lower relative abundances of saprotroph. Concerning functional guilds (Fig.2.6), the relative abundance of ectomycorrhizal fungi was significantly higher in the moss transplantation treatment and control moss than in control lichen and lichen removal treatments in both organic layer and rhizosphere. Furthermore, we noted that ericoid mycorrhizal fungi had a lower relative abundance, and orchid mycorrhizal fungi had the highest relative abundance in moss transplantation treatment in the organic layer. In the mineral soil, lichen removal treatment had the highest relative abundance of endophyte among forest ground cover treatments. Undefined saprotroph had a higher relative abundance in treatments with a thick organic layer: control moss and moss transplantation. In the rhizosphere, moss transplantation treatment and control moss had lower relative abundance of ericoid mycorrhizal fungi compared to control lichen and lichen removal treatment.

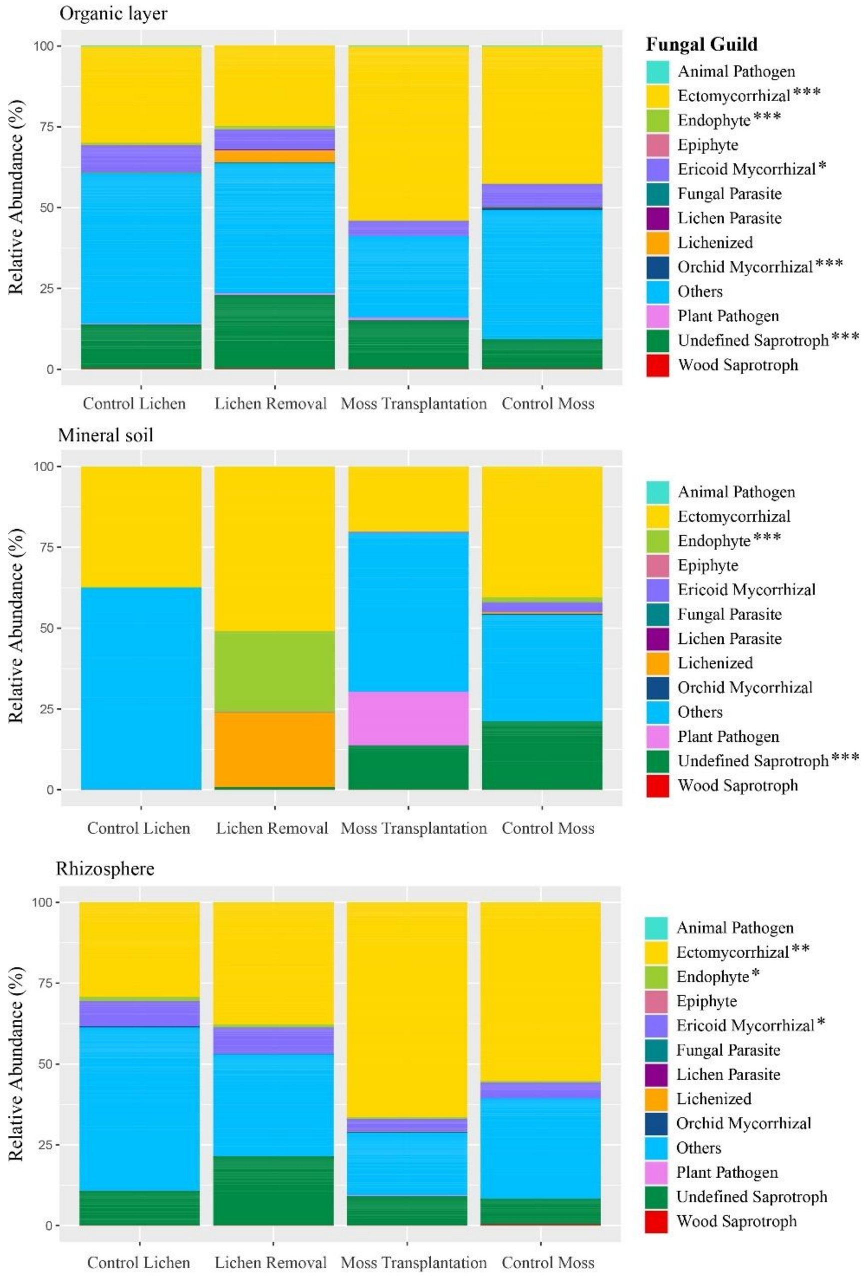


Figure 2.6 Compositions of main fungal functional guild inferred by FUNGuild. ASVs assigned to a guild with the confidence ranking of "Highly probable" and "Probable" were retained for further use, whereas those with the "possible" confidence ranking were classified as 'Others'. Stars indicate significant differences in the relative abundance of a functional guild among forest cover treatments ($p < 0.001$*, $p < 0.01$**, $p < 0.05$*).**

2.6 Discussion

Using soils collected from a 10-year moss transplantation trial set up in a boreal open-lichen woodland, this study clearly showed the important role of the forest ground cover in structuring soil microbial communities. Our results supported our three hypotheses: the soil microbial communities varied with treatment; distinct communities were related to soil conditions, and, finally, moss cover treatments harbored a larger proportion of copiotrophic and beneficial taxa. In the following sections we discuss the implication of the results at the community, taxonomic, and functional levels.

2.6.1 Effects of forest ground covers on soil microbial communities, and factors shaping these communities

In line with previous studies indicating that soil fungal communities are more impacted by vegetation than bacterial communities (Urbanová et al. 2015), the effects of the forest ground cover on fungal alpha diversity were much stronger than those observed for bacterial alpha diversity. Interestingly, the soil fungal diversity of the control moss treatment appeared to be lower than that of other forest ground cover treatments, for both the organic layer and the rhizosphere. Previous studies have shown that the soil fungal communities are associated with stand or tree age and that soil fungal diversity may decrease with increasing tree age (Wu et al. 2013; Kyaschenko et al. 2017; Sun et al. 2017). This is consistent with our results as the stand of control moss site was older (65-70 yr.) than that of the lichen site (c. 40 yr.). Of note, for some alpha diversity indices (*e.g.,* Shannon diversity in organic and mineral soils), the moss transplantation treatment led to alpha diversity values closer to those observed in control moss plots. Similarly, the fungal community composition of moss transplantation and control moss were more similar, but significantly different from those of the lichen removal and control lichen treatments, especially in the organic layer. It is well documented that the soil bacterial and fungal community are sensitive to changes of soil properties (Delgado-Baquerizo et al. 2017; Ma et al. 2022). In our previous study (Gao et al. 2023), we found that ground cover treatments affected soil nutrient availability (*e.g.*, phosphorus, potassium, calcium and available N) as well as soil temperature and moisture content. Moss transplantation treatment and control moss soils were cooler, moister, and nutrient richer than that of control lichen, while the lichen removal treatment had the poorest soil conditions. In addition, DNA concentrations of soils from the moss transplantation treatment were significantly higher than those of soils from the control lichen and lichen removal treatments, but close to those of soil from the control moss plots (Fig.S2.4), indicating that the soil conditions found in the moss treatments allowed to support a more abundant microbial community with a different composition than that of lichen covers.

Further analysis of the linkages between microbial communities and environmental factors identified soil moisture and temperature as potential drivers of soil microbial community composition in our study (Fig.2.2 and Fig.2.3). Our findings are consistent with other studies (Zogg et al. 1997; Fierer et al. 2003; Brockett et al. 2012). Soil warming was previously shown to lead to a reduction in microbial biomass and to changes in the composition of microbial communities (Frey et al. 2008). We also found that soil nutrients, including available N, TC, TN, Mg, K, and Ca, were significantly related with the soil microbial composition as observed by Liu et al. (2020) and Wang et al. (2020). Jiang et al. (2021) suggested that the microbial response to N is related to the N forms. We found that the soil microbial community composition was significantly correlated with soil DON, TDN and NH_4^+-N, and that N forms changed with the ground cover treatments. Soil pH is often found as a dominant factor affecting the composition of microbial communities (Lauber Christian et al. 2009; Zinger et al. 2011), but it was not the case in our study. A restricted pH range (organic layer: 3.74 - 4.15; mineral soil: 4.9 - 5.1) across treatments may explain why this factor did not come into play here.

2.6.2 Taxa-specific changes of soil microbial communities in forest ground covers

At the bacterial phylum level, *Proteobacteria* and *Acidobacteria* were found to be the most abundant phylum regardless of habitat type (Fig.S2.1). These two groups are very common and ubiquitous in soils (Kim et al. 2014; Jiang et al. 2021). *Proteobacteria* are known to be mainly copiotrophic, and prefer living in nutrient-sufficient environments enriched with nitrogen and carbon, while *Acidobacteria* (oligotrophic bacteria) prefer nutrient-limited environments (Fierer et al. 2007; Koyama et al. 2014; Dai et al. 2018) and can decompose complex C substrates derived from the recalcitrant soil organic matter (SOM) pool (Rasche et al. 2011). Of note, *Proteobacteria* include many N-fixing bacteria (Spain et al. 2009). Consistently, our results indicated that *Acidobacteria* was associated with control lichen, and *Proteobacteria* was related to moss transplantation treatment and control moss. Furthermore, our results showed that *Chloroflexi*, *Firmicutes*, *Actinobacteriota*, and the *WPS_2* candidate phylum were associated to the lichen removal treatment (Fig.2.2). It is noteworthy that *Actinobacteria* and *Firmicutes* contain many spore-forming and Gram-positive species which are capable of surviving under extreme conditions (Smith et al. 2008; Smith et al. 2013). These traits may confer advantages in systems where bare soils experience extreme conditions of moisture and temperature and low nutrient supply. Previous studies have shown that members of the *WPS_2* prefer organic-poor and bare soil habitats (Sheremet et al. 2020). *Chloroflexi* is prevalent in nutrient-poor environments (Gómez-Acata et al. 2016; Lee et al. 2020) and can survive in such environments by generating energy through solar radiation and 3-hydroxypropionate bi-cycle (Klatt et al. 2013; Liu et al. 2020). Thus, the increase in *Chloroflexi* relative abundance observed in the lichen removal treatment may be due to greater light availability and low available organic C sources compared to other treatments.

At the fungal phylum level, *Basidiomycota* were associated with the control moss and moss transplantation treatments, and *Ascomycota* were associated with control lichen and lichen removal treatments (Fig.2.3). Several studies have suggested that *Basidiomycota* is associated with high soil nutrients (*e.g.*, SOC and N content), whereas *Ascomycota* are known to thrive in barren soil (Zumsteg et al. 2012; Yan et al. 2020). Our results are consistent with these observations, in that there was a higher soil nutrient availability (*e.g.*, CEC, available N) in the moss transplantation and control moss treatments, and poorer soil conditions in the control lichen and lichen removal treatments (Gao et al. 2023).

Overall, at the phylum level, our results showed that soil habitats of moss transplantation and control moss are more suitable for copiotrophic microbes, while soils of control lichen and lichen removal treatments are preferred by oligotrophic microbes. Previous studies have shown that in environments where microorganisms are exposed to sustained environmental stress, particularly where the stress stems from low resource concentrations, oligotrophs are likely to outcompete copiotrophs (Fierer et al. 2007; Ho et al. 2017). Our results suggest that moss transplantation on open-lichen woodlands could change the nutrient status of the soil, and in turn the microbial community structure, with potential beneficial impacts on soil functions.

2.6.3 Functional changes in fungal communities between forest ground covers

In boreal forest ecosystems, soil fungal communities play a dominant role in driving soil functions (Clemmensen et al. 2013), including the decomposition of accumulated recalcitrant organic matter (Rousk et al. 2016) and the contribution to nutrient acquisition by trees (Cheeke et al. 2017). In our study, the relative abundance of different microbial trophic modes of fungi (saprotroph, symbiotroph and pathogen, Fig.S2.3) changed under the influence of the different forest ground covers. The moss transplantation and control moss treatments were mainly dominated by symbiotic fungi. It is well known that symbiotic fungi facilitate the uptake of water and essential mineral nutrients by host plants, enhancing their resilience and adaptability (Smith and Read 2010; Kolaříková et al. 2017). Remarkably, moss transplantation treatment was dominated by ectomycorrhizal fungi in the organic layer and the rhizosphere, similarly to control moss. This finding indicates that plant hosts of the moss transplantation treatment were able to develop abundant ectomycorrhizal fungi communities that are known to be highly efficient in nutrient acquisition, particularly in forest soils that are low in available nutrients (Lõhmus 2006). By extending their mycelial network throughout the soil, ectomycorrhizal fungi enable tree fine roots to tap into otherwise inaccessible sources of water and nutrients (Lindahl and Tunlid 2015; McCormack and Iversen 2019). This mycelial network also facilitates the transfer of nutrients from the soil to the fine roots, enhancing the efficiency of nutrient uptake, especially nitrogen (Cheeke et al. 2017). Moreover, ectomycorrhizal fungi have the ability to acquire nitrogen from the organic compounds present in SOM (Nicolás et al. 2019).

A classification system of ectomycorrhizal mycelial systems according to their patterns of differentiation and putative ecological importance was developed by Agerer (2001). It suggests two main ectomycorrhizal strategies for growth and nitrogen acquisition, one focusing on uptake of labile nitrogen forms such as amino acids, ammonium, and nitrate, and one focusing on insoluble, complex organic resources (Agerer 2001; Lilleskov et al. 2011). The second strategy might be enhanced by hydrophobic rhizomorphs to prevent leakage of solutes during medium- to long-distance transport. Such medium- to long-distance exploration types are unlikely to rely on labile substrates under conditions of low nutrient availability, as such substrates are too scarce (*e.g.*, free amino acids) to make the exploratory investment worthwhile (Lilleskov et al. 2011). This system provides a framework for discussing anatomical features that influence fungal exploration of the soil, and in our case, it facilitates examination of the consequences of forest ground cover manipulation for soil exploration. In our study, *Piloderma*, a taxa in the medium-distance fringe category of exploration types (Agerer 2001) which has the ability to use organic nitrogen and deliver it to trees (Heinonsalo et al. 2015), was responsive to forest ground covers, with higher relative abundance in the moss transplantation treatment compared to control lichen and lichen removal treatments. It's hydrophobic exploration subtype typically involves a dense proliferation of hyphae into loose, relatively undifferentiated rhizomorphs that ramify with high hyphal density around patches of organic matter, often in organic horizons (Lilleskov et al. 2011). This exploration type is well-adapted morphologically to explore for organic N under N-limited conditions (Lilleskov et al. 2011), and could help trees have access to N when mineral N forms are not available.

We also noted a higher relative abundance of ericoid mycorrhizal fungi in the control lichen and lichen removal treatments, especially in the rhizosphere. In boreal forests, ericoid mycorrhizal fungi usually form symbioses with ericoid plant roots, and compete with other boreal trees for soil resources (Fanin et al. 2022). Previous studies have identified that ericoid mycorrhizal fungi associated with ericaceous plants as able to take up N from organic matter and inhibit decomposition of soil organic matter, which would further increase nutrient limitation in boreal forests (Clemmensen et al. 2015; Adamczyk et al. 2016; Fanin et al. 2022; Ward et al. 2022).These results may better explain the poorer soil quality and weaker tree growth under the control lichen and lichen removal treatments than under moss transplantation (Gao et al. 2023). Surprisingly, lichenized fungi only represented a very small proportion of the sequences in the control lichen treatment, while the proportion was higher (although not significantly) in the lichen removal treatment. This was surprising as we were expecting higher proportions of lichen DNA in soils underlying the control lichen treatment. Further work, including the testing for potential methodological biases, will be needed to determine the reason for this discrepancy.

2.6.4 Implications for boreal forest dynamics

Global change is bringing increasing pressures on boreal biomes (Scheffer et al. 2012). Our findings suggested that forest ground covers drive changes in microbial communities, which could then alter soil functions. For example, the environment created by feather moss can increase the proportion of ectomycorrhizal fungi and decrease that of ericoid mycorrhizal fungi. Some studies have shown that mycorrhizal fungal communities are closely linked to stand productivity in boreal forests (Nagati et al. 2018; 2019). In the context of climate change, the expansion of the proportion of open-canopy lichen woodlands observed in Quebec, Canada (Jasinski and Payette 2005; Girard et al. 2008) may be amplified by plant-microbe-soil positive feedback loops which maintain low forest productivity and slow nutrient cycling in lichen-dominated stands, and the opposite conditions in feather moss forests (Gao et al. 2023). The presence of such positive feedback loops suggests that boreal forest management should pay attention to impacts of forestry practices on the ground cover layer.

The progression of lichen-woodlands in Quebec is thought to be linked to an increase in fire frequency and insect outbreaks (Girard et al. 2008; Côté et al. 2013; Veraverbeke et al. 2017; Baltzer et al. 2021). Studies in northern and Temperate Europe have indicated an opposite trend with a decrease in the lichen cover of forests and woodlands (Reinecke et al. 2014; Sandström et al. 2016; Tonteri et al. 2022). However, the drivers of such changes also appear to be related to the disturbance history of the sites, with a role for forest management and reduce forest fire frequency (Tonteri et al. 2022). Our results suggest, in agreement with Jasinski and Payette (2005) that once changed to a different alternative stable state, the biota, including plants and soil microbes, confer a resistance to further changes in composition and function.

2.7 Conclusions

Experimental modifications of forest ground covers had significant impact on the diversity, composition and function of soil microbial communities. Fungal diversity was found to be more sensitive to changes of forest ground cover, compared to bacterial diversity. More importantly, changes of forest ground cover significantly affected the structure of microbial communities and fungal functional groups. This study demonstrated the ground cover type plays an important role in shaping distinct soil microbial communities in the boreal forest and may help maintaining two contrasting stable alternate ecosystem states. It reveals the potential for management activities that favor a moss ground cover in boreal regions to regulate and modify the composition, structure, and function of microbial communities with potential benefits for soil-plant conditions.

2.8 Acknowledgements

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2.9 Data availability statement

The data presented in this study are deposited in the NCBI-SRA (Sequence Read Archive) repository, accession number PRJNA983772. The data is available in Genbanck (https://www.ncbi.nlm.nih.gov/bioproject/?term=PRJNA983772).

3. RELATIONS ENTRE LES PROPRIÉTÉS DU CHARBON DE BOIS ET L'ÉTAT DE L'ÉCOSYSTÈME DANS LA FORÊT BORÉALE

RELATIONSHIPS BETWEEN CHARCOAL PROPERTY AND ECOSYSTEM STATUS IN THE BOREAL FOREST

(Manuscript)

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3.1 Résumé

Les perturbations dues aux incendies augmentent dans le cadre du changement climatique mondial et des transformations écologiques des forêts se produisent. Plus précisément, dans les forêts boréales de l'est du Canada, on a observé le passage de forêts productives à couvert fermé de mousses hypnacées à des forêts peu productives à couvert ouvert de lichens (*Cladonia* spp.). On a émis l'hypothèse qu'une forte intensité des incendies serait à l'origine de ce changement, mais cette hypothèse est difficile à valider *a posteriori* sur des peuplements forestiers matures. Les propriétés du charbon de bois étant affectées par la sévérité des incendies, nous avons émis l'hypothèse que la quantité et les propriétés chimiques et physiques du charbon de bois (C, N, H, O, cendres, surface) seraient différentes et indicatives d'une plus grande sévérité des incendies dans les forêts à couvert ouvert par rapport aux forêts à couvert fermé. Notre hypothèse a été partiellement validée dans la mesure où la quantité de charbon de bois trouvée sur le sol des forêts à canopée fermée était supérieure à celle des forêts à canopée ouverte. Cependant, les propriétés chimiques et physiques n'étaient pas différentes, bien qu'il y ait une plus grande variabilité des propriétés du charbon de bois pour les peuplements à canopée ouverte. Ces résultats ne nous permettent pas de valider ou de rejeter complètement notre hypothèse sur le rôle de la sévérité du feu dans le passage entre les peuplements à canopée ouverte et fermée. Cependant, ils suggèrent que la variabilité des conditions d'incendie ainsi que la quantité de charbon de bois produite sont différentes entre les types d'écosystèmes. En outre, compte tenu du rôle que le biochar peut jouer dans l'amélioration des conditions du sol et la promotion de la restauration de la végétation, nos résultats suggèrent que le charbon de bois peut jouer un rôle dans le maintien de ces deux états alternatifs stables de l'écosystème.

Mots-clés: Charbon de bois; Mousse hypnacées; Lichen; Héritage du feu; Écosystème forestier; Forêt boréale

3.2 Abstract

Fire disturbances are increasing under global climate change and ecological transformations of forests are occurring. Specifically, shifts from productive closed-canopy feather moss forests to low-productivity open-canopy lichen (*Cladonia* spp.) woodlands have been observed in boreal forests of eastern Canada. It has been hypothesized that high severity of fires would be the cause of this change, but this is difficult to validate *a posteriori* on mature forest stands. Because charcoal properties are affected by fire severity, we have put forward the hypothesis that the amount and chemical and physical properties of charcoal (C, N, H, O, ash, surface area) would be different and indicative of a greater fire severity for open-canopy forests compared to closed canopy ones. Our hypothesis was partly validated in that the amount of charcoal found on the ground of closed-canopy forests was greater than that of open-canopy forests. However, the chemical and physical properties were not different, albeit a greater variability of charcoal properties for open canopy stands. These results do not allow us to fully validate or reject our hypothesis on the role of fire severity in the shift between open and closed canopy stands. However, they suggest that the variability in fire conditions as well as the amount of charcoal produced are different between ecosystem types. Furthermore, considering the role that biochar may play in improving soil conditions and promoting vegetation restoration, our results suggest that charcoal may play a role in maintaining these two stable alternative ecosystem states.

Keywords: Charcoal; Feather moss; Lichen; Fire legacy; Forest ecosystem; Boreal forest

3.3 Introduction

Increasing fire activity in the boreal forest of North America (Boulanger et al. 2014; Ellis et al. 2022; Davis et al. 2023), are making the forest more vulnerable to ecological transformations (Baltzer et al. 2021; Davis et al. 2023). Shifts from productive closed-canopy feather moss forests to low-productivity open lichen (Cladonia spp.) woodlands have been observed in boreal forests of eastern Canada (Girard et al. 2008; Pacé et al. 2020a). These two forest types (moss vs lichen) are considered as distinct alternative stable states (Payette et al. 2000; Jasinski and Payette 2005), with the moss cover favoring productive forests and the lichen cover usually occurring in low-productivity open-canopy woodlands (Pacé et al. 2020a; Gao et al. 2023). However, the causes for the transformation from a closed moss-canopy forests to a lichen-open woodlands are poorly known. A high fire intensity, consecutive fires leading to tree regeneration failure and/or lower tree growth as well as the combined effects of insects and fire are possible causes (Girard et al. 2008; Bergeron et al. 2010; Pacé et al. 2020a). Soil nutrient losses due to a high fire intensity (Bormann et al. 2008) could also induce low stand productivity even when stand stocking is adequate.

Fire disturbance can produce fire-related pyrogenic substances such as charcoal. Charcoal is then deposited locally on the forest floor and incorporated into the soil. Charcoal is a C-enriched, nitrogen (N)-depleted pyrogenic material with a highly aromatic molecular structure (Makoto and Koike 2021). Because of its aromatic structure, it is highly recalcitrant to decomposition, which explains its long-lasting presence in the soil for hundreds or even thousands of years (DeLuca and Aplet 2008). Ohlson et al. (2009) estimated that a total of approximately 1 Pg of carbon is contained in the soil charcoal of the world's boreal forests, an amount equivalent to about 15% of annual anthropogenic emissions from fossil fuel burning. Along similar lines, Jones et al. (2019) suggested that cumulative pyrogenic charcoal production is 60 Pg since 1750 which could be equivalent to as much as 40% of the global anthropogenic carbon lost through land use change in this period.

Charcoal is characterized by its highly porous structure and high adsorption capacity (Brimmer 2006; Preston and Schmidt 2006). The porous structure of charcoal increases its surface area, facilitating the adsorption of substances (Hart and Luckai 2013), that may offer several physical benefits to soil, such as enhanced soil-water holding capacity and reduced soil bulk density (Gundale and DeLuca 2006; Lehmann et al. 2006). Charcoal also acts as a reservoir for alkaline metals and phosphate, providing cation exchange sites (Liang et al. 2006). Studies on *terra preta* soils, have demonstrated a significant impact of charcoal on soil productivity and ecosystem diversity (Glaser et al. 2001). Additionally, the adsorption capacity of charcoal prevents the inhibition of plants or microorganisms by adsorbing potentially inhibitory organic compounds (DeLuca and Aplet 2008). Moreover, charcoal's hydrophobic properties are believed to play a role in soil humus formation (Piccolo et al. 2004). Overall, these properties of charcoal may have an essential role to play in the re-establishment and growth of post-fire vegetation after a fire (Zackrisson et al. 1996; Wardle et al. 1998; Robertson et al. 2012; Licht and Smith 2020).

While charcoal accumulation has been found to be highly variable across stands and plots likely due to heterogeneity of fire severity (Brimmer 2006; Preston et al. 2017) and stand biomass spatial variation, some studies have shown that charcoal amount may decrease with increasing fire severity (Pingree et al. 2012; Buma et al. 2014). Fire severity is also related to charcoal physical properties such as porosity (Shetty et al. 2021), and to chemical properties such as carbon content and pH (Lehmann et al. 2011). It was shown that the maximum temperatures reached during fire and the associated prolonged heating times were associated with greater C enrichment, increased thermal recalcitrance and degree of aromaticity of the charcoal (Soucémarianadin et al. 2015; Santín et al. 2016). Moreover, previous studies have shown that it seems possible to reconstruct fire regimes based on the sensitivity of charcoal properties (e.g., C, N, H, O content, surface area) to pyrolysis conditions (Wolf et al. 2013; Li et al. 2023). Generally, high severity fires would reduce the nitrogen content of charcoal due to volatilization of nitrogen at high temperatures (typically, >200°C) (DeLuca et al. 2009). Similarly, a greater depletion of O and H from organic bonds occurs, and the H and O content of charcoal decreases (Keiluweit et al. 2010). Such changes indicate that the degree of carbonization is accelerated with increasing fire severity (Chen et al. 2012). By contrast, the carbon content and specific surface area of charcoal are typically greater in high severity fires than in low severity fires (Mukherjee et al. 2011; Kim et al. 2012).

In this study, we examined the amount and quality of pieces of charcoal found in the soil of open and closed canopy forests to investigate what these indices can tell us about the nature of the fires that occurred in these two forest types, and how can the soil carbon legacy play a role in shaping these ecosystems. Our objective was to assess the differences in charcoal amount as well as properties (charcoal C, N, H, O, ash, surface area) between moss forests and lichen woodlands. Thus, here we hypothesize that 1) the dry forest conditions and the presumably high fire intensity in lichen woodland leads to lower amounts of charcoal in the soil of lichen woodlands compared to moss forests. 2) Charcoal properties such as N, H, O content will be higher in moss forests than in lichen woodlands, and ash, C content and surface area will be higher in lichen woodlands than in moss forests indicating the prevalence of higher severity fires in lichen woodlands.

3.4 Materials and methods

3.4.1 Study area

Our study area is located in the boreal forest of eastern Canada, dominated by the spruce-moss bioclimatic zone. Twenty-seven pure Jack pine (*Pinus banksiana* Lamb.) stands with lichen and feather moss ground cover were selected and sampled. See Pacé et al. (2020a) for more information, as several sites that the last major disturbance is harvesting, so we do not include these sites. We only selected that the last major disturbance is fire. Ten sites were located near the locality of La Sarre (48° 48′N; 79°12′W), whereas the other seventeen sites were located near the locality of Chibougamau (49° 53′N; 74° 20′W), yielding twelve open canopy lichen sites and fifteen closed-canopy moss sites. The last major disturbance in all sites was a forest wildfire. Average annual temperatures are 0 ± 2.9 °C and average annual precipitations are 909.1 mm in the sites near La Sarre, and average annual temperatures are 0.2 ± 3.7 °C and average annual precipitations are 995.8 mm in the sites near Chibougamau (Environment Canada 2018). All soils are humo-ferric and ferro-humic podzolic soils (Soil Classification Working Group 1998). The understory was composed of a few common species, including *Epigaea repens* (L.), *Vaccinium angustifolium* (Ait.), and *Kalmia angustifolia* (L.). Ground cover is mainly composed of terricolous lichens, including *Cladonia stellaris* (Opiz) Pouzar & Veda, *C. rangiferina* (L.) F.H. Wigg. and *C. mitis* Sandst, and feather mosses, including *Pleurozium schreberi* (Brid.) Mitt., *Dicranum polysetum* Swartz, *D. undulatum* Schra. Ex Brid.

3.4.2 Sampling design

In 2022, a 400 m^2 circular plot was delineated at each site. Soil samples were collected from four separate locations set at pre-determined distances from the center of each plot. The north, east, south and west cardinal points were used as directions when measuring distances. A distance of 2m from the plot center was measured following the north point, a distance of 5m following the east point, a distance of 8m following the south point and a distance of 11m following the west point. Then two soil samples were collected at each distance point from the OF horizon to mineral layer (depth around 12cm) using a tube (diameter 8.8cm). Of these, four soil samples from each site are mixed and used for chemical analysis; the other four samples, at each site, will be kept independently and used for visual analysis. So, a total of four independent samples and one mixed sample were collected per site. All samples were oven dried at 70°C for 24 hours.

3.4.3 Charcoal analysis

Charcoal amounts

After drying, samples for visual analysis were weighed and then sieved to 2mm. Charcoal particles visible to the naked eye were collected from the >2mm portion of the soil and weighed. A ratio of collected charcoal amount, in *mg*, to initial sample mass, in *g*, was calculated for the four independent samples per site. Then an average of the four ratios was calculated to obtain a unique charcoal amount at each site. Note that these charcoal particles were retained for the charcoal properties analysis below. We use the term visual-charcoal for these determinations.

For chemical analysis of charcoal content, after drying, the mixed soil samples were re-mixed to maximize their homogeneity. A subsample then was taken from the mixed soil samples using a 473ml container for each site. These subsamples were ground with a mortar grinder (Retsch RM 200, Verder Scientific Inc., PA, USA), set at level 2 for 60 seconds. The hydrogen/weak nitric acid digestion method described in Kurth et al. (2006), was used to determine the charcoal content in soil. 1g soil samples were taken from the crushed subsamples and placed in borosilicate tubes for distillation. A 20 ml volume of H_2O_2 of 30% concentration and a 10 ml volume of HNO_3 of 10% concentration were added to the distillation tubes. The tubes were vortexed for 10 seconds and placed on a heating block at 100 °C for 16 hours. After cooling, the tubes were vortexed again to resuspend the solid portion. The mixtures were then filtered through Whatman NO.3 filters and collected in a container. Five rinses and filtrations were performed to recover as many solids as possible. Then, carbon content was measured by dry combustion using an elemental analysis instrument (LECO 928 Series C/N/S Analyzer, St-Joseph, MI, USA). Charcoal amount was estimated from the measured carbon mass by assuming that the carbon content of the charcoal is equal to 55% (Bélanger and Pinno 2008). We refer to this charcoal as chemical-charcoal.

Charcoal properties

The charcoal particles were collected visually, and we measured the specific surface area (SSA), carbon (C), nitrogen (N), oxygen (O), hydrogen (H), sulfur (S) and ash of the charcoal particles in Centre technologique des résidus industriels (CTRI), Rouyn-Noranda, Quebec. Ash content is calculated by subtracting the total C, N, H, O, S content from 1. The reason we chose these charcoal properties is because studies have shown that these charcoal indicators are more applicable in responding to fire severity (pyrolysis process). Due to project funding constraints, we chose a total of 16 sites we chose (8 lichen woodlands + 8 moss woodlands), e.g., 7 sites near to La Sarre (3 lichen woodlands + 4 moss woodlands), 9 sites near Chibougamau (5 lichen woodlands + 4 moss woodlands). For C, N, O, H, S analysis, charcoal samples were oven-dried at 105°C and then ground prior to analysis on a Thermo Flash*Smart* elemental analyzer. Surface area was measured using CO_2 sorptometry on a Quantachrome Autosorb1 (Mukherjee et al. 2011). All charcoal samples were de-gassed under vacuum at least 24 h at 180 °C prior to analysis.

3.4.4 Other biotic and abiotic factors

In this study, stands productivity, tree height, the degree of canopy opening, and site index of each site (SI) and tree regeneration density was measured in each site. The site index of each site was calculated by using the height of the ten most dominant trees per site (Pacé et al. 2020a). In addition, the aboveground dry biomass of the understory vegetation, including feather moss, lichen, and Ericaceae, was estimated in each site. More detailed information, see Pacé et al. (2020a).

3.4.5 Statistical analysis

Differences in charcoal amount (chemical/visual) and charcoal properties (C, N, H, O, S, ash and surface area) between lichen woodlands and moss forests were analyzed by one-way ANOVA with Tukey post-hoc tests ($\alpha = 0.05$). Here, we considered the ground cover mainly dominated by lichen as lichen woodland and mainly dominated by feather moss as feather moss forest. Also, the charcoal S content was not detected in any of charcoal samples, so it is not presented in the results. To meet the assumptions of normality and homogeneity of variance, the data of chemical charcoal amount and visual charcoal amount were log transformed. Using linear regression models and generalized linear model (GLM), we tested the correlations between charcoal amount and vegetation status, including lichen biomass, moss biomass, Ericaceae biomass , the degree of canopy opening, tree density, and SI. To select the model with the best fit by lowest AIC and standard errors. All analyses were performed on R software, version 4.2.2 (R Development Core Team 2022).

3.5 Results

3.5.1 Charcoal amount in lichen woodlands and feather moss forests

The amount of post-fire charcoal estimated by chemical and visual methods were positively correlated at $p < 0.001$ (Fig.3.1).

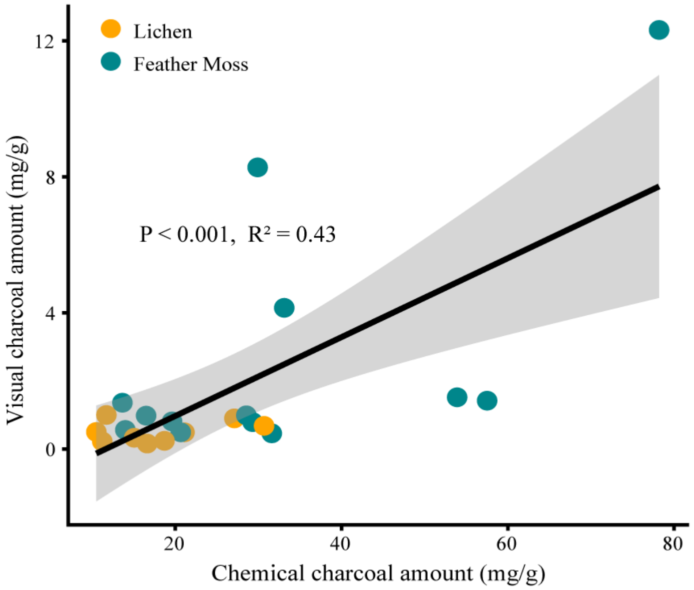

Figure 3.1 Linear relationships between chemical charcoal and visual charcoal amounts. The regression line and 95% confidence interval with shaded area are shown. The R^2 denotes the regression between log-transformed chemical charcoal amount and log-transformed visual charcoal amount. The p values denote the significance of the bivariate regression relationships.

Further, our one-way ANOVA showed that the charcoal amount differed noticeably between lichen woodlands and moss forests (Fig.3.2). Consistently, both methods showed a significantly higher charcoal amount in moss forests than in lichen woodlands. The average amount of charcoal (visual) was 2.79 mg/g in moss forests and 0.60 mg/g in lichen woodlands (Table S3.1). The amounts of charcoal estimated by the chemical method was about ten time higher than that estimated by the visual method.

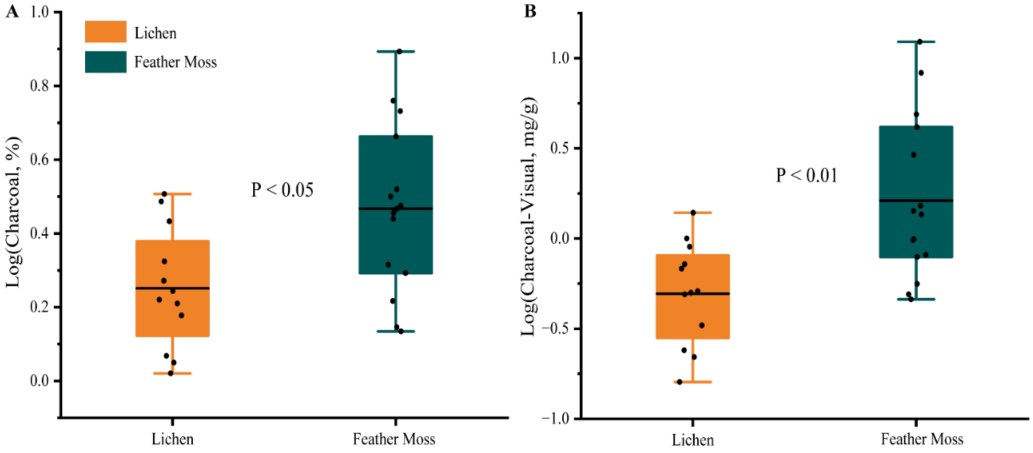

Figure 3.2 Differences in charcoal amount between lichen woodlands and feather moss forests, A) chemical charcoal amount, B) visual charcoal amount. The p values denote the significance of the difference between lichen and moss. The line represents the mean value. Detailed descriptive statistics for all observations across studies are provided in Supplementary Information (Table S3.1).

3.5.2 The relationship between charcoal amount and aboveground vegetation

Several linear relationships were found between charcoal amount and lichen and moss ground covers (Fig. 3.3). There were negative linear relationships between visual and chemical charcoal amounts and lichen biomass with lichen biomass decreasing significantly with increasing charcoal amounts (Fig. 3.3A). In contrast, feather moss biomass was positively related to visual charcoal amounts and this relationship barely missed the 5% significance threshold (Fig. 3.3B). In addition, we found that the amount of understory vegetation mostly composed of ericaceous shrubs, was not significantly correlated with charcoal amount, even though there was a positive trend (Fig. 3.3C).

We assessed the relationships between charcoal amount and several indicators of forest productivity state using a polynomial function (Fig. 3.4). The results indicated a relationship between visual charcoal amounts and the degree of canopy opening. There was a general downward trend in visual charcoal amounts with canopy opening, i.e., opening degree decreases with increasing charcoal amount, while charcoal amounts firstly increase then decrease with tree density. The amount of charcoal was not significantly related to site index (SI), but showed a positive trend.

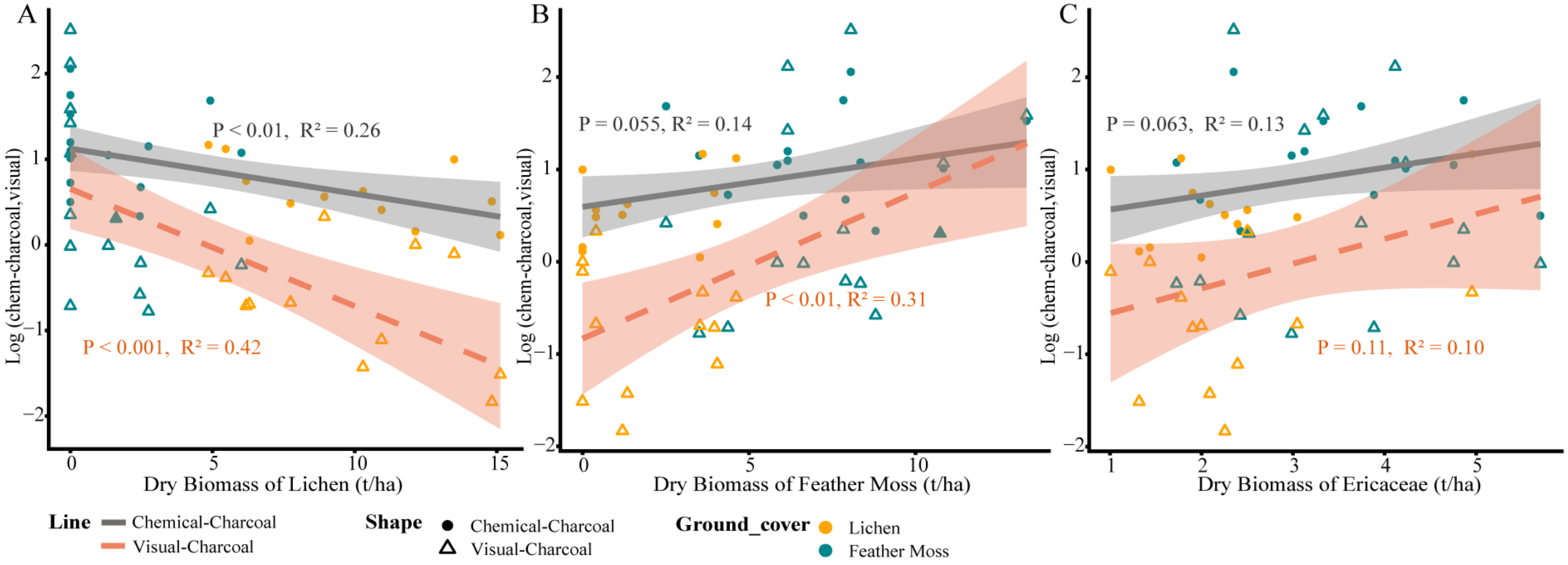

Figure 3.3 The relationships between chemical/visual charcoal amount and A) dry biomass of lichen, B) dry biomass of moss and C) dry biomass of ericaceae. The regression line and 95% confidence interval with shaded area are shown. The R2 denotes the regression between log-transformed chemical/visual charcoal amount and above vegetation biomass. The p values denote the significance of the bivariate regression relationships.

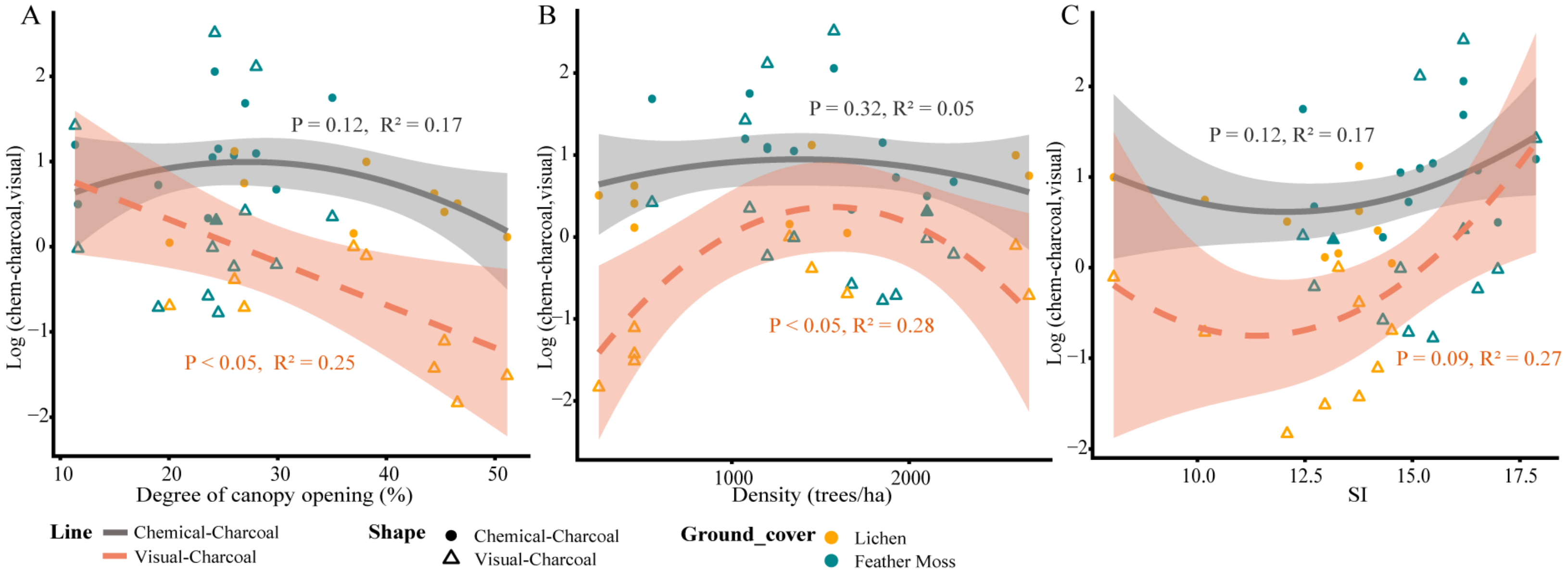

Figure 3.4 The relationships between chemical/visual charcoal amount and A) degree of canopy opening, B) density and C) site index (SI). The regression line and 95% confidence interval with shaded area are shown. The R^2 denotes the regression between log-transformed chemical/visual and site productivity status. The p values denote the significance of the bivariate regression relationships.

3.5.3 Differences in charcoal properties between lichen and moss forests

Mean values of charcoal chemical and physical properties did not differ significantly between lichen and moss forests (Fig.3.5). However, the variability in charcoal properties N, C, H, O, ash and specific surface area seems to be much larger in lichen woodlands than moss forests. For example, the standard deviation and variance of the charcoal N, C, H, O, ash and surface area reflected higher values in lichen woodlands than in moss forests (Table S3.2).

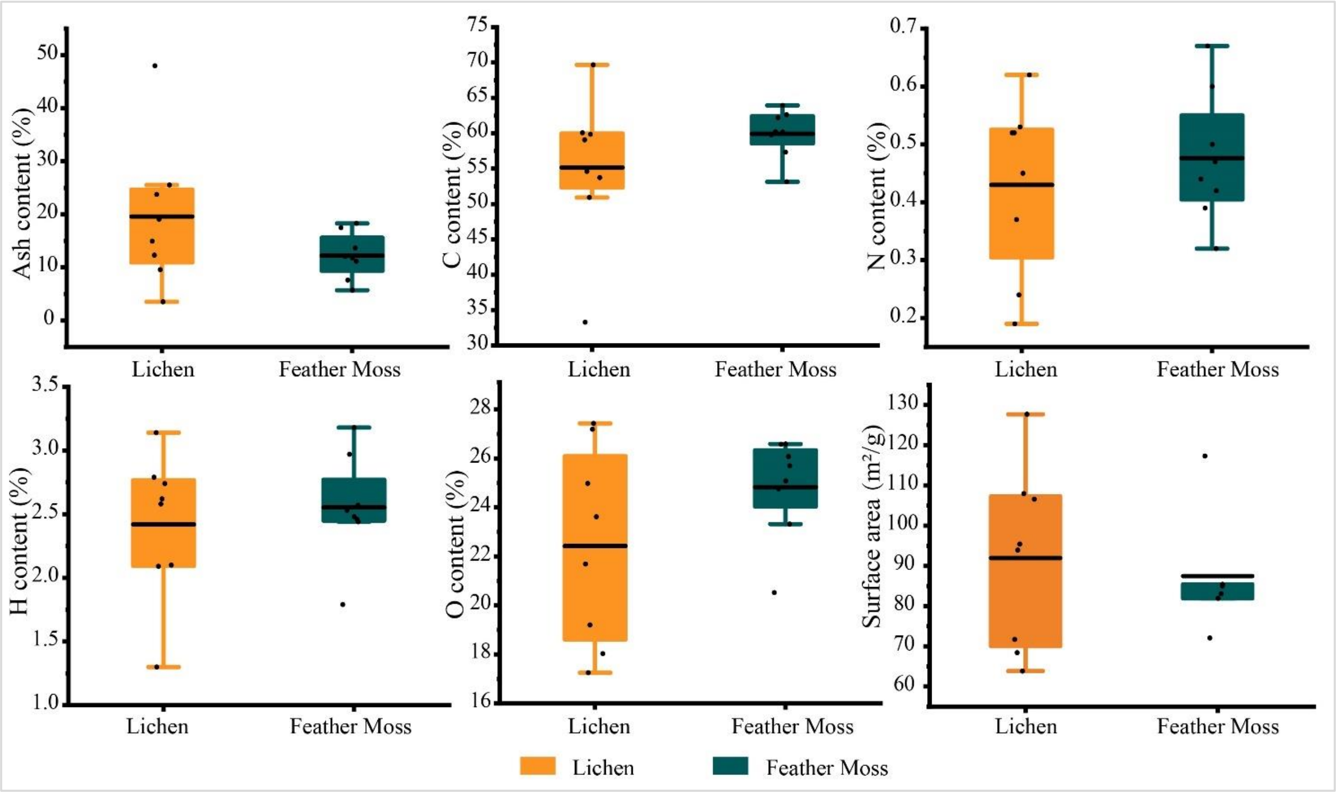

Figure 3.5 Differences in charcoal properties between lichen and feather moss forests, including ash content, carbon content (C), nitrogen content (N), hydrogen content (H), oxygen content (O) and surface area. The line represents the mean value.

3.6 Discussion

3.6.1 The soil charcoal pool in lichen woodlands and moss forests

Both the chemical and visual content of charcoal consistently showed significantly higher amounts of charcoal in moss forests than in lichen woodlands. It has been suggested that pre-fire vegetative biomass as fuel may be correlated with charcoal amount, i.e. it is possible that more live biomass leads to more charcoal (Ohlson et al. 2013). Furthermore, it is likely that most sites have been in their current status for several rotations, i.e. the charcoal conditions are inherited from a long history, not necessarily from the last fire event. A previous study in western Montana showed a post-fire charcoal content of around 200 g/m^2 (DeLuca et al. 2020). Other studies in the boreal region showed that the amount of soil charcoal after forest fires averaged 179 g/m^2 (Ohlson et al. 2013), and a mean of 162 ± 44 g/m^2 (Ohlson et al. 2016). Here, the amount of charcoal in the moss forests is closed to these results with values averaging 146 ±161 g/m^2 for moss forests and 39.5 ± 23.8 g/m^2 for lichen woodlands using the visual method (Table S3.1). Additionally, the chemical amount of charcoal ranges from 0.1%-2.6% in the study of Bélanger and Pinno (2008), which is close to ours, with averages 1.90% in lichen woodlands and 3.33% in moss forests (Table S3.1), but overall our mean value is higher than theirs which may be related to the type of aboveground vegetation (such as the dominant tree species) as well as the fire regime characteristics, or maybe because they didn't include charcoal from the forest floor.

Fire severity can influence the amount of charcoal. Typically, charcoal content decreases as fire severity increases (Knicker et al. 2006; Pingree et al. 2012; Buma et al. 2014), presumably due to more biomass becoming ash and/or volatiles as a result of complete combustion (Demirbas 2004). The lower amounts of charcoal in lichen woodlands could indicated that fires of high intensities generate these forest conditions. However, we found no significant relationships between charcoal chemical or physical properties and forest ecosystem type, albeit a greater variability of charcoal properties for open canopy stands. A study demonstrated that intense wildfires can cause significant losses of soil organic and inorganic matter, leading to erosion and substantial releases of carbon and nitrogen (Bormann et al. 2008). Also, several studies have shown that N, H, O content of charcoal decreases with increasing pyrolysis temperature, while ash, C content and specific surface area increase (Weber and Quicker 2018; El-Naggar et al. 2019). Our results do not allow us to fully validate or reject our hypothesis on the role of fire severity in the shift between open and closed canopy stands. However, they suggest that the variability in fire conditions as well as the amount of charcoal produced are different between ecosystem types.

Nevertheless, a parameter that distinguished the nature of charcoal between the two-forest type is the greater variability of the values for N, H, O, ash and surface area that was observed for charcoal from lichen woodlands, that may be due to the heterogeneity of fire severity (Brimmer, 2006; Preston et al., 2017) and reflects the patchiness of charcoal distribution at the spatial scale. The high variability in charcoal properties appears to be common on wildfire sites. Preston et al. (2017) also found a very wide range for ash from 46 to 311 mg/g with 26 samples in the boreal forest.

3.6.2 Potential role for charcoal in maintaining the two alternative ecosystem stable states.

Soil charcoal can have a stimulating effect on moss growth and cover, and on fern establishment (Wardle et al. 1998). Some studies have suggested that charcoal can promote tree growth, and soil moisture retention (Makoto et al. 2010; Licht and Smith 2020). A recent study in the Canadian boreal forest showed that charcoal enhances plant growth after fire disturbance up to an optimum point (peak value of polynomial), and that this effect was greater in acidic and nutrient-poor boreal soils (Gale and Thomas 2021). Another study in the Russian Far East, showed that the presence of charcoal was correlated with tree root vitality (Bryanin and Makoto 2017). Our results are basically consistent with these studies. However, Kasin et al. (2016) found no correlation between tree density and charcoal amount. Additionally, tree species may differ in their response to charcoal additions, for example, fire-adapted species (e.g., *P. banksiana*) may show a positive response to fire residues (Gale and Thomas 2021), however, neutral to negative effects of biochar and wood ash additions were observed in a field experiment with *Picea glauca* (Bieser and Thomas 2019).

Charcoal can improve soil conditions, for example, it may increase the soil sorption capacity as a powerful adsorbent for organic compounds such as phenolics that may suppress N mineralization and nitrification in the forest floor (Zackrisson et al. 1996; Pingree et al. 2016; Makoto and Koike 2021). Previous studies have also shown that charcoal can improve soil quality, leading for example to higher CEC (Liang et al. 2006), improved soil P availability (Makoto and Koike 2021), extended periods of high P availability, high concentrations of Ca and Mg (Makoto et al. 2012; Pluchon et al. 2014), and higher soil pH (Gale and Thomas 2021). Thus, the presence of charcoal may contribute to the recovery and growth of vegetation by improving soil conditions. In this context abundance of charcoal may play an important role in explaining the co-occurrence of two alternative forest stable states.

3.7 Conclusion

Charcoal in the top soil layer can play an important role in boreal forest ecosystems. Overall, the amount of charcoal in closed-canopy moss forests was significantly higher than that in lichen woodlands, while the chemical and physical properties of charcoal were not different. The charcoal amount could be linked to the biomass of the previous stand, hence the results cannot be used to firmly confirmed that a high fire severity is at the origin of open canopy stands. The results suggest that dense stands, undergoing low intensity fire may produce more charcoal and that charcoal could contribute to maintaining a productive state. More broadly, modern forestry in the boreal region is associated with logging as well as fire suppression (Boucher et al. 2017; Rolstad et al. 2017), which necessarily reduce the production and deposition of new charcoal. Further investigations on the amounts, distribution and the role of charcoal in managed boreal forest are needed.

GENERAL CONCLUSION

This doctoral thesis contributes to a better understanding of the presence of two ecosystem stable states in the boreal forest (open canopy woodland, and closed canopy moss forests) and on the mechanisms that maintain these stable states and the ones that can initiate the transition from one to the other. Here we used an experimental approach that allow to discriminate the effect of ground cover (moss vs lichen) from other factors such as soil and site properties. To our knowledge, this is the first experiment using moss transplantation, the 10-years effects of moss transplantation in an open lichen woodland on soil conditions, and soil microbial communities, including bacteria and fungi, tree growth and foliar nutrients were assessed. Another aspect of this research was to investigate the role of fire history on the occurrence of one or the other of the stable alternative states by investigating the physical and chemical characteristics as well as the amounts of soil charcoal. The results of the study would contribute to a better understanding of the mechanisms of ecological transitions between lichen and moss forest ecosystems, and the relationship between the charcoal and these two forest ecosystems.

In Chapter 1, we used moss transplantation treatments, lichen removal treatments, and lichen and moss control treatments to determine the survival potential and impact of moss invasion in lichen woodlands. This chapter confirms that feather moss is an important component in boreal forests, contributing to boreal forest growth and improving soil properties. Our 10-year results indicate that it is possible to replace a lichen cover with a feather moss cover and that feather moss can establish, survive, and remain healthy in an environment previously occupied by lichen. In view of the positive role of feather moss transplants in enhancing soil conditions and tree growth and given the fact that soil microorganisms are closely interconnected with soils and plants, the effects of moss transplantation, as well as other treatments, on the diversity, composition, and function of soil microorganisms, including bacteria and fungi, are covered in Chapter 2. This chapter confirms that changing the forest ground cover has a significant impact on the diversity, composition and function of soil microbial communities. Fungal diversity was more sensitive to changes in lichen and moss cover, compared to bacterial diversity. Soil microbial community composition showed significant differences among forest ground covers, but that of moss transplantation and control moss showed greater similarities. More importantly, changes of forest ground cover significantly affected the structure of microbial communities and fungal functional groups. Moss transplantation significantly increased the relative abundance of the organic nitrogen-scavenging fungal genus, *Piloderma*. Soil moisture and temperature were the main environmental variables associated to the shift in microbial community composition. The ground cover type plays an important role in shaping distinct soil microbial communities in the boreal forest and may help maintaining two contrasting stable alternate ecosystem states. Based on the results of the previous two chapters, we concluded that lichen woodland and moss forests are two different ecosystems in which the ground cover together with the soil microbial populations that they support, contribute to maintaining these ecosystems in their distinct status by influencing the soil conditions for tree growth. An aspect that is still poorly understood is the transition between one stable state and the other and specifically the role of fire severity. Chapter 3 investigates how the characteristics of soil charcoal could inform on past fire severity. Our findings validated in that the amount of charcoal found on the ground of closed-canopy forests was greater than that of open-canopy forests. However, the chemical and physical properties were not different, albeit a greater variability of charcoal properties for open canopy stands. These results do not allow us to fully validate or reject our hypothesis on the role of fire severity in the shift between open and closed canopy stands. However, they suggest that the variability in fire conditions as well as the amount of charcoal produced are different between ecosystem types.

Characteristics of ecosystem transformation and fire legacies in the boreal forest. The overall goals and objectives of this project have been met in the three main chapters (Chapters 1, 2, and 3), but a synthesis of the results from each chapter helps us to better understand the role of these effects in the dynamics of the boreal forest.

The role of moss in the recovery of the boreal forest productivity

The results of 10 years of experiment indicates that feather moss plays a crucial role in supporting tree growth (Fig.4.1), possibly through its influence on water regulation and litter decomposition rates in low-productivity lichen woodlands. Mansuy et al. (2013) suggested that afforestation of open lichen woodlands in boreal forests can be a means of increasing forest productivity. However, without appropriate soil conditions, productive closed-canopy forest conditions may not be achieved. Notably, our study is the first to propose that manipulating the ground layer could lead to a shift from open-lichen woodland conditions to a more productive closed-canopy moss forest environment. Furthermore, our results of no moss expansion after 10 years suggest that the effect may be local, at least in the short term. Therefore, the feasibility of transplanting moss over large areas and the long-term survival of moss need to be evaluated. Moreover, lichen removal can adversely affect tree growth, so removing lichen as an alternative to increasing productivity does not seem to be a good approach, probably because lichen cover prevents direct soil exposure, which in turn maintains soil moisture. Therefore, we conclude that the replacement of lichen by feather moss establishes soil conditions that appear conducive to better tree growth and has the potential of restoring the productivity of boreal forests in open-canopy lichen woodlands. Productive forest conditions can be maintained by promoting low light conditions in the understory that are favorable to feather mosses rather than to lichens. Dense plantations or restocking natural stands could be possible solutions.

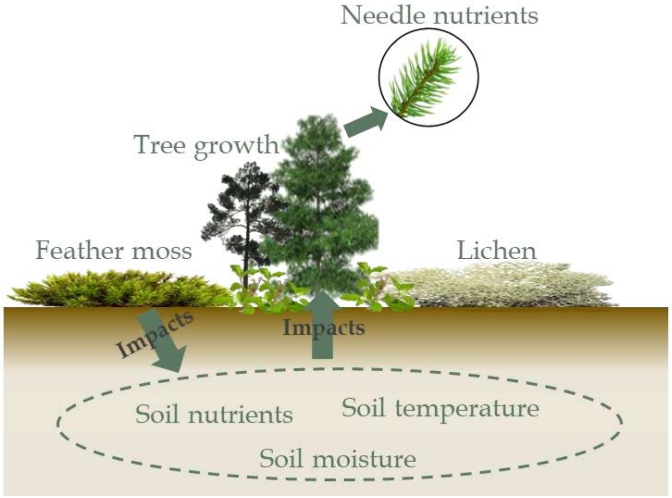

Figure 4.1 The role of feather moss in the recovery of boreal forests productivity. Feather moss enhances soil conditions to promote tree growth and regulates foliar nutrient concentrations.

Links between soil microbiomes and boreal ecosystems

This study demonstrates the ground cover type (i.e., moss or lichen) plays an important role in shaping distinct soil microbial communities in the boreal forest and may help maintaining two contrasting stable alternate ecosystem states (Fig.4.2). This is closely interlinked with our findings in Chapter 1. These two forest ecosystems could maintain different soil functions by driving soil microbial communities, such as the environment created by feather moss can increase the proportion of ectomycorrhizal fungi which are closely linked to stand productivity in boreal forests (Nagati et al. 2018; Nagati et al. 2019). Under climate change, the expansion of the proportion of open-canopy lichen woodlands may be amplified by plant-microbe-soil positive feedback loops which maintain low forest productivity and slow nutrient cycling in lichen-dominated stands, and the opposite conditions in feather moss forests. Therefore, the role of soil microbes should be taken into account in future work on assessing ecosystem transformation.

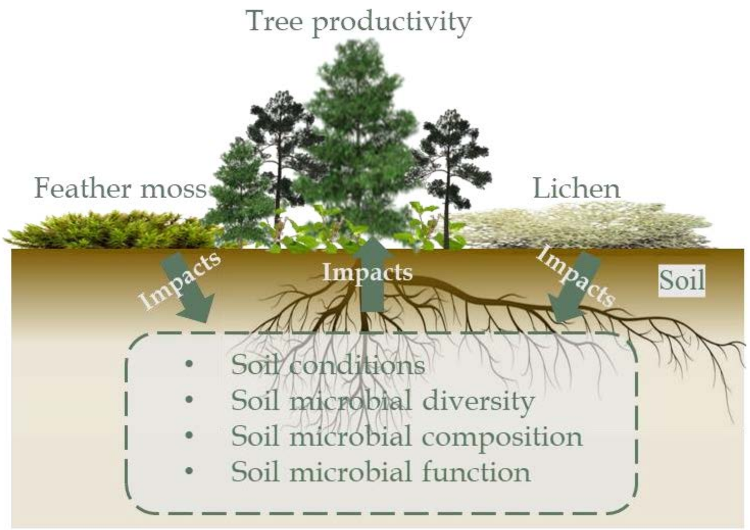

Figure 4.2 Links between soil microbiomes and the two forest ecosystems, i.e., lichen and moss forests.

The potential impacts of charcoal on boreal forests

Based on the findings in Chapters 1, 2, and 3, it is possible to link charcoal pools to the two alternative boreal forest ecosystems, including moss and lichen forest ecosystems (Fig.4.3). First, in experiments with ground cover manipulation, moss cover showed improvement in soil conditions and tree growth (Chapters 1), as well as alteration of soil function through microbial communities (Chapters 2), whereas significant differences of charcoal amounts were also found in observations between lichen woodlands and moss forests. Fire severity may influence the amount of charcoal, typically, charcoal content decreases as fire severity increases (Knicker et al. 2006; Pingree et al. 2012; Buma et al. 2014), presumably due to more biomass becoming ash and/or volatiles as a result of complete combustion (Demirbas 2004). The lower amounts of charcoal in lichen woodlands could indicated that fires of high severity generate these forest conditions. Second, charcoal properties indicated greater variability in charcoal properties in lichen woodlands than in moss forests, that may be due to the heterogeneity of fire severity. Finally, charcoal pools were positively associated with moss biomass, and negatively associated with lichen. Also, charcoal could promote forest growth and regeneration under an optimum point of charcoal amount. These linkages suggest that fire disturbance acts as a driver in boreal forest dynamics, particularly the role of charcoal in post-fire re-establishment and restoration deserves more attention, and demonstrate the potential role of charcoal in explaining the co-occurrence of two stable alternative states.

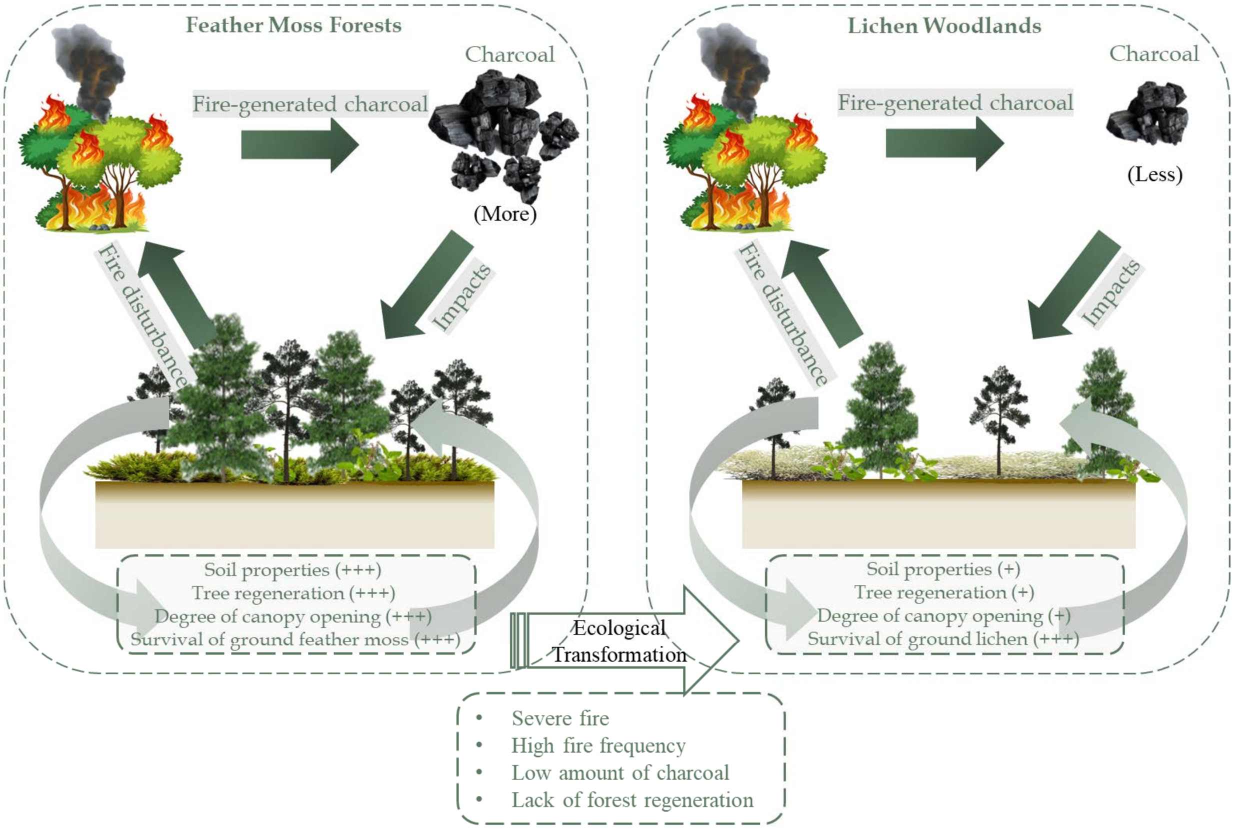

Figure 4.3 The potential ecological role of charcoal on the maintenance of two stable alternative forest states in boreal forests and the potential mechanisms for transition of these two ecosystems. (+++) represents potentially more positive effects of charcoal on forest ecosystems, (+) represents potentially fewer positive effects of charcoal on forest ecosystems.

What charcoal can reveal about the origin of open lichen woodland and their progression in the closed canopy moss forest

Charcoal, as a product of fire disturbances, plays a crucial role in unraveling the origin and progression of open lichen woodlands within closed canopy moss forests in the boreal region. The observed shift between these alternative stable states has been linked to increased fire disturbances, including both severity and frequency, influenced by global climate change. By investigating the amount and properties of charcoal, our research provides insights into the ecological transformations occurring in these ecosystems.

Our findings showed that the amount of charcoal is significantly higher in closed-canopy moss forests compared to open lichen woodlands. This discrepancy supports the hypothesis that a greater severity of fires contributes to the transition between these two states. While the chemical and physical properties of charcoal did not exhibit significant differences, the observed variability in open canopy stands implies heterogeneity in fire severity, emphasizing the intricate nature of these ecological shifts. Through ground cover manipulation experiments, specifically moss transplantation, we demonstrated that moss cover positively influences soil conditions, tree growth, and microbial communities. The positive association between charcoal pools and moss biomass further highlights the intricate relationship between fire disturbances, charcoal dynamics, and ecosystem states. Our research underscores the potential of charcoal to act as a driver in boreal forest dynamics, influencing post-fire re-establishment and restoration.

In summary, the amount and properties of charcoal provide potential indicators of fire severity and ecosystem changes, revealing on the factors contributing to the transition from closed-canopy moss forests to open lichen woodlands. Understanding the role of charcoal in these alternative states enhances our knowledge of boreal forest dynamics and emphasizes the importance of considering fire disturbances in ecosystem management and conservation strategies.

Implications for boreal forest management

The findings from this study carry significant implications for forest management in boreal ecosystems. Firstly, the observed transition from open lichen woodlands to closed-canopy feather moss forests underscore the importance of understanding and mitigating the factors driving ecological transformations in these regions. As global change continues to increase the frequency and severity of natural disturbances, such as fires and insect outbreaks, forest managers must prioritize strategies that promote resilience and adaptability in forest ecosystems.

The successful transplantation of feather moss into open lichen woodlands highlights the potential for targeted interventions to influence ground cover vegetation and, consequently, soil conditions, microbial communities, and tree growth. This suggests that forest management practices aimed at favoring feather moss over lichen could contribute to maintaining or restoring productive woodland conditions. To address the dominance of mosses and maintain closed-canopy conditions, it is important to reconsider current forestry practices. While thinning is often recommended to reduce competition for light and promote tree growth, this approach can exacerbate the problem. Instead, high-density planting or filling in regenerating stands may be more effective strategies. Specifically, implementing a strategy firstly to quickly identify regeneration failures and replant these sites to restore vegetation cover is crucial. Secondly, planting at greater densities on these sites can help maintain a dense canopy. Additionally, encouraging the growth of companion species that contribute to restoring vegetation cover is beneficial.

Furthermore, the observed differences in charcoal amounts between moss and lichen ecosystems, along with the potential role of charcoal in promoting vegetation recovery and moss survival, emphasize the importance of considering fire dynamics in forest management planning. While the study did not conclusively validate the hypothesis regarding fire severity's role in the transition between forest states, it underscores the need for further research to understand the complex interactions between fire regimes, ground cover vegetation, and ecosystem dynamics.

In summary, this research underscores the importance of considering ground cover vegetation, particularly the role of feather moss and lichen, in boreal forest management strategies. By manipulating ground cover composition, forest managers may have the opportunity to promote conditions conducive to productivity and resilience, ultimately contributing to the long-term sustainability of boreal forest ecosystems. Further studies investigating the relationship between fire dynamics, charcoal properties, and forest soil will be essential for developing informed forest management strategies in the face of ongoing global change.

Contributions This thesis proposes the possibility of restoring open lichen woodlands to closed-canopy forests and determines the relationship of fire legacy (i.e., charcoal) to these two ecosystems and its ecological roles in boreal forests.

1) Confirmation that moss transplantation can survive in open lichen woodlands. it is possible to replace a lichen cover with a feather moss cover and that feather moss can establish, survive in an environment previously occupied by lichen.

2) Identification of the positive role of moss transplantation in improving soil conditions, promoting tree growth, and altering microbial community function in open lichen woodlands. The replacement of lichen by feather moss establishes soil conditions and microbial community that appear conducive to better tree growth and has the potential of restoring the productivity of boreal forests in open-canopy lichen woodlands. Thus, our findings suggest that productive forest conditions can be maintained by promoting conditions that are favorable to feather moss rather than lichen, or that dense plantations are possible solutions. Finally, forest managers can prioritize sites already dominated by feather moss, which may favor tree growth during reforestation.

3) Understanding the role of charcoal in boreal forests. Our results indicate that the amount of charcoal is higher in moss forests than lichen woodlands. The literature shows that the amount of charcoal on the ground can be beneficial to the recovery and regeneration of vegetation up to an optimum point and to favor the survival of mosses. This study suggests a role for charcoal in the establishment and subsequent maintenance of vegetation after fire and in explaining the co-occurrence of two stable alternative states.

4) Charcoal properties as an indicator of the nature of previous fire. Although our study found no significant differences in charcoal properties between open-canopy lichen woodlands and closed-canopy moss forests, it's crucial for future investigations to approach this analysis with caution. The absence of significant differences in our study emphasizes the need for careful consideration and robust methodologies when assessing charcoal properties as indicators of past fire events.

Limitations and future research

Research limitations

1) Short-term observations: In Chapter 1, it was mentioned that, with regard to the effects of mosses in open lichen woodland restoration, the results of the study suggest that the impacts of mosses are likely to be localized and non-diffuse in the short term. This means that we need more long-term studies to determine the lasting effects of mosses in ecosystem restoration.

2) Biodiversity effects: The ecological effects of mosses and lichens have been the main focus of research, but their impact on biodiversity has not been explored in depth. Future research could focus on the effects of mosses and lichens on other groups of organisms, such as insects and birds, in order to more fully understand their roles in the ecosystem.

3) Effects of charcoal: Charcoal has a very high potential to improve soil quality, however we did not explore the association between the forest soil and charcoal due to project resource constraints. Therefore, the relationship between the quantity and physicochemical properties of charcoal and soil quality after fire could be focused on in future studies.

Future research

1) Future studies could expand time scales and conduct longer-term experiments to assess

the long-term ecosystem impacts of moss replacement of lichens. In addition, cross-scale studies could cover different geographic regions to determine the applicability of this alternative strategy.

2) Studies have shown that moss helps improve ecosystem conditions and increase tree growth rates. Future research could consider applying this strategy in practical forest ecological engineering projects, such as reforestation in the moss-dominated areas or through intensive planting that are favorable to feather moss, to restore and enhance forests in less productive areas.

3) Evaluate the relationship between fire characteristics and charcoal properties and amount. Fire disturbances would affect boreal forest ecosystems, and fire severity and frequency may determine the amount and properties of charcoal (fire legacy). Also, we know that the amount of charcoal may alter soil conditions and vegetation growth and recovery. Therefore, it is necessary to clarify the relationship between fire characteristics and charcoal, which in turn has constructive implications for future forest fire management.

4) Evaluate the role of charcoal in the early stages of post-fire re-establishment of vegetation as well as dynamics in the long term. In particular, the relationship with moss forests and lichen woodlands needs to be further defined. Furthermore, assessing the role of charcoal in forest soil.

APPENDICE A - SUPPLEMENTARY MATERIAL FROM CHAPTER 1

Table S1.1 Foliar nutrient content and unit needle dry mass in current-year needles of Jack pine tree (Turkey test).

	Control Lichen	Lichen removal	Moss Transplantation	*p*
Mass per needle (mg)	8.08	7.57	7.87	0.842
N content (ug) per needle	74.37	77.91	76.84	0.948
P content (ug) per needle	7.60	7.41	7.82	0.921
K content (ug) per needle	20.90	20.04	21.95	0.871

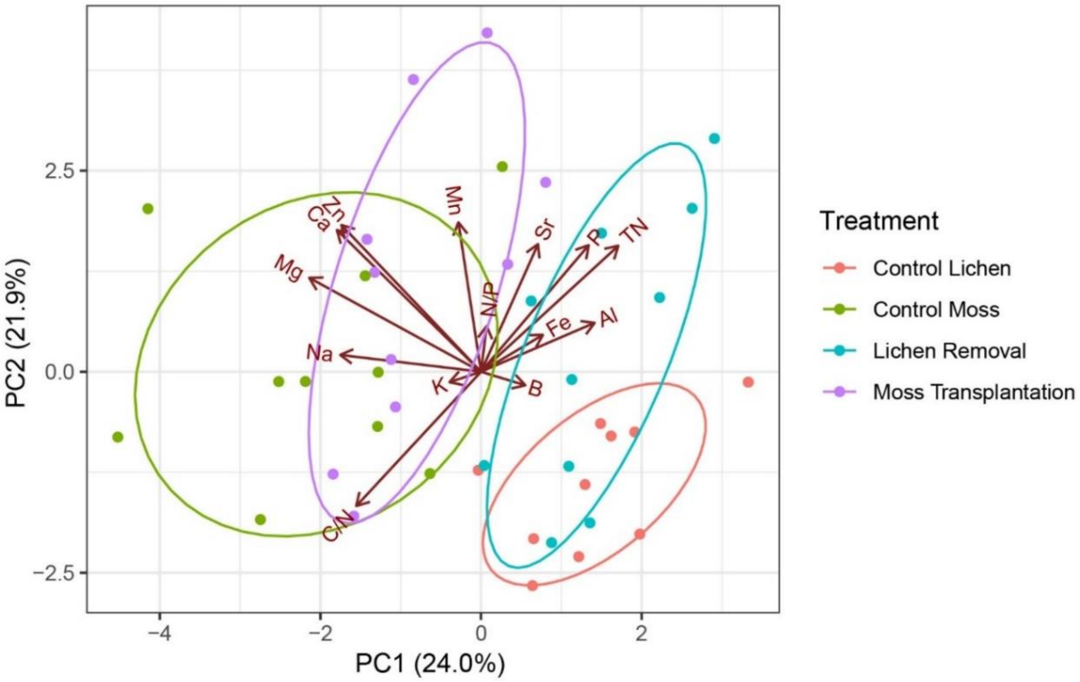

Figure S1.1 Principal component analysis (PCA) biplot of individuals (i.e., treatment plots, *n* = 40) and explanatory variables (i.e., older foliar nutrients, *n* = 14). The biplot shows the PCA scores of the explanatory variables as vectors (dark-red arrows) and individuals of each forest-ground treatment (circles), of the first (x-axis) and second (y-axis) principal components (PCs). Individuals on the same side as a given explanatory variable should be interpreted as having a high contribution on it. The magnitude of the vectors (length of lines) shows the strength of their contribution to each PC. Vectors pointing in similar directions indicate positively correlated variables, vectors pointing in opposite directions indicate negatively correlated variables, and vectors at approximately right angles indicate low or no correlation.

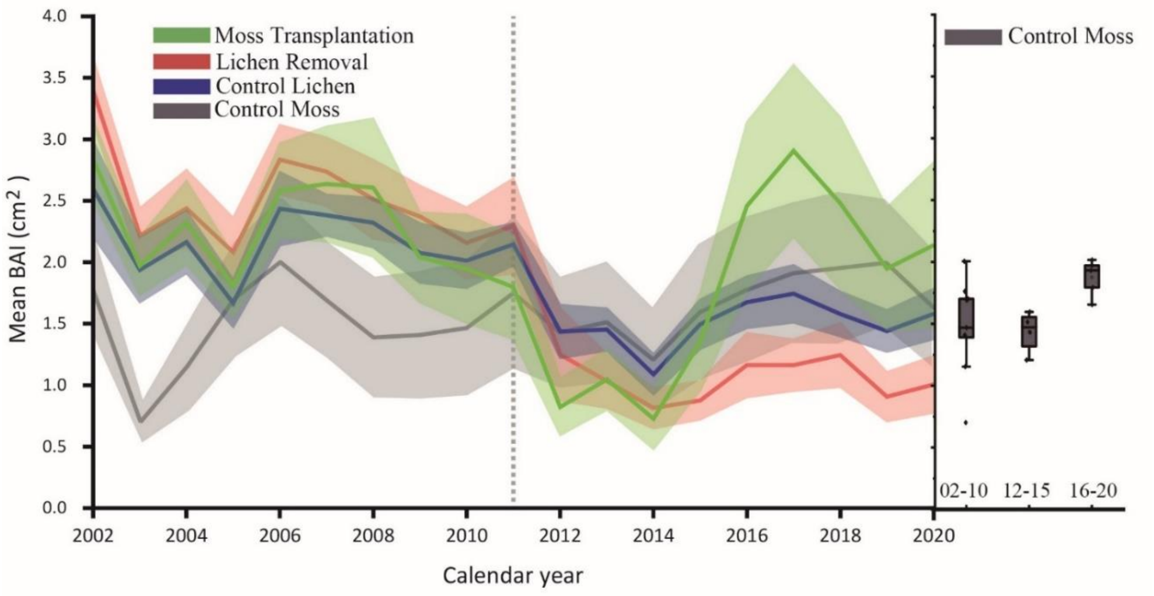

Figure S1.2 Jack pine mean basal area increment (BAI) during 2002–2020. The legend is as follows: Green line, transplanted moss; Blue line: no treatment, control lichen; Red line: lichen removal; Grey line: control moss. The shaded area represents the standard error of the mean. The subplot on the right side shows the median and confidence intervals for different time periods from 2002-2010, 2012-2015 and 2016-2020 for control moss only. Boxplots show the five-number summary of a set of data, including the minimum score, first (lower) quartile, median, third (upper) quartile, and maximum score. the black points represent the distribution of the sample.

APPENDICE B - SUPPLEMENTARY MATERIAL FROM CHAPTER 2

Table S2.1 Bacterial and Fungal alpha diversity among forest cover treatments in forest soil. Forest cover treatments dominated by control lichen (CL), lichen removal (RL), moss transplantation (MT) and control moss (CM). Values are means ± standard errors. Significant differences between ground-cover treatments are represented by different letters. Significant P values (ANOVA with Tukey test) are shown in bold.

		CL	**RL**	**MT**	**CM**	**P value**
				Bacterial		
Organic layer	Observed index	437.2 (89.89)	425.6 (105.57)	391.8 (127.2)	451.0 (99.77)	0.642
	Shannon index	5.22 (0.16)	5.27 (0.25)	5.13 (0.35)	5.31 (0.21)	0.419
	InvSimpson index	76.33 (19.14)	80.99 (29.67)	75.87 (29.77)	96.96 (29.68)	0.29
Mineral soil	Observed index	390.9 (48.30)ab	357.4 (31.78)b	361.3 (74.22)ab	476.1 (171.55)a	**0.0354**
	Shannon index	5.67 (0.11)	5.59 (0.10)	5.54 (0.27)	5.66 (0.34)	0.547
	InvSimpson index	230.98 (25.64)	212.82 (34.81)	189.29 (67.52)	174.36 (59.77)	0.074
Rhizosphere	Observed index	323.67 (99.99)	288.50 (40.55)	376.10 (73.64)	319.75 (121.66)	0.175
	Shannon index	4.83 (0.42)	4.70 (0.27)	5.06 (0.17)	4.82 (0.52)	0.161
	InvSimpson index	58.41 (23.79)	46.31 (16.02)	68.42 (14.03)	63.07 (26.15)	0.11
				Fungal		
Organic layer	Observed index	189.6 (24.89)ab	217.7 (20.73)a	196.4 (32.66)a	163.4 (18.53)b	**<0.001**
	Shannon index	4.13 (0.24)ab	4.36 (0.20)a	3.89 (0.57)b	3.78 (0.21)b	**<0.01**
	InvSimpson index	34.89 (11.41)ab	41.75 (12.13)a	23.62 (12.01)bc	20.65 (6.97)c	**<0.001**
Mineral soil	Observed index	9.5 (3.94)	8.25 (3.40)	35.44 (60.70)	57 (51.56)	0.205
	Shannon index	0.98 (0.49)b	0.91 (0.61)b	1.85 (0.92)ab	2.37 (1.07)a	**<0.05**
	InvSimpson index	2.32 (0.89)	2.20 (1.22)	5.51 (4.97)	8.73 (6.74)	0.0672
Rhizosphere	Observed index	139.4 (26.41)a	120.5 (22.83)a	142.9 (36.30)a	77.2 (28.95)b	**<0.0001**
	Shannon index	3.54 (0.49)a	3.44 (0.33)a	3.15 (0.66)a	2.25 (0.72)b	**<0.0001**
	InvSimpson index	18.99 (13.83)a	17.20 (7.46)a	11.24 (10.62)ab	5.25 (3.23)b	**<0.05**

Table S2.2 Permutational multivariate analysis of variance (PERMANOVA) table showing the significant effect of ground cover treatments on soil microbial community (bacterial and fungal) in different soil layers. The post hoc pairwise PERMANOVAs with a multiple comparison correction was tested based on Benjamini–Hochberg method to compare differences between ground cover treatments.

	Df	**F**	**P value**	**Treatment pairs**	**Df**	**Sums of Sqs**	**F**	**P value**	**P adjusted**
Bacterial									
Organic layer	3	5.34	**<0.001**	CL vs MT	1	0.432	3.019	0.001	**<0.01**
				CL vs RL	1	0.684	4.132	0.001	**<0.01**
				CL vs CM	1	0.636	4.601	0.001	**<0.01**
				MT vs RL	1	0.872	5.916	0.001	**<0.01**
				MT vs CM	1	0.620	5.157	0.001	**<0.01**
				RL vs CM	1	1.332	9.332	0.001	**<0.01**
Mineral soil	3	1.33	**<0.001**	CL vs MT	1	0.413	1.018	0.306	0.306
				CL vs RL	1	0.444	1.070	0.028	**<0.05**
				CL vs CM	1	0.752	1.880	0.001	**<0.01**
				MT vs RL	1	0.453	1.100	0.01	**<0.05**
				MT vs CM	1	0.546	1.377	0.029	**<0.05**
				RL vs CM	1	0.639	1.576	0.002	**<0.01**
Rhizosphere	3	1.83	**<0.01**	CL v MT	1	0.352	1.808	0.019	**<0.05**
				CL vs RL	1	0.303	1.426	0.1	0.1031
				CL vs CM	1	0.295	1.299	0.132	0.1340
				MT vs RL	1	0.489	2.675	0.01	**<0.05**
				MT vs CM	1	0.327	1.702	0.049	0.0513
				RL vs CM	1	0.453	2.140	0.022	**<0.05**
Fungal									
Organic layer	3	3.24	**<0.001**	CL vs MT	1	0.864	3.103	0.001	**<0.01**
				CL vs RL	1	0.594	2.164	0.002	**<0.01**
				CL vs CM	1	0.737	2.800	0.001	**<0.01**
				MT vs RL	1	1.183	4.256	0.001	**<0.01**
				MT vs CM	1	0.690	2.585	0.001	**<0.01**
				RL vs CM	1	1.198	4.558	0.001	**<0.01**
Mineral soil	3	1.13	**<0.01**	CL vs MT	1	0.518	1.071	0.127	0.1309
				CL vs RL	1	0.490	0.990	0.614	0.614
				CL vs CM	1	0.572	1.219	0.024	**<0.05**
				MT vs RL	1	0.491	1.014	0.465	0.4721
				RL vs CM	1	0.570	1.223	0.07	0.0733
				MT vs CM	1	0.566	1.215	0.028	**<0.05**
Rhizosphere	3	2.00	**<0.001**	CL vs MT	1	0.569	1.678	0.01	**<0.05**
				CL vs RL	1	0.504	1.460	0.061	0.0649
				CL vs CM	1	0.666	1.701	0.003	**<0.01**
				MT vs RL	1	0.799	2.460	0.006	**<0.01**
				MT vs CM	1	0.830	2.238	0.003	**<0.01**
				RL vs CM	1	0.923	2.446	0.001	**<0.01**

Table S2.3 The differential abundance analysis for forest cover treatments at genus level (Bacterial and Fungal) using the glmmTMB method (*P* < 0.05). Significant differences among forest cover treatments in bold. NA indicates that comparisons between treatments corresponding to each soil layer are not valid, due to the fact that in some treatments are zero values.

Taxon	Organic layer			Mineral soil			Rhizosphere		
	Chisq	Df	P value	Chisq	Df	P value	Chisq	Df	P value
Bacterial Genus									
Acidibacter	1.7801	3	0.6193				4.1378	3	0.247
Acidipila	10.723	3	**0.01332**	0.2903	3	0.9618	8.2768	3	**0.0406**
Acidocella	21.582	3	**7.967e-05**				6.9986	3	0.0719
Acidothermus	6.442	3	0.09198	0.6628	3	0.8819	6.2364	3	0.1007
Aquisphaera	35.575	3	**9.209e-08**	6.5797		0.0865	7.1367		0.0677
Bradyrhizobium	17.024	3	**0.0006986**	11.756	3	**0.0082**	3.625		0.3049
Bryobacter	44.805	3	**1.018e-09**	9.2536		**0.0261**	0.5152	3	0.9155
Burkholderia_Caballeronia_Paraburkholderia	12.458	3	**0.005967**	2.7152	3	0.4377	6.1588	3	0.1041
Candidatus_Xiphinematobacter	47.547	3	**2.659e-10**				11.793	3	**0.0081**
Conexibacter	133.46	3	**2.2e-16**	0.3945	3	0.9414			
Granulicella	32.723	3	**3.682e-07**	3.8691	3	0.276	0.7998	3	0.8495
Mycobacterium	69.114	3	**6.608e-15**	2.6646	3	0.4446	17.552	3	**0.0005**
Occallatibacter	33.458	3	**2.579e-07**				4.6613	3	0.1983
Puia	14.803	3	**0.001993**	6.4875	3	0.0901	2.8425	3	0.4166
Roseiarcus	44.546	3	**1.156e-09**	2.972	3	0.396	11.227	3	**0.0106**
Subgroup_2	14.925	3	**0.001882**	4.58	3	0.2053	1.6385	3	0.6507
Tundrisphaera	49.103	3	**1.24e-10**				17.889	3	**0.0005**
WD2101_soil-group	16.735	3	**0.000801**	0.3378	3	0.9528	5.9448	3	0.1143
WD260	27.055	3	**5.733e-06**	2.2639	3	0.5195	4.1595	3	0.2447
WPS_2	52.503	3	**2.34e-11**				1.5971	3	0.66
AD3				14.86	3	**0.001**			

				2		**9**			
Ellin6067				6.0306	3	0.1101			
Flavobacterium				8.2305	3	**0.0414**			
Mucilaginibacter				10.9	3	**0.0122**			
RCP2_54				13.899	3	**0.0030**	5.7681	3	0.1235
Subgroup_7				18.002	3	**0.0004**			

Fungal Genus	**Organic layer**			**Mineral soil**			**Rhizosphere**		
Archaeorhizomyces	16.567	3	**0.00086**				0.8634	3	0.8343
Cantharellula	NA	3	NA						
Cenococcum	15.27	3	**0.0016**						
Clavaria	0.8651	3	0.8338						
Cortinarius	2.0677	3	0.5585				3.9308	3	0.272
Hyaloscypha	9.8479	3	**0.0199**	NA	3	NA	8.4258	3	**0.0379**
Hydnellum	1.0916	3	0.7791				8.3855	3	**0.0387**
Hygrophorus	2.5623	3	0.4641				5.2636	3	0.1535
Luellia	6.4646	3	0.0910						
Meliniomyces	33.311	3	**2.769e-07**						
Mortierella	26.698	3	**6.812e-06**	NA	3	NA	18.484	3	**0.00035**
Mycosymbioces	6.1785	3	0.1032				3.6957	3	0.2962
Oidiodendron	8.7258	3	**0.03317**				8.6994	3	**0.03357**
Penicillium	2.4663	3	0.4814						
Pezoloma	34.342	3	**1.677e-07**				26.207	3	**8.633e-06**
Phellodon	2.8	3	0.4235				2.4559	3	0.4833
Piloderma	35.683	3	**8.737e-08**	1.3643	3	0.7139	16.721	3	**0.00081**
Sclerococcum	6.8414	3	0.0771	3.1358	3	0.3712	4.6167	3	0.2021
Tricholoma	1.2239	3	0.7473	NA	3	NA	7.0135	3	0.07147
Umbelopsis	25.346	3	**1.307e-05**	NA	3	NA	9.9102	3	**0.0194**
Amphinema				NA	3	NA			
Cadophora				NA	3	NA			
Ciliophora				NA	3	NA			

Cladosporium	NA	3	NA			
Hannaella	NA	3	NA			
Helicodendron	NA	3	NA			
Hymenula	NA	3	NA			
Polyscytalum	NA	3	NA			
Ramularia	NA		NA			
Russula	15.065	3	**0.0017**			
Sebacina	NA	3	NA			
Sphaerulina	NA	3	NA			
Suillus	NA	3	NA	6.1725	3	0.1035
Tylospora	NA	3	NA			
Infundichalara				1.7196	3	0.6326
Mycena				0.8934	3	0.827
Rhizopogon				4.7779	3	0.1888
Sistotrema				0.3293	3	0.9544
Xeromphalina				1.8651	3	0.6009

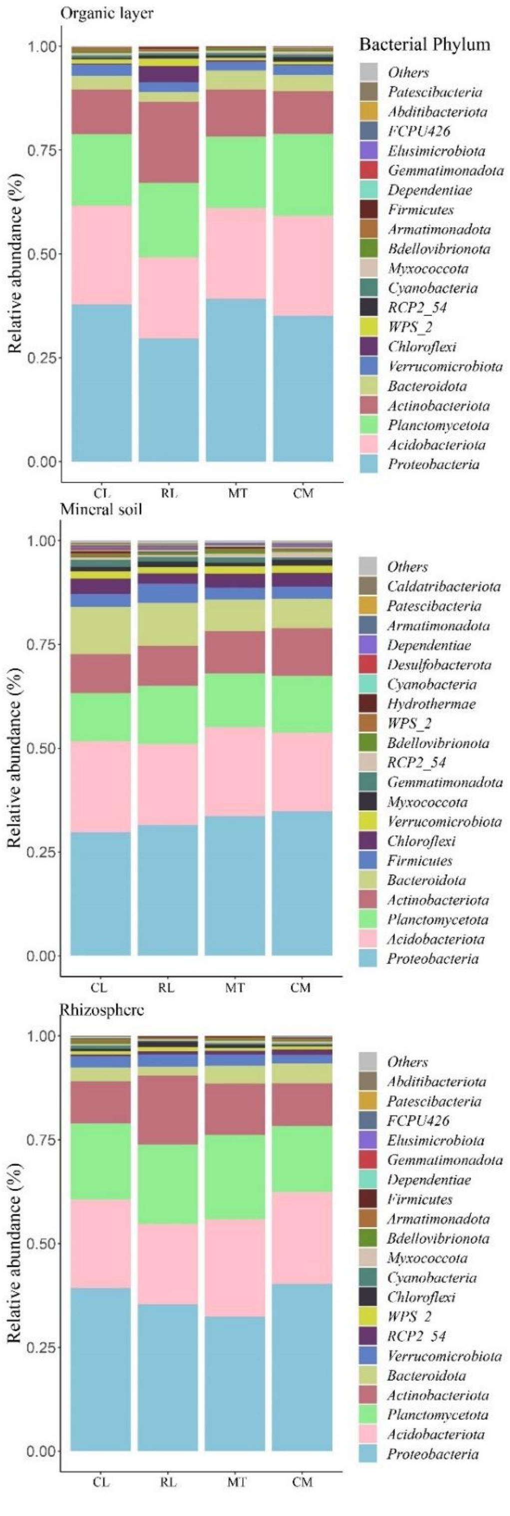

Figure S2.1 Differences in relative abundance (%) of ASVs assigned to Top 20 most relatively abundant bacterial phylum for each soil layer among the forest cover treatments: control lichen (CL), lichen removal (RL), moss transplantation (MT) and control moss (CM).

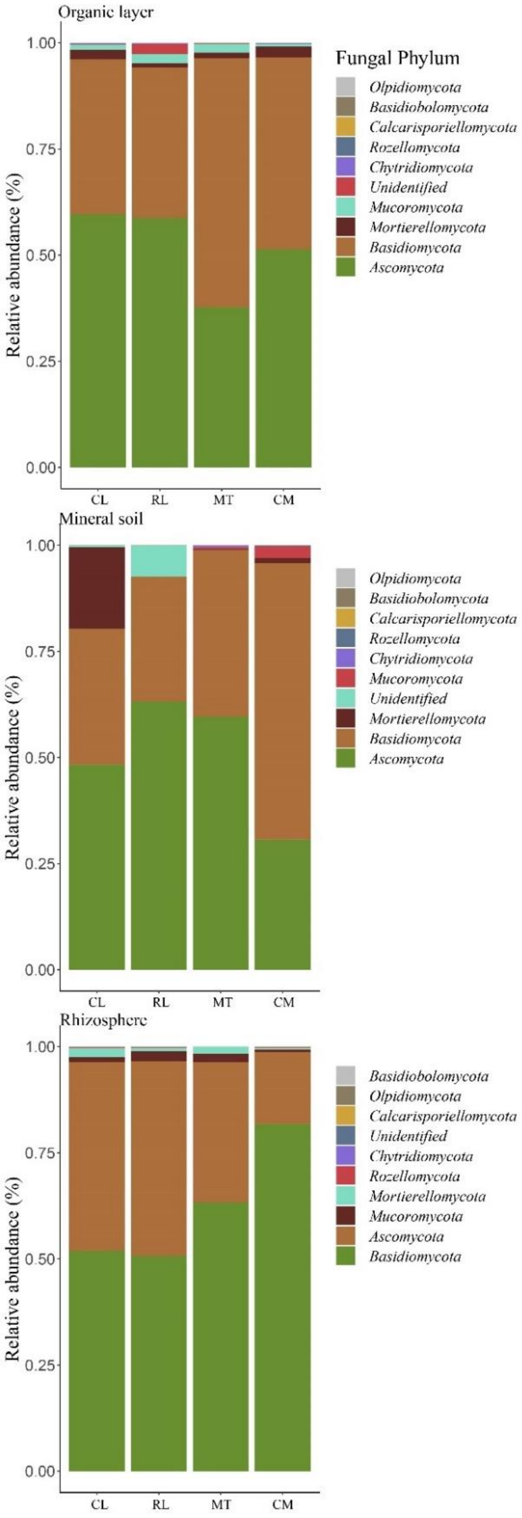

Figure S2.2 Differences in relative abundance (%) of ASVs assigned to fungal phylum for each soil layer among the forest cover treatments: control lichen (CL), lichen removal (RL), moss transplantation (MT) and control moss (CM).

Table S2.4 The differential abundance analysis for forest cover treatments at functional trophic mode and guild level (Klaubauf et al.) using the glmmTMB method (P < 0.05). Significant differences among forest cover treatments in bold. NA indicates that comparisons between treatments corresponding to each soil layer are not valid, due to the fact that values in some treatments are zero.

Fungal Taxon	Organic layer			Mineral soil			Rhizosphere		
	Chisq	**Df**	**P value**	**Chisq**	**Df**	**P value**	**Chisq**	**Df**	**P value**
Trophic Mode									
Pathotroph	2.67	3	0.4453	NA	3	NA	11.871	3	**0.0078**
Symbiotroph	26.604	3	**7.128e-06**	12.054	3	**0.0072**	13.9	3	**0.0030**
Saprotroph	10.971	3	**0.01188**	11.599	3	**0.0088**	5.9831	3	0.1124
Guild									
Animal Pathogen	NA	3	NA	NA	3	NA	NA	3	NA
Ectomycorrhizal	35.299	3	**1.053e-07**	0.4227	3	0.9355	14.403	3	**0.0024**
Endophyte	28.659	3	**2.641e-06**	40.168	3	**9.81e-09**	10.363	3	**0.01572**
Epiphyte	NA	3	NA	NA	3	NA	NA	3	NA
Ericoid Mycorrhizal	8.2496	3	**0.0411**	NA	3	NA	8.0771	3	**0.0444**
Fungal Parasite	NA	3	NA	NA	3	NA	NA	3	NA
Lichen Parasite	NA	3	NA	NA	3	NA	NA	3	NA
Lichenized	NA	3	NA	NA	3	NA	4.3188	3	0.229
Orchid Mycorrhizal	41.594	3	**4.893e-09**	NA	3	NA	0.1703	3	0.9822
Plant Pathogen	0.8064	3	0.8479	NA	3	NA	NA	3	NA
Soil Saprotroph	NA	3	NA	NA	3	NA	NA	3	NA
Undefined Saprotroph	17.516	3	**0.0005**	23.053	3	**3.94e-05**	6.4555	3	0.0914
Wood Saprotroph	1.6379	3	0.6508	NA	3	NA	1.1693	3	0.7604

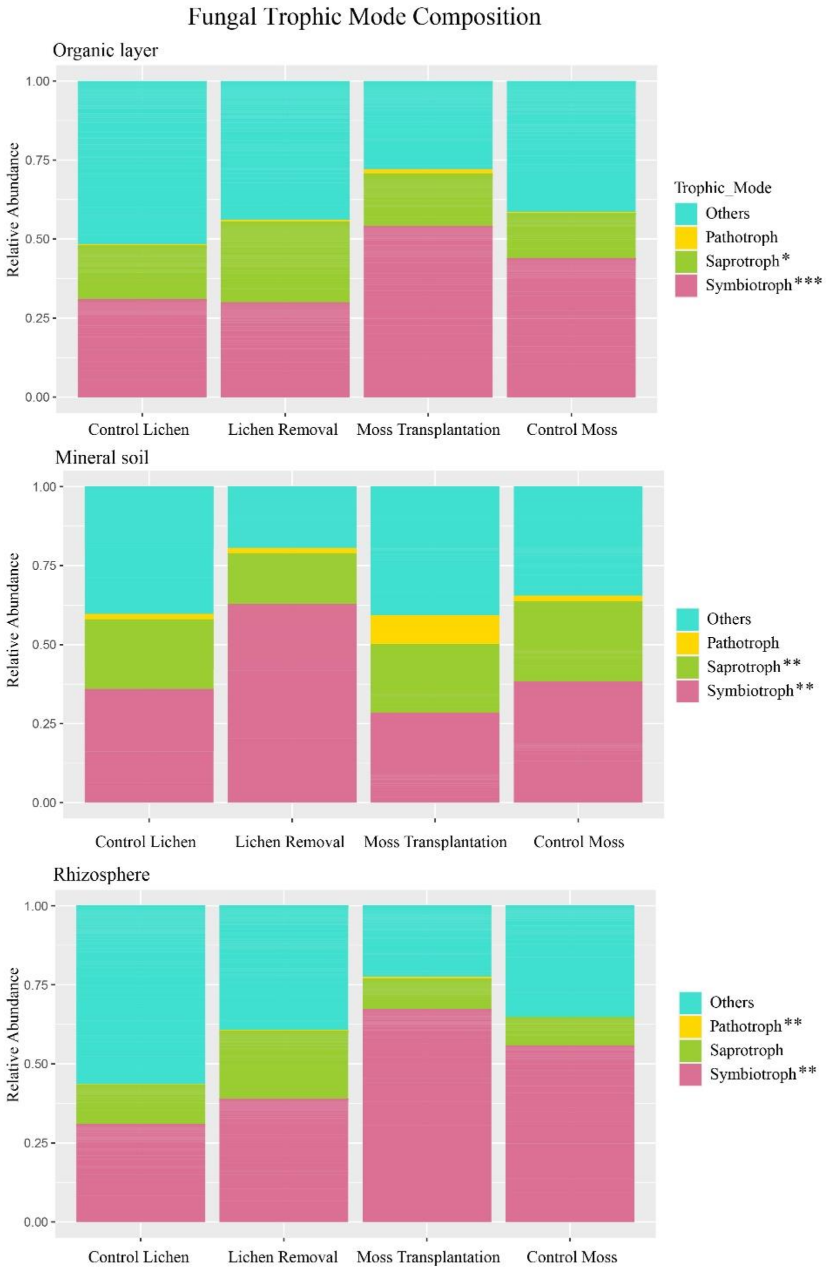

Figure S2.3 Compositions of main fungal functional trophic mode inferred by FUNGuild. ASVs assigned to a guild with the confidence ranking of "Highly probable" and "Probable" were retained for further use, whereas those with the "possible" confidence ranking were classified as 'Others'. Stars indicate significant differences in the relative abundance of a functional guild among forest cover treatments ($p < 0.001$*, $p < 0.01$**, $p < 0.05$*).**

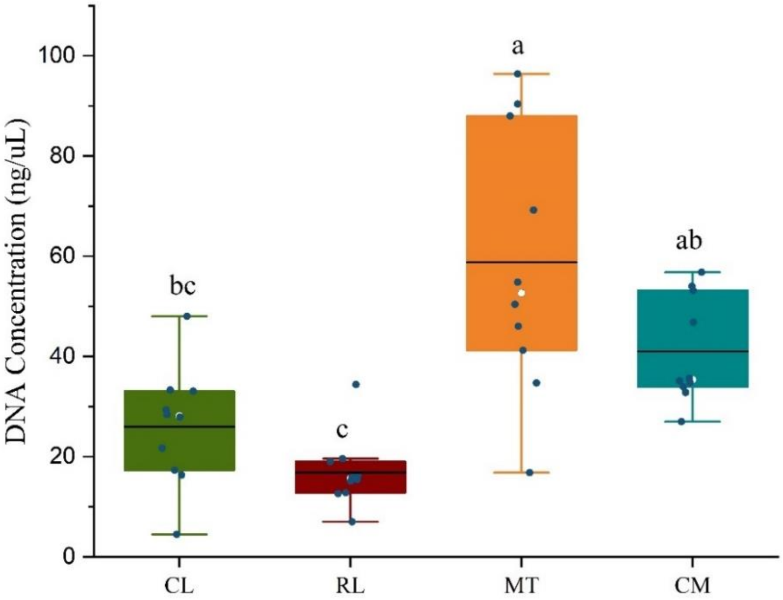

Figure S2.4 DNA concentrations (ng/uL) in humus soils of different forest ground covers. Forest cover treatments dominated by control lichen (CL), lichen removal (RL), moss transplantation (MT) and control moss (CM). Significant differences between ground-cover treatments are represented by different letters. The line represents the mean value. The white circle represents the median.

APPENDICE C - SUPPLEMENTARY MATERIAL FROM CHAPTER 3

Table S3.1 Visual and chemical amounts of charcoal in lichen woodlands and moss forests, respectively.

	Mean	Media	SD	Var	Df
		Chemical-Charcoal (%)			
Lichen woodlands	1.90	1.71	0.736	0.542	11
Moss woodlands	3.33	2.93	1.83	3.33	14
		Visual-Charcoal (mg/g)			
Lichen woodlands	0.595	0.505	0.365	0.133	11
Moss woodlands	2.79	1.36	3.41	11.6	14
		Visual-Charcoal (g/m^2)			
Lichen woodlands	39.5	35.4	23.8	566	11
Moss woodlands	146	68.6	161	25970	14

Table S3.2 Chemical and physical properties of charcoal in lichen woodlands and moss forests, respectively. Lichen woodlands are abbreviated as "L" and feather moss forests are abbreviated as '"FM".

	Char-N		Char-C		Char-H		Char-O		Ash		Surface area	
	L	FM	L	FM	L	FM	L	FM	L	FM	L	FM
Mean	0.43	0.48	55.1	59.9	2.42	2.55	22.4	24.8	19.6	12.2	91.9	87.5
Median	0.49	0.46	56.8	60.2	2.6	2.50	22.7	25.4	17	11.9	94.7	84.0
SD	0.15	0.11	10.5	3.41	0.57	0.41	4.01	2.05	13.6	4.34	22.4	15.4
Var	0.02	0.01	110	11.6	0.33	0.17	16.1	4.20	185	18.9	502	238
Df	7	7	7	7	7	7	7	7	7	7	7	5

LIST OF REFERENCES

Abarenkov K, Henrik Nilsson R, Larsson KH, Alexander IJ, Eberhardt U, Erland S, Høiland K, Kjøller R, Larsson E, Pennanen T (2010) The UNITE database for molecular identification of fungi–recent updates and future perspectives. New Phytologist 186: 281-285. doi: https://www.jstor.org/stable/27797548.

Adamczyk B, Ahvenainen A, Sietiö O-M, Kanerva S, Kieloaho A-J, Smolander A, Kitunen V, Saranpää P, Laakso T, Straková P, Heinonsalo J (2016) The contribution of ericoid plants to soil nitrogen chemistry and organic matter decomposition in boreal forest soil. Soil Biology and Biochemistry 103: 394-404. doi: https://doi.org/10.1016/j.soilbio.2016.09.016.

Agerer R (2001) Exploration types of ectomycorrhizae. Mycorrhiza 11: 107-114. doi: https://doi.org/10.1007/s005720100108.

Arseneault D (2001) Impact of fire behavior on postfire forest development in a homogeneous boreal landscape. Canadian Journal of Forest Research 31: 1367-1374. doi: 10.1139/x01-065.

Augusto L, De Schrijver A, Vesterdal L, Smolander A, Prescott C, Ranger J (2015) Influences of evergreen gymnosperm and deciduous angiosperm tree species on the functioning of temperate and boreal forests. Biological reviews 90: 444-466. doi: https://doi.org/10.1111/brv.12119.

Bahram M, Netherway T, Hildebrand F, Pritsch K, Drenkhan R, Loit K, Anslan S, Bork P, Tedersoo L (2020) Plant nutrient-acquisition strategies drive topsoil microbiome structure and function. New Phytologist 227: 1189-1199. doi: https://doi.org/10.1111/nph.16598.

Balshi MS, McGuire AD, Duffy P, Flannigan M, Kicklighter DW, Melillo J (2009a) Vulnerability of carbon storage in North American boreal forests to wildfires during the 21st century. Global Change Biology 15: 1491-1510. doi: https://doi.org/10.1111/j.1365-2486.2009.01877.x.

Balshi MS, McGuire AD, Duffy P, Flannigan M, Walsh J, Melillo J (2009b) Assessing the response of area burned to changing climate in western boreal North America using a Multivariate Adaptive Regression Splines (MARS) approach. Global Change Biology 15: 578-600. doi: https://doi.org/10.1111/j.1365-2486.2008.01679.x.

Baltzer JL, Day NJ, Walker XJ, Greene D, Mack MC, Alexander HD, Arseneault D, Barnes J, Bergeron Y, Boucher Y, Bourgeau-Chavez L, Brown CD, Carrière S, Howard BK, Gauthier S, Parisien M-A, Reid KA, Rogers BM, Roland C, Sirois L, Stehn S, Thompson DK, Turetsky MR, Veraverbeke S, Whitman E, Yang J, Johnstone JF (2021) Increasing fire and the decline of fire adapted black spruce in the boreal forest. Proceedings of the National Academy of Sciences 118: e2024872118. doi: 10.1073/pnas.2024872118.

Bastianelli C, Ali AA, Beguin J, Bergeron Y, Grondin P, Hély C, Paré D (2017) Boreal coniferous forest density leads to significant variations in soil physical and geochemical properties. Biogeosciences 14: 3445-3459. doi: https://doi.org/10.5194/bg-14-3445-2017.

Bay G, Nahar N, Oubre M, Whitehouse MJ, Wardle DA, Zackrisson O, Nilsson MC, Rasmussen U (2013) Boreal feather mosses secrete chemical signals to gain nitrogen. New Phytologist 200: 54-60. doi: https://doi.org/10.1111/nph.12403.

Beisner BE, Haydon DT, Cuddington K (2003) Alternative stable states in ecology. Frontiers in Ecology and the Environment 1: 376-382. doi: https://doi.org/10.1890/1540-9295(2003)001[0376:ASSIE]2.0.CO;2.

Bélanger N, Pinno BD (2008) Carbon sequestration, vegetation dynamics and soil development in the Boreal Transition ecoregion of Saskatchewan during the Holocene. CATENA 74: 65-72. doi: https://doi.org/10.1016/j.catena.2008.03.005.

Benjamini Y, Hochberg Y (1995) Controlling the False Discovery Rate: A Practical and Powerful Approach to Multiple Testing. Journal of the Royal Statistical Society: Series B (Methodological) 57: 289-300. doi: https://doi.org/10.1111/j.2517-6161.1995.tb02031.x.

Bergeron Y, Cyr D, Girardin MP, Carcaillet C (2010) Will climate change drive 21st century burn rates in Canadian boreal forest outside of its natural variability: collating global climate model experiments with sedimentary charcoal data. International Journal of Wildland Fire 19: 1127-1139. doi: https://doi.org/10.1071/WF09092.

Berner LT, Goetz SJ (2022) Satellite observations document trends consistent with a boreal forest biome shift. Global Change Biology 28: 3275-3292. doi: https://doi.org/10.1111/gcb.16121.

Bieser JMH, Thomas SC (2019) Biochar and high-carbon wood ash effects on soil and vegetation in a boreal clearcut. Canadian Journal of Forest Research 49: 1124-1134. doi: https://doi.org/10.1139/cjfr-2019-0039.

Boiffin J, Munson AD (2013) Three large fire years threaten resilience of closed crown black spruce forests in eastern Canada. Ecosphere 4: art56. doi: https://doi.org/10.1890/ES13-00038.1.

Bolyen E, Rideout JR, Dillon MR, Bokulich NA, Abnet CC, Al-Ghalith GA, Alexander H, Alm EJ, Arumugam M, Asnicar F (2019) Reproducible, interactive, scalable and extensible microbiome data science using QIIME 2. Nature Biotechnology 37: 852-857. doi: https://doi.org/10.1038/s41587-019-0209-9.

Bonan GB, Shugart HH (1989) Environmental factors and ecological processes in boreal forests. Annual review of ecology systematics 1-28. doi: https://doi.org/10.1111/nph.12403.

Bond-Lamberty B, Peckham SD, Ahl DE, Gower ST (2007) Fire as the dominant driver of central Canadian boreal forest carbon balance. Nature 450: 89-92. doi: 10.1038/nature06272.

Bormann BT, Homann PS, Darbyshire RL, Morrissette BA (2008) Intense forest wildfire sharply reduces mineral soil C and N: the first direct evidence. Canadian Journal of Forest Research 38: 2771-2783. doi: https://doi.org/10.1139/X08-136.

Boucher Y, Perrault-Hébert M, Fournier R, Drapeau P, Auger I (2017) Cumulative patterns of logging and fire (1940–2009): consequences on the structure of the eastern Canadian boreal forest. Landscape ecology 32: 361-375. doi: https://doi.org/10.1007/s10980-016-0448-9.

Boudreault C, Bergeron Y, Gauthier S, Drapeau P (2002) Bryophyte and lichen communities in mature to old-growth stands in eastern boreal forests of Canada. Canadian Journal of Forest Research 32: 1080-1093. doi: https://doi.org/10.1139/x02-027.

Boulanger Y, Gauthier S, Burton PJ (2014) A refinement of models projecting future Canadian fire regimes using homogeneous fire regime zones. Canadian Journal of Forest Research 44: 365-376. doi: 10.1139/cjfr-2013-0372.

Boulanger Y, Pascual Puigdevall J (2021) Boreal forests will be more severely affected by projected anthropogenic climate forcing than mixedwood and northern hardwood forests in eastern Canada. Landscape Ecology 36: 1725-1740. doi: https://doi.org/10.1007/s10980-021-01241-7.

Boulanger Y, Puigdevall JP (2021) Boreal forests will be more severely affected by projected anthropogenic climate forcing than mixedwood and northern hardwood forests in eastern Canada. Landscape Ecology 36: 1725-1740. doi: https://doi.org/10.1007/s10980-021-01241-7.

Brady NC, Weil RR, Weil RR (2008) The nature and properties of soils. Prentice Hall Upper Saddle River, NJ.

Brais S, David P, Ouimet R (2000) Impacts of wild fire severity and salvage harvesting on the nutrient balance of jack pine and black spruce boreal stands. Forest Ecology and Management 137: 231-243. doi: https://doi.org/10.1016/S0378-1127(99)00331-X.

Brandt JP, Flannigan MD, Maynard DG, Thompson ID, Volney WJA (2013) An introduction to Canada's boreal zone: ecosystem processes, health, sustainability, and environmental issues. Environmental Reviews 21: 207-226. doi: 10.1139/er-2013-0040.

Brimmer, R.J. Charcoal Quantity and Adsorptive Activity in Ponderosa Pine Ecosystems of Western Montana; the University of Montana: Missoula, MT, USA, 2006. doi: https://scholarworks.umt.edu/etd/6936.

Brockett BFT, Prescott CE, Grayston SJ (2012) Soil moisture is the major factor influencing microbial community structure and enzyme activities across seven biogeoclimatic zones in western Canada. Soil Biology and Biochemistry 44: 9-20. doi: https://doi.org/10.1016/j.soilbio.2011.09.003.

Brooks ME, Kristensen K, Van Benthem KJ, Magnusson A, Berg CW, Nielsen A, Skaug HJ, Machler M, Bolker BM (2017) glmmTMB balances speed and flexibility among packages for zero-inflated generalized linear mixed modeling. The R journal 9: 378-400. doi: https://doi.org/10.3929/ethz-b-000240890.

Brown CD, Johnstone JF (2011) How does increased fire frequency affect carbon loss from fire? A case study in the northern boreal forest. International Journal of Wildland Fire 20: 829-837. doi: https://doi.org/10.1071/WF10113.

Brown CD, Johnstone JF (2012) Once burned, twice shy: Repeat fires reduce seed availability and alter substrate constraints on Picea mariana regeneration. Forest Ecology and Management 266: 34-41. doi: https://doi.org/10.1016/j.foreco.2011.11.006.

Bryanin SV, Makoto K (2017) Fire-derived charcoal affects fine root vitality in a post-fire Gmelin larch forest: field evidence. Plant and soil
416: 409-418. doi: https://doi.org/10.1007/s11104-017-3217-x.

Buma B, Brown CD, Donato DC, Fontaine JB, Johnstone JF (2013) The Impacts of Changing Disturbance Regimes on Serotinous Plant Populations and Communities. BioScience 63: 866-876. doi: 10.1525/bio.2013.63.11.5.

Buma B, Poore RE, Wessman CA (2014) Disturbances, their interactions, and cumulative effects on carbon and charcoal stocks in a forested ecosystem. Ecosystems 17: 947-959.

Bunn AG (2008) A dendrochronology program library in R (dplR). Dendrochronologia 26: 115-124. doi: https://doi.org/10.1016/j.dendro.2008.01.002.

Cabrera M, Beare M (1993) Alkaline persulfate oxidation for determining total nitrogen in microbial biomass extracts. Soil Science Society of America Journal 57: 1007-1012. doi: https://doi.org/10.2136/sssaj1993.03615995005700040021x.

Callahan BJ, McMurdie PJ, Rosen MJ, Han AW, Johnson AJA, Holmes SP (2016) DADA2: High-resolution sample inference from Illumina amplicon data. Nature Methods 13: 581-583. doi: https://doi.org/10.1038/nmeth.3869.

Carter MR, Gregorich EG (2007) Soil sampling and methods of analysis. CRC press.

Certini G (2005) Effects of fire on properties of forest soils: a review. Oecologia 143: 1-10. doi: https://doi.org/10.1007/s00442-004-1788-8.

Cheeke TE, Phillips RP, Brzostek ER, Rosling A, Bever JD, Fransson P (2017) Dominant mycorrhizal association of trees alters carbon and nutrient cycling by selecting for microbial groups with distinct enzyme function. New Phytologist 214: 432-442. doi: https://doi.org/10.1111/nph.14343.

Chen Y, Yang H, Wang X, Zhang S, Chen H (2012) Biomass-based pyrolytic polygeneration system on cotton stalk pyrolysis: influence of temperature. Bioresource Technology 107: 411-418. doi: https://doi.org/10.1016/j.biortech.2011.10.074.

Clemmensen K, Bahr A, Ovaskainen O, Dahlberg A, Ekblad A, Wallander H, Stenlid J, Finlay R, Wardle D, Lindahl B (2013) Roots and associated fungi drive long-term carbon sequestration in boreal forest. Science 339: 1615-1618. doi: 10.1126/science.1231923.

Clemmensen KE, Finlay RD, Dahlberg A, Stenlid J, Wardle DA, Lindahl BD (2015) Carbon sequestration is related to mycorrhizal fungal community shifts during long-term succession in boreal forests. New Phytologist 205: 1525-1536. doi: https://doi.org/10.1111/nph.13208.

Connell JH, Sousa WP (1983) On the Evidence Needed to Judge Ecological Stability or Persistence. The American Naturalist 121: 789-824. doi: 10.1086/284105.

Cornelissen J, Lavorel S, Garnier E, Díaz S, Buchmann N, Gurvich D, Reich P, Ter Steege H, Morgan H, Van Der Heijden M (2003) A handbook of protocols for standardised and easy measurement of plant functional traits worldwide. Australian Journal of Botany 51: 335-380. doi: https://doi.org/10.1071/BT02124.

Côté D, Girard F, Hébert F, Bouchard S, Gagnon R, Lord D (2013) Is the closed-crown boreal forest resilient after successive stand disturbances? A quantitative demonstration from a case study. Journal of Vegetation Science 24: 664-674. doi: https://doi.org/10.1111/j.1654-1103.2012.01488.x.

Dai Z, Su W, Chen H, Barberán A, Zhao H, Yu M, Yu L, Brookes PC, Schadt CW, Chang SX, Xu

J (2018) Long-term nitrogen fertilization decreases bacterial diversity and favors the growth of Actinobacteria and Proteobacteria in agro-ecosystems across the globe. Global Change Biology 24: 3452-3461. doi: https://doi.org/10.1111/gcb.14163.

Davis KT, Dobrowski SZ, Higuera PE, Holden ZA, Veblen TT, Rother MT, Parks SA, Sala A, Maneta MP (2019) Wildfires and climate change push low-elevation forests across a critical climate threshold for tree regeneration. Proceedings of the National Academy of Sciences 116: 6193-6198. doi: https://doi.org/10.1073/pnas.1815107116.

Davis KT, Robles MD, Kemp KB, Higuera PE, Chapman T, Metlen KL, Peeler JL, Rodman KC, Woolley T, Addington RN, Buma BJ, Cansler CA, Case MJ, Collins BM, Coop JD, Dobrowski SZ, Gill NS, Haffey C, Harris LB, Harvey BJ, Haugo RD, Hurteau MD, Kulakowski D, Littlefield CE, McCauley LA, Povak N, Shive KL, Smith E, Stevens JT, Stevens-Rumann CS, Taylor AH, Tepley AJ, Young DJN, Andrus RA, Battaglia MA, Berkey JK, Busby SU, Carlson AR, Chambers ME, Dodson EK, Donato DC, Downing WM, Fornwalt PJ, Halofsky JS, Hoffman A, Holz A, Iniguez JM, Krawchuk MA, Kreider MR, Larson AJ, Meigs GW, Roccaforte JP, Rother MT, Safford H, Schaedel M, Sibold JS, Singleton MP, Turner MG, Urza AK, Clark-Wolf KD, Yocom L, Fontaine JB, Campbell JL (2023) Reduced fire severity offers near-term buffer to climate-driven declines in conifer resilience across the western United States. Proceedings of the National Academy of Sciences 120: e2208120120. doi: 10.1073/pnas.2208120120.

Delgado-Baquerizo M, Reich PB, Khachane AN, Campbell CD, Thomas N, Freitag TE, Abu Al-Soud W, Sørensen S, Bardgett RD, Singh BK (2017) It is elemental: soil nutrient stoichiometry drives bacterial diversity. Environmental Microbiology 19: 1176-1188. doi: https://doi.org/10.1111/1462-2920.13642.

DeLuca T, Zackrisson O, Bergman I, Hörnberg G (2013) Historical land use and resource depletion in spruce-*Cladina* forests of subarctic Sweden. Anthropocene 1: 14-22. doi: https://doi.org/10.1016/j.ancene.2013.03.002.

DeLuca TH, Aplet GH (2008) Charcoal and carbon storage in forest soils of the Rocky Mountain West. Frontiers in Ecology and the Environment 6: 18-24.

DeLuca TH, Gundale MJ, Brimmer RJ, Gao S (2020) Pyrogenic carbon generation from fire and forest restoration treatments. Frontiers in Forests Global Change 3: 24. doi: https://doi.org/10.3389/ffgc.2020.00024.

DeLuca TH, MacKenzie MD, Gundale MJ, Lehmann J, Joseph S (2009) Biochar for environmental management: Science and technology. Lehmann J, Joseph S Biochar Effects on Soil Nutrient Transformations London: Earthscan: 251-270.

DeLuca TH, Zackrisson O, Gundale MJ, Nilsson M-C (2008) Ecosystem feedbacks and nitrogen fixation in boreal forests. Science 320: 1181-1181. doi: https://doi.org/10.1126/science.1154836.

DeLuca TH, Zackrisson O, Nilsson M-C, Sellstedt A (2002) Quantifying nitrogen-fixation in feather moss carpets of boreal forests. Nature 419: 917-920. doi: https://doi.org/10.1038/nature01051.

DeLuca TH, Zackrisson O, Nilsson M-C, Sun S, Arróniz-Crespo M (2022) Long-term fate of nitrogen fixation in *Pleurozium schreberi* Brid (Mit.) moss carpets in boreal forests. Applied Soil Ecology 169: 104215. doi: https://doi.org/10.1016/j.apsoil.2021.104215.

Demirbas A (2004) Effects of temperature and particle size on bio-char yield from pyrolysis of agricultural residues. Journal of Analytical and Applied Pyrolysis 72: 243-248. doi: https://doi.org/10.1016/j.jaap.2004.07.003.

Duncan RP (1989) An evaluation of errors in tree age estimates based on increment cores in kahikatea (Dacrycarpus dacrydioides). New Zealand Natural Sciences 16: 31-37.

Egli M, Mirabella A, Sartori G, Zanelli R, Bischof S (2006) Effect of north and south exposure on weathering rates and clay mineral formation in Alpine soils. Catena 67: 155-174. doi: https://doi.org/10.1016/j.catena.2006.02.010.

El-Naggar A, El-Naggar AH, Shaheen SM, Sarkar B, Chang SX, Tsang DCW, Rinklebe J, Ok YS (2019) Biochar composition-dependent impacts on soil nutrient release, carbon mineralization, and potential environmental risk: A review. Journal of Environmental Management 241: 458-467. doi: https://doi.org/10.1016/j.jenvman.2019.02.044.

Ellis TM, Bowman DMJS, Jain P, Flannigan MD, Williamson GJ (2022) Global increase in wildfire risk due to climate-driven declines in fuel moisture. Global Change Biology 28: 1544-1559. doi: https://doi.org/10.1111/gcb.16006.

Fanin N, Clemmensen KE, Lindahl BD, Farrell M, Nilsson M-C, Gundale MJ, Kardol P, Wardle DA (2022) Ericoid shrubs shape fungal communities and suppress organic matter decomposition in boreal forests. New Phytologist 236: 684-697. doi: https://doi.org/10.1111/nph.18353.

Fauria MFM, Helle T, Niva A, Posio H, Timonen M (2008) Removal of the lichen mat by reindeer enhances tree growth in a northern Scots pine forest. Canadian Journal of Forest Research 38: 2981-2993. doi: https://doi.org/10.1139/X08-135.

Fierer N, Bradford MA, Jackson RB (2007) Toward an ecological classification of soil bacteria. Ecology 88: 1354-1364. doi: https://doi.org/10.1890/05-1839.

Fierer N, Schimel J, Holden P (2003) Influence of drying-rewetting frequency on soil bacterial community structure. Microbial Ecology: 63-71. doi: https://doi.org/10.1007/s00248-002-1007-2.

Forbes M, Raison R, Skjemstad J (2006) Formation, transformation and transport of black carbon (charcoal) in terrestrial and aquatic ecosystems. Science of the Total Environment 370: 190-206. doi: https://doi.org/10.1016/j.scitotenv.2006.06.007.

Frey SD, Drijber R, Smith H, Melillo J (2008) Microbial biomass, functional capacity, and community structure after 12 years of soil warming. Soil Biology and Biochemistry 40: 2904-2907. doi: https://doi.org/10.1016/j.soilbio.2008.07.020.

Gale NV, Thomas SC (2021) Spatial heterogeneity in soil pyrogenic carbon mediates tree growth and physiology following wildfire. Journal of Ecology 109: 1479-1490. doi: https://doi.org/10.1111/1365-2745.13571.

Gao L, Paré D, Chavardès RD, Bergeron Y (2023) Initiating the transition from open-canopy lichen woodland to productive forest by transplanting moss, results from a 10-year experiment. Plant and Soil 488: 363–376. doi: https://doi.org/10.1007/s11104-023-05977-

w.

Girard F, Payette S, Gagnon R (2008) Rapid expansion of lichen woodlands within the closed-crown boreal forest zone over the last 50 years caused by stand disturbances in eastern Canada. Journal of Biogeography 35: 529-537. doi: https://doi.org/10.1111/j.1365-2699.2007.01816.x.

Glaser B, Haumaier L, Guggenberger G, Zech W (2001) The 'Terra Preta' phenomenon: a model for sustainable agriculture in the humid tropics. Naturwissenschaften 88: 37-41. doi: 10.1007/s001140000193.

Gómez-Acata ES, Valencia-Becerril I, Valenzuela-Encinas C, Velásquez-Rodríguez AS, Navarro-Noya YE, Montoya-Ciriaco N, Suárez-Arriaga MC, Rojas-Valdez A, Reyes-Reyes BG, Luna-Guido M, Dendooven L (2016) Deforestation and Cultivation with Maize (Zea mays L.) has a Profound Effect on the Bacterial Community Structure in Soil. Land Degradation & Development 27: 1122-1130. doi: https://doi.org/10.1002/ldr.2328.

Gordon SJ (2005) Effect of environmental factors on the chemical weathering of plagioclase in Hawaiian basalt. Physical Geography 26: 69-84. doi: https://doi.org/10.2747/0272-3646.26.1.69.

Gornall JL, Woodin SJ, Jónsdóttir IS, van der Wal R (2011) Balancing positive and negative plant interactions: how mosses structure vascular plant communities. Oecologia 166: 769-782. doi: https://doi.org/10.1007/s00442-011-1911-6.

Greene DF, Noël J, Bergeron Y, Rousseau M, Gauthier S (2004) Recruitment of *Picea mariana*, *Pinus banksiana*, and *Populus tremuloides* across a burn severity gradient following wildfire in the southern boreal forest of Quebec. Canadian Journal of Forest Research 34: 1845-1857. doi: 10.1139/x04-059.

Group SCW (1998) The Canadian system of soil classification. Agriculture agri-food Canada publication 1646: 187.

Gundale MJ, DeLuca TH (2006) Temperature and source material influence ecological attributes of ponderosa pine and Douglas-fir charcoal. Forest Ecology and Management 231: 86-93.

Gundale MJ, Nilsson M, Bansal S, Jäderlund A (2012) The interactive effects of temperature and light on biological nitrogen fixation in boreal forests. New Phytologist 194: 453-463. doi: https://doi.org/10.1111/j.1469-8137.2012.04071.x.

Hart S, Luckai N (2013) Charcoal function and management in boreal ecosystems. Journal of Applied Ecology 50: 1197-1206.

Hart, S. C., Stark, J. M., Davidson, E. A., & Firestone, M. K. (1994). Nitrogen Mineralization, Immobilization, and Nitrification. In R. V. Weaver et al. (Eds.), Methods of Soil Analysis, Part 2, SSA Book Series 5 (pp. 985-1018). Madison, WI: SSSA. https://doiorg/102136/sssabookser52c42.

Haughian SR, Burton PJ (2015) Microhabitat associations of lichens, feathermosses, and vascular plants in a caribou winter range, and their implications for understory development. Botany 93: 221-231. doi: https://doi.org/10.1139/cjb-2014-0238.

Hawkes CV, Menges ES (2003) Effects of lichens on seedling emergence in a xeric Florida shrubland. Southeastern Naturalist 2: 223-234. doi: https://doi.org/10.1656/1528-

7092(2003)002[0223:EOLOSE]2.0.CO;2.

Heinonsalo J, Sun H, Santalahti M, Bäcklund K, Hari P, Pumpanen J (2015) Evidences on the ability of mycorrhizal genus Piloderma to use organic nitrogen and deliver it to Scots pine. PLoS One 10: e0131561. doi: https://doi.org/10.1371/journal.pone.0131561.

Héon J, Arseneault D, Parisien M-A (2014) Resistance of the boreal forest to high burn rates. Proceedings of the National Academy of Sciences 111: 13888-13893. doi: 10.1073/pnas.1409316111.

Hervé M (2014) RVAideMemoire: Diverse Basic Statistical and Graphical Functions, Version 0.9-63.

Ho A, Di Lonardo DP, Bodelier PL (2017) Revisiting life strategy concepts in environmental microbial ecology. FEMS microbiology ecology 93. doi: https://doi.org/10.1093/femsec/fix006.

Holmes RL (1983) Computer-assisted quality control in tree-ring dating and measurement. Tree-Ring Bulletin 43: 51-67.

Houle D, Lajoie G, Duchesne L (2016) Major losses of nutrients following a severe drought in a boreal forest. Nature Plants 2: 16187. doi: https://doi.org/10.1038/nplants.2016.187.

Ilek A, Kucza J, Szostek M (2015) The effect of stand species composition on water storage capacity of the organic layers of forest soils. European Journal of Forest Research 134: 187-197. doi: https://doi.org/10.1007/s10342-014-0842-2.

Ilisson T, Chen HYH (2009) The direct regeneration hypothesis in northern forests. Journal of Vegetation Science 20: 735-744. doi: https://doi.org/10.1111/j.1654-1103.2009.01066.x.

Illumina IJPSRRGAftIMS (2013) 16S Metagenomic sequencing library preparation. : 1-28.

Imo M, Timmer VR (1998) Vector competition analysis: a new approach for evaluating vegetation control methods in young black spruce plantations. Canadian Journal of Soil Science 78: 3-15. doi: https://doi.org/10.4141/S97-020.

Jasinski JPP, Payette S (2005) The creation of alternative stable states in the southern boreal forest, Quebec, Canada. Ecological Monographs 75: 561-583. doi: https://doi.org/10.1890/04-1621.

Jean M, Fenton NJ, Bergeron Y, Nilsson M-C (2021) *Sphagnum* and feather moss-associated N_2 fixation along a 724-year chronosequence in eastern boreal Canada. Plant Ecology 222: 1007-1022. doi: https://doi.org/10.1007/s11258-021-01157-x.

Jiang S, Xing Y, Liu G, Hu C, Wang X, Yan G, Wang Q (2021) Changes in soil bacterial and fungal community composition and functional groups during the succession of boreal forests. Soil Biology Biochemistry 161: 108393. doi: https://doi.org/10.1016/j.soilbio.2021.108393.

Johnstone JF, Chapin FS (2006) Fire Interval Effects on Successional Trajectory in Boreal Forests of Northwest Canada. Ecosystems 9: 268-277. doi: 10.1007/s10021-005-0061-2.

Johnstone JF, Chapin FS, Hollingsworth TN, Mack MC, Romanovsky V, Turetsky MJCJoFR (2010a) Fire, climate change, and forest resilience in interior Alaska. 40: 1302-1312.

Johnstone JF, Hollingsworth TN, Chapin Iii FS, Mack MC (2010b) Changes in fire regime break the legacy lock on successional trajectories in Alaskan boreal forest. Global Change

Biology 16: 1281-1295. doi: https://doi.org/10.1111/j.1365-2486.2009.02051.x.

Jones MW, Santín C, van der Werf GR, Doerr SH (2019) Global fire emissions buffered by the production of pyrogenic carbon. Nature Geoscience 12: 742-747. doi: 10.1038/s41561-019-0403-x.

Kalra, Y.P. Hand Book of Reference Methods for Plant Analysis; CRC Press: Boca Raton, FL, USA, 1997; pp. 75–92.

Kang S, Kimball JS, Running SW (2006) Simulating effects of fire disturbance and climate change on boreal forest productivity and evapotranspiration. Science of The Total Environment 362: 85-102. doi: https://doi.org/10.1016/j.scitotenv.2005.11.014.

Kasin I, Ellingsen VM, Asplund J, Ohlson M (2016) Spatial and temporal dynamics of the soil charcoal pool in relation to fire history in a boreal forest landscape. Canadian Journal of Forest Research 47: 28-35. doi: 10.1139/cjfr-2016-0233.

Kasischke ES, Turetsky MR (2006) Recent changes in the fire regime across the North American boreal region—Spatial and temporal patterns of burning across Canada and Alaska. Geophysical Research Letters 33. doi: https://doi.org/10.1029/2006GL025677.

Keiluweit M, Nico PS, Johnson MG, Kleber M (2010) Dynamic molecular structure of plant biomass-derived black carbon (biochar). Environmental Science & Technology 44: 1247-1253. doi: https://doi.org/10.1021/es9031419.

Kim HM, Jung JY, Yergeau E, Hwang CY, Hinzman L, Nam S, Hong SG, Kim O-S, Chun J, Lee YK (2014) Bacterial community structure and soil properties of a subarctic tundra soil in Council, Alaska. FEMS Microbiology Ecology 89: 465-475. doi: https://doi.org/10.1111/1574-6941.12362.

Kim KH, Kim J-Y, Cho T-S, Choi JW (2012) Influence of pyrolysis temperature on physicochemical properties of biochar obtained from the fast pyrolysis of pitch pine (Pinus rigida). Bioresource Technology 118: 158-162. doi: https://doi.org/10.1016/j.biortech.2012.04.094.

Klatt CG, Liu Z, Ludwig M, Kühl M, Jensen SI, Bryant DA, Ward DM (2013) Temporal metatranscriptomic patterning in phototrophic Chloroflexi inhabiting a microbial mat in a geothermal spring. The ISME Journal 7: 1775-1789. doi: https://doi.org/10.1038/ismej.2013.52.

Klaubauf S, Inselsbacher E, Zechmeister-Boltenstern S, Wanek W, Gottsberger R, Strauss J, Gorfer M (2010) Molecular diversity of fungal communities in agricultural soils from Lower Austria. Fungal Diversity 44: 65-75. doi: https://doi.org/10.1007/s13225-010-0053-1.

Knicker H, Almendros G, González-Vila FJ, González-Pérez JA, Polvillo O (2006) Characteristic alterations of quantity and quality of soil organic matter caused by forest fires in continental Mediterranean ecosystems: a solid-state 13C NMR study. European Journal of Soil Science 57: 558-569. doi: https://doi.org/10.1111/j.1365-2389.2006.00814.x.

Kolaříková Z, Kohout P, Krüger C, Janoušková M, Mrnka L, Rydlová J (2017) Root-associated fungal communities along a primary succession on a mine spoil: Distinct ecological guilds assemble differently. Soil Biology and Biochemistry 113: 143-152. doi:

https://doi.org/10.1016/j.soilbio.2017.06.004.

Koyama A, Wallenstein MD, Simpson RT, Moore JC (2014) Soil bacterial community composition altered by increased nutrient availability in Arctic tundra soils. Frontiers in Microbiology 5: 516. doi: https://doi.org/10.3389/fmicb.2014.00516.

Kranabetter JM, McKeown K, Hawkins B (2021) Post-disturbance conifer tree-ring δ15N reflects openness of the nitrogen cycle across temperate coastal rainforests. Journal of Ecology 109: 342-353. doi: https://doi.org/10.1111/1365-2745.13482.

Kurth V, MacKenzie M, DeLuca T (2006) Estimating charcoal content in forest mineral soils. Geoderma 137: 135-139. doi: https://doi.org/10.1016/j.geoderma.2006.08.003.

Kyaschenko J, Clemmensen KE, Hagenbo A, Karltun E, Lindahl BD (2017) Shift in fungal communities and associated enzyme activities along an age gradient of managed Pinus sylvestris stands. The ISME Journal 11: 863-874. doi: https://doi.org/10.1038/ismej.2016.184.

Kytöviita M-M, Stark S (2009) No allelopathic effect of the dominant forest-floor lichen *Cladonia stellaris* on pine seedlings. Functional Ecology: 435-441. doi: https://doi:10.1111/j.1365-2435.2008.01508.x.

Landuyt D, De Lombaerde E, Perring MP, Hertzog LR, Ampoorter E, Maes SL, De Frenne P, Ma S, Proesmans W, Blondeel H, Sercu BK, Wang B, Wasof S, Verheyen K (2019) The functional role of temperate forest understorey vegetation in a changing world. Global Change Biology 25: 3625-3641. doi: https://doi.org/10.1111/gcb.14756.

Larsson L (2020) CDendro package version 9.6. Available online: http://www.cybis.se/forfun/dendro/index. Accessed 10 Aug 2022

Lauber Christian L, Hamady M, Knight R, Fierer N (2009) Pyrosequencing-Based Assessment of Soil pH as a Predictor of Soil Bacterial Community Structure at the Continental Scale. Applied and Environmental Microbiology 75: 5111-5120. doi: https://doi.org/10.1128/AEM.00335-09.

Lavoie M, Paré D, Bergeron Y (2006) Relationships between microsite type and the growth and nutrition of young black spruce on post-disturbed lowland black spruce sites in eastern Canada. Canadian Journal of Forest Research 37: 62-73. doi: https://doi.org/10.1139/x06-196.

Lee SA, Kim JM, Kim Y, Joa J-H, Kang S-S, Ahn J-H, Kim M, Song J, Weon H-Y (2020) Different types of agricultural land use drive distinct soil bacterial communities. Scientific Reports 10: 17418. doi: https://doi.org/10.1038/s41598-020-74193-8.

Lehmann, J., Gaunt, J. & Rondon, M (2006). Bio-char sequestration in terrestrial ecosystems – a review. Mitig Adapt Strat Glob Change 11, 403–427. https://doi.org/10.1007/s11027-005-9006-5

Lehmann J, Rillig MC, Thies J, Masiello CA, Hockaday WC, Crowley D (2011) Biochar effects on soil biota–a review. Soil biology and biochemistry 43: 1812-1836. doi: https://doi.org/10.1016/j.soilbio.2011.04.022.

Li L, Long A, Fossum B, Kaiser M (2023) Effects of pyrolysis temperature and feedstock type on biochar characteristics pertinent to soil carbon and soil health: A meta-analysis. Soil Use and Management 39: 43-52. doi: https://doi.org/10.1111/sum.12848.

Liang B, Lehmann J, Solomon D, Kinyangi J, Grossman J, O'Neill B, Skjemstad JO, Thies J, Luizão FJ, Petersen J (2006) Black carbon increases cation exchange capacity in soils. Soil Science Society of America Journal 70: 1719-1730. doi: https://doi.org/10.2136/sssaj2005.0383.

Licht J, Smith NG (2020) Pyrogenic carbon increases pitch pine seedling growth, soil moisture retention, and photosynthetic intrinsic water use efficiency in the field. Frontiers in Forests and Global Change 3: 31. doi: https://doi.org/10.3389/ffgc.2020.00031.

Lilleskov EA, Hobbie EA, Horton TR (2011) Conservation of ectomycorrhizal fungi: exploring the linkages between functional and taxonomic responses to anthropogenic N deposition. Fungal Ecology 4: 174-183. doi: https://doi.org/10.1016/j.funeco.2010.09.008.

Lindahl BD, Tunlid A (2015) Ectomycorrhizal fungi – potential organic matter decomposers, yet not saprotrophs. New Phytologist 205: 1443-1447. doi: https://doi.org/10.1111/nph.13201.

Liu T, Wu X, Li H, Alharbi H, Wang J, Dang P, Chen X, Kuzyakov Y, Yan W (2020) Soil organic matter, nitrogen and pH driven change in bacterial community following forest conversion. Forest Ecology and Management 477: 118473. doi: https://doi.org/10.1016/j.foreco.2020.118473.

Lõhmus K, Truu, J., Truu, M., Kaar, E., Ostonen, I., Alama, S., Kuznetsova, T., Rosenvald, K., Vares, A., Uri, V., & Mander, Ü. (2006) Black alder as a promising deciduous species for the reclaiming of oil shale mining areas. In C. A. Brebbia & Ü. Mander (Eds.), WIT transactions on ecology and the environment, Brownfield sites iii: Prevention, assessment, rehabilitation and development of brownfield sites. Wit Press: Southampton, England, 2006; pp. 87–97.

Ma S, Chen X, Su H, Xing A, Chen G, Zhu J, Zhu B, Fang J (2022) Phosphorus addition decreases soil fungal richness and alters fungal guilds in two tropical forests. Soil Biology and Biochemistry 175: 108836. doi: https://doi.org/10.1016/j.soilbio.2022.108836.

Macias Fauria M, Helle T, Niva A, Posio H, Timonen M (2008) Removal of the lichen mat by reindeer enhances tree growth in a northern Scots pine forest. Canadian Journal of Forest Research 38: 2981-2993. doi: 10.1139/X08-135.

Makoto K, Koike T (2021) Charcoal ecology: Its function as a hub for plant succession and soil nutrient cycling in boreal forests. Ecological Research 36: 4-12. doi: https://doi.org/10.1111/1440-1703.12179.

Makoto K, Shibata H, Kim Y, Satomura T, Takagi K, Nomura M, Satoh F, Koike T (2012) Contribution of charcoal to short-term nutrient dynamics after surface fire in the humus layer of a dwarf bamboo-dominated forest. Biology and Fertility of Soils 48: 569-577. doi: https://doi.org/10.1007/s00374-011-0657-y.

Makoto K, Tamai Y, Kim Y, Koike T (2010) Buried charcoal layer and ectomycorrhizae cooperatively promote the growth of Larix gmelinii seedlings. Plant and Soil 327: 143-152. doi: https://doi.org/10.1007/s11104-009-0040-z.

Mallik A, Kayes I (2018) Lichen mated seedbeds inhibit while moss dominated seedbeds facilitate black spruce (*Picea mariana*) seedling regeneration in post-fire boreal forest. Forest Ecology and Management 427: 260-274. doi: https://doi.org/10.1016/j.foreco.2018.05.064.

Mansuy N, Gauthier S, Bergeron Y (2013) Afforestation opportunities when stand productivity is driven by a high risk of natural disturbance: a review of the open lichen woodland in the eastern boreal forest of Canada. Mitigation and Adaptation Strategies for Global Change 18: 245-264. doi: https://doi.org/10.1007/s11027-012-9362-x.

Martin M (2011) Cutadapt removes adapter sequences from high-throughput sequencing reads. EMBnet journal 17: 10-12. doi: https://doi.org/10.14806/ej.17.1.200.

Maynard D, Paré D, Thiffault E, Lafleur B, Hogg K, Kishchuk B (2014) How do natural disturbances and human activities affect soils and tree nutrition and growth in the Canadian boreal forest? Environmental Reviews 22: 161-178. doi: https://doi.org/10.1139/er-2013-0057.

McCormack ML, Iversen CM (2019) Physical and Functional Constraints on Viable Belowground Acquisition Strategies. Frontiers in Plant Science 10. doi: https://doi.org/10.3389/fpls.2019.01215.

McGonigle T, Grant C (2015) Variation in potassium and calcium uptake with time and root depth. Canadian Journal of Plant Science 95: 771-777. doi: https://doi.org/10.4141/cjps-2014-227.

McMurdie PJ, Holmes S (2013) phyloseq: an R package for reproducible interactive analysis and graphics of microbiome census data. PloS One 8: e61217. doi: https://doi.org/10.1371/journal.pone.0061217.

Heydari M, Salehi A, Mahdavi A, Adibnejad M (2012) Effects of different fire severity levels on soil chemical and physical properties in Zagros forests of western Iran. Folia Forestalia Polonica 54:241–250.

Mekonnen ZA, Riley WJ, Randerson JT, Grant RF, Rogers BM (2019) Expansion of high-latitude deciduous forests driven by interactions between climate warming and fire. Nature Plants 5: 952-958. doi: 10.1038/s41477-019-0495-8.

Mengel K, Kirkby E (2001) Principles of plant nutrition., 5th edn (Kluwer Academic Publishers: Dordrecht, The Netherlands). pp. 849. doi:10.1093/aob/mch063

Menkis A, Burokienė D, Gaitnieks T, Uotila A, Johannesson H, Rosling A, Finlay RD, Stenlid J, Vasaitis R (2012) Occurrence and impact of the root-rot biocontrol agent *Phlebiopsis gigantea* on soil fungal communities in *Picea abies* forests of northern Europe. FEMS Microbiology Ecology 81: 438-445. doi: https://doi.org/10.1111/j.1574-6941.2012.01366.x.

Merilä P, Malmivaara-Lämsä M, Spetz P, Stark S, Vierikko K, Derome J, Fritze H (2010) Soil organic matter quality as a link between microbial community structure and vegetation composition along a successional gradient in a boreal forest. Applied Soil Ecology 46: 259-267. doi: https://doi.org/10.1016/j.apsoil.2010.08.003.

MFFP (2012) Ministère des Forêts, de la Faune et des Parcs (MFFP). Data from: Insectes, maladies et feux dans les forêts québécoises. In: dlFedP Ministère des Forêts, Direction des Inventaires Forestiers, Québec, Canada. https://mffp.gouv.qc.ca/documents/forets/fimaq/bilan2012-g.pdf (ed). pp. 1-66. Accessed 15 Aug 2022

MFFP. (2022) Ministère des Forêts, de la Faune et des Parcs (MFFP). Data from: Cartographie du 5e inventaire écoforestier du Québec méridional – Méthodes et données associées. . In: dlFedP Ministère des Forêts, Direction des Inventaires Forestiers, Québec, Canada. https://www.donneesquebec.ca/recherche/fr/dataset/resultats-d-inventaire-et-carte-ecoforestiere/resource/1ea8bc6b-18e9-4676-8aba-c1f3edbcbc0e (ed). pp. 2-91. Accessed 15 Aug 2022

Morris EK, Caruso T, Buscot F, Fischer M, Hancock C, Maier TS, Meiners T, Müller C, Obermaier E, Prati D (2014) Choosing and using diversity indices: insights for ecological applications from the German Biodiversity Exploratories. Ecology and Evolution 4: 3514-3524. doi: https://doi.org/10.1002/ece3.1155.

Mukherjee A, Zimmerman AR, Harris W (2011) Surface chemistry variations among a series of laboratory-produced biochars. Geoderma 163: 247-255. doi: https://doi.org/10.1016/j.geoderma.2011.04.021.

Mundra S, Kauserud H, Økland T, Nordbakken J-F, Ransedokken Y, Kjønaas OJ (2022) Shift in tree species changes the belowground biota of boreal forests. New Phytologist 234: 2073-2087. doi: https://doi.org/10.1111/nph.18109.

Nagati M, Roy M, Desrochers A, Manzi S, Bergeron Y, Gardes M (2019) Facilitation of balsam fir by trembling aspen in the boreal forest: do ectomycorrhizal communities matter? Frontiers in Plant Science 10: 932. doi: https://doi.org/10.3389/fpls.2019.00932.

Nagati M, Roy M, Manzi S, Richard F, Desrochers A, Gardes M, Bergeron Y (2018) Impact of local forest composition on soil fungal communities in a mixed boreal forest. Plant and Soil 432: 345-357. doi: https://doi.org/10.1007/s11104-018-3806-3.

Newton P, Amponsah I (2006) Systematic review of short-term growth responses of semi-mature black spruce and jack pine stands to nitrogen-based fertilization treatments. Forest Ecology Management 237: 1-14. doi: https://doi.org/10.1016/j.foreco.2006.10.009.

Nguyen NH, Song Z, Bates ST, Branco S, Tedersoo L, Menke J, Schilling JS, Kennedy PG (2016) FUNGuild: an open annotation tool for parsing fungal community datasets by ecological guild. Fungal Ecology 20: 241-248. doi: https://doi.org/10.1016/j.funeco.2015.06.006.

Nicolás C, Martin-Bertelsen T, Floudas D, Bentzer J, Smits M, Johansson T, Troein C, Persson P, Tunlid A (2019) The soil organic matter decomposition mechanisms in ectomycorrhizal fungi are tuned for liberating soil organic nitrogen. The ISME Journal 13: 977-988. doi: https://doi.org/10.1038/s41396-018-0331-6.

Nilsson RH, Larsson K-H, Taylor AFS, Bengtsson-Palme J, Jeppesen TS, Schigel D, Kennedy P, Picard K, Glöckner FO, Tedersoo L (2019) The UNITE database for molecular identification of fungi: handling dark taxa and parallel taxonomic classifications. Nucleic Acids Research 47: D259-D264. doi: https://doi.org/10.1093/nar/gky1022.

Ohlson M, Dahlberg B, Økland T, Brown KJ, Halvorsen R (2009) The charcoal carbon pool in boreal forest soils. Nature Geoscience 2: 692-695.

Ohlson M, Ellingsen VM, del Olmo MV, Lie MH, Nybakken L, Asplund J (2016) Late-Holocene fire history as revealed by size, age and composition of the soil charcoal pool in neighbouring beech and spruce forest landscapes in SE Norway. The Holocene 27: 397-403. doi: 10.1177/0959683616660174.

Ohlson M, Kasin I, Wist AN, Bjune AE (2013) Size and spatial structure of the soil and lacustrine charcoal pool across a boreal forest watershed. Quaternary Research 80: 417-424. doi: https://doi.org/10.1016/j.yqres.2013.08.009.

Oksanen J, Blanchet FG, Kindt R, Legendre P, Minchin PR, O'hara RB, Simpson GL, Solymos P, Stevens MHH, Wagner H (2013) Vegan: community ecology package version 2.0-10. Journal of Statistical Software 48: 103-132.

Ortiz-Rivero J, Garrido-Benavent I, Heiðmarsson S, de los Ríos A (2023) Moss and Liverwort Covers Structure Soil Bacterial and Fungal Communities Differently in the Icelandic Highlands. Microbial Ecology. doi: https://doi.org/10.1007/s00248-023-02194-x.

Ouimet R, Boucher J-F, Tremblay P, Lord D (2018) Comparing soil profiles of adjacent forest stands with contrasting tree densities: lichen woodlands vs. black spruce–feathermoss stands in the continuous boreal forest. Canadian Journal of Soil Science 98: 458-468. doi: https://doi.org/10.1139/cjss-2018-0017.

Pacé M, Fenton NJ, Paré D, Bergeron Y (2016) Ground-layer composition affects tree fine root biomass and soil nutrient availability in jack pine and black spruce forests under extreme drainage conditions. Canadian Journal of Forest Research 47: 433-444. doi: 10.1139/cjfr-2016-0352.

Pacé M, Fenton NJ, Paré D, Stefani FO, Massicotte HB, Tackaberry LE, Bergeron Y (2019) Lichens contribute to open woodland stability in the boreal forest through detrimental effects on pine growth and root ectomycorrhizal development. Ecosystems 22: 189-201. doi: https://doi.org/10.1007/s10021-018-0262-0.

Pacé M, Gadet B, Beguin J, Bergeron Y, Paré D (2020a) Drivers of boreal tree growth and stand opening: the case of jack pine on sandy soils. Ecosystems 23: 586-601. doi: https://doi.org/10.1007/s10021-019-00425-2.

Pacé M, Paré D, Fenton NJ, Bergeron Y (2020b) Effects of lichen, *Sphagnum* spp. and feather moss leachates on jack pine and black spruce seedling growth. Plant and Soil 452: 441-455. doi: https://doi.org/10.1007/s11104-020-04587-0.

Paquin R, Margolis HA, Doucet R (1998) Nutrient status and growth of black spruce layers and planted seedlings in response to nutrient addition in the boreal forest of Quebec. Canadian Journal of Forest Research 28: 729-736. doi: https://doi.org/10.1139/x98-041.

Parada AE, Needham DM, Fuhrman JA (2016) Every base matters: assessing small subunit rRNA primers for marine microbiomes with mock communities, time series and global field samples. Environmental microbiology 18: 1403-1414. doi: https://doi.org/10.1111/1462-2920.13023.

Payette S (1992) Fire as a controlling process in the North American boreal forest. A systems analysis of the global boreal forest (ed. by H.H. Shugart, R. Leemans and G.B. Bonan), pp. 144–169. Cambridge University Press, Cambridge. : 144-169.

Payette S, Bhiry N, Delwaide A, Simard M (2000) Origin of the lichen woodland at its southern range limit in eastern Canada: the catastrophic impact of insect defoliators and fire on the spruce-moss forest. Canadian Journal of Forest Research 30: 288-305. doi: https://doi.org/10.1139/x99-207.

Peng X, Li G, Liu Z (2016) Zero-inflated beta regression for differential abundance analysis with metagenomics data. Journal of Computational Biology 23: 102-110. doi: https://doi.org/10.1089/cmb.2015.0157.

Philippot L, Raaijmakers JM, Lemanceau P, van der Putten WH (2013) Going back to the roots: the microbial ecology of the rhizosphere. Nature Reviews Microbiology 11: 789-799. doi: https://doi.org/10.1038/nrmicro3109.

Phillips LA, Ward V, Jones MD (2014) Ectomycorrhizal fungi contribute to soil organic matter cycling in sub-boreal forests. The ISME Journal 8: 699-713. doi: https://doi.org/10.1038/ismej.2013.195.

Piccolo A, Spaccini R, Nieder R, Richter J (2004) Sequestration of a Biologically Labile Organic Carbon in Soils by Humified Organic Matter. Climatic Change 67: 329-343. doi: 10.1007/s10584-004-1822-1.

Pingree MR, DeLuca EE, Schwartz DT, DeLuca TH (2016) Adsorption capacity of wildfire-produced charcoal from Pacific Northwest forests. Geoderma 283: 68-77. doi: https://doi.org/10.1016/j.geoderma.2016.07.016.

Pingree MR, Homann PS, Morrissette B, Darbyshire R (2012) Long and short-term effects of fire on soil charcoal of a conifer forest in southwest Oregon. Forests 3: 353-369. doi: https://doi.org/10.3390/f3020353.

Pluchon N, Gundale MJ, Nilsson MC, Kardol P, Wardle DA (2014) Stimulation of boreal tree seedling growth by wood-derived charcoal: effects of charcoal properties, seedling species and soil fertility. Functional Ecology 28: 766-775. doi: https://doi.org/10.1111/1365-2435.12221.

Preston CM, Schmidt MW (2006) Black (pyrogenic) carbon: a synthesis of current knowledge and uncertainties with special consideration of boreal regions. Biogeosciences 3: 397-420.

Preston CM, Simard M, Bergeron Y, Bernard GM, Wasylishen RE (2017) Charcoal in organic horizon and surface mineral soil in a boreal forest fire chronosequence of western Quebec: stocks, depth distribution, chemical properties and a synthesis of related studies. Frontiers in Earth Science 5: 98. doi: https://doi.org/10.3389/feart.2017.00098.

Quast C, Pruesse E, Yilmaz P, Gerken J, Schweer T, Yarza P, Peplies J, Glöckner FO (2012) The SILVA ribosomal RNA gene database project: improved data processing and web-based tools. Nucleic Acids Research 41: D590-D596. doi: https://doi.org/10.1093/nar/gks1219.

Rasche F, Knapp D, Kaiser C, Koranda M, Kitzler B, Zechmeister-Boltenstern S, Richter A, Sessitsch A (2011) Seasonality and resource availability control bacterial and archaeal communities in soils of a temperate beech forest. The ISME journal 5: 389-402. doi:

https://doi.org/10.1038/ismej.2010.138.

Reinecke J, Klemm G, Heinken T (2014) Vegetation change and homogenization of species composition in temperate nutrient deficient Scots pine forests after 45 yr. Journal of Vegetation Science 25: 113-121. doi: https://doi.org/10.1111/jvs.12069.

Renaudin M, Laforest-Lapointe I, Bellenger JP (2022) Unraveling global and diazotrophic bacteriomes of boreal forest floor feather mosses and their environmental drivers at the ecosystem and at the plant scale in North America. Science of the Total Environment 837. doi: https://doi.org/10.1016/j.scitotenv.2022.155761.

Robertson SJ, Rutherford PM, Lopez-Gutierrez JC, Massicotte HB (2012) Biochar enhances seedling growth and alters root symbioses and properties of sub-boreal forest soils. Canadian Journal of Soil Science 92: 329-340. doi: https://doi.org/10.4141/cjss2011-066.

Rodríguez-Rodríguez JC, Fenton NJ, Bergeron Y, Kembel SW (2023) Soil and tree phyllosphere microbial communities differ between coniferous and broadleaf deciduous boreal forests. Plant and Soil 488: 233-253. doi: https://doi.org/10.1007/s11104-023-05959-y.

Rolstad J, Blanck Y-l, Storaunet KO (2017) Fire history in a western Fennoscandian boreal forest as influenced by human land use and climate. Ecological Monographs 87: 219-245. doi: https://doi.org/10.1002/ecm.1244.

Rousk K, Jones DL, DeLuca TH (2013) Moss-cyanobacteria associations as biogenic sources of nitrogen in boreal forest ecosystems. Frontiers in Microbiology 4: 150. doi: https://doi.org/10.3389/fmicb.2013.00150.

Sandström P, Cory N, Svensson J, Hedenås H, Jougda L, Borchert N (2016) On the decline of ground lichen forests in the Swedish boreal landscape: Implications for reindeer husbandry and sustainable forest management. Ambio 45: 415-429. doi: https://doi.org/10.1007/s13280-015-0759-0.

Santín C, Doerr SH, Merino A, Bryant R, Loader NJ (2016) Forest floor chemical transformations in a boreal forest fire and their correlations with temperature and heating duration. Geoderma 264: 71-80. doi: https://doi.org/10.1016/j.geoderma.2015.09.021.

Saucier JP, Robitaille A, Grondin P, Bergeron Y, Gosselin J (2011) Les régions écologiques du Québec méridional. In: DdIF Quebec: Ministère des Ressources Naturelles et de la Faune (ed). pp. 3-29.

Scheffer M, Hirota M, Holmgren M, Van Nes EH, Chapin FS (2012) Thresholds for boreal biome transitions. Proceedings of the National Academy of Sciences 109: 21384-21389. doi: https://doi.org/10.1073/pnas.1219844110.

Sedia EG, Ehrenfeld JG (2003) Lichens and mosses promote alternate stable plant communities in the New Jersey Pinelands. Oikos 100: 447-458. doi: https://doi.org/10.1034/j.1600-0706.2003.12058.x.

Sedia EG, Ehrenfeld JG (2005) Differential effects of lichens, mosses and grasses on respiration and nitrogen mineralization in soils of the New Jersey Pinelands. Oecologia 144: 137-147. doi: https://doi.org/10.1007/s00442-005-0037-0.

Sedia EG, Ehrenfeld JG (2006) Differential effects of lichens and mosses on soil enzyme activity and litter decomposition. Biology and Fertility of Soils 43: 177-189. doi:

https://doi.org/10.1007/s00374-006-0077-6.

Sheremet A, Jones GM, Jarett J, Bowers RM, Bedard I, Culham C, Eloe-Fadrosh EA, Ivanova N, Malmstrom RR, Grasby SE (2020) Ecological and genomic analyses of candidate phylum WPS-2 bacteria in an unvegetated soil. Environmental Microbiology 22: 3143-3157. doi: https://doi.org/10.1111/1462-2920.15054.

Shetty R, Vidya CS-N, Prakash NB, Lux A, Vaculik M (2021) Aluminum toxicity in plants and its possible mitigation in acid soils by biochar: A review. Science of the Total Environment 765: 142744. doi: https://doi.org/10.1016/j.scitotenv.2020.142744.

Smith DJ, Timonen HJ, Jaffe DA, Griffin DW, Birmele MN, Perry KD, Ward PD, Roberts MS (2013) Intercontinental Dispersal of Bacteria and Archaea by Transpacific Winds. Applied and Environmental Microbiology 79: 1134-1139. doi: https://doi.org/10.1128/AEM.03029-12.

Smith NR, Kishchuk BE, Mohn WW (2008) Effects of wildfire and harvest disturbances on forest soil bacterial communities. Applied and Environmental Microbiology 74: 216-224. doi: https://doi.org/10.1128/AEM.01355-07.

Smith RJ, Jovan S, Gray AN, McCune B (2017) Sensitivity of carbon stores in boreal forest moss mats - effects of vegetation, topography and climate. Plant and Soil 421: 31-42. doi: https://doi.org/10.1007/s11104-017-3411-x.

Smith SE, Read DJ (2010) Mycorrhizal symbiosis. Academic press.

Soucémarianadin LN, Quideau SA, Wasylishen RE, Munson AD (2015) Early-season fires in boreal black spruce forests produce pyrogenic carbon with low intrinsic recalcitrance. Ecology 96: 1575-1585. doi: https://doi.org/10.1890/14-1196.1.

Spain AM, Krumholz LR, Elshahed MS (2009) Abundance, composition, diversity and novelty of soil Proteobacteria. The ISME journal 3: 992-1000. doi: https://doi.org/10.1038/ismej.2009.43.

Splawinski TB, Cyr D, Gauthier S, Jetté J-P, Bergeron Y (2018) Analyzing risk of regeneration failure in the managed boreal forest of northwestern Quebec. Canadian Journal of Forest Research 49: 680-691. doi: 10.1139/cjfr-2018-0278.

Stark S, Hyvärinen M (2003) Are phenolics leaching from the lichen *Cladina stellaris* sources of energy rather than allelopathic agents for soil microorganisms? Soil Biology Biochemistry 35: 1381-1385. doi: https://doi.org/10.1016/S0038-0717(03)00217-7.

Stark S, Kytöviita M-M, Neumann AB (2007) The phenolic compounds in *Cladonia* lichens are not antimicrobial in soils. Oecologia 152: 299-306. doi: https://doi.org/10.1007/s00442-006-0644-4.

Stokes MA (1996) An introduction to tree-ring dating. University of Arizona Press.

Sun S, Li S, Avera BN, Strahm BD, Badgley BD (2017) Soil bacterial and fungal communities show distinct recovery patterns during forest ecosystem restoration. Applied and Environmental Microbiology 83: e00966-00917. doi: https://doi.org/10.1128/AEM.00966-17.

Tedersoo L, Bahram M (2019) Mycorrhizal types differ in ecophysiology and alter plant nutrition and soil processes. Biological Reviews 94: 1857-1880. doi:

https://doi.org/10.1111/brv.12538.

Thacker SJ, Quideau SA (2021) Rhizosphere response to predicted vegetation shifts in boreal forest floors. Soil Biology and Biochemistry 154: 108141. doi: https://doi.org/10.1016/j.soilbio.2021.108141.

Thompson I, Mackey B, McNulty S, Mosseler A (2009) Forest resilience, biodiversity, and climate change. A synthesis of the biodiversity/resilience/stability relationship in forest ecosystems Secretariat of the Convention on Biological Diversity, Montreal Technical Series no. 43, pp 1–67.

Tonteri T, Hallikainen V, Merilä P, Miina J, Rautio P, Salemaa M, Tolvanen A (2022) Response of ground macrolichens to site factors, co-existing plants and forestry in boreal forests. Applied Vegetation Science 25: e12690. doi: https://doi.org/10.1111/avsc.12690.

Turetsky MR (2003) The role of bryophytes in carbon and nitrogen cycling. The Bryologist 106: 395-409. doi: https://www.jstor.org/stable/3244721.

Turetsky MR, Kane ES, Harden JW, Ottmar RD, Manies KL, Hoy E, Kasischke ES (2011) Recent acceleration of biomass burning and carbon losses in Alaskan forests and peatlands. Nature Geoscience 4: 27-31. doi: 10.1038/ngeo1027.

Urbanová M, Šnajdr J, Baldrian P (2015) Composition of fungal and bacterial communities in forest litter and soil is largely determined by dominant trees. Soil Biology and Biochemistry 84: 53-64. doi: https://doi.org/10.1016/j.soilbio.2015.02.011.

Vallicrosa H, Sardans J, Maspons J, Peñuelas J (2022) Global distribution and drivers of forest biome foliar nitrogen to phosphorus ratios (N:P). Global Ecology and Biogeography 31: 861-871. doi: https://doi.org/10.1111/geb.13457.

Van Der Heijden MGA, Bardgett RD, Van Straalen NM (2008) The unseen majority: soil microbes as drivers of plant diversity and productivity in terrestrial ecosystems. Ecology Letters 11: 296-310. doi: https://doi.org/10.1111/j.1461-0248.2007.01139.x.

Veraverbeke S, Rogers BM, Goulden ML, Jandt RR, Miller CE, Wiggins EB, Randerson JT (2017) Lightning as a major driver of recent large fire years in North American boreal forests. Nature Climate Change 7: 529-534. doi: https://doi.org/10.1038/nclimate3329.

Wang K, Zhang Y, Tang Z, Shangguan Z, Chang F, Jia Fa, Chen Y, He X, Shi W, Deng L (2019) Effects of grassland afforestation on structure and function of soil bacterial and fungal communities. Science of The Total Environment 676: 396-406. doi: https://doi.org/10.1016/j.scitotenv.2019.04.259.

Ward EB, Duguid MC, Kuebbing SE, Lendemer JC, Bradford MA (2022) The functional role of ericoid mycorrhizal plants and fungi on carbon and nitrogen dynamics in forests. New Phytologist 235: 1701-1718. doi: https://doi.org/10.1111/nph.18307.

Wardle DA, Zackrisson O, Nilsson M-C (1998) The charcoal effect in Boreal forests: mechanisms and ecological consequences. Oecologia 115: 419-426. doi: https://doi.org/10.1007/s004420050536.

Weber K, Quicker P (2018) Properties of biochar. Fuel 217: 240-261. doi: https://doi.org/10.1016/j.fuel.2017.12.054.

Wheeler JA, Hermanutz L, Marino PM (2011) Feathermoss seedbeds facilitate black spruce seedling recruitment in the forest-tundra ecotone (Labrador, Canada). Oikos 120: 1263-1271. doi: https://doi.org/10.1111/j.1600-0706.2010.18966.x.

White TJ, Bruns T, Lee S, Taylor J (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics[J]. PCR protocols: a guide to methods applications 18: 315-322.

Whitman E, Parisien M-A, Thompson DK, Flannigan MD (2019) Short-interval wildfire and drought overwhelm boreal forest resilience. Scientific Reports 9: 18796. doi: 10.1038/s41598-019-55036-7.

Wolf M, Lehndorff E, Wiesenberg GLB, Stockhausen M, Schwark L, Amelung W (2013) Towards reconstruction of past fire regimes from geochemical analysis of charcoal. Organic Geochemistry 55: 11-21. doi: https://doi.org/10.1016/j.orggeochem.2012.11.002.

Wu YT, Wubet T, Trogisch S, Both S, Scholten T, Bruelheide H, Buscot F (2013) Forest age and plant species composition determine the soil fungal community composition in a Chinese subtropical forest. PLoS One 8: e66829. doi: https://doi.org/10.1371/journal.pone.0066829.

Xiao R, Man X, Duan B, Cai T, Ge Z, Li X, Vesala T (2022) Changes in soil bacterial communities and nitrogen mineralization with understory vegetation in boreal larch forests. Soil Biology and Biochemistry 166: 108572. doi: https://doi.org/10.1016/j.soilbio.2022.108572.

Yan B, Sun L, Li J, Liang C, Wei F, Xue S, Wang G (2020) Change in composition and potential functional genes of soil bacterial and fungal communities with secondary succession in Quercus liaotungensis forests of the Loess Plateau, western China. Geoderma 364: 114199. doi: https://doi.org/10.1016/j.geoderma.2020.114199.

Zackrisson O, Nilsson M-C, Dahlberg A, Jäderlund A (1997) Interference mechanisms in conifer-Ericaceae-feathermoss communities. Oikos: 209-220. doi: https://doi.org/10.2307/3546287.

Zackrisson O, Nilsson M-C, Wardle DA (1996) Key ecological function of charcoal from wildfire in the Boreal forest. Oikos: 10-19. doi: https://doi.org/10.2307/3545580.

Zhang S, Yang X, Li D, Li S, Chen Z, Wu J (2022) A meta-analysis of understory plant removal impacts on soil properties in forest ecosystems. Geoderma 426: 116116. doi: https://doi.org/10.1016/j.geoderma.2022.116116.

Zhou G, Lucas-Borja ME, Eisenhauer N, Eldridge DJ, Liu S, Delgado-Baquerizo M (2022) Understorey biodiversity supports multiple ecosystem services in mature Mediterranean forests. Soil Biology and Biochemistry 172: 108774. doi: https://doi.org/10.1016/j.soilbio.2022.108774.

Zinger L, Lejon DP, Baptist F, Bouasria A, Aubert S, Geremia RA, Choler P (2011) Contrasting diversity patterns of crenarchaeal, bacterial and fungal soil communities in an alpine landscape. PLoS One 6: e19950. doi: https://doi.org/10.1371/journal.pone.0019950.

Zogg GP, Zak DR, Ringelberg DB, White DC, MacDonald NW, Pregitzer KS (1997) Compositional and functional shifts in microbial communities due to soil warming. Soil Science Society of America Journal 61: 475-481. doi:

https://doi.org/10.2136/sssaj1997.03615995006100020015x.

Zumsteg A, Luster J, Göransson H, Smittenberg RH, Brunner I, Bernasconi SM, Zeyer J, Frey B (2012) Bacterial, archaeal and fungal succession in the forefield of a receding glacier. Microbial ecology 63: 552-564. doi: https://doi.org/10.1007/s00248-011-9991-8.