Permission Statement

Additional information on the report may be obtained by contacting COSIA.

The report may be cited as:

> Landhäusser Research Group and Karst Lab. 2018. *The Roots of Succession: Relations among Plants, Soil, and Mycorrhizal Fungi in a Reclaimed Site. The Aurora Soil Capping Study: The First Five Years*. Canada's Oil Sands Innovation Alliance, 2021, www.cosia.ca.

The Roots of Succession: Relations among Plants, Soils, and Mycorrhizal Fungi in a Reclaimed Site

The Aurora Soil Capping Study: The First Five Years

June 14, 2018

Prepared for Syncrude Canada Ltd
Syncrude Canada Ltd – Research
9421 17 Ave NW
Edmonton, AB T6N 1H4

Prepared by the Landhäusser Research Group and Karst Lab
4-42 Earth Sciences Building
Department of Renewable Resources
University of Alberta
Edmonton, AB T6G 2E3

Acknowledgements

Preparation of this report was led by Caren Jones, Shauna Stack, and Simon Landhäusser with the data and help of members of the Landhäusser Research Group and Karst Lab, Department of Renewable Resources, University of Alberta. This report includes data summarizing tree, vegetation, and mycorrhizal development over the first five growing seasons at the Aurora Soil Capping Study.

Individuals who contributed significantly to this report include Justine Karst, Gregory Pec, Simon Bockstette, Jana Bockstette, Natalie Scott, Stefan Hupperts, and Shanon Hankin.

Support is acknowledged from the countless summer research assistants and staff at Syncrude. In particular, we thank Eckehart Marenholtz, Fran Leishman, and Marty Yarmuch for their support. This research was financially supported by Canadian Natural Resources Ltd., Syncrude Canada Ltd., Total E&P Canada Ltd., Imperial Oil Resources Ltd., and Shell Canada Energy, as part of Canada's Oil Sands Innovation Alliance (COSIA).

Table of Contents

Highlights

- Using different soil cover designs can provide a wide range of (early) tree and vegetation growth.
- Early growth of Aspen and Pine were more sensitive to the different soil placement treatments, while spruce was relatively indifferent.
- Salvaged and directly placed upland forest floor material used as a coversoil was associated with higher nutrient availability and a richer propagule bank, which supported accelerated tree growth and of colonizing vegetation development more closely resembling upland forest communities.
- Imbalances of nutrient availability (particularly P), high water content during wet years, and cooler soil temperatures during the growing season limited tree growth on Peat when placed at 30cm. Colonizing vegetation cover on peat was very low and was dominated by introduced ruderal species; however, some lowland species likely from the peat seedbank were also present.
- Water holding capacity of the Peat increased water availability compared to the other sandier soils, which buffered against dry conditions during drought years, allowing for continued tree growth.
- Texture and nutrient availability of subsoils affect tree growth (i.e. in coarse textured soils even slight changes in the proportions of the clay and loam fraction appear to influence the water holding capacity).
- Increasing capping thickness of subsoil placed under a Peat coversoil allowed excess soil water in the coversoil to infiltrate deeper into the soil profile and lower the water content of the surface soil.
- Shifts in the community composition of ectomycorrhizal fungi were driven more by disturbance severity than by tree species planted or the surface soil material placed.
- Disturbances that remove or disrupt the surface organic layer of soils substantially affect the composition of ectomycorrhizal fungal communities.
- The effect of tree species planted and soil placement on early development of ectomycorrhizal fungal communities establishing at the Aurora Soils Capping Study is dynamic through time.

- Mixing tree species had an additive effect on ectomycorrhizal; however, it did not result in greater synergistic effects on ectomycorrhizal diversity this early in succession.

Summary

The type of coversoil material placed at the soil surface had the greatest impact on early seedling growth and vegetation development at the Aurora Soil Capping Study (ASCS). Generally, aspen and pine responded more strongly to the different soil treatments compared to white spruce, which appears to be more tolerant to most soil conditions tested at ASCS. This was somewhat expected, as spruce is a later successional, relatively stress tolerant species, while aspen and pine are early successional species that can take readily advantage of high initial resource availability. Aspen and pine seedlings grew best on the salvaged forest floor material (FFM) compared to Peat or no coversoil treatments. These differences were likely driven by resource availability, mostly nutrients where the availability of P, K, and NH_4^+ was greatest in the FFM coversoil. A more detailed analysis of the data also indicates that the availability of micronutrients such as Mn combined with low Ca concentrations might have played a role. This observation is somewhat in contrast to the Peat coversoil, which was associated with high concentrations of NO_3^-, K, S, Fe, Ca, Mg, and Zn, but a low availability of P. High levels of Ca and Mg cations may have exacerbated P limitations in Peat, as they can form complex compounds and precipitates that reduce P availability. Furthermore, foliar analysis in 2013 indicated that S levels in aspen leaves were potentially high enough to slow tree metabolism and growth. The two Subsoil coversoil treatments had overall lower levels of all essential nutrients, which likely limited tree growth on these soils.

Seasonal soil temperature in the rooting zone was very different between the Peat and mineral coversoils, and this might have played a role in the growth limitation of seedlings. The warming of Peat in the spring was consistently delayed, lowering the number of days for root growth, impacting nutrient and water uptake. An explanation for the cooler temperatures on Peat is the insulating effects of the undecomposed organic matter. This relationship became apparent when comparing tree growth, particularly aspen, between the shallow and the deep Peat coversoil treatments, where aspen negatively responded to the thicker 30cm placement of Peat. Further lower soil temperature can also be associated with higher water holding capacity of Peat compared to mineral coversoils. While the greater water holding capacity can have a positive effect by storing and supplying water to vegetation, it comes with a negative trade-off that requires greater energy input to warm the soil in the spring. This trade off was evident during the very dry conditions of 2015, where trees had noticeably less growth in the thinly placed peat (10 cm) and the mineral coversoils, compared to the thick placed peat (30 cm). This suggests that the shallow placement of peat on the surface and the mineral soils did not hold enough water to mediate against the dry growing conditions. White spruce showed the greatest positive response to the thicker

layer of Peat indicating its overall ability to continue growing in cooler soil temperatures and higher soil water availability. Interestingly, pine grew expressively more in the dry season of 2015 when compared to the other years measured. However, due to its determinate growth strategy, pine height growth is largely dependent on the early growing season conditions, and in 2014 the area received above average precipitation in the early and mid-growing season when the formation of the buds for 2015 occurred and these favourable soil conditions lasted into the early portion of the growing season on 2015. Therefore, the early spring in 2015 coupled with the effects of a wetter 2014 season might have been the cause for better growth of pine in 2015; however, the effects of the dry conditions later in the growing season of 2015 were then noticeable in 2016 when pine growth decreased.

The underlying subsoil materials also appear to influence tree growth. However, the response was dependent on the type of coversoil, as that affected the accessibility of the subsoil due to root system size. When Peat coversoil was used, aspen and pine had somewhat taller trees with a selectively salvaged Subsoil Bm horizon, although these differences were not statistically significant. These differences might be driven by the greater availability of P, total N, and NO_3^- in the Subsoil Bm. Growth responses to underlying Subsoils C and BC were similar for all tree species, which may be due to the similar chemical and physical characteristics of these two soils. Water and temperature were likely not driving the subsoil treatment differences because the 30cm of Peat maintained high water availability and cool soil temperatures in the rooting zone. When capped with a FFM coversoil, water availability rather than nutrients appear to drive tree growth. Slight variation in soil texture might have changed water availability, which was particularly evident during the dry year of 2015. Although the three subsoils were all considered sandy, average daily water content during the growing season was consistently higher in the Subsoil C treatment within the top 15cm of soil than in the treatments with Subsoil Bm or BC, the latter of which had the lowest average daily water content. During 2015 water content in the Subsoil BC treatment remained below permanent wilting point for the entire growing season in both 2015 and 2016, although it had the smallest trees and leaf area. Interestingly, although seedlings grew negligibly in height, aspen grew in stem diameter during that time indicating preferential allocation to roots. Although the different subsoils had similar overall textures and bulk densities, there were slight differences in silt content; however, it remains unclear whether these subtle differences significantly influenced soil water infiltration and storage abilities and may warrant further investigation.

The importance of both nutrient and water availability is also evident in a comparison of tree responses to total capping material depth. All tree species responded poorly to the 30cm of Peat directly over lean oil sand (LOS) overburden. The placement of a subsoil layer under the Peat coversoil, no matter the thickness, influenced soil water and chemical dynamics of the Peat layer. The higher variability in water content observed with increasing soil cap thickness is likely related to the depth of the soil-to-LOS boundary and the fluctuations of a temporary water table above this boundary. Seasonal water table fluctuations can create ideal moisture conditions for tree growth and allow for the movement of nutrients such as S and P. The high soil water content with little variation found in the 30cm Peat over LOS may have developed temporarily saturated and anaerobic conditions due to decomposition of the peat that could have reduced root growth. Trees with roots solely grown in the Peat layer would have also been limited by low P supply and critically high S levels, which likely further slowed metabolism and overall growth.

At this early stage in development, interactions among tree seedlings appear to be minimal. There were no differences detected among trees planted in single-species versus mixed stands, and this applied to all species. One observation that was unique to the FFM covered plots with pine was a noticeable draw-down of water in the soil each growing season, which may have a greater impact on the growth of single-pine versus pine mixed with other species in the future. Pine was the only tree species that responded this early on to planting density, where trees grew taller when planted at low densities in FFM cover soils. Given the water limitations of the sandy FFM coversoil, the lower planting density may have reduced the competition for water among trees compared to the higher density plots.

Coversoil material type mainly influenced composition and cover of colonizing vegetation in the first four years. Differences in the vegetation communities of the four coversoil materials were expected due to the assumed different propagule banks of the materials. As FFM was salvaged and directly placed from an upland forest, the propagule bank was anticipated to contain species that are more suited to the drier conditions of upland areas, particularly for coarse textured soils. Propagules contained within the Peat material were predictably of species adapted to the wetter conditions common in lowland areas and subsequently were not well suited for the drier conditions of upland sites. Additionally, the Peat material was salvaged up to three meters deep; this significantly diluted the viable propagules throughout the material, as most propagules are found within the surface 20cm of soil. The community developing on FFM was characterized by native forest forb and shrub species such as *Epilobium angustifolium, Vaccinium myrtilloides,* and *Geranium bicknellii*, while grasses and annual forb species dominated the Peat and no coversoil treatments. The functional groups to which the indicator species belong is an important aspect of characterizing the plant communities, as they potentially affect the successional trajectory of the reclamation sites.

While vegetation cover increased in Peat between 2013 and 2015, cover was still less than one percent. The low cover of colonizing vegetation in the Peat could be driven by a large range of variables such as propagule availability, nutrient availability, and/or lack of suitable microsites for establishment. The lack of a viable propagule bank in the Peat and no coversoil treatments resulted in the poor early plant community development, lacking in cover and diversity. However, the plant community in these materials will most likely change as the site and tree canopy develops in time. Dispersal of propagules from nearby forested areas and established reclamation areas are currently affecting vegetation development at ASCS. At this early stage of vegetation development, an impact of the tree species composition and cover on the colonizing plant community has not been detected, but it is expected that they will become dominant drivers in the forest development of these areas.

In the soils, ectomycorrhizal fungal (EcM) community composition varied across the ASCS site and at a benchmark reference site. For both sites, disturbance severity accounted for a greater total variation (18%) in EcM fungal community composition than host identity (7%). Ectomycorrhizal fungal community composition differed among the three host species. For a given host species, EcM fungal composition did not differ among coversoils on one- and two-year old seedlings. However, the effect of coversoil changed with time as EcM fungal community composition differed among FFM, Peat, and no coversoil on four-year old seedlings. At year four, mixed species plots had a higher diversity of EcM fungi, but that was strictly related to the greater number of host species relatively to single species plots, indicating that using mixed species plots have an additive rather than a synergistic effect on EcM communities at this stage in forest development. Ectomycorrhizal fungi of seedlings planted in soils at the reference site where mature trees were removed, but forest floor and soils were left intact, did not differ in composition from seedlings planted into soils of a mature intact forest.

Implications

- This remarkable study design has provided an opportunity and the options of potentially chosing future soil cover designs and depths appropriate for specific site conditons, with predictable outcomes of tree growth and vegetation development for these salvaged material types.

- Conditions provided by the coversoils are the most important factors driving early seedling and colonizing vegetation establishment. This study highlights that composition of coversoil need to provide a balance between water and nutrient availability. Therefore mixing of organic and mineral materials, particularly when using Peat as an upland coversoil material, could improve some of the limiting conditions associated with only using Peat or a coarse mineral substrate as a coversoil. In this study, mixing a coarse mineral soil material with Peat to a depth of 20 cm would likely have provided higher water holding capacity for trees during dry years compared to the coarse mineral material alone, while providing the benefits of greater soil warming and nutrient availability compared to Peat only coversoil. However, some of these relationships still need to be explored in greater depth.

- To evaluate the effects of capping thickness, subsoil selection, and LOS overburden material interactions with root systems and overall tree and stand performance, longer-term growth data is necessary. At this stage of the study the root systems of these seedlings are just starting to explore the deeper soil layers (particularly in the 30 cm peat); however, there are some indications that the type of subsoil material will influence the growth of trees and likely the resistance of them to stresses such as drought. Since some of these treatments have a thick Peat coversoil, the response of trees might take longer to detect in these treatments.

- The aboveground effects of canopy tree selection and planting density on soil and understory development are not identifiable at this early stage in development. However, some trees in some treatments have reached canopy closure in the high-density plots. Clearly, higher-density tree planting is a valuable tool for reaching canopy closure quickly. The next few years will show whether these areas will maintain a greater diversity of understory species, compared to areas that have had an open canopy for longer times.

- The effects of canopy and litter type and its input into the soil will likely also have a measurable impact on soil development and overall plant diversity. The next 5-

10 years will likely give an indication on these medium-term developments and better define stand trajectories on these coarse soil materials.

- The dynamic effects of tree species and soil placement suggests that early assessments of the ecological factors influencing EcM fungal communities following disturbance should be interpreted cautiously. Similar to understory vegetation communities, EcM communities will continue to change in both species composition and relative abundance across the maturing reclaimed area. Longer term monitoring should be incorporated to better understand and track how differences in host and coversoils interact to affect belowground EcM communities.
- EcM fungal communities were likely similar across coversoils when initially placed at the ASCS and fungi colonizing roots were initially sorted by host identity. This initial pool of fungi were those that were able to withstand soil handling such as displacement, mixing, and transport. Importantly, coversoil type seemed to influence which fungi persisted following these processes, dormancy, and dispersal.
- The influence of coversoil on EcM fungal community composition became more pronounced with time; however, the strength of host identity was consistent across the study duration and explained a considerably greater portion of variance. Interestingly, it seems those fungi able to withstand severe disturbance are not host-generalists.
- Shifts in the community composition of EcM fungi were driven to a greater extent by disturbance severity than either host identity or coversoil used in soil placement at the ASCS. Though planting trees of different species on the reclaimed site recovered slightly different EcM fungal communities across the reconstructed soils, there remained a wide gap between communities assayed at the ASCS and those of ecological benchmarks representing less soil disturbance. This suggests that the disturbance effects from reclamation likely have the most pronounced influence on EcM fungal communities, while manipulations of coversoil type and tree species planted played a relatively minor role.
- Results suggest management objectives should include a shift from a focus on historic fidelity of community composition to one of ecosystem function. Regardless of the differences in the composition of EcM fungal communities across disturbances investigated in our study, an unresolved question is whether

EcM function similarly to natural sites are developed and maintained. An emphasis on function should guide future research in this area.

Terminology and Abbreviations

AOSR	Athabasca Oil Sands Region
ASCS	Aurora Soil Capping Study
Coversoil	Soil material placed at the soil surface of reconstructed soil profiles
CS	Coversoil
EcM	Ectomycorrhizal fungi
FFM	Forest floor material
LOS	Lean oil sands material; type of overburden material
OB	Overburden material
OKC	O'Kane Consultants Inc., Saskatoon, SK
OM	Organic matter
OTU	Operational taxonomic unit
Overburden	Geologic material removed during the mining process that is not considered suitable for root growth
PRS	Plant root simulator
Subsoil	Soil material placed below the coversoil, but above the overburden material of reconstructed soil profiles
Total capping material thickness	Thickness of both the coversoil and subsoil combined over overburden

List of Tables

Project 1

Project 2

List of Figures

Project 1

Site Layout

Results

Coversoil Material Type

Coversoil Placement Depth

Underlying Subsoil Material

Total Capping Material Depth

Tree Species Composition

Tree Planting Density

Project 2

Restoration of Belowground Fungal Communities

Early Ectomycorrhizal Fungal Community and Mixed Tree Plantings

List of Appendices A: Supplemental Material of Climatic and Soil variables

List of Appendices B: Supplemental Material of Tree Performance and Analyses

List of Appendices C: Supplemental Material of Vegetation Development and Analyses

List of Appendices D: Supplemental Material of Mycorrhizal Fungi Community Development and Analysis

Introduction

Canopy cover and vertical structure are essential elements of forests that create conditions necessary for forest diversity and associated biogeochemical cycling between soils and plants (Macdonald and Fenniak 2007, Hart and Chen 2008). One of the priorities of forest land reclamation is the establishment of a tree cover; however, prior to planting a major challenge is the reconstruction of soil profiles that have the ability to provide both a suitable growing medium and the resources necessary to allow for the establishment and sustainability of planted and volunteer vegetation. Soils play a particularly important role in forest restoration as they have to sustain long-lived plants (i.e. trees) over decades or centuries all while these ecosystems can be exposed to a wide range of climatic conditions, disturbances, and other biotic or abiotic stresses. While it is impossible to recreate the exact soils with all its distinct horizons and differences in properties, reclamation practices can attempt to emulate some of the characteristics that provide short- and long-term benefits (Hobbs et al. 2009, 2013).

During the reclamation of a mined landscape there are many types of landforms that are constructed and need to be reclaimed. Overburden (OB) dumps are large upland landscape features created from the geologic material that was removed during mining operations and were situated between soil materials and the desired resource. A common type of OB in the Athabasca Oil Sands Region (AOSR) is lean oil sands (LOS) material. This material can contain up to a seven percent concentration of bitumen (below the economical ore grade concentration (Ansley 1963)) and is generally not considered suitable for vegetation establishment due to various physical and chemical characteristics and therefore needs to be capped with suitable rooting materials (Conly et al. 2002, Macyk et al. 2004). For soil reclamation, subsoil materials are placed on top of the OB material and subsequently covered with a salvaged coversoil (surface soil) material (Macyk et al. 2004, Alberta Environment 2010). The salvaged soil materials can either be placed directly on a reclamation area after salvaging or stockpiled and stored for later use. Stockpiling of these materials has shown to significantly reduce soil quality, especially of coversoils (Johnson et al. 1991, Harris et al. 1993, Sheoran et al. 2010). Direct-placement has been found not only to be beneficial for forest restoration, as these materials contain more viable soil organisms and propagules than stockpiled material (Harris et al. 1989, Koch et al. 1996, Mackenzie and Naeth 2010, Sheoran et al. 2010, Béasse 2011, Naeth et al. 2013), but also to be more cost efficient due to reduced transportation.

Along with a continued emphasis on tree cover and survival, there has been an increased focus on the re-establishment of understory species in reclaimed forests

(Macdonald et al. 2015a). Having diverse forest communities are important as they contribute to a number of ecosystem functions, including soil stabilization, nutrient cycling, water filtration, and food sources (Carroll et al. 2000, Nilsson and Wardle 2005, Gilliam 2007). Propagule banks contained within salvaged coversoil material have been found to be one of the most significant resources for initial vegetation community establishment (Paré et al. 1993, Koch et al. 1996, Mackenzie and Naeth 2010, Macdonald et al. 2015b, Schott et al. 2016).

Soil microbes play a significant role in the function of soils and in the interaction between soils and plants, yet their role in restoration is often overlooked (Callaham et al. 2008, Wubs et al. 2016, Strickland et al. 2017). In particular, ectomycorrhizal (EcM) fungi are essential to ecosystem functions, such as carbon flow and nutrient cycling, as well as forest regeneration and succession (Jumpponen and Egerton-Warbuton 2005, Peay et al. 2008, Clemmensen et al. 2013). Ectomycorrhizal fungi, which acquire carbon from and enhance the water and nutrient uptake of their hosts (Peay et al. 2008), are recognized as playing a critical role in seedling survival (Hawkins et al. 2015), nutrition (Jones et al. 2010), and mediating the fertility, structure and turnover of organic matter (OM) in soils (Johnson et al. 2016). Thus, EcM fungi can potentially facilitate revegetation and the development of soils, two key goals in restoration.

In 2011, the Aurora Soil Capping Study (ASCS) was established to test a range of questions related to the impact and efficacy of soil capping cover designs (soil cap) on early upland forest establishment at an operational scale. Soil caps in this study differed in the type, arrangement and application thickness of coversoil and subsoil materials. The research presented here explores the effect of different coversoil and subsoil material types and their application sequence and thickness on early tree establishment, development of colonizing vegetation, and EcM communities. The impact of these different soil capping treatments on soil physical (soil temperature and water content) and chemical (i.e. nutrient availability) properties were also assessed and related to early seedling growth of trembling aspen (*Populus tremuloides* Michx.), jack pine (*Pinus banksiana* Lamb.) and white spruce (*Picea glauca* (Moench) Voss.).

Ectomycorrhizal fungi communities at the ASCS were also compared to those from sites with varying levels of disturbances (reference sites). This was done to address whether disturbance intensity trumps common management interventions to restore belowground fungal communities of boreal forests in post-mined landscapes.

The ASCS research focused on exploring the links between plants and fungi and the rooting behavior of trees and other plants in order to assess the suitability of different capping treatments (depths, types, and composition of coversoils and subsoil as outlined in the capping treatments) over LOS for the successful reclamation of these areas to functioning ecosystems. The first chapter of this report (p.5) includes results from the first proposed project at the ASCS focusing on the development of trees and vegetation while the second chapter (p.94) includes results of the second project proposed at the ASCS focusing on EcM fungi development at both the reclaimed and reference sites. These projects from the ASCS research proposal are summarized below.

Project 1: Impact of different capping treatments on tree establishment, growth, and root development

Overall, this project explored the early development of planted tree seedlings, above- and belowground, in response to different capping material treatments. This project was expanded further to also include the early development of the colonizing vegetation communities in response to the capping treatments and tree seedling planting prescriptions.

The impact of six main variables on tree performance and vegetation were explored in this project:

1) coversoil material type,
2) coversoil material placement depth,
3) subsoil material type,
4) total capping material depth,
5) tree species composition, and
6) tree planting density.

Project 2: Impact of different capping treatments and tree species selection on ectomycorrhizal community development

The second project explored the development of EcM communities in response to three common management tools used in restoration–selection of planted tree species, soil placement, and time – at the ASCS. These EcM community assemblies were also compared to natural forest ecosystems with disturbance benchmarks. This project was also expanded to include the assessment of the EcM community development in response to mixed versus single species plantation (i.e. species composition).

Three main aspects related to EcM communities were explored:

1) coversoil soil material type selection,

2) tree species selection, and
3) tree species composition.

Project 1: Tree & Vegetation Development

Methods

Site Description

The ASCS was constructed on an OB material dump at the Syncrude Aurora North Mine (57°20'01"N, 111°31'58"W). The mine is located within the Alberta Central Mixedwood Natural Subregion, an area made up of upland and lowland forests. Upland forests include mixedwood stands of white spruce and trembling aspen on Luvisolic soils or relatively pure jack pine stands on Brunisolic soils (Soil Classification Working Group 1998, Downing and Pettapiece 2006). Lowlands are bogs and fens dominated by black spruce (*Picea mariana* Mill.) and tamarack (*Larix laricina* (Du Roi) K. Koch) and have developed on poorly drained Organic soils (Soil Classification Working Group 1998, Downing and Pettapiece 2006).

Climate normals during the growing season (May-September) for the region (1981-2010) were an average daily temperature of 13.4°C and precipitation totalling 284.3mm (Government of Canada 2018). At the ASCS, average daily air temperatures during the growing seasons (beginning 10 days after first daily average temperature >5˚C until first frost) were 15.5°C (2012), 16.9°C (2013), 15.1°C (2014), 14.5°C (2015), and 15.2°C (2016). Total growing season precipitation was 253.2mm (2012), 266.9mm (2013), 315.4mm (2014), 209.1mm (2015), and 342.8mm (2016). More detailed information on growing season climate conditions are provided in Table A1.

Experimental Design

Thirteen soil covers were randomly assigned across the experimental site. Each soil treatment was applied in one-hectare cells and replicated three times (Figure 1.1). The thirteen cover treatments varied in coversoil material type over one or two subsoil layers (Figure 1.2).

One of the coversoil materials, Peat, was salvaged to a maximum depth of approximately three meters. Another coversoil, forest floor material (FFM), was salvaged to a maximum depth of 15cm from a dominantly pine stand with sandy, Brunisolic soils. Both the Peat and FFM were directly placed on the site following salvage.

The subsoil materials used in the study were also salvaged from Brunisolic soils. Subsoils Bm1 and Bm2 were both salvaged from a depth of 15 to 50cm but have different Pleistocene parent geologic materials (Bm2 comprised of larger sand grain size). Subsoil BC was salvaged from a depth of 50 to 100cm from the same location as Subsoil Bm. These materials were salvaged in the winter of 2007/08 and were stockpiled until relocated to the ASCS in the winter of 2011/12. The Subsoil C material was salvaged from a depth of 15 to 250cm and came from the same salvage area as the FFM. As was done with the FFM and Peat, Subsoil C was directly placed on the ASCS after salvaging.

Depending on the assigned soil treatment, the Peat coversoil material was placed at a target thickness of 10 or 30cm and the FFM coversoil was placed at 10 or 20cm. Coversoils were underlain by different subsoil material types and configurations. Apart from three soil treatments, which varied in total soil cover depths (i.e. total capping thickness) of 30, 60, and 100cm, the total soil cover depth was 150cm in all other soil treatments (Figure 1.2).

Four tree plots (25m × 25m) were established with a minimum buffer of 10m in each treatment cell (Figure 1.1). In May 2012 trees were planted in all treatments and tree plots. Three of the plots were planted each with a single species of trembling aspen, jack pine, or white spruce while the fourth tree plot was planted with an even mixture of all three species. Commercially grown, one-year-old seedlings of the three species, grown from a local seed source, were hand-planted at a regular 1 × 1m spacing (10,000 seedlings ha^{-1} (sph)). In treatments 8 and 10, an additional set of four tree plots were established which were planted at a low planting density (2,000sph) (i.e. these treatments contained a total of eight tree plots in each cell). Areas outside the tree plots were planted with a mixture of the same three tree species at a density of approximately 2,000sph. Additionally, these areas outside the tree plots were planted with three native shrub species (*Prunus pensylvanica* L.f., *Alnus crispa* Chaix., *Amelanchier alnifolia* (Nutt.) Nutt. ex M. Roem.) at a density of approximately 800sph.

Figure 1.1: Map of the Aurora Soil Capping Study (ASCS). The soil layering treatment (Figure 1.2) is designated in each cell within circles and tree plots are designated as squares with their planting treatment identified within. Tree plots that were planted at low densities (2,000sph) rather than high densities (10,000sph) are designated with a subscript 2. Note: ‘A’ refers to trembling aspen, ‘S’ refers to white spruce, ‘P’ refers to jack pine, and ‘M’ refers to mixed tree plots.

Figure 1.2: The 13 soil layering treatments at the Aurora Soil Capping Study (ASCS): Treatment 1 is 30cm of Peat over 120cm of Subsoil C; treatment 2 is 10cm of forest floor material (FFM) over 140cm of Subsoil C; treatment 3 is 10cm of Peat over 140cm of Subsoil C; treatment 4 is 30cm of Peat over 30cm of Subsoil BC; treatment 5 is 30cm of Peat; treatment 6 is 30cm of Peat over 30cm of Subsoil Bm1 over 90cm of Subsoil C; treatment 7 is 20cm of FFM over 130cm of Subsoil C; treatment 8 is 20cm of FFM over 130cm of Subsoil BC; treatment 9 is 20cm of FFM over 30cm of Subsoil Bm1 over 100cm of Subsoil C; treatment 10 is 30cm of Peat over 120cm of Subsoil BC; treatment 11 is 30cm of Peat over 70 cm of Subsoil BC; treatment 12 is 150cm of Subsoil Bm2; treatment 13 is 150cm of Subsoil C. All treatments were placed over lean oil sands (LOS) overburden material. Physical and chemical characteristics of these materials are presented in Table 1.2.

Measurements

Climate

Air temperature, relative humidity, wind speed and direction, net radiation, rainfall, snowpack depth, and barometric pressure were monitored at the ASCS. The following sensors were used: a CS model HMP45C probe with a protective radiation shield was used to measure air temperature and RH; wind speed and direction was measured using a Young model 05103AP-10 wind monitor; a Kipp and Zonen model NR-LITE net radiometer with a thermopile sensor was used to measure net radiation; rainfall was recorded by a Texas Electronics TR525M tipping bucket rain gauge; a CS 61302V sensor measured barometric pressure; lastly, snowpack depth was measured by a CS model SR50AT sonic ranging sensor. All meteorological sensors were connected to a CR1000 datalogger powered by a solar panel and rechargeable battery source, which recorded data every 60 seconds and output hourly and daily averages for subsequent data collection (O'Kane Consultants Inc. (OKC) 2017).

Soil Characteristics

Prior to the first growing season, soil samples of each type of soil material, including the LOS OB material, were collected in each cell by NorthWind Land Resources Ltd. Soil texture, bulk density, bitumen content, pH, electrical conductivity (EC), sodium absorption ratio (SAR), OM content, total organic carbon content (TOC), total organic nitrogen content (TON), and availability of nitrate (NO_3^-), ammonium (NH_4^+), phosphorous (P), potassium (K), and sulphate (SO_4^-) were tested on all samples.

Multiple monitoring systems were installed to monitor vadose zone water dynamics in the soil materials. Soil moisture and temperature were installed in each cell. Two types of soil sensors were used to collect moisture and temperature data: 1) Campbell Scientific Model 616 time domain reflectometry (TDR) sensors were used to continuously monitor the in situ water content, and 2) Campbell Scientific Model 229 thermal conductivity (TC) sensors were used to monitor the in situ temperatures. Each sensor was calibrated and connected to CR1000 dataloggers, which recorded the sensor data every four hours (O'Kane Consultants Inc. (OKC) 2017). In addition, MPS-2 soil water potential sensors and EM50 data loggers (Decagon, Pullman, WA, USA) were installed in the centre of all tree plots in treatments 1, 5, 6, 7, and 9 in 2013 to measure soil water availability and soil temperature. Sensors were inserted at the centre depth of each reconstructed soil horizon, and soil water potentials and soil temperature were recorded every two hours over the course of each growing season (May – September).

Nutrient availability was also assessed in treatments 1, 5, 6, 7, and 9 in 2013 using ion-exchange resin membranes (PRS^{TM} probes, WesternAG, Saskatoon, SK, Canada). PRS-probes were inserted at centre depth of each reconstructed soil horizon. One probe pair (cation + anion) was placed in each horizon in each of the four tree plots of a cell. Probes remained in the soil for 34 days (July 04-August 07, 2013). Following retrieval, they were cleaned with de-ionized water to remove any adhering soil particles. Probes were pooled by soil horizon for each cell and subsequently sent to WesternAG for analysis.

Tree Performance

To assess tree performance, three sub-plots were established in each tree plot. Circular sub-plots (radius=1.99m) were located in the north-west and in the south-east corners of the tree plot and a single square sub-plot (5 × 5m) was located in the center of the tree plot (Figure 1.3). In the square sub-plot all 16 seedlings were individually tagged. Seedling heights were measured from ground to bud tip and root collar diameter (RCD) was measured at ground level in late August for five growing seasons (2012 to 2016). In the circular sub-plots only height was measured while both height and RCD were measured in the center sub-plot.

To allow for a non-destructive observation of root growth, minirhizotron tubes were installed in all tree plots in treatments 1, 6, 7, and 9 in 2012. Minirhizotrons could not be installed in treatment 5, as the underlying LOS proved to be too compacted to penetrate with the coring equipment. All minirhizotron tubes were installed in the center of four 1 × 1m spaced trees and the tube was randomly pointed towards one of the four seedlings. To do so the tube was inserted at a 45° angle to a maximum depth of 85cm beneath a seedling. Between May 2013 and October 2015 the minirhizotrons were visited several times during growing season and 85 images in 1cm increments were taken along each tube using the BTC-2 ICAP camera system (Bartz Technology Corp., Carpinteria, CA, USA). The resulting images were analyzed using Rootfly (Wells and Birchfield, Clemson University, SC, USA) to determine total root length. Root-length density, i.e. root length per volume of soil (RLD, cm cm^{-3}) was then calculated for each image using the known width and height (18 × 13mm) with a depth of field of 0.785mm, as suggested by Taylor et al. (2014). Total root length observed along each minirhizotron tube, i.e. the sum of all root length (cm) measured in all 85 images was calculated; this value was used to estimate the fraction of observed root-length contained within the coversoil, as well as the depth required to account for 90% of the total observed root-length.

To further explore the root system development of the trees in some of the soil treatments, seedlings were excavated in each single-species tree plot in soil treatments 1, 3, 6, 9 and 12 in September, 2014, and again in 2015 in soil treatments 1, 5, 6, 7, and 9. To identify representative seedlings for excavation in each treatment, the average RCD was determined for each tree plot and cell, and three seedlings with the average RCD were then selected from within each tree plot, but outside of the tree sampling sub-plots. Seedlings were carefully excavated to capture the majority of the root system. The excavated seedlings were cold stored in the field and then frozen when in the laboratory until processing. Roots, stems, and leaves were separated, and roots were carefully washed. The different organs (leaves/needles, stems, and roots) were then dried at 70°C to constant weight and their dry mass was determined. Leaf mass fraction (LMF), stem mass fraction (SMF), and root mass fraction (RMF) were calculated to determine allocation to leaves, stem, and roots by dividing leaf, stem, or root dry mass by the total dry mass. A sample of the leaves/needles was also used to determine seedling leaf/needle area (cm^2). The lengths of the three longest lateral roots from the seedlings excavated in 2015 were averaged and used as an estimate of the radial extend of each root-system (root radius).

Foliar Nutrients

To assess foliar nutrient concentrations, leaves were collected in May 2013 (pine and spruce only) and in August of 2013 and 2014 (all three species). Leaves from 9-16 seedlings were collected in the single-species tree plots (but outside the sub-plots) and combined into one sample per tree plot. Aspen leaves were randomly collected from within the entire crown, while pine and spruce needles were collected from the current year growth of the terminal leader of branches. Leaves and needles were then cold stored in the field and then frozen in the laboratory before processing. They were dried at 70°C to constant weight and subsequently ground with a Wiley mill to 40 mesh (0.4mm) and sent for nutrient analysis to a commercial laboratory (Natural Resources Analytical Laboratory (NRAL), University of Alberta, Edmonton, AB, Canada). The samples were analyzed for, aluminum (Al), boron (B), calcium (Ca), copper (Cu), iron (Fe), potassium (K), magnesium (Mg), manganese (Mn), phosphorous (P), sulfur (S), total nitrogen (N), and zinc (Zn) concentrations. Nitrogen was measured by combustion while all other nutrients were measured by microwave digestion (using HNO_3) followed by inductively coupled plasma optical emission spectrometry (ICP-OES). In order to identify possible nutrient deficiencies, those values were compared to foliar nutrient concentrations indicated by Paré et al. 2013.

Vegetation

Colonizing vegetation was assessed in each tree plot by measuring percent cover and the presence/absence of species. In each tree plot two vegetation sub-plots (1.41m × 1.41m) were established, one in the northeast and one in the southwest corner (Figure 1.3). Percent cover was assessed for each individual species in July 2013 and 2015. Percent cover was measured to the nearest 1% when less than 10%, otherwise it was rounded to the nearest 5% (Macdonald et al. 2015b). Presence/absence of species was assessed by performing walkthroughs of the entire tree plot. Of the 119-species observed across the ASCS, 16 could not be identified to species, in these cases specimens were given a consecutive number behind the Genus (ex. Salix sp1) and the sample was kept on file to allow for comparison with other unidentified specimens. All scientific species nomenclature is based on Flora of Alberta (Moss 1983) and lists of all species identified at the ASCS are provided in Table C3, Table C4, and Table C5.

Figure 1.3: Tree plot (25m x 25m) layout. Dashed lines show tree measurement plots and solid lines represent vegetation measurement quadrats.

Statistical Analyses

All analyses, unless otherwise mentioned, were executed using R software, v 3.4.0, 64 bit (R Core Team 2018).

To analyze the effects of all 13 treatments on seedling height in 2016 of aspen, pine, and spruce as well as on colonizing vegetation cover and species richness in 2015, linear mixed effect models (LMM), with treatment as the fixed effect and cell as the random effect, were used with α=0.1 in the R *nlme* package (v 3.1-131; Pinheiro et al. 2017). When significant effects were found, pair-wise comparisons were done with a Holm-Bonferonni adjusted α using the contrast function from the R *car* package (v 2.1-6; Fox and Weisberg 2017). Model residuals were tested for normality using the Shapiro-Wilk test from the R *stats* package and homogeneity of variance using Levene's test in the R *car* package (v 2.1-6; Fox and Weisberg 2017); when data did not meet assumptions of normality or homogeneity they were logarithmically transformed.

Soil Characteristics

Comparison of all initial soil physical and chemical characteristics between the seven types of soil materials at the ASCS were done with two-way ANOVAs based on a full factorial design with α=0.1 in the R *stats* package (v 3.4; R Foundation for Statistical Computing, Vienna, Austria). Pair-wise comparisons were done with a Bonferroni adjusted α in the R *agricolae* package (v.1.2-3; de Mendiburu 2015). The ANOVA residuals were tested with the same analyses mentioned above. Two-way ANOVAs were also used to compare the average seasonal soil temperature, soil temperature, volumetric water content (VWC), and the number of days with an average soil temperature above 5°C in 2013 and 2014 between the treatments with FFM and Peat as the coversoils. This analysis was also done comparing average growing season soil temperature and number of days with an average soil temperature above 5°C in 2013 and 2014 in treatments with 10cm of Peat, 30cm of Peat, 10cm of FFM, and 20cm of FFM. Average growing season water potential (averaged from 2013-2015) in single-species tree plots in both the Peat and FFM treatments were also compared with a two-way ANOVA.

Differences in overall soil characteristics (initial soil nutrients, physical characteristics, and soil nutrients in 2013) between the treatments was analyzed using a permutational multivariate ANOVA (PERMANOVA) with a Benjamini and Hochberg adjusted α for pair-wise comparisons in the R *vegan* package (v 3.2.2; Oksanen et al. 2015). This analysis was run twice, once comparing the four coversoil material types and again comparing the characteristics of subsoil materials underlying both FFM and Peat coversoils. For these analyses, treatment was used as the fixed effect and cell as the random effect. Soil characteristics were also used in non-metric multidimensional scaling (NMDS) analysis to produce ordinations; the NMDS were executed on untransformed data matrices with a random starting configuration, the Bray distance measure, and the Wisconsin-style double standardized scaling. The final NMDS graphs used two dimensions and included ellipses showing 90% confidence intervals around the centroid of each group and vectors showing characteristics significantly associated with the ordination.

Seedling Performance

To test the effects of coversoil material type, coversoil placement depth, underlying subsoil in treatments with Peat, underlying subsoil in treatments with FFM, and total capping material depth, repeated measure linear mixed effect models (LMM), with treatment as the fixed effect and cell as the random effect, were used to analyze tree height and RCD (of each species separately) in 2016 with α=0.1 in the R *nlme* package (v 3.1-131; Pinheiro et al. 2017). When significant effects were found, pair-wise comparisons were done with a Holm-Bonferonni adjusted α using the contrast function from the R *car* package (v 2.1-6; Fox and Weisberg 2017). Model residuals were tested with the same analyses mentioned above; when data did not meet assumptions of normality or homogeneity they were logarithmically transformed.

Total seedling biomass, LMF, SMF, and RMF, LA, CD, and RR were compared between Peat and FFM treatments (not the two Subsoil coversoils) and analysed with a one-way ANOVA using a full factorial design in the R *stats* package (v 3.4; R Foundation for Statistical Computing, Vienna, Austria). Pair-wise comparisons and testing of model residuals were done the same way as described above in section 1.4.1 and all tests were conducted using α=0.1. Total biomass, LA, CD, and RR were also compared between treatments with 30cm of Peat (treatment 5) and 150cm of total capping material (treatment 1).

To explore and visualize the underlying relationships between reconstructed soil profiles, edaphic factors, and the mean root-length density for the three tree species, principal component analysis (PCA) were run comparing the four coversoil materials for each species separately. The edaphic variables included nutrient availability data from the PRS™-probes averaged for the entire soil profile in each main plot, growing season averages (2013-2015) of soil water potential and soil temperature, and the mean root-length densities observed in each single-species tree plot in 2015. The three-year averages were chosen because they eliminate some of the annual variability and are thus a better representation of the average soil conditions under which the roots were developing. The PCA was conducted in SPSS 20.0 (IBM Corp., 2011) using the Kaiser-Meyer-Olkin measure of sampling adequacy and Bartlett's test of sphericity to test underlying assumptions. The direct oblimin technique was used to rotate the data and only principal components with eigenvalues > 1 were selected. For visualization purposes, the analysis was repeated in R using the princomp function in the *stats* package (v 3.4; R Foundation for Statistical Computing, Vienna, Austria). The resulting ordination was overlaid with vectors, representing edaphic factors and root-densities for all species, as well as ellipses representing ordination space in the R *vegan* package (v 2.4-4; Oksanen et al. 2017).

Differences in foliar nutrient concentrations among soil treatments for each tree species were analyzed using NMDS (see section 1.4.1 for description of analysis). The final graphs for each tree species also used two dimensions, ellipses showing the 90% confidence intervals around the centroid of each group, and vectors showing foliar nutrients that were significantly associated with the ordination. NMDS were made for seedlings in the four coversoil materials, in Peat treatments with different underlying subsoil materials, and FFM treatments with different underlying subsoil materials.

In order to get a broad idea of what site factors were driving differences in tree performance, tree height in 2016 was also analyzed with a regression tree analysis in the R *mvpart* package (v 1.6-1; Therneau et al. 2013). The predictor dataset consisted of 107-122 variables (depending on the species), including initial soil nutrients and physical characteristics, soil nutrients from PRS probes in 2013, soil temperature and water content during the 2013-2016 growing seasons, and various foliar nutrients (Table B1). Regression tree analysis was run in R software, v 2.15.3, 64 bit (R Core Team 2013). This analysis was run for each tree species separately and was done comparing the different coversoil materials, Peat treatments with different underlying subsoil materials, FFM treatments with different underlying subsoil materials, and treatments with different total capping material depths.

Linear mixed-effect models were also used to compare the effect of planting composition and planting density on tree performance in 2016. However, while cell was still the random factor for these analyses, the fixed effects were coversoil material type (FFM and Peat) and either planting composition (single or mixed) or planting density (high or low). While height was used as the response variable for the analysis comparing the impact of planting density, for the effect of planting density, the response variable was a ratio calculated for each tree species comparing the average tree height in mixed plots to average height in single-species plots.

Colonizing Vegetation

Differences in colonizing vegetation cover and species richness between the four coversoil materials were analyzed with a LMM model in the R *nlme* package (v 3.1-131; Pinheiro et al. 2017) as a repeated measures split-plot design. In the model, the repeated measures fixed effect was year, the main-plot fixed effect was treatment, the split-plot fixed effect was the planted tree species, and cell was the random factor. Cover and richness data that did not meet the assumptions of normality and homogeneity and was either logarithmically or boxcox transformed prior to analyses. When statistically significant effects were found at $\alpha=0.1$, pair-wise comparisons were conducted using the Holm α adjustment method. As the split-plot fixed effect in this analysis was the planted tree species, this analysis was used for comparing impact of the planted tree seedlings on colonizing vegetation cover and richness.

The vegetation cover community in the four coversoil materials was analyzed with a PERMANOVA in the R *vegan* package (v 2.4-4; Oksanen et al. 2017); year, treatment, and planted tree species were the fixed effects and cell the random effect. When statistically significant effects were found at $\alpha=0.1$, pair-wise comparisons were done using the Holm α adjustment method. Indicator species analysis was run to determine which species were representative of the significant relationships identified in the PERMANOVA with the *labdsv* package (v 1.8-0; Roberts 2016) with an $\alpha=0.1$. Community composition was also analyzed by running a NMDS ordination on the vegetation cover data. This NMDS was run in R the *vegan* package (v 2.4-4; Oksanen et al. 2017) using the same parameters are discussed in above in section 1.4.1 with vectors showing species significantly associated with the ordination at $\alpha=0.05$.

Differences in vegetation cover and richness in treatments with different Peat depth, different FFM depth, Peat treatments with different underlying subsoil material, FFM treatments with different underlying subsoil material, and treatments with different total capping material depth were analyzed using the same methods as described above; however, they were only analyzed on the data collected in 2015. As a result, the models were not repeated measures analyses and there was no fixed effect of year.

The effect of planting density on colonizing vegetation cover and richness was also only run on 2015 data; however, the effect of soil treatment was eliminated by analyzing plots in FFM and Peat treatments separately. The fixed effects for all vegetation analyses for this question were tree species and planting density, cell was still the random effect.

Results

Following five growing seasons, there were marked differences in tree and colonization vegetation performance between the 13 treatments at the ASCS. Aspen grew the tallest in treatments with FFM as the coversoil material (Figure 1.4a). Treatments with no coversoil material or 30cm of Peat as the coversoil materil resulted in the shortest aspen seedlings, although some 30cm Peat treatments had aspen seedlings with intermediate heights following five growing seasons (Figure 1.4a). Jack pine also generally grew taller in treatments with FFM as the coversoil material and shorter in treatments with no coversoil; however, the shortest pine trees were found in the treatment with 30cm of Peat placed directly over overburden (Figure 1.4b). There was less variability in white spruce height between the three treatments compared to the aspen and pine responses; however, the shortest spruce trees were also found in the treatments with no coversoil material and the treatment with 30cm of Peat placed directly on overburden (Figure 1.4c). After four growing seasons, colonizing vegetation cover and species richness were higher in the treatments with FFM as the coversoil material than those with Peat as the coversoil or no coversoil at all (Figure 1.5).

Figure 1.4: Average and standard error of tree seedling height (trembling aspen (a), jack pine (b), white spruce (c)) in 2016 in all 13 soil layering treatments at the ASCS (n=3). A break down of all 13 treatments can be found in Figure 1.2 (p 8) and Table 1.2 (p. 87). Uppercase letters indicate significant differences within the heights of each species (α=0.1) following pair-wise comparisons that were adjusted using the Holms-Bonferonni method.

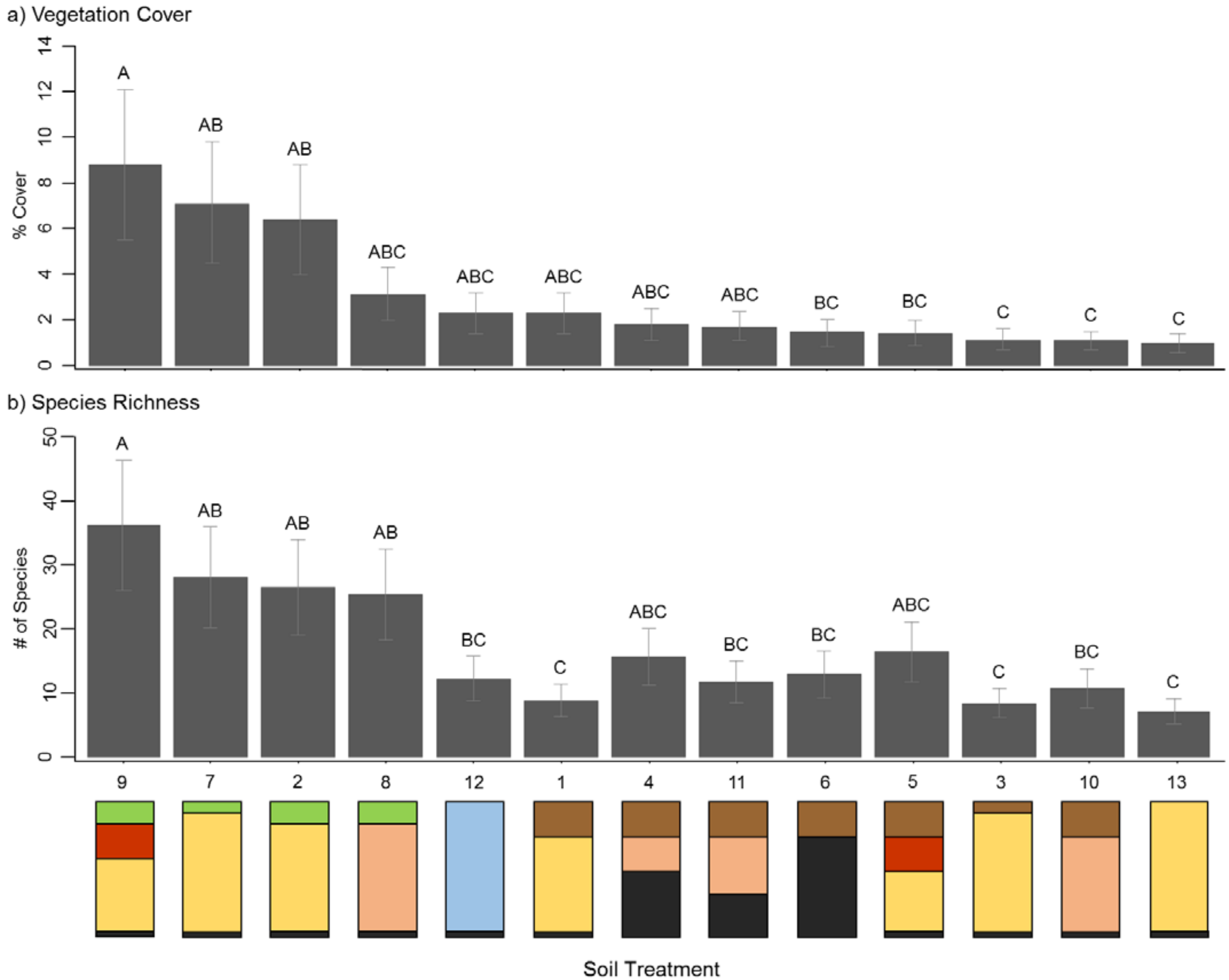


Figure 1.5: Lsmeans adjusted average colonizaing vegetation cover (a) and richness (b) in 2015 in all 13 soil layering treatments at the ASCS (n=3). Bars represent 90% confidence intervals. A break down of all 13 treatments can be found in Figure 1.2 (p. 8) and Table 1.2 (p. 87). Uppercase letters indicate significant difference (α=0.1) following pair-wise comparisons that were adjusted using the Holms-Bonferonni method.

Coversoil Material Type

Figure 1.6: To compare effects of coversoil material type on tree performance and colonizing vegetation development we compared treatment 1 (30cm of Peat over 120cm of Subsoil C); treatment 7 (20cm of FFM over 130cm of Subsoil C); treatment 12 (150cm of Subsoil Bm2); and treatment 13 (150cm of Subsoil C). Treatments 1 and 7 were chosen as they represent current operational procedures. All treatments were placed over lean oil sands (LOS) overburden material. Physical and chemical characteristics of these materials are presented in Table 1.2.

Soil Characteristics

We chose the 30 cm and 20 cm application of coversoil for Peat and FFM, respectively as they reflect the current operational procedures for placing coversoils on reclamation sites. When comparing the four coversoil materials (Figure 1.6), the 30cm Peat had lower annual and seasonal soil temperatures at 15cm depth than the 20cm FFM ($p < 0.05$) in 2013 and 2014 (Table 1.1). In these two growing seasons, the Peat treatment was on average 2.6°C colder than the FFM treatment; as a result the Peat material had 11 fewer days where the daily average soil temperature was above 5°C ($p < 0.005$; Table 1.1). The subsoils were not significantly different from the FFM (data not shown).

Volumetric soil water content (VWC) at 15cm depth was also significantly higher in the 30cm Peat than the 20cm FFM in both 2013 and 2014 ($p < 0.001$); although there were no significant differences between the two years. In the 2015 and 2016 growing seasons, water content at 15cm in the Peat material never dropped below the wilting point for plants (25% vwc; Ojekanmi and Chang 2014), whereas water content in the sandy FFM and Subsoil Bm2 treatments dropped below the wilting point (5% vwc; Saxton and Rawls 2006) for extended periods (Figure 1.7). Water content in the Subsoil C treatment came close to the wilting point several times during the growing seasons in 2015 and 2016, but never dropped below it (Figure 1.7).

There were several significantly different physical and chemical characteristics among the four coversoil materials (Table 1.2). The NMDS shows that all soils were significantly different from one another ($p < 0.1$). Peat material was associated with higher soil Zn, S, Fe, and NO_3^- concentrations and higher soil EC; Subsoil Bm2 was associated with higher soil pH, Cu concentrations, and bulk density; Subsoil C was also associated with higher bulk density; while FFM was associated with higher soil NH_4^+, K, and P concentrations (Figure 1.8).

Aspen Response

While aspen seedlings in all four coversoil materials grew similar amounts in the first growing season, by the second growing season trees in the FFM and Subsoil Bm2 grew substantially more than in the Peat and Subsoil C (Figure 1.9a). In 2015, a notably dry growing season, growth in the FFM and both Subsoils decreased, while it continued to increase in the Peat (Figure 1.9a). Aspen seedlings were significantly taller in the FFM than the other three coversoil materials after the fifth growing season ($p < 0.01$; Figure 1.9b). Trees in Subsoil C were significantly shorter than all other treatments in 2016 ($p < 0.01$; Figure 1.9b). Trees in the Peat and Subsoil Bm2 were not significantly different from one another after five growing seasons. Root collar diameter followed a similar trend (data not shown).

After three growing seasons, aspen seedlings growing in FFM had significantly more biomass, more than double the leaf area, and twice as wide crowns than seedlings growing in Peat. Aspen growing on FFM also allocated relatively more to leaves and roots, but less to stems than aspen growing on Peat (Figure 1.10). Biomass allocation to leaves (LMF) was significantly greater on Peat than on FFM ($p < .05$). However, SMF, RMF, and root system radii in aspen seedlings were similar between the two coversoil materials (Table 1.3).

Aspen trees in FFM were associated with high levels of foliar Mg (Figure 1.11). Trees in Subsoil C were associated with high P foliar concentrations in 2013, whereas aspen trees in Subsoil Bm2 were not significantly associated with any foliar nutrients. Foliar nutrients of aspen trees in the Peat was associated with high foliar concentrations of Zn, B, and N:P in 2013 as well as B in 2014. There was more variability in foliar nutrients in the Peat material than in the FFM and Subsoil Bm2 treatments (larger ellipses; Figure 1.11).

Aspen mean root length density (RLD) did not significantly differ between the Peat and FFM after three growing seasons (Figure 1.12a). However, principal components analysis (PCA) showed a positive correlation between aspen RLD and P and K availability as well as warmer soil temperatures associated with FFM (Figure 1.13). Coversoil material type had a strong impact on aspen root distribution ($p < 0.01$; Figure 1.10b). Aspen growing on Peat had a clear majority (~60%) of their total observed root length in the coversoil horizon, whereas aspen growing on FFM only had about 20% of their total root length in the coversoil horizon (Figure 1.12b). Root systems of aspen grew much deeper in FFM as the 90% quantile depth, i.e. the soil depth that contained 90% of the observed total root length, was significantly greater ($p < 0.05$) for aspen growing on FFM (~70 cm) than on Peat (~40 cm; Figure 1.12c).

When aspen total height in 2016 was analyzed with a regression tree, three significant splits were found to best fit the data (Figure 1.14; error=0.1, CV error=1.05, SE=0.47). The first split was driven by nutrients; specifically, high concentrations of both foliar and soil Mn, initial soil P concentrations, and low soil Ca concentrations were drivers of the first split and were associated with the tallest aspen trees. The second split was also driven by soil nutrients, with the smallest aspen trees associated with having high foliar Al, Fe, and N:K, as well as low initial soil K and EC. The third and final split was again driven by foliar P as well as water content (WC) in August 2014, with the taller trees in this split being in cells with high soil P and low WC (Figure 1.14).

Pine Response

Pine seedlings in the FFM grew more annually than seedlings in the other three coversoils (Figure 1.15a). Whereas aspen seedlings decreased in growth in the sandy treatments during the dry 2015 growing season, pine seedlings continued to have positive growth in 2015 (Figure 1.15a). However, sharp decreases in growth in all four treatments was observed in 2016 (Figure 1.15a). After the fifth growing season (2016), pine seedlings were significantly taller in the FFM than trees in the three other coversoils ($p < 0.1$; Figure 1.15b). Trees in Peat and Subsoil C were not different in height, but seedlings in Peat were significantly taller than those in Subsoil Bm2 ($p < 0.5$; Figure 1.15b). Root collar diameter followed a similar trend (data not shown).

Pine seedlings had significantly greater total biomass on FFM than when growing on Peat after three growing seasons ($p < .05$). Allocation to needles, stem, and roots was not significantly different between FFM and Peat (Table 1.3). The crowns of pine growing on FFM were only 25% wider than those on Peat; however, pine on FFM had nearly double the leaf area than pine on Peat. Similar to the response shown by aspen, pine growing on FFM also allocated relatively more to leaves and roots, but less to stems than pine growing on Peat (Figure 1.10).

Pine trees in Subsoil Bm2 were associated with high foliar Ca concentrations in 2013 while trees in Subsoil C were associated with high 2013 foliar Fe and Al concentrations. Trees in Peat were significantly associated with high concentrations of N in 2014 and high N:P in both 2013 and 2014. Pine trees in FFM were significantly associated with high Zn concentrations in 2013 and high foliar Mn concentrations in fall 2014 and spring 2013 (Figure 1.16).

After three growing seasons, pine seedlings on FFM had significantly greater ($p < 0.05$) mean RLD than pine growing on Peat (Figure 1.12a). Principal components analysis showed a strong positive correlation between pine RLD, P and K availability, as well as warmer soil temperatures associated with FFM coversoil (Figure 1.13). Coversoil material type had a significant impact on pine root distribution ($p < 0.01$). Pine growing on Peat had a clear majority (~60%) of the total observed root length in the first coversoil horizon. In contrast, pine growing on FFM only had about 20% of their total root length in the coversoil horizon (Figure 1.12b). The 90% quantile depth, i.e. the soil depth that contained 90% of the observed total root length, was significantly greater ($p < 0.01$) for pine growing on FFM (~70 cm) than on Peat (~40 cm; Figure 1.12c).

Regression tree analysis indicates two splits that were found to best fit the data (Figure 1.17; error=0.2, CV error=0.58, SE=0.17). The first split was driven by foliar Fe concentrations at the end of the 2013 growing season and by the coversoil temperature in June 2015. Lower concentrations and temperatures were associated with taller pine trees. The second split was driven by soil temperature and nutrients. Within this split, taller trees were associated with cooler coversoil temperatures in July 2013 and August 2013 and 2015, high foliar B concentrations at the end of the 2014 growing season, and high soil Mn availability in 2013 (Figure 1.17).

Spruce Response

Annual growth was greater for spruce seedlings in FFM than the other three coversoils for the first three growing seasons (Figure 1.18a). In 2015 and 2016, growth for spruce seedlings were highest in Peat (Figure 1.18a). In 2016, spruce trees in FFM and Peat were significantly taller than trees in both Subsoils ($p < 0.001$; Figure 1.18b). Trees in Peat and FFM were not significantly different and neither were the trees in the two Subsoils (Figure 1.18b). Root collar diameter followed a similar trend (data not shown).

Spruce seedling mass, crown diameter, leaf area, SMF, and RMF did not significantly differ between treatments with either FFM or Peat as the coversoil (Table 1.3). However, LMF and mean root system radius were significantly greater in spruce seedlings on FFM than Peat ($p < 0.01$; Table 1.3).

There were few nutrients significantly associated with the NMDS of spruce foliar nutrients. High concentrations of foliar N and Mn in 2013 and Zn in 2014 were significantly associated with spruce trees in the FFM (Figure 1.19). There were no foliar nutrients significantly associated with spruce trees in the Peat or Subsoil treatments.

After three growing seasons, spruce mean RLD did not significantly differ between Peat or FFM (Figure 1.12a). Principal components analysis showed no clear correlation between spruce RLD and the abiotic conditions associated with either coversoil material (Figure 1.13). Coversoil material type had a significant impact on spruce root distribution ($p < 0.001$). Spruce growing on Peat had about 60% of the total observed root length in the coversoil horizon. In contrast, spruce growing on FFM only had about 25% of their total root length in the first horizon (Figure 1.12b). The 90% quantile depth, i.e. the soil depth that contained 90% of the observed total root length, did not differ among spruce growing in either the FFM or Peat treatment (Figure 1.12c).

The regression tree analysis of total spruce tree height in 2016 identified only one split that best fit the data. (Figure 1.20; error=0.1, CV error=0.75, SE=0.64). The split was driven by a mixture of soil temperatures, soil physical characteristics, and both foliar and soil nutrients (Figure 1.20). Taller trees were associated with cooler coversoil temperatures in May 2013 and August 2016, low coversoil bulk density, high soil total N, and high foliar N:K.

Colonizing Vegetation Response

Cover of the colonizing vegetation significantly increased in Peat ($p < 0.001$) and significantly decreased in FFM ($p < 0.05$) between 2013 and 2015, while total cover did not significantly change in either of the Subsoil treatments (year:coversoil $p < 0.001$; Table 1.4). Although there was a decrease in vegetation cover in 2015, cover was still higher in FFM than in Peat ($p < 0.01$) and both subsoils (both $p < 0.01$). Subsoil C also had significantly lower vegetation cover than Peat and Subsoil Bm2 ($p < 0.05$; Table 1.4).

Species richness significantly increased between 2013 and 2015 in FFM ($p < 0.01$), but did not significantly change Peat or the two Subsoils (Table 1.4). There were no significant differences between richness in Peat and the Subsoils in 2015; however, FFM had significantly more species than the other three coversoils (all $p < 0.001$).

The overall vegetation community was significantly different in 2013 and 2015 across the entire site ($p < 0.001$) and there was a significant interaction between year and treatment ($p < 0.001$). The vegetation communities in FFM and Peat were significantly different between 2013 and 2015 ($p < 0.05$) but did not change between the two Subsoils. In 2015, the vegetation communities in each of the coversoil materials were significant different from one another ($p < 0.005$).

The vegetation community in the FFM was significantly characterized by herbaceous annuals such as *G. bicknelii*, *Polygonum convolvulus*, and *E. angustifolium* as well as graminoids such as *Oryzopsis pungens* in 2013, and shrub species *V. myrtilloides, Rosa acicularis*, and *Prunus pensylvanica* as well as the graminoid *Elymus innovatus* in 2015 (Table 1.5). There were no species significantly associated with the vegetation community in Peat in 2013, although by 2015 *Muhlenbergia glomerata* and *Salsola pestifer* were significant indicator species (Table 1.5). Subsoil Bm2 was significantly associated with *Lepidium densiflorum* and *Melilotus alba*, while Subsoil C had no associated species (Table 1.5).

These trends in the vegetation community are represented in the NMDS by the changes in the size and positions of the ellipses for each coversoil in 2013 and 2015 (Figure 1.21). The ellipses for the 2013 communities in Peat and FFM were larger than their 2015 counterparts. The ellipses for the two Subsoils were not drastically different between the two years. In addition to the species identified in the indicator species analysis, from the NMDS, Subsoil Bm2 was also associated with several graminoids, including *Hordeum jubatum, Agropyron trachycaulum* var. *trachycaulum*, and *Koeleria macrantha*, and Peat with *Salix candida* (Figure 1.21). Forest floor material was associated with several additional species in the NMDS, including *Crepis tectorum*, particularly for the 2013 community, and *Aster laevis* and *Amelanchier alnifolia* for the 2015 community (Figure 1.21).

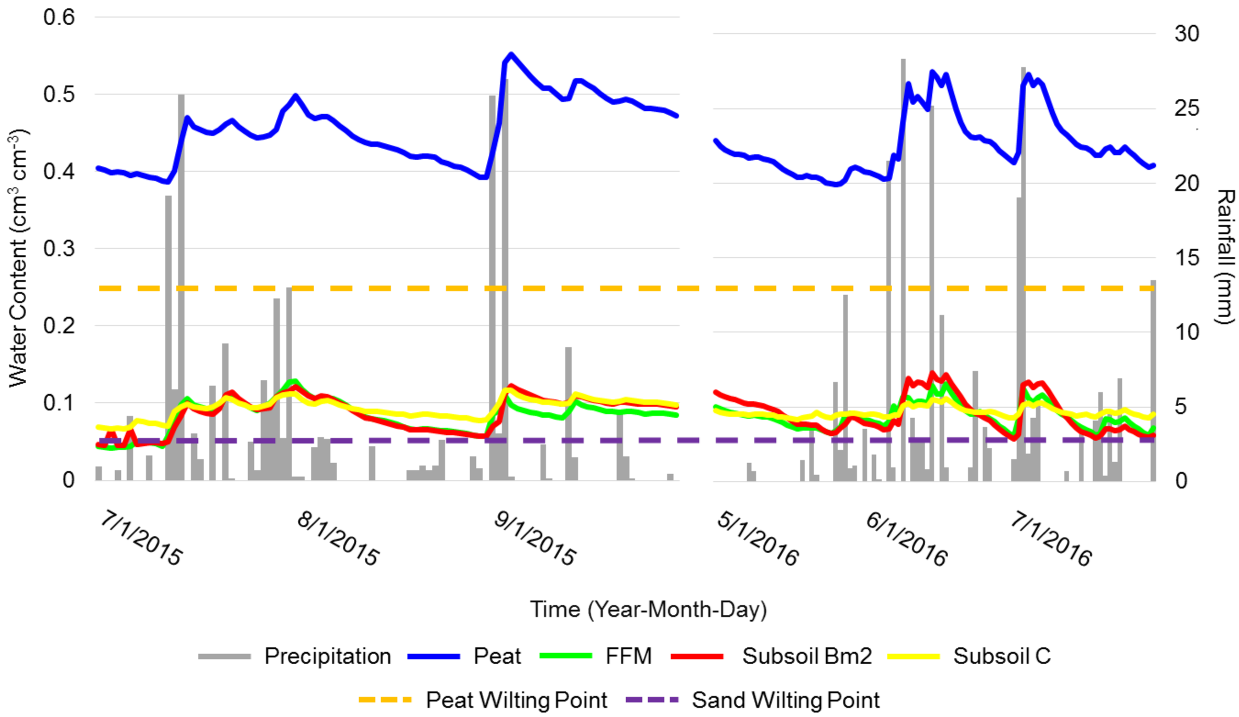


Figure 1.7: Average daily water content 15cm below the surface and rainfall in the 2015 and 2016 growing seasons. Included is the water content wilting points in soils that are predominantly Peat (25%) and sand (5%) (Saxton and Rawls 2006, Ojekanmi and Chang 2014).

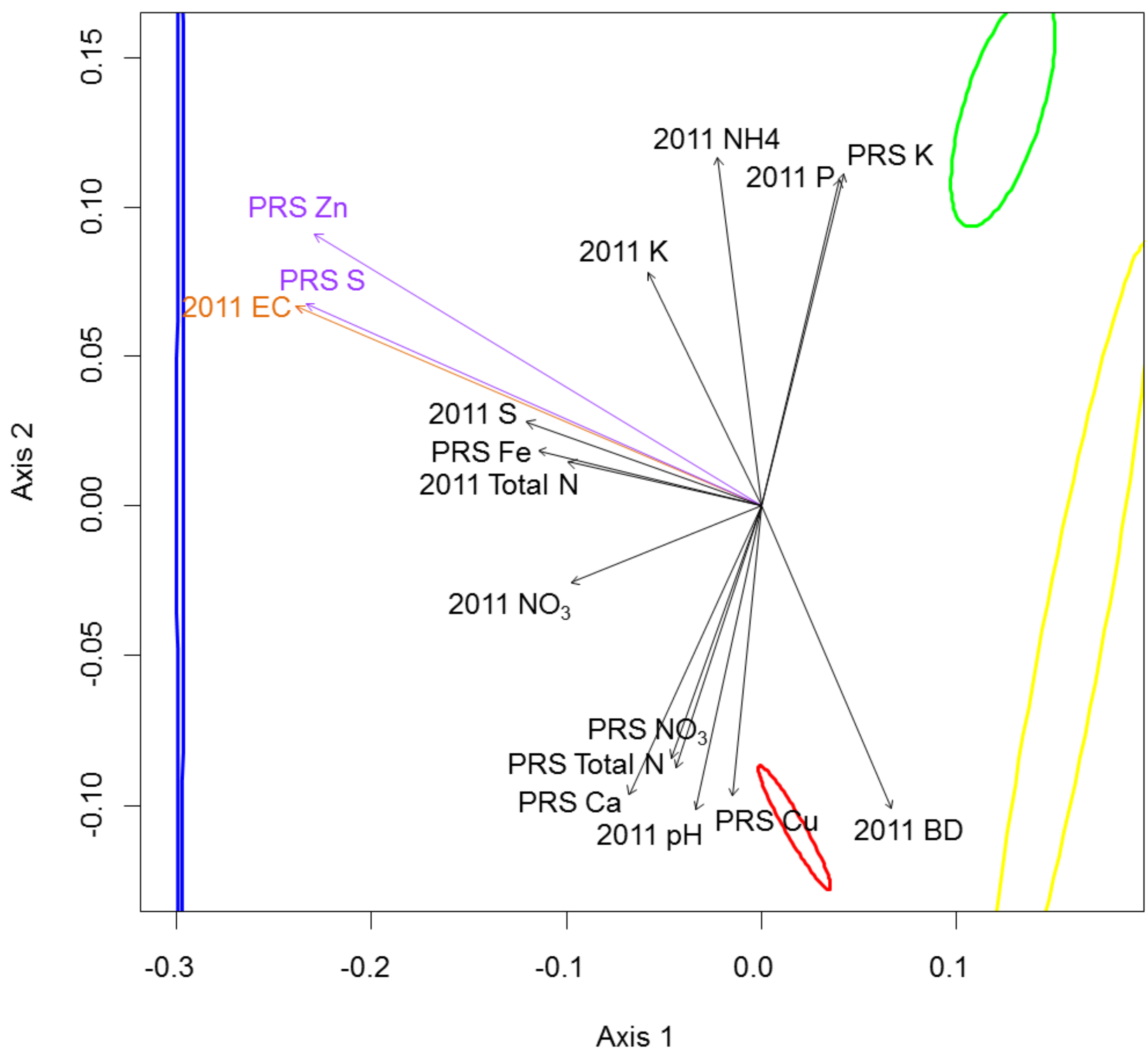


Figure 1.8: NMDS of the physical and chemical characteristics as well as the nutrients (initial and in 2013 of the four coversoil materials. Ellipses indicate a 90% confidence interval for each coversoil (blue=Peat, green=FFM, red=subsoil Bm2, yellow=subsoil C). Arrows indicate characteristics that are significantly associated with the NMDS orientation (orange α=0.0001, purple α=0.001, black α=0.01).

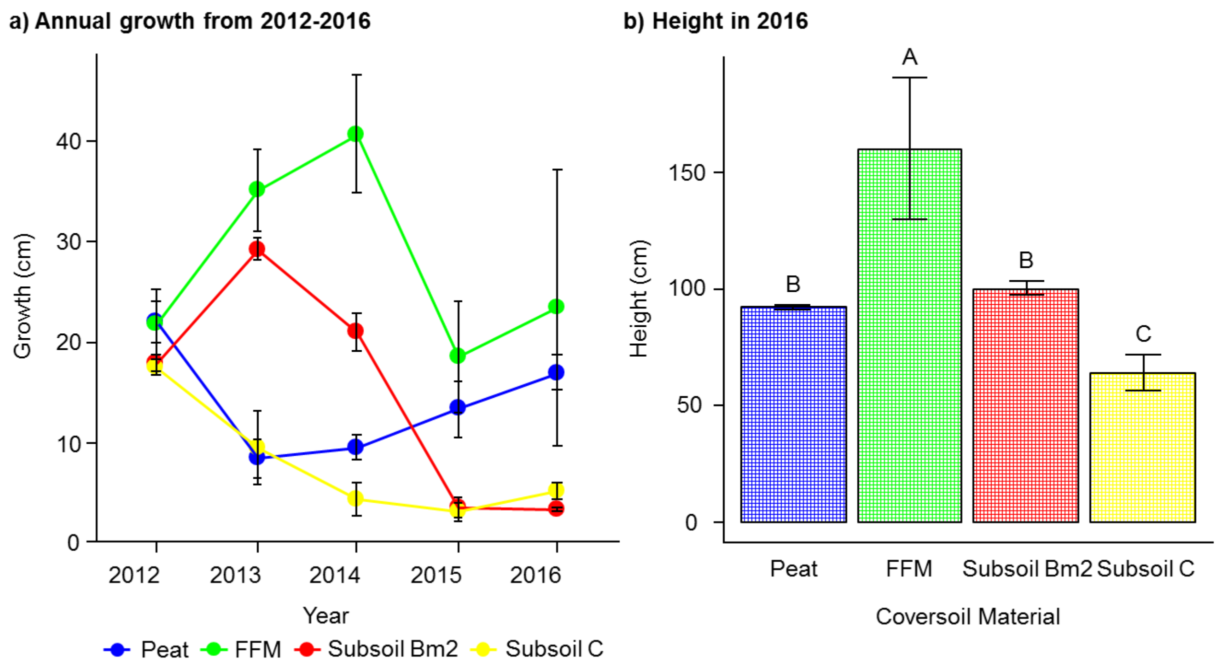


Figure 1.9: Average and standard error of aspen growth in the first five growing seasons (a) as well as height in 2016 (b) in the four coversoil material types (n=3). Uppercase letters indicate significant differences in the 2016 heights (α=0.1) following pair-wise comparisons that were adjusted using the Holms-Bonferonni method.

Figure 1.10: Average leaf mass fraction (LMF), root mass fraction (RMF), and stem mass fraction (SMF; g g-1) for aspen, pine and spruce seedlings in treatments with either FFM or Peat as the coversoil (n=3). Uppercase letters indicate significant differences for each species (α=0.1). Error bars represent 95% CI.

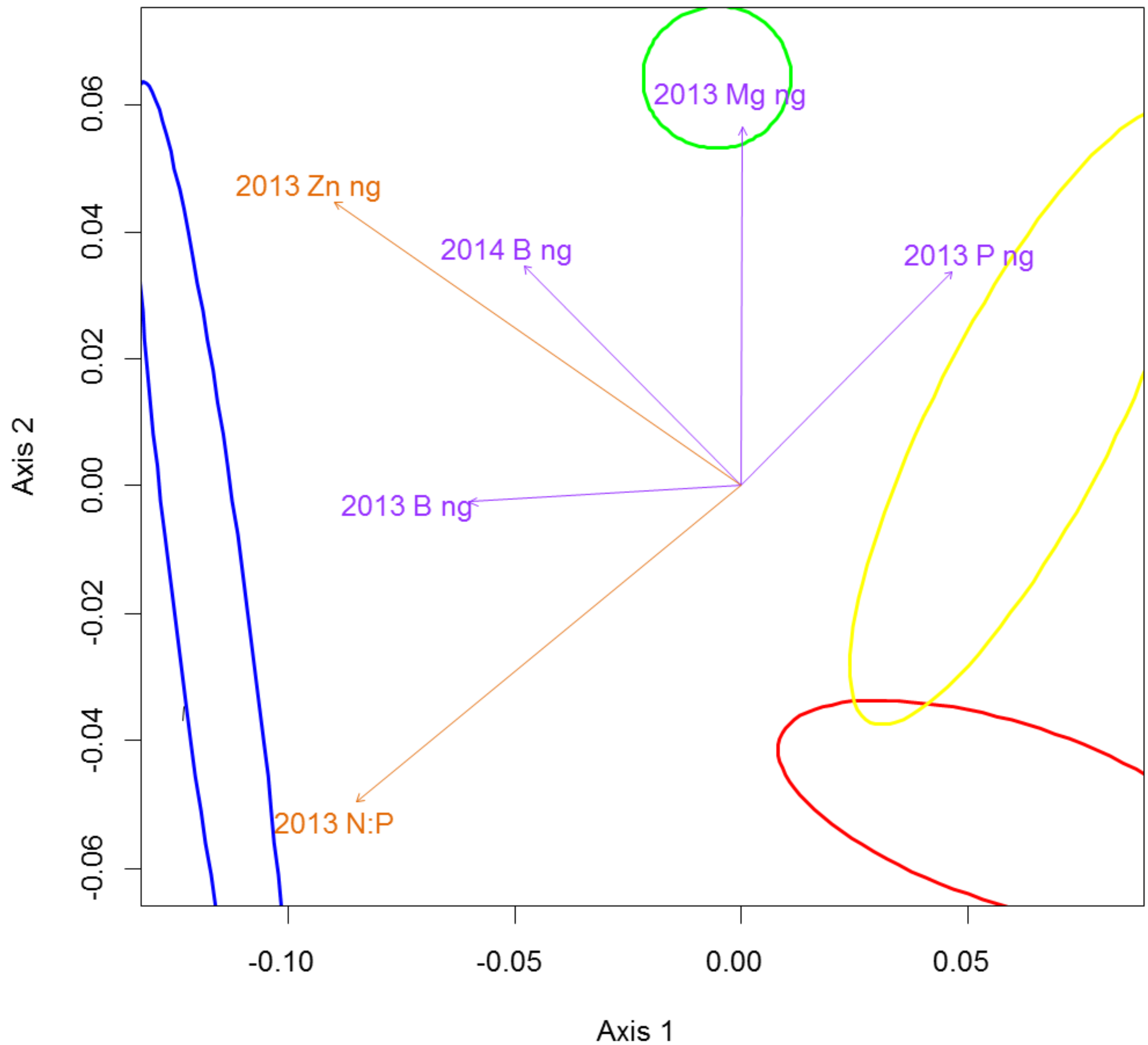


Figure 1.11: NMDS of aspen foliar nutrients in the four coversoil materials. Ellipses indicate 90% confidence intervals for each coversoil (blue=Peat, green=FFM, red=subsoil Bm2, yellow=subsoil C). Arrows indicate foliar nutrients that are significantly associated with the NMDS (orange α=0.0001, purple α=0.001). All 2013 values shown are from the fall collection.

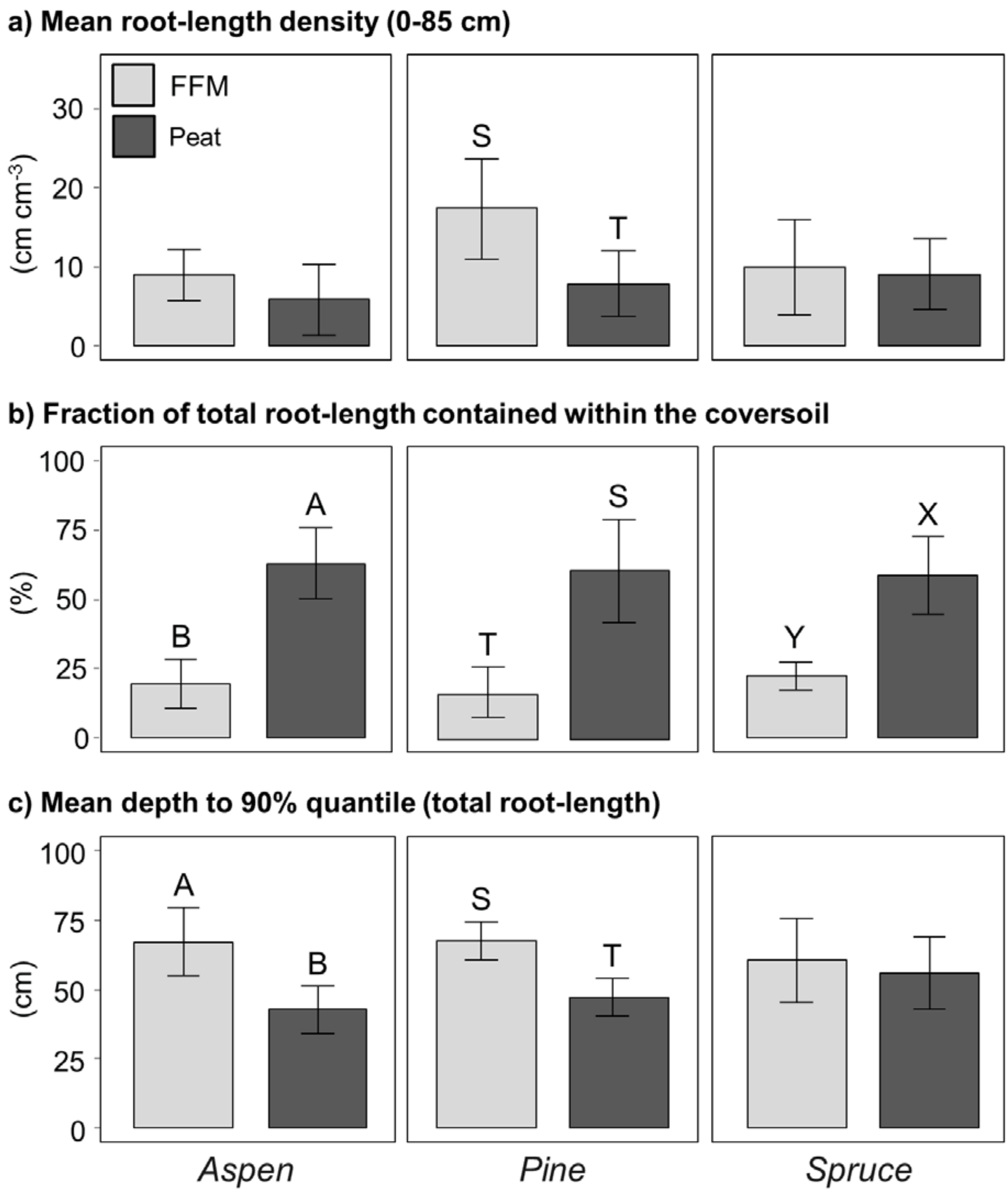


Figure 1.12: Root-length data from minirhizotron images taken at the end of the third (2015) growing season: a) mean root-length densities (cm cm^{-3}) averaged over the entire depth of each minirhizotron (85cm); (b) fraction of the total observed root-length (cm) contained within the coversoil; (c) mean soil depth containing 90% of the total observed root-length (cm). Error bars represent 95% CI (n=3).

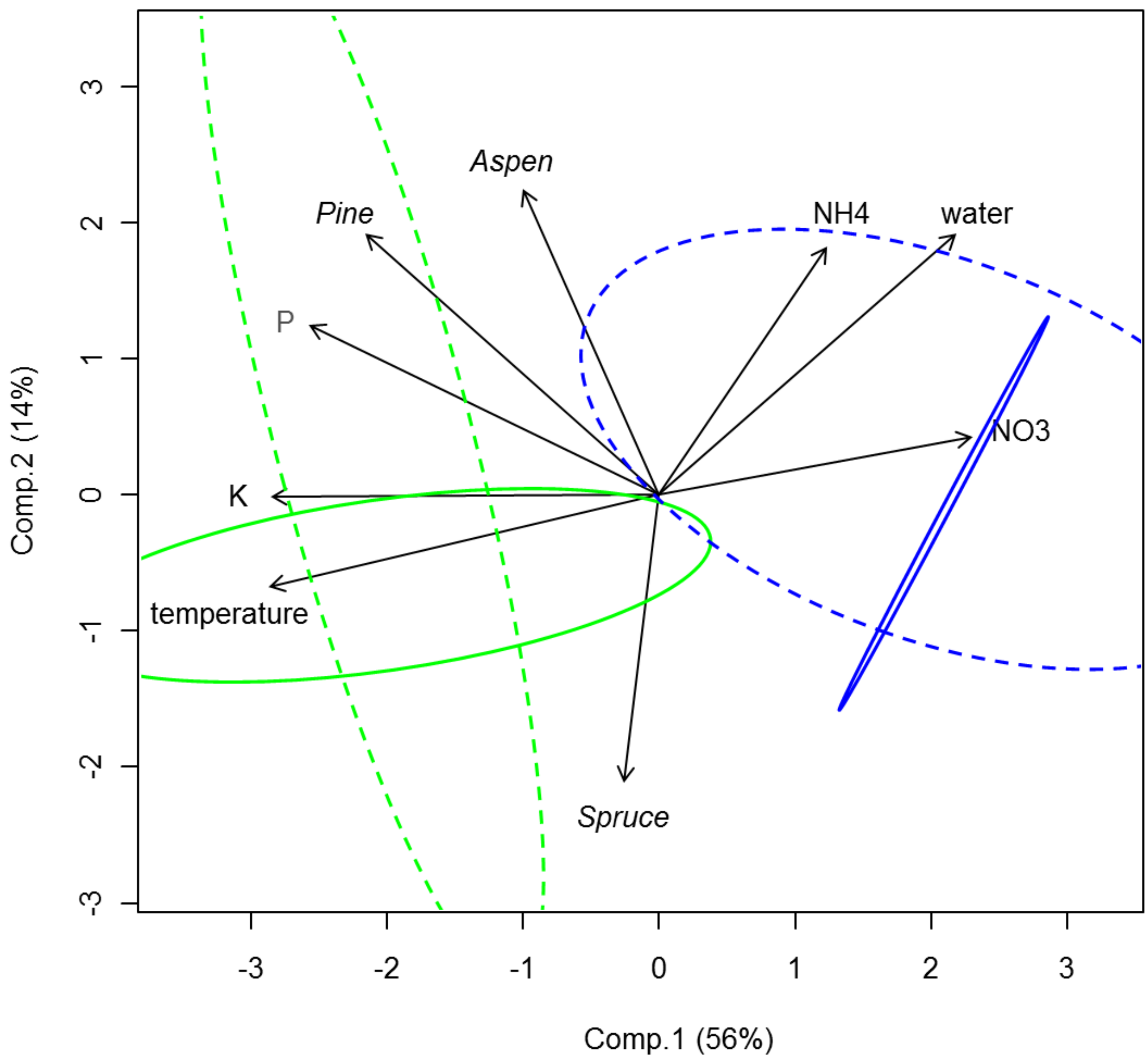


Figure 1.13: Biplot showing the results of a principal component analysis (PCA) linking soil properties associated with the different soil profiles and the observed mean root-length densities for all three tree species after the third growing season (2015). Green denotes treatments with FFM as the coversoil, blue treatments with Peat as the coversoil, dashed lines treatments with Subsoil Bm1 as the underlying subsoil material, and solid lines treatments with Subsoil C as the underlying material.

Figure 1.14: Regression tree of 2016 Aspen heights (cm) in four different coversoil materials. Full descriptions for each of the significant variables shown above can be found in Table B1.

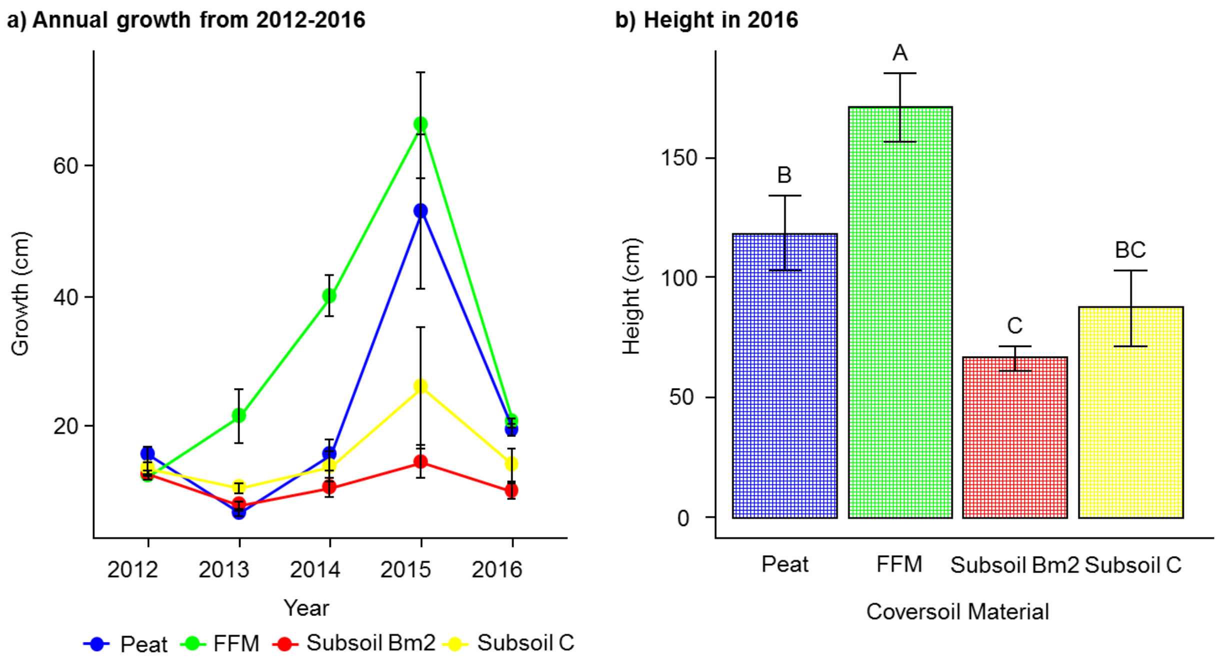


Figure 1.15: Average and standard error of pine growth in the first five growing seasons (a) as well as height in 2016 (b) in the four coversoil material types (n=3). Uppercase letters indicate significant differences in the 2016 heights (α=0.1) following pair-wise comparisons that were adjusted using the Holms-Bonferonni method.

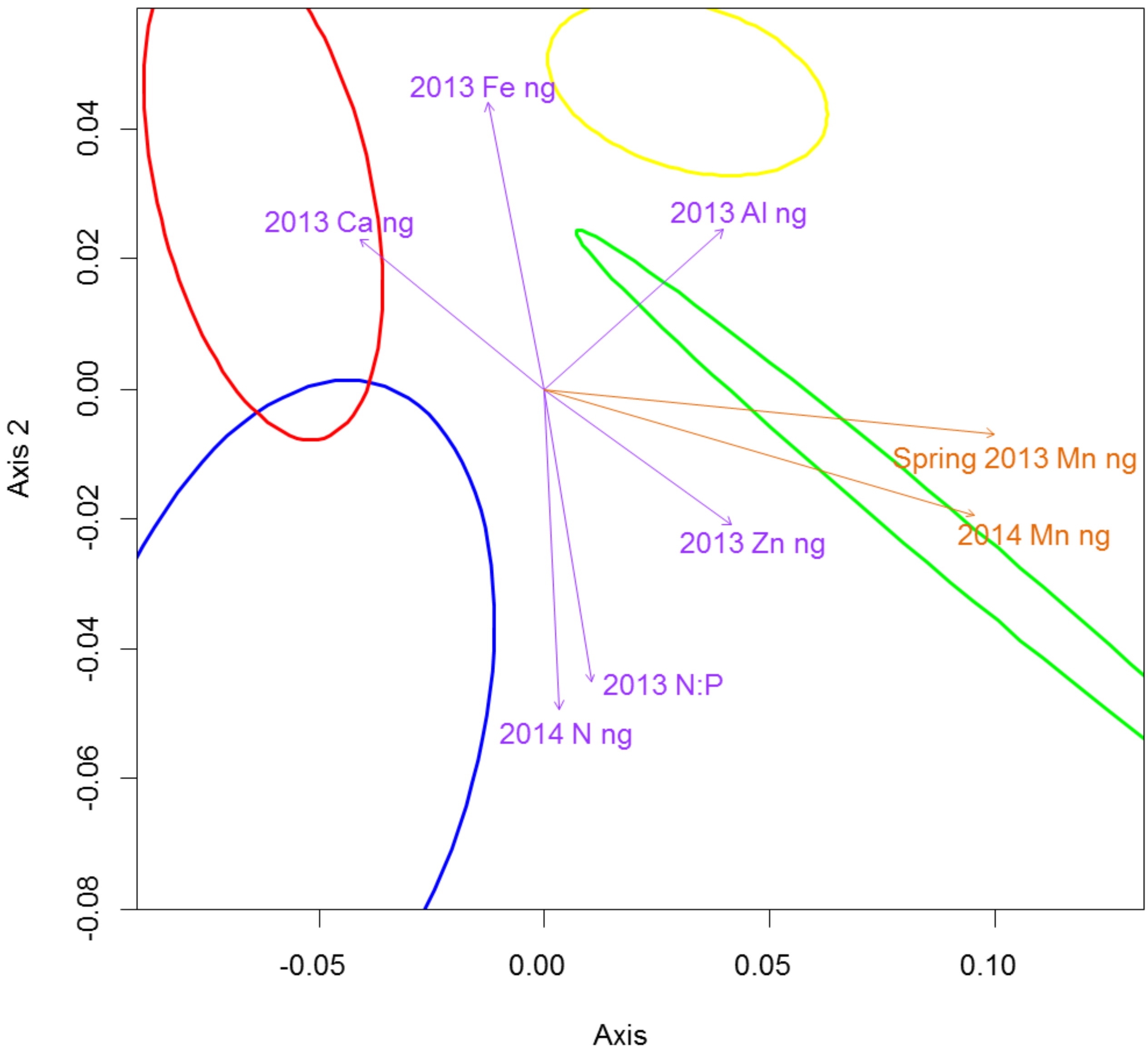


Figure 1.16: NMDS of pine foliar nutrients in the four coversoil materials. Ellipses indicate 90% confidence intervals for each coversoil (blue=Peat, green=FFM, red=subsoil Bm2, yellow=subsoil C). Arrows indicate foliar nutrients that are significantly associated with the NMDS (orange α=0.0001, purple α=0.001). All 2013 values shown are from the fall collection unless specified as being from the spring collection.

Figure 1.17: Regression tree of 2016 Pine heights (cm) in four different coversoil materials. Full descriptions for each of the significant variables shown above can be found in Table B1.

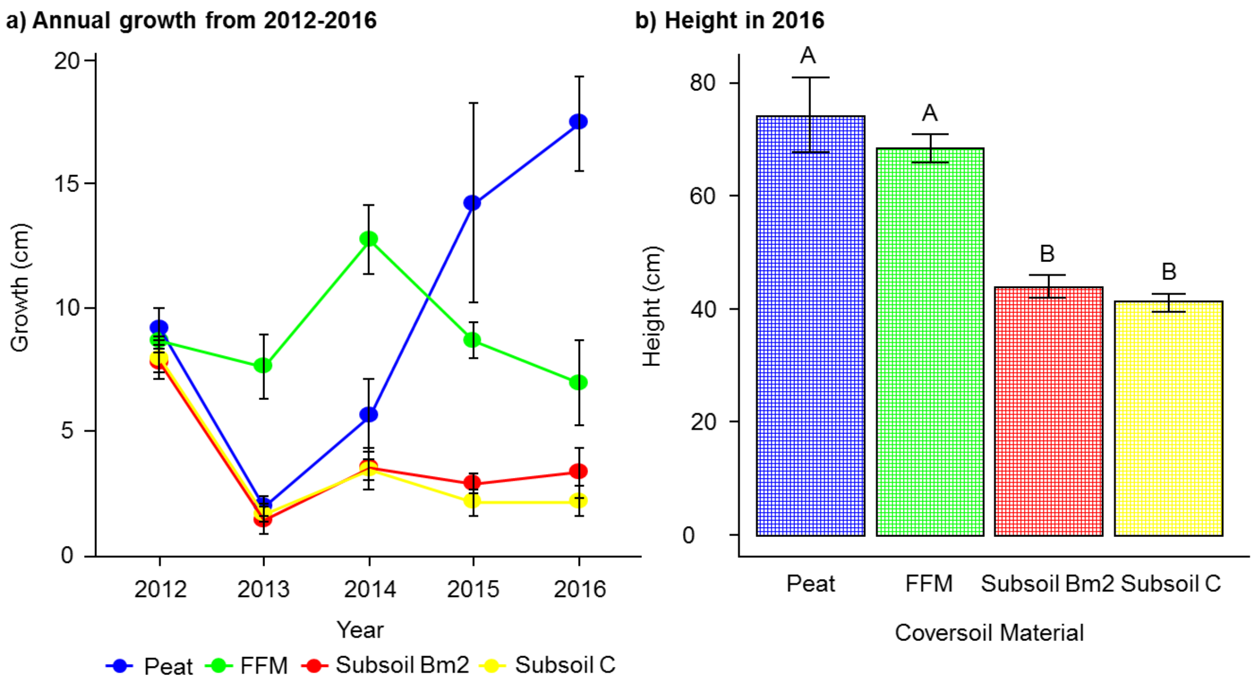


Figure 1.18: Average and standard error of spruce growth in the first five growing seasons (a) as well as height in 2016 (b) in the four coversoil material types (n=3). Uppercase letters indicate significant differences in the 2016 heights (α=0.1) following pair-wise comparisons that were adjusted using the Holms-Bonferonni method.

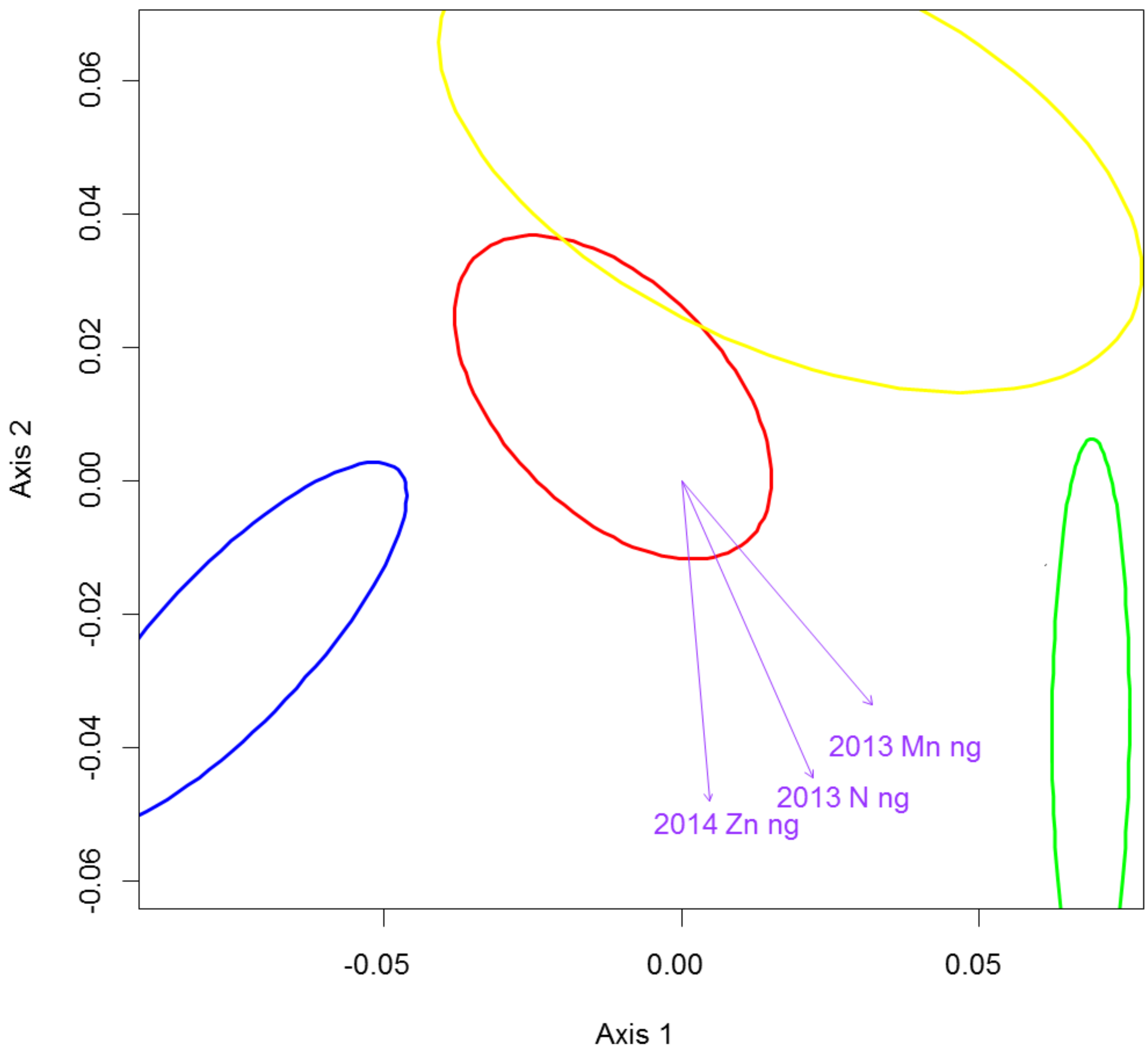


Figure 1.19: NMDS of spruce foliar nutrients in the four coversoil materials. Ellipses indicate 90% confidence intervals for each coversoil (blue=Peat, green=FFM, red=subsoil Bm2, yellow=subsoil C). Arrows indicate foliar nutrients that are significantly associated with the NMDS (α=0.001). All 2013 values shown are from the fall collection.

Figure 1.20: Regression tree of 2016 Spruce heights (cm) in four different coversoil materials. Full descriptions for each of the significant variables shown above can be found in Table B1.

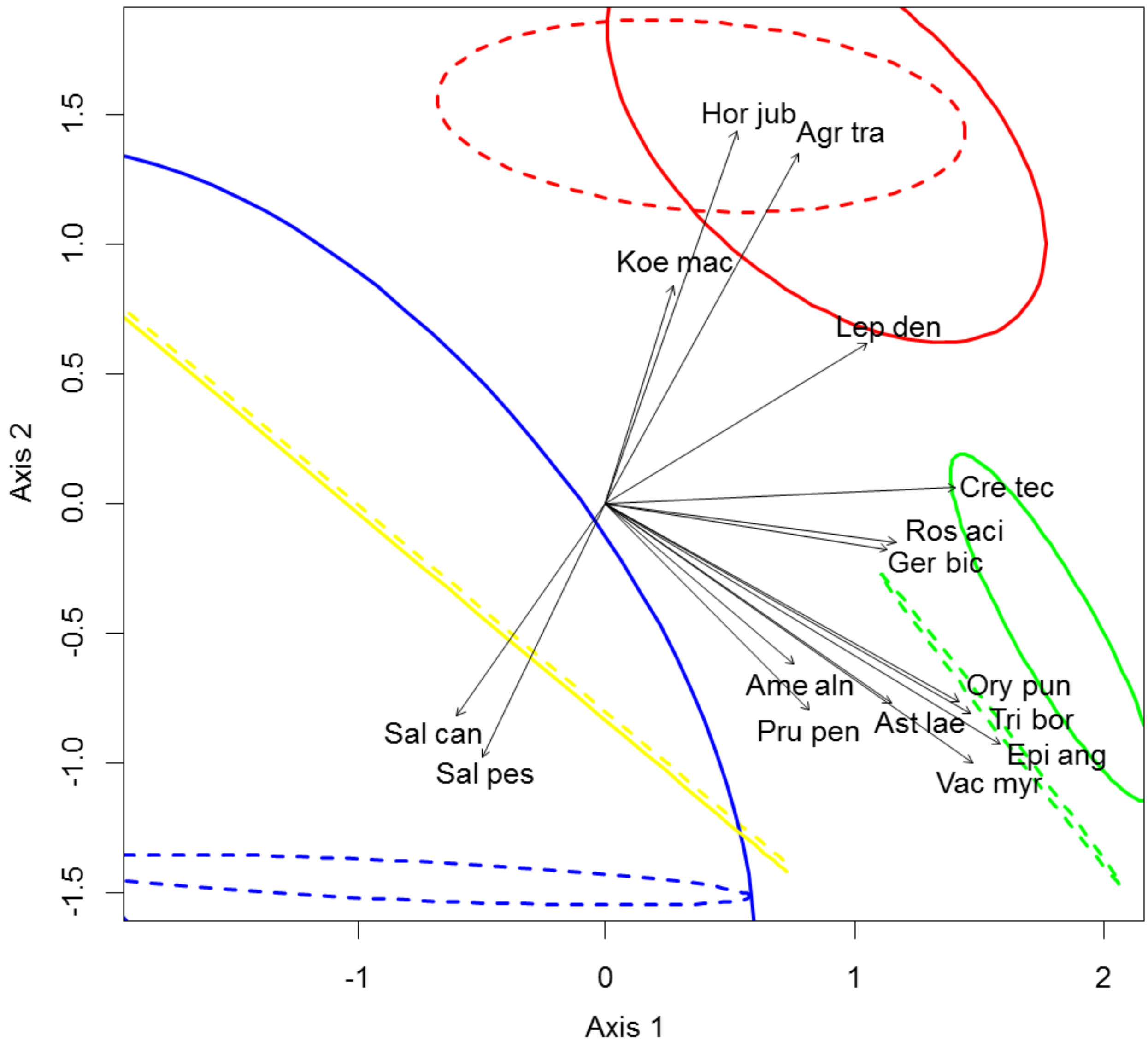


Figure 1.21: NMDS of the vegetation cover community of the four coversoil materials (blue=Peat, green=FFM, red=Subsoil Bm2, yellow=Subsoil C) in 2013 (solid lines) and 2015 (dashed lines). Vectors indicate species that were significantly associated with the ordination (α=0.05).

Coversoil Placement Depth

Peat Depth

Figure 1.22: The two treatments used to compare the effect of Peat placement depth on tree performance and colonizing vegetation. Treatment 1 (30cm of Peat over 120cm of Subsoil C) and treatment 3 (10cm of Peat over 140cm of Subsoil C). All treatments were placed over lean oil sands (LOS) overburden material. Physical and chemical characteristics of these materials are presented in Table 1.2.

Treatment Characteristics

When comparing treatments with 30cm or 10cm of Peat over Subsoil C, application thickness of Peat significantly affected surface soil temperature measured at 35cm below the soil surface during the growing seasons (Table 1.6; Figure 1.22). Soil temperature in the thicker Peat application treatment (30cm) was approximately 2°C cooler in 2014, 2015, and 2016 compared to the shallow Peat application of 10cm (Table 1.6). The number of days where the average daily soil temperature was above 5°C was not significantly different in 2013 or 2016 between the two Peat application thickness treatments; however, in 2014 and 2015 this period was shorter by 15 and 10 days in the deeper Peat treatment (Table 1.6). Volumetric water content (VWC) 15cm below the soil surface was not significantly different between the two Peat thickness treatments in 2013 or 2014; however, in both 2015 and 2016 VWC was significantly higher in the 30cm Peat treatment than in the 10cm Peat treatment (Table 1.6).

Specifically during the 2016 growing season, soil temperature 15cm below the soil surface in the 30cm Peat treatment was cooler until September (Figure 1.23), and average daily VWC during the growing season was consistently around 4.5 times higher in the 30cm Peat thickness treatment than in the 10cm treatment (Figure 1.24).

Aspen Response

Aspen seedling growth was greater in the shallow, 10cm Peat treatment from the second to the fourth growing seasons (Figure 1.25a). In the fourth growing season, 2015, while the shallow treatment still had more growth, it was substantially less than the previous year, unlike in the 30cm treatment where growth in 2015 was slightly greater than in 2014 (Figure 1.25a). In the fifth growing season growth in the deep treatment continued to increase relative to the previous year and growth in the shallow treatment continued to decrease (Figure 1.25a). However, by the end of the fifth growing season, aspen seedlings were significantly taller in the shallow Peat treatment than the deep placement treatment ($p < 0.05$; Figure 1.25b). Root collar diameter followed a similar trend (data not shown).

Pine Response

Growth of pine seedlings in the 30cm and 10cm Peat treatments followed similar trends in all years measured (Figure 1.26a). There was no significant difference between pine heights in the two Peat placements by the end of the fifth growing season (Figure 1.26b). Root collar diameter followed a similar trend (data not shown).

Spruce Response

Growth of spruce seedlings in the two Peat placement depths were similar for the first two growing seasons; in the third growing season growth increased in both treatments, mostly notably in the shallow treatment (Figure 1.27a). In 2015 and 2016, growth decreased in the shallow treatment, but increased in the deeper 30cm treatment (Figure 1.27a). However, despite the different growth in the deeper treatment there was no significant difference between spruce heights in the treatments in 2016 (Figure 1.27b). Root collar diameter followed a similar trend (data not shown).

Colonizing Vegetation Response

Cover was significantly higher in the 30cm Peat treatment (1.5±0.1) than in the 10cm Peat treatment (0.3±0.0) in 2015 (treatment effect $p < 0.01$). Species richness and the community composition were not significantly different between the two treatments in 2015 (data not shown).

Figure 1.23: Daily soil temperature 15cm below the soil surface in the 2016 growing season in treatments with either 10 or 30cm of Peat.

Figure 1.24: Average and standard error of the daily water content 15cm below the surface during the growing season in treatments with two Peat depths in 2013, 2014, 2015, and 2016 (n=3).

Figure 1.25: Average and standard error of aspen growth in the first five growing seasons (a) as well as height in 2016 (b) in treatments with different Peat depths (n=3). Uppercase letters indicate significant differences in the 2016 heights (α=0.1).

Figure 1.26: Average and standard error of pine growth in the first five growing seasons (a) as well as height in 2016 (b) in in treatments with different Peat depths (n=3).

Figure 1.27: Average and standard error of spruce growth in the first five growing seasons (a) as well as height in 2016 (b) in treatments with different Peat depths (n=3).

FFM Depth

Figure 1.28: The two treatments used to compare the effect of FFM placement depth on tree performance and colonizing vegetation. Treatment 2 (10cm of FFM over 140cm of Subsoil C) and treatment 7 (20cm of FFM over 130cm of Subsoil C). All treatments were placed over lean oil sands (LOS) overburden material. Physical and chemical characteristics of these materials are presented in Table 1.2.

Treatment Characteristics

When comparing treatments with either 20cm or 10cm of FFM over Subsoil C, the application thickness of FFM (Figure 1.28) did not significantly affect surface soil temperature during the 2013 and 2014 growing seasons. The number of days where the average daily soil temperature was above 5°C was also not significantly different between the two FFM placement depths (Table 1.6).

Aspen Response

Growth of aspen seedlings in treatments with different placement depths of FFM were similar in all five years measured (Figure 1.29a), and by the end of the fifth growing season there was no significant difference in aspen heights (Figure 1.29b). Root collar diameter followed a similar trend (data not shown).

Pine Response

Similarly to aspen seedlings, pine seedlings in the treatments with 10cm or 20cm of FFM had similar growth in all five years measured (Figure 1.30a). There also was no significant difference in pine height between the two treatments in 2016 (Figure 1.30b). Root collar diameter followed a similar trend (data not shown).

Spruce Response

Growth of spruce seedlings in treatments with either 10cm or 20cm of FFM material were similar in the first five growing seasons (Figure 1.31a), and seedling height was not significantly different in the two treatments in 2016 (Figure 1.31b). Root collar diameter followed a similar trend (data not shown).

Colonizing Vegetation Response

There were no significant differences in total vegetation cover, species richness, or the vegetation community between the two FFM placement depth treatments in 2015 (data not shown).

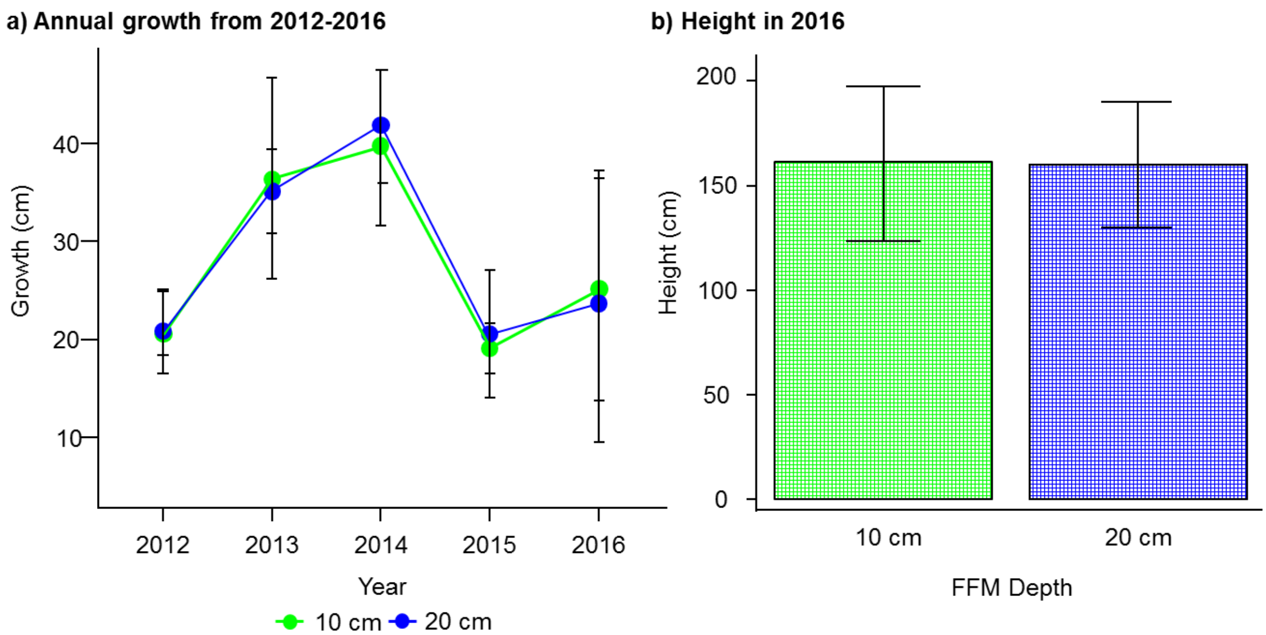


Figure 1.29: Average and standard error of aspen growth in the first five growing seasons (a) as well as height in 2016 (b) in treatments with different FFM depths (n=3).

Figure 1.30: Average and standard error of pine growth in the first five growing seasons (a) as well as height in 2016 (b) in in treatments with different FFM depths (n=3)

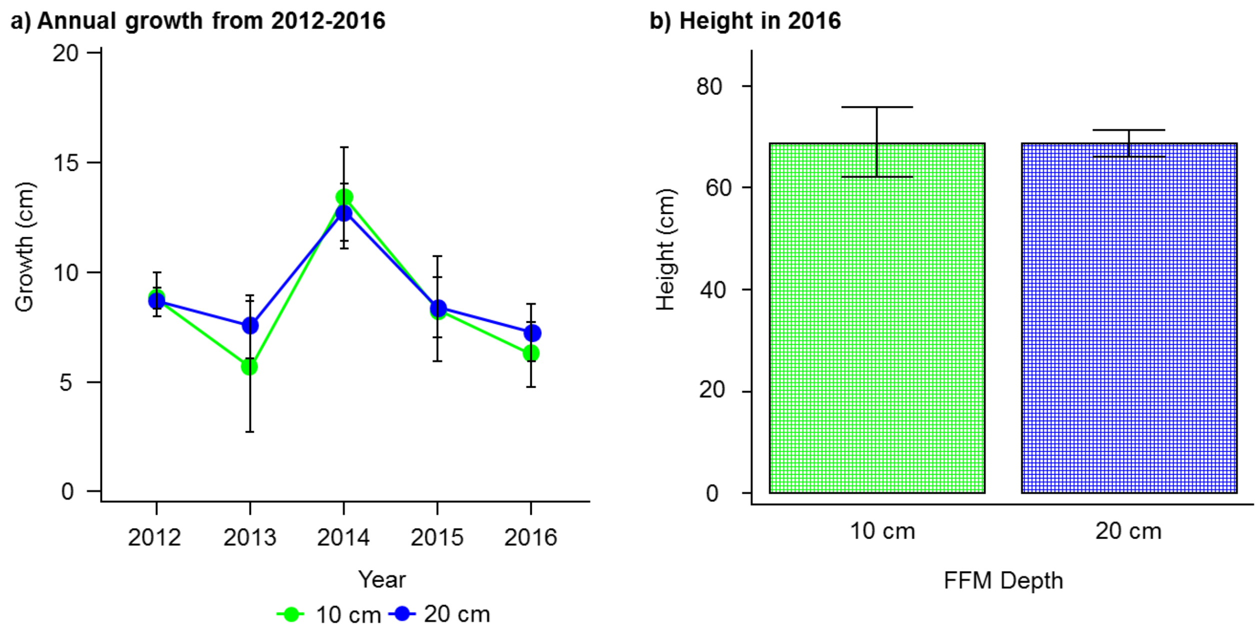


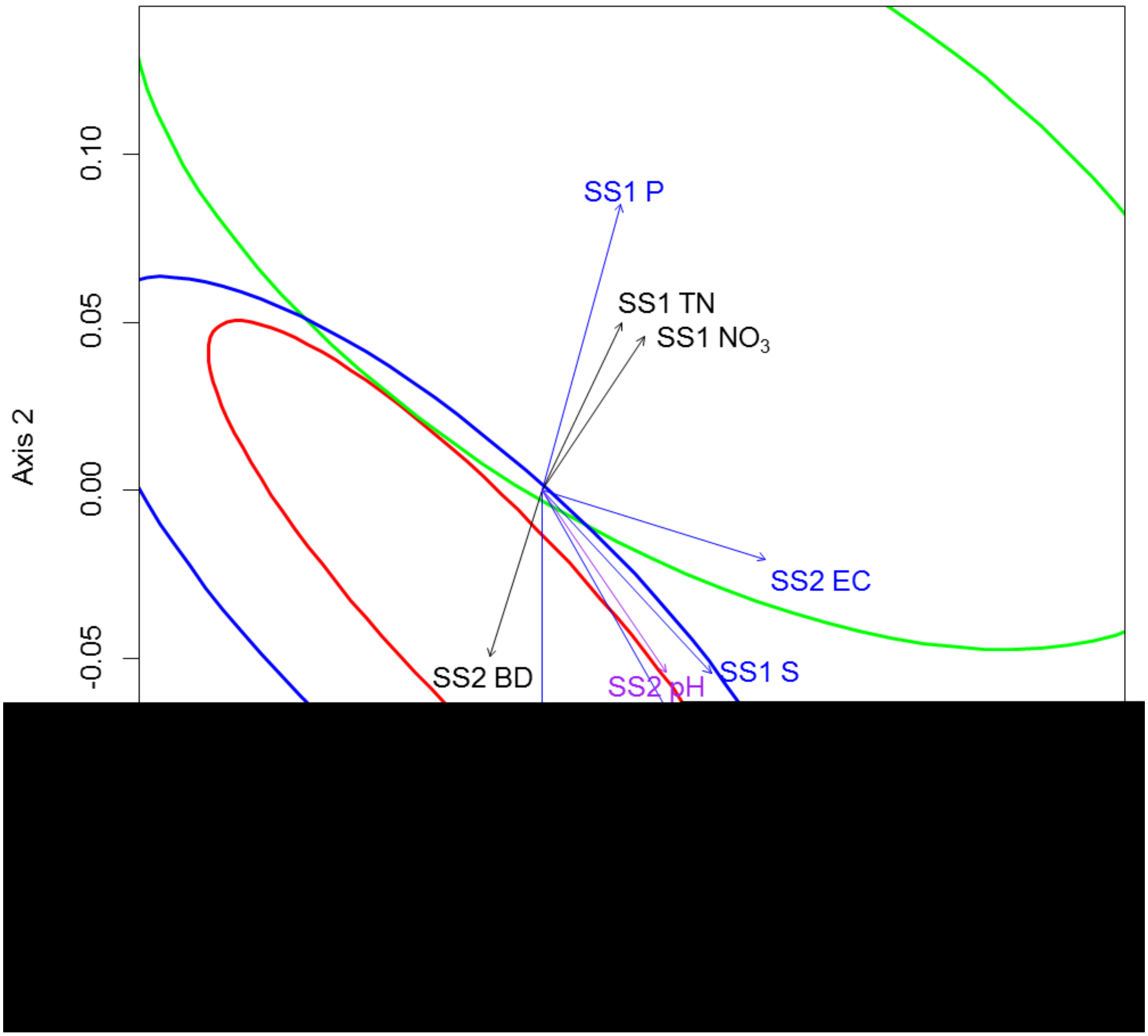
Figure 1.31: Average and standard error of spruce growth in the first five growing seasons (a) as well as height in 2016 (b) in treatments with different FFM depths (n=3).

Underlying Subsoil Material

Figure 1.32: The six treatments used to compare the physical and chemical differences between the three types of underlying subsoil materias. Treatment 1 (30cm of Peat over 120cm of Subsoil C); treatment 6 (30cm of Peat over 30cm of Subsoil Bm1 over 90cm of Subsoil C); treatment 10 (30cm of Peat over 120cm of Subsoil BC), treatment 7 (20cm of FFM over 130cm of Subsoil C), treatment 8 (20cm of FFM over 130cm of Subsoil BC), and treatment 9 (20cm of FFM over 30cm of Subsoil Bm1 over 100cm of Subsoil C). All treatments were placed over lean oil sands (LOS) overburden material. Physical and chemical characteristics of these materials are presented in Table 1.2.

Subsoil Material Characteristics

When the subsoil materials in six treatments were compared (Figure 1.32), Subsoil Bm1 had significantly different chemical properties and initial nutrient composition than both Subsoil C ($p < 0.01$) and Subsoil BC ($p < 0.1$). When plotted on an NMDS, the ellipses representing Subsoil C and Subsoil BC overlapped, reflecting their similarities (Figure 1.33). These treatments were significantly associated with high pH, EC, and bulk density ($p < 0.1$). Subsoil Bm1 was significantly associated with high P, total N, and NO_3^- ($p < 0.1$; Figure 1.33).



[redacted]

Figure 1.33: NMDS of the chemical characteristics and initial soil nutrients from 2011 in the subsoil material from three treatments with Peat and three with FFM (Figure 1.32). Ellipses indicate a 90% confidence interval for each treatment (blue=treatments1&7, Subsoil C; green=treatments 6&9, Subsoil Bm1 and Subsoil C; red=treatments 8&10, Subsoil BC). Arrows indicate characteristics that are significantly associated with the NMDS orientation (blue α=0.0001, purple α=0.01, black α=0.1). Note: ‘SS1’ refers to subsoil between 30 and 60cm below the surface; ‘SS2’ refers to subsoil from 60 to 150cm below the surface.