Reindeer Lichen Transplant Feasibility for Reclamation of Lichen Ecosites on Alberta's Athabasca Oil Sand Mines

by

Sara Duncan
B.Sc., University of British Columbia, 2004

A Thesis Submitted in Partial Fulfillment
of the Requirements for the Degree of

Master of Science

in the School of Environmental Studies

Supervisory Committee

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Abstract

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This project is a pilot study to assess the viability of transplantation as a technique to establish reindeer lichens on reclaimed areas of oil sands surface mines in the Athabasca region of Alberta. There were two components to this study: a) a lichen transplant trial, where I investigated which commonly available substrates found in reclaimed forest sites would promote the best lichen fragment survival and vigour for a lichen 'seeding' program; and b) a diversity assessment of the reclaimed site to compare the existing cryptogam community with the expected community for the target ecosite based on published descriptions from the surrounding native forests and documented chronosequences for terrestrial lichen communities. In July 2009, *Cladonia mitis* was transplanted into 54 plots on three sites that were planted with jack pine or spruce 12 or 24 years ago, respectively, on the Suncor Millenium/Steepbank Mine (Suncor Mine).

This trial was designed to investigate possible short-term indicators of successful lichen establishment and the effect of substrate (moss, litter, or soil) on the establishment of transplanted lichen thallus fragments. The indicators of lichen establishment evaluated were vigour, movement from plots, photographic areal cover, and microscopic growth (hyphal growth, annual growth and lateral branching). After two growing seasons, the effect of substrate on lichen transplant survival varied by site; there was no significant difference in lichen fragment retention in plots by substrate on the 24-year old sites, but median fragment retention was significantly higher on moss and litter substrates than soil on the 12-year old site. There was also no significant difference in fragment vigour between substrates on each site, except on the south-facing 24-year-old forest site where average vigour was significantly higher on moss plots than on soil plots. Photographic areal measurement is not recommended as a short-term lichen establishment monitoring

tool for transplanted fragments based on the difficulties encountered using the method for this trial.

Forty-one percent of the fragments collected for microscopic assessment after the first growing season had grown hyphae, 23 percent of the fragments collected during September 2009 and September 2010 had formed apothecia, and 31 percent of the fragments collected in September 2010 had grown lateral branches.

The results of the biodiversity assessment were compared with the successional communities previously described for spruce- and pine-lichen boreal forests. There were no lichens found on the 12-year-old site, though the cup lichens were common to abundant on the 24-year-old sites, which is consistent with the cryptogammic community expected for a regenerating natural site of that age. *Cladonia mitis* was also present but rare to uncommon on the 24-year-old site, while *Cladonia stellaris*, *Cladonia rangiferina* and *Cladonia stygia* that, together with *C. mitis*, are indicative of the al and c1 ecosites of the Central Mixedwood Boreal forest, were not present.

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Acknowledgements

This thesis would not have been possible without the generous funding support of Suncor Energy Inc. and Shell Canada Energy through the Canadian Oilsands Network for Research and Development's Environmental and Reclamation Research Group. Thanks to Dr. Francis Salifu at Suncor in particular, for supporting me through all the steps of the CONRAD research process and site access hurdles, and for having faith in the value of lichen reclamation research from the beginning. Thanks also to Xiao Tan at Shell for her helpful questions.

For introducing me to the site and taking me on a whirlwind tour of lichen-jackpine stands in the Fort McMurray region, I am grateful to Justin Straker at Integral Ecology Group Ltd, and to Wayne Tedder of Golder Associates Ltd. for letting us all in on the existence of the lichen stand at Wood Creek. I am indebted to Seonaid Duffield and Caitlin Currey (Uvic students), Alex Fox (Suncor co-op student), and Joshua Martin (Suncor Reclamation Research Assistant) for assisting me in the installation and monitoring of the trials – I couldn't have got it all done on time without them.

I'd also like to acknowledge the help of Dr. Dale Vitt at Southern Illinois University and Dr. Shanti Berryman at Integral Ecology Ltd. for agreeing to confirm my moss and lichen species identifications, respectively; and also thanks to Shanti for helping me track down the elusive chemicals needed to identify the *Cladonia*s – that was no mean trick.

Thanks also to Debra Stokes at First Coal Corporation, and Jennifer Turner and Sean Sharpe at AECOM, for providing me the invaluable opportunity to participate in the planning and installation of high elevation lichen reclamation research trials at First Coal's Central South property – while not included in this thesis, those trips were integral to my learning experience.

I would also like to express my gratitude to my committee. Thanks to my supervisor, Dr. Val Schaefer, for being accessible and enthusiastic at all times, and particularly for managing to wade through all the contracts and expenses and agreements to ensure I could do the research. Thanks also to Dr. Brian Starzomski for agreeing to heroically wade in part way through my program, to provide thorough feedback on the drafts of this thesis and advice on the difficult world of statistics.

Last but not least, I owe thanks to my husband Donal for helping me figure out how to create those graphs in *R*, saving me from hours of programming misery, and for generally putting up with my enthusiasm for lichens.

1.0 Introduction

1.1 Lichens as Potential Reclamation Species

The need to develop reclamation techniques to restore a wide range of native species from the boreal forest is becoming more important in the Athabasca region as the number of mines approved increases and operating areas expand. According to the Alberta Department of Energy (GIS Services 2010a), there were four producing surface mines operating in the Athabasca oil sand region as of July 2010 for a total disturbed area of approximately 602 km^2: Suncor's Steepbank/Millenium mine (Suncor Oil Sands), Syncrude mine, Shell Canada's Muskeg River mine (also known as Albian Sands), and CNRL's Horizon mine. Shell Canada's Jack pine mine and Imperial Oil's Kearl mine are under construction, and four more projects have been proposed; Fort Hills (Suncor), Voyageur South (Suncor), Pierre River (Shell) and Joslyn Mine (Total). However, only approximately 3 percent (4,750 km^2) of the mapped Athabasca deposit, in a region roughly following the banks of the Athabasca River, is surface mineable; there are also 49 producing and proposed *in situ*[1] mines outside of the surface mineable area, many of which are within threatened boreal caribou range (GIS Services 2010b; Alberta Energy 1991; Boreal Caribou Committee 2001; Alberta Department of Energy 1996).

Research into techniques to use reindeer lichens in oil sands reclamation addresses mine permit requirements for re-establishment of pre-disturbance biodiversity on reclaimed sites, and the need to ensure that the resulting reclaimed ecosystems include critical terrestrial lichen forage for caribou.

1.2 Reclamation Requirements for Biodiversity

Each mining project requires a permit from Alberta Environment to proceed. Section 2 of the *Alberta Environmental Protection and Enhancement Act* (AR115/93 s2; 167/93)

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[1] Most of the bitumen from the Athabasca oil sands will be recovered using *in situ* technologies, which remove the bitumen from the oil sand without removing the sand from the ground, creating less surface disturbance than surface mining (Government of Alberta 2007).

states that, "The objective of conservation and reclamation of specified land is to return the specified land to an equivalent land capability."

The specific manner in which the operator of an individual project proposes to define and achieve equivalent land capability for their site has generally been by using forest productivity as an indicator (e.g. Cumulative Environmental Management Association 2007; Oil Sands Vegetation Reclamation Committee 1998). Lichen ecosites are not frequent targets for reclamation. This is in part due to the focus on forest productivity as a measure of equivalent land capability; lichen ecosites are by definition poor sites for forest production, and their existence on a reclaimed site lowers the average land capability achieved. It is also due to the suitability of the post-closure landforms and reconstructed soils themselves, which are capable of supporting richer ecosites on all but the driest tailings and dump crests (Cumulative Environmental Management Association 2007; Oil Sands Vegetation Reclamation Committee 1998).

The stated revegetation objectives for the Athabasca oil sands are: to provide erosion-resistant plant covers, utilize native woody-stemmed species, establish a diverse range of plant species to recreate the level of biodiversity on the pre-disturbed site, and establish a "viable plant community capable of developing into a self-sustaining cover of species suitable for commercial forest, wildlife habitat, traditional land uses, and with possibilities for recreation and other end uses" (Oil Sands Vegetation Reclamation Committee 1998 p. 27). These recommendation and guidelines have been incorporated into the permits for each of the major surface mine producers (Syncrude, Suncor, and Shell) as follows:

> "The approval holder shall reclaim the land so that the reclaimed soils and landforms are capable of supporting a self-sustaining, locally common boreal forest, regardless of the end land use" which is "…integrated with the surrounding area...". It must also "re-establish the capability for long term biodiversity consistent with the [operator's commitment to the Biodiversity Program recommended by the Biodiversity and Wildlife subgroup of the Reclamation Working Group of CEMA][2].

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[2] E.g. Suncor Energy Inc. Approval No. 94-02-00, Sections: 6.1.6, 6.1.7, 6.1.45(a)(ii), 6.1.86(a) under Province of Alberta R.S.A. 2000, c.E-12, as amended; Suncor Energy Inc. Approval No. 80105-00-00, Section 5.1.13 under Province of Alberta S.A. 1992, c.E-13.3, as amended,

Thus, while a "viable plant community" is not explicitly defined to include lichens, lichens are implicitly included within the requirements for the creation of a community that reflects the locally common boreal forest.

In addition, the Guidelines for Reclamation to Forest Vegetation in the Athabasca Oil Sands Region suggest that future reclamation research should develop "techniques for propagation and promotion of lesser vegetation species in the pursuit of greater levels of biodiversity" (Alberta Environment 1999 p. 4), and that "methods to enhance the establishment of native understory species to achieve greater biodiversity than is possible through seeding/planting need to be developed" (Oil Sands Vegetation Reclamation Committee 1998 p. iv). Adding reindeer lichens to the species mix for reclaimed sites in the a1 (lichen-jack pine), c1 (Labrador tea mesic – jack pine/ black spruce) g1 (Labrador tea subhygric – jack pine/ black spruce), i (bog) and j (poor fen) ecosites would help to meet objectives for greater biodiversity (see Appendix A for ecosite descriptions). Currently, even where the target ecosite is lichen-jack pine, and reindeer lichens are a key component of that ecosite, lichens are not included in the planting prescriptions (see Oil Sands Vegetation Reclamation Committee 1998, p. 29).

The lichen-jack pine ecosites are too dry to have commercial forest as an end land use objective, but these low productivity forested sites are opportunities to increase the biodiversity of the reclaimed ecosystem by including species like the reindeer lichens, which will not be found on the higher productivity pine and spruce forest sites.

Syncrude Canada Limited Approval No. 26-02-00, Sections 6.1.7, 6.1.44(a)(ii), 6.81(a) under Province of Alberta R.S.A. 2000, c.E-12, as amended,
Albian Sands Energy Inc (Shell Canada) Approval No: 20809-01-00, Sections 6.1.7, 6.1.44(a)(ii), 6.81(a); and Shell Canada Limited Approval No 153125-00-00, Sections: 5.1.27(ii), 5.1.44 under Province of Alberta R.S.A. 2000, c.E-12, as amended.

1.2.1 Woodland Caribou Range Recovery

Species of the *Cladonia* genus are referred to collectively as 'reindeer lichens' (synonym: *Cladina*[3]), and are so named because they are important winter forage for caribou in North America, and reindeer in northern Europe and Asia.

Woodland caribou of the boreal ecotype reside year-round in northern Alberta (Bradshaw et al. 1995; Dzus 2001; Edmonds 1991). Woodland caribou have been listed as 'threatened' by the Committee on the Status on Endangered Wildlife in Canada (COSEWIC 2002; COSEWIC 2008a) and are Schedule 1 species under the *Species at Risk Act* (COSEWIC 2008b). Within Alberta, as with many caribou ranges in Canada, caribou populations are threatened by a combination of large-scale habitat changes caused by wildfires and human land use, predation, hunting, poaching, and vehicle collisions (Alberta Woodland Caribou Recovery Team 2005).

Six populations of boreal caribou have been identified in Alberta. All but one of the six herds are known to be at immediate risk of extirpation or are in decline – data for the remaining herd, (the Richardson herd) is insufficient to be able to determine a population trend (Alberta Woodland Caribou Recovery Team 2005).

There are 15 *in situ* projects producing bitumen in the Athabasca region, with five more existing but not producing, 24 more proposed, and four under construction (GIS Services 2010b). The primary effects of *in situ* developments on caribou are fragmentation of their habitat, and the creation of road and pipeline corridors that increase predator and poacher access to caribou on their ranges (Boreal Caribou Committee 2001; Dzus 2001; James and Stuart-Smith 2000). All industrial operations (including forestry and oil and gas pipelines, as well as oil sand mining) require Caribou Protection Plans (CPPs) and adherence to Best Practices for Caribou Ranges according to guidelines set out by the

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[3] Authors referring to the reindeer lichens as *Cladina* base their subgenus classification on morphological characteristics these lichens (e.g. *C. mitis*, *C. arbuscula*, *C. stygia*, *C. stellaris*, and *C. rangiferina*) share (erect, branching and shrub-like, uncommon sexual reproduction, lack of specialized asexual reproductive propagules) which make them visually different from other *Cladonia* lichens (such as the 'cup lichens') (see Ahti's (1961) monograph on North American reindeer lichens). However, molecular, chemical, and morphological data shows that these lichen species are not in fact different enough to warrant a separate subgenus, and are properly classified as *Cladonia* (Ahti and Depriest 2001; Brodo 1978; Stenroos et al. 2002). Synonyms used herein are listed in Esslinger (2009).

Boreal Caribou Committee (Boreal Caribou Committee 2001). At present, CPPs do not stipulate caribou-specific reclamation strategies to be used in the reclamation of disturbed lands in caribou ranges, though research on caribou range recovery strategies is being piloted. To date, range recovery research has focused on promoting rapid revegetation of linear disturbances to reduce human and predator movement (Szkorupa 2002).

To avoid predators, caribou space themselves widely on the landscape and forage within spruce forested peat complexes, where other ungulates typically do not go (Bergerud, Jakimchuk and Carruthers 1984; Bradshaw et al. 1995; James et al. 2004; Sanchez-Azofeifa and Bechtel 2001; Thomas and Gray 2002). Revegetated industrial disturbances frequently support an early successional vegetation community, with dense undergrowth and lush forage, frequently due to the practice of seeding agronomic grasses and legumes during reclamation. These vegetation communities attract deer and moose, which support higher densities of predators (primarily wolves) than caribou populations alone (Gustine et al. 2006; James and Stuart-Smith 2000; James et al. 2004; McLoughlin et al. 2003). The increase in predator densities causes increased predation on the caribou, forcing them to avoid areas where deer and moose cohabitate (Gustine et al. 2006; James et al. 2004; McLoughlin et al. 2003).

Thus, while not the most critical aspect of caribou population recovery plans, caribou range recovery can only be improved by research into reclamation using caribou forage species that will not act as an attractant to other ungulates. The most critical species in that regard are the terrestrial lichens that support caribou during the winter, but that other ungulates are not known to consume.

1.2.2 Conclusions

Lichen-dominated ecosites are a minor component on reclaimed mine sites, but represent opportunities for operators to increase the biodiversity on their sites by introducing terrestrial lichens, which are not found on sites with higher forest productivity. Ecosite phases c1, g1 or any of the i, or j ecosite phases have minor lichen components, and biodiversity on reclaimed sites intended to support these communities could also be enhanced by the addition of reindeer lichens to the list of species that can be used in reclamation. On sites being reclaimed within caribou management areas, inclusion of lichens in the reclamation species list may also address wildlife habitat reclamation goals, as they are important caribou forage and do not attract other ungulates that serve as alternate prey for the caribou's predators.

1.3 Reindeer Lichen Autecology

1.3.1 Substrate Ecology

Reindeer lichen podetia[4] are loosely attached to their substrates via hyphae[5]; in older mats that blanket forest floors, they may not even be attached to the substrate by living tissue at all, and are held in place by entanglement or fusion with other podetia in the mat. Despite this tenuous attachment to the substrate, reindeer lichen abundance is still strongly correlated with certain substrates: coarse, well-drained mineral soils (Ahti 1977; Brodo, Duran Sharnoff, and Sharnoff 2001; Sulyma 2000; Thomson 1967), and dry peat hummocks in bogs (Beckingham and Archibald 1996; Kershaw 1977). Lichen-dominated sites are associated with coarse soils primarily because lichens are extreme stress tolerators - able to tolerate xeric conditions on these soils that bryophytes and higher plants are not, and thus thriving in the absence of competition (Ahti 1959; Ahti 1977; Brodo 1973; Kershaw 1977; Thomson 1967). However, reindeer lichens are still present on mesic sites and finer textured soils within low productivity forests as lesser components of the understory (Botting and Fredeen 2006; Brulisauer, Bradfield and Maze 1996).

While lichen abundance may be correlated with soil texture, reindeer lichens are seldom found growing directly on mineral soils; the most frequently cited substrates are moss, litter, stumps and decaying logs on the forest floor (Brodo, Duran Sharnoff, and Sharnoff 2001; McCune and Geiser 1997; Pope 2005; Smith 1921; Tolpysheva and Timofeeva 2008; Vitt, Marsh and Bovey 1988).

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[4] stalk-like body on which the lichen fruiting bodies are typically borne.
[5] Thread-like fungal structures.

One researcher claims to have demonstrated that mat-forming *Cladonia* lichens obtain nutrients from their substrate - even through dead podetial bases to live tissue (e.g. Barashkova 1963 in Tolpysheva and Timofeeva 2008). Other authors, however, using radioactive tracers, have found that nutrients are translocated from dead tissues and old live tissues to new tissues (Crittenden, Katucka and Oliver 1994 in Crittenden 2000; Ellis et al. 2005; Hyvarinen and Crittenden 2000), but not from the soil (Crittenden, Scrimgeour and Ellis 2004), and the dominant position is that nutrients are not obtained from the substrate in any appreciable amount, and that the reindeer lichens meet their nutrient requirements from atmospheric deposition.

Lichen researchers also frequently stress the association of reindeer lichens with acidic and oligotrophic soils (the most frequently cited research being that of Ahti (1961) and Brodo (1973); and Kershaw (1977) provides a possible mechanism through which soil pH may affect the lichens: during snow melt, when the surface soil is saturated and water is pooled at the surface due to frozen soils below the shallow thawed surface, the pH of the soil solution may affect lichen nutrient uptake. However, there have been no controlled studies to determine whether reindeer lichen development actually requires low soil pH, or whether this association is due to other factors correlated with low pH soils such as reduced productivity of competing vegetation.

1.3.2 Reproduction and Dispersal

The reindeer lichens are not known to produce soredia, ‘rarely’ or ‘uncommonly’ produce apothecia[6] (apothecia were found on approximately 21 percent of *Cladonia mitis* podetia by Kotelko, Doering, and Piercy-Normore (2008), and ‘frequently’ produce pycnidia[7] (Ahti 1961; Brodo, Duran Sharnoff and Sharnoff 2001; Goward 1999; Smith 1921; Thomson 1967). The primary mode of reproduction for the reindeer lichens is via

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[6] a type of fruiting body borne on the surface of the thallus that opens at maturity to expose the sexual spores (ascospores) within(Ulloa and Hanlin 2000). In the case of the reindeer lichens, the apothecia are borne on the tips of the branches.

[7] a type of fruiting body bearing conidiophores (a type of asexual fungal spore) which is also borne on the tips of the branches, but lies beneath the surface of the thallus, opening to the surface through a pore (Ulloa and Hanlin 2000).

fragmentation of the thallus; a trait shared with other fruticose species such as *Usnea* and *Bryoria*. Reproduction via fragmentation has the benefit of ensuring both symbionts are disseminated, and that new podetia reach a relatively large size rapidly; but it has only been shown to be effective as a means of dispersal over short distances by wind and animals. Heinken et al. (1999) mimicked the effects of trampling on lichen mats, and mapped the distribution of *Cladonia* spp. fragments around the mats after 15 days. The majority of the *C. arbuscula*[8] fragments, of all visible size ranges, stayed within 70 cm of the trampled mats.

1.3.3 Growth Characteristics

Growth rates for reindeer lichen podetia have been reported for Alaska, Newfoundland, Russia, and the Northwest Territories, and varies between an average of 3 and 6 mm/year; with growth rate varying by latitude and forest cover (e.g. Ahti 1957; Ahti 1959; Helle, Aspi and Tarvainen 1983; Pegau 1968; Scotter 1963).

Krabbe (1891 in Smith 1921) reportedly observed lichen mats 4 to 5 cm tall growing on a soil burned only 10 years previously in a German forest, and reindeer lichens in northern Alberta have been found to regenerate from bare soil to mature mats on a fire-disturbed site within approximately 40 - 45 years (Carroll and Bliss 1982; Dunford et al. 2006). The ‘typical’ post-fire succession sequence for terrestrial lichens in the boreal forest, described by Ahti (1959), follows a similar pattern to that in northern Alberta, where the first reindeer lichen dominant stage (*C. mitis*) occurs within 30 to 50 years of bare soil stage; in the absence of fire, the second reindeer lichen stage, dominated by *C. stellaris*, occurs after 50 to 80 years and may last 40 years or more. The time to return of reindeer lichen cover can be drastically reduced where propagules are abundant and the forest floor bryophytes and litter are relatively intact; in the dry pine-lichen forests of British Columbia, Sulyma (2000) found that lichen mats returned to their previous cover only 17 years after disturbance during logging, and Webb (1998) recorded a return to average

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[8] Morphologically identical to *C. mitis*. Some authors treat *C. arbuscula* and *C. mitis* separately (e.g. Brodo, Duran Sharnoff and Sharnoff 2001; Goward 1999) while others treat them as subspecies, one species, or two species which intergrade to such a degree wherever they are conspecific there is little utility in trying to separate them (e.g. Ahti 1959; Kotelko, Doering and Piercy-Normore 2008; Osyczka 2006).

lichen cover of approximately 20% on sites that were logged 2 to 16 years previously, compared to 25% cover on undisturbed sites.

1.3.4 Conclusions

The *Cladonia* lichens referred to collectively as “reindeer” lichens are not typically associated with early seral ecosystems, such as those forming on reclaimed oil sands sites; particularly those with soils with neutral to basic pH and relatively high nutrient availability. However, since the lichens are shallowly attached to the top few millimeters of the soil (or more commonly, the overlying humus or moss), their establishment should be less subject to the effect of differing soil conditions than other species found in their ecosites which have nevertheless been found to be suitable for reclamation (e.g. jack pine, white spruce). The naturally colonizing mosses and developing litter and humus layers found on reclaimed forest sites may be suitable substrates for the reindeer lichens, which have the potential to establish under various canopy closure conditions; though they will be most successful where trees are widely spaced and there is little shade. It remains to be seen whether the difference in initial soil chemistry on reclaimed sites will alter forest succession trajectories for dry sites, resulting in higher site productivity and competitive exclusion of the reindeer lichens by vascular vegetation even at later seral stages.

Reindeer lichen annual growth rate is low - 4 to 5 mm/year in northern Alberta; however, a mature mat can develop from bare soil within 50 years, since the height of a mature lichen mat is also small. Natural dispersal of the reindeer lichens is thought to occur largely through thallus fragmentation up to distances of only a few metres, with wind-borne sexual diaspore production being relatively rare. It is not known whether any long distance dispersal occurs with enough frequency to assure a timely return of these lichens to reclaimed sites, which may exist at large distances from lichen source populations. Thus, the time to achieve a reindeer lichen cover may have less to do with the inherent growth rates of the lichens, but the rate of their arrival on the site; a factor which transplantation programs may overcome (barriers to dispersal as a limitation in lichen colonization are discussed further in Sections 1.4.1 and 1.4.2, below).

1.4 Lichen Restoration Via Transplantation in the Literature

Some of the drier reclaimed sites in the oil sands may be suitable for reindeer lichen establishment, but reindeer lichens are not currently known to be present on reclaimed areas. Without transplantation experiments, we do not know whether these lichens are absent due to dispersal limitations or inhospitable site conditions, e.g. competitive exclusion by bryophytes and vascular plants, or unsuitable moisture and light regimes.

The practice of artificially dispersing lichens for mine reclamation has not been documented in the academic literature, conference proceedings, or mine annual reports I searched. Thus, this project was based on lichen restoration studies I could find that were conducted for the purpose of endangered species conservation, maintenance of biodiversity in managed forests, and restoration of degraded domestic reindeer range.

1.4.1 Arboreal Transplants: Applications for Endangered Species

Two transplant experiments aimed at developing methods of preserving endangered lichen species that have methodologies or results relevant to reindeer lichen transplantation on reclaimed mine sites are described here.

Liden et al. (2004) transplanted fragments of endangered old-growth forest arboreal lichens *Evernia divaricata* (L.) Ach. and *Ramalina dilacerata* (Hoffm.) Hoffm. to trees in three Norway spruce stands within their range in northern Sweden. Like the reindeer lichens, these fruticose lichens are also thought to reproduce primarily by thallus fragmentation, and so may be limited to old forests due to slow dispersal rates into new forests rather than site limitations. Liden et al. (2004) sought to evaluate the effect of aspect (north or south), forest stand type (within old growth source site, mature forest, or mature forest adjacent to clearcut), and protection (with or without plastic shield cover to exclude gastropods) on the survival and vigour of the transplants over one year. The authors found that transplant survival was high in all locations (85 – 97.5 percent) after one year, and that most loss of transplants was due to human error – in this case, fragments being tied too loosely to the twigs. As with other forest lichen transplant studies (see discussion of work by Dettki, Klintberg and Esseen 2000; Hilmo and Såstad

2001; Hilmo 2002; Sillett and McCune 1998; Stevenson 1988 in Section 1.4.2, below), Liden et al. (2004) found that, at least in the short term, survival of the lichen fragments they transplanted remained high even in different forest stand types, suggesting that dispersal ability may be more limiting to forest lichen distribution than site conditions.

A limitation of lichen transplant experiments in the literature is the lack of long-term monitoring. The longest monitored lichen transplant experiment I could find was that of Gilbert (Gilbert 1991; Gilbert 2002), who followed the success of the foliose forest lichen, *Lobaria amplissima*, transplants for 20 years. He found that transplants which survived the first year had an excellent chance of becoming "vigorous colonies", and that there was no apparent correlation between initial thallus size and survival, or rate of growth of transplants and their subsequent survival. He concluded that it might have been better to transplant many small thalli that grow quickly (relative to size), than few large thalli that grow slowly (Gilbert 2002). While most of Gilbert's conclusions may not be directly translatable to reindeer lichens, it is important to note a finding of first-year establishment of lichen transplants correlated to long-term success.

1.4.2 Arboreal Transplants: Applications for Forestry

The limitations to survival and dispersal of old forest macrolichens to young forest environments has been investigated largely for arboreal lichens, which are threatened by forestry practices that produce large clearcuts that reduce connectivity in the landscape, and rotation lengths which are too short to allow for the development of very old forests that support large lichen biomass (Esseen, Renhorn, Pettersson 1996; Goward and Campbell 2005). Researchers have hypothesized that the relative lack of arboreal lichens in young forests is due to the unsuitable substrate properties of the young trees themselves (e.g. Armstrong 1988; Dettki, Klintberg and Esseen 2000; Sillett and McCune 1998), microclimatic effects of the young forest stand (e.g. susceptibility to increased insolation described by Hilmo 2002; Gauslaa and Solhaug 1996 and susceptibility to wind damage as described by Boudreault et al. 2008; Coxson, Stevenson and Campbell 2003; Demmig-Adams et al. 1990), limited dispersal capability of the lichens themselves that require long time periods to establish in a new environment, particularly at any distance from a well-established source population (Dettki, Klintberg and Esseen 2000; Esseen, Renhorn, Pettersson 1996; Sillett and McCune 1998; Stevenson 1988) , or slow growth rate of the lichens that do establish (Dettki, Klintberg and Esseen 2000).

Hilmo (2002) found that several foliose lichens (*Hypogymnia physodes, Platismatia glauca, P. norvegica* and *Lobaria scrobiculata*) grew at least as well or better in young spruce forests in Norway than the old forests they were transplanted from, despite prolonged exposure to high light levels. Sillett and McCune (1998) had findings similarly contrary to expectation; cyanolichen transplants (lichens with cyanobacteria as photobiont instead of algae, typically found in the canopies of old-growth forest) survived and grew as well in young forests (35 to 40 years old) as in mature (140 to 150 years old) or old-growth forests (400 to 700 years old); however, mortality was high in clearcuts. Finally, Hazell and Gustafsson (1999) found that *Lobaria pulmonaria*, a foliose lichen considered indicative of old forests and landscape continuity, had high survival and vitality after 2 years following transplantation to clustered and solitary trees retained in logged sites. These studies, taken together, support the hypothesis that lichen species found to be most abundant in old forests are not restricted to those forests by site conditions, but are instead slow to arrive due to dispersal limitations, and then take many years to accumulate biomass. While slow biomass accumulation rates are characteristic of most lichens, transplantation may be a method of reducing the lag time between forest stand regeneration and lichen establishment.

Establishment of forest lichens, particularly fruticose lichens that propagate primarily by fragmentation and wind dispersal, may depend on both distance to and density of source populations. Stevenson (1988) found that establishment of *Bryoria* spp., *Alectoria* spp., and *Usnea* spp. in second growth stands declined from abundant colonization of 2-year-old twigs at a distance of 100 to 150 meters from old forest edges, to low numbers of fragments colonizing twigs at 300 to 400 meters from the forest edge. Stevenson (1988) also found that young forests next to stands with abundant lichens had high lichen establishment rates, but that young forests next to old forest stands with low to moderate lichen abundance did not have high lichen establishment rates, regardless of proximity to source. Similarly, in the Scots pine (*Pinus sylvestris*) forests of northern Sweden, Dettki et al. (2000) found that *Alectoria sarmentosa*, while not found on young pine trees in young forests, was abundant on young pine in old forests, suggesting that it was not the characteristics of the young tree itself (bark roughness, branch size, etc.) that limited old-forest lichen establishment, but distance to abundant source populations.

Enns (1988 reported in Stevenson et al. 2001) attempted to determine whether transplantation might speed the accumulation of arboreal lichen biomass in second growth stands, and transplanted *Alectoria sarmentosa* fragments to young trees with a leafblower. As with Krekula's (2007) experimentation with similar techniques for terrestrial caribou forage lichens (see Section 1.4.3, below), this experiment suggests that transplanting lichen thalli may be relatively simple, as well as potentially successful.

Lichen transplant experiments with arboreal lichens have indicated that a) lichen species associated with mature forest types may not be restricted to them, and may be able to establish well in younger forests, b) much of the distribution of forest lichens can be explained by dispersal limitations rather than site conditions, and c) transplanting lichen thalli can be simple and result in high initial survival and vigour.

1.4.3 Terrestrial Transplants: Applications for Reindeer Husbandry

As with many studies on the changes in arboreal lichen abundance, studies on the recovery of terrestrial reindeer lichens have focused on the ability of lichens to re-establish in forests disrupted by harvesting. Logging results in the disturbance or removal of the duff layer and moss and lichen mats, and burial of mats under soil and debris; but it also fragments existing mats and disperses them throughout the site with machinery (Sulyma 2000 and Webb 1998). Because the lichen mats and their substrates are not usually wholly removed from logged sites or caribou/reindeer range, investigations of reindeer lichen recovery have concentrated largely on observing the recovery of mats from existing fragments (e.g. Boudreau and Payette 2004; Coxson and Marsh 2001; Gaio-Oliveira et al. 2006; Webb 1998), rather than the feasibility of re-introduction of the lichens to the site. A notable exception is one undergraduate thesis by Krekula (2007), who examined the technical feasibility of dispersing reindeer lichen thallus fragments using leafblowers in a reindeer range degraded by soil scarification during forest harvesting, similar to Enns' (1988 in Stevenson et al. 2001) attempt to bolster arboreal lichen biomass in B.C (see Section 1.4.2). Krekula (2007) found that his technique was cost-efficient for small-scale application; he was able to consistently spread lichen fragments ranging in size from a few millimetres to five centimetres at a rate of 10 g/m^2 over a five hectare area in under eight hours.

Reindeer lichen transplant studies have been conducted on degraded reindeer ranges in Sweden, where reindeer lichen mats have been overgrazed, and where the objective is to increase the rate of lichen mat recovery in second-growth forests to support reindeer husbandry, though few of these studies have been published in English. The effect of substrate and fragment size on reindeer lichen thallus establishment was evaluated in a clearcut and 40-year-old second-growth forest (Roturier et al. 2007), and the effect of dispersal method (clump vs. fragment) on establishment was evaluated in winter reindeer range (Roturier and Bergsten 2009). Roturier and Bergsten (2009) found that plots with transplanted lichen mat clumps had higher covers than plots with dispersed fragments after 6 years, but after an initial loss of material over the first 3 years of study, the lichen cover in both treatments increased at the same rate for the last 3 years (i.e., the dispersed fragment plots had a higher initial loss of cover than the clumped plots in the first year, but the fragments that remained in all plots grew at the same rate). Much of the removal of material from the clumped plots appeared to be due to reindeer grazing.

Fragment size (1-cm or 3-cm length) affected the ability of the thallus fragments to stay in place on the clearcut (1-cm fragments were subject to being blown from plots), but not in the forest (Roturier et al. 2007). Similarly, the type of substrate (moss, bark, twigs, or soil) had an effect on fragment retention in the clearcut (where moss and twigs promoted the greatest retention), but not in the forest.

1.5 Transplanting Established Techniques to Mining Reclamation Applications

Through transplanting/ artificial dispersal, dispersal limitations can be overcome, and the number of safe sites for lichen establishment can be influenced through selection or preparation of substrate. Reindeer lichen transplant studies conducted in Swedish forests indicate that the relative importance of the substrate may depend on the type of site (e.g. Roturier et al. 2007).

The influence that environment has on lichen growth and distribution is critical (Armstrong 1988), and there have been a number of lichen transplant studies conducted to investigate the tolerance and adaptability of forest lichen species to different forest types. These studies have demonstrated that transplants of lichen species associated with mature forest types can establish in younger forests, and survive at least one growing season (though few studies have been followed past two years).

2.0 Ecological Restoration Transplant Study

2.1 Thesis Objectives

The primary objective of this research was to investigate whether artificial introduction of reindeer lichen fragments could be a feasible part of the revegetation plan for reclaimed sites where these lichens form a significant part of the community. Years or decades of research are usually required to properly assess the long-term viability of a reclamation technique; however, since there has been no prior investigation into this question, I sought to provide a pilot study to open avenues of investigation for further reindeer lichen research by reclamation practitioners.

Revegetation research for land reclamation usually focuses on one of three main issues: species selection, site preparation, and determining reclamation success. Reclamation practitioners want to know which species might be appropriate for planting on their sites based on the species assemblages present on that landscape prior to disturbance, species assemblages that exist under similar edaphic conditions to those which have been created on the reclaimed site, and prior planting program experience on similar sites. They also want to know how to introduce those species (by seed, cutting, seedling, etc.), and how the site should be prepared (usually involving surface treatments like organic matter addition, contouring, mounding, or ripping of the soil) in order to provide the most favourable conditions for re-establishment of the desired species. Determining reclamation success is the most difficult issue, because it requires not just demonstrating that indicators of ‘success’ have been met, but determining what those indicators should be.

For example, indicators of successful forest re-establishment on reclaimed oil sands are achieving species mixes, stocking densities and site indices for commercial forest production (Oil Sands Vegetation Reclamation Committee 1998). The methods for determining stocking and site index are adapted from those developed for the forest industry (e.g. B.C. Ministry of Forests 1995; B.C. Ministry of Forests 2009).

Since reindeer lichens are currently not included in the species mix used for reclamation, there are no established techniques for assessing reclamation success using these species. In Chapter 1, I outlined the reasons that reindeer lichens are potential reclamation species. This trial was designed to investigate possible methods for determining successful lichen establishment, and one aspect of site preparation on successful lichen establishment (substrate) by one method of introduction (fragments).

I chose fragments as the method of introduction for this trial, over other methods such as transplanting clumps or entire lichen mats, because I started with the assumption that a technique of ‘seeding’ sites with lichen thallus fragments would be most cost-effective in terms of labour, harvest impacts on source sites, and amount of land that could be reclaimed with lichens for a given amount of harvested source material. In addition, fragmentation is the primary method of propagation of the reindeer lichens, and fragment dispersion was the primary method used in lichen transplant studies in the literature (e.g. Roturier et al. 2007; Liden et al. 2004; Enns 1988 reported in Stevenson et al. 2001).

I chose substrate as the site factor to investigate since it is the most easily manipulated for experimentation compared to other site factors that may affect lichen establishment, such as canopy closure and moisture regime.

The substrate treatments I chose were bare soil, moss, and conifer litter. These substrates are all readily available on reclaimed sites, requiring no new materials to be imported to the site, and are common substrates of lichen mats in the surrounding boreal forest. Reindeer lichens have been found growing on virtually all terrestrial substrates available in the boreal forests except bare rock (though moss and humus appear to be preferred) (Brodo, Duran Sharnoff and Sharnoff 2001; Pope 2005; Tolpysheva and Timofeeva 2008).

Mosses may provide a good substrate as they enable the stabilization of dispersed thallus fragments required for continued growth (Brodo 1973; Roturier 2009; Webb 1998), and prolong the active metabolic period of the lichens by retaining then releasing moisture like a sponge (Sillett and McCune 1998; Topham 1977). Litter should provide the best substrate for lichen fragment establishment, since it provides a textured surface that should result in good lichen retention, like moss, without the potential to grow over the lichen fragments and compete for light. Soil is the least suitable substrate from the perspective of lichen retention, but it is worth assessing as a potential substrate since the vigour and growth benefits of lack of competition from other vegetation may outweigh the lower retention rates.

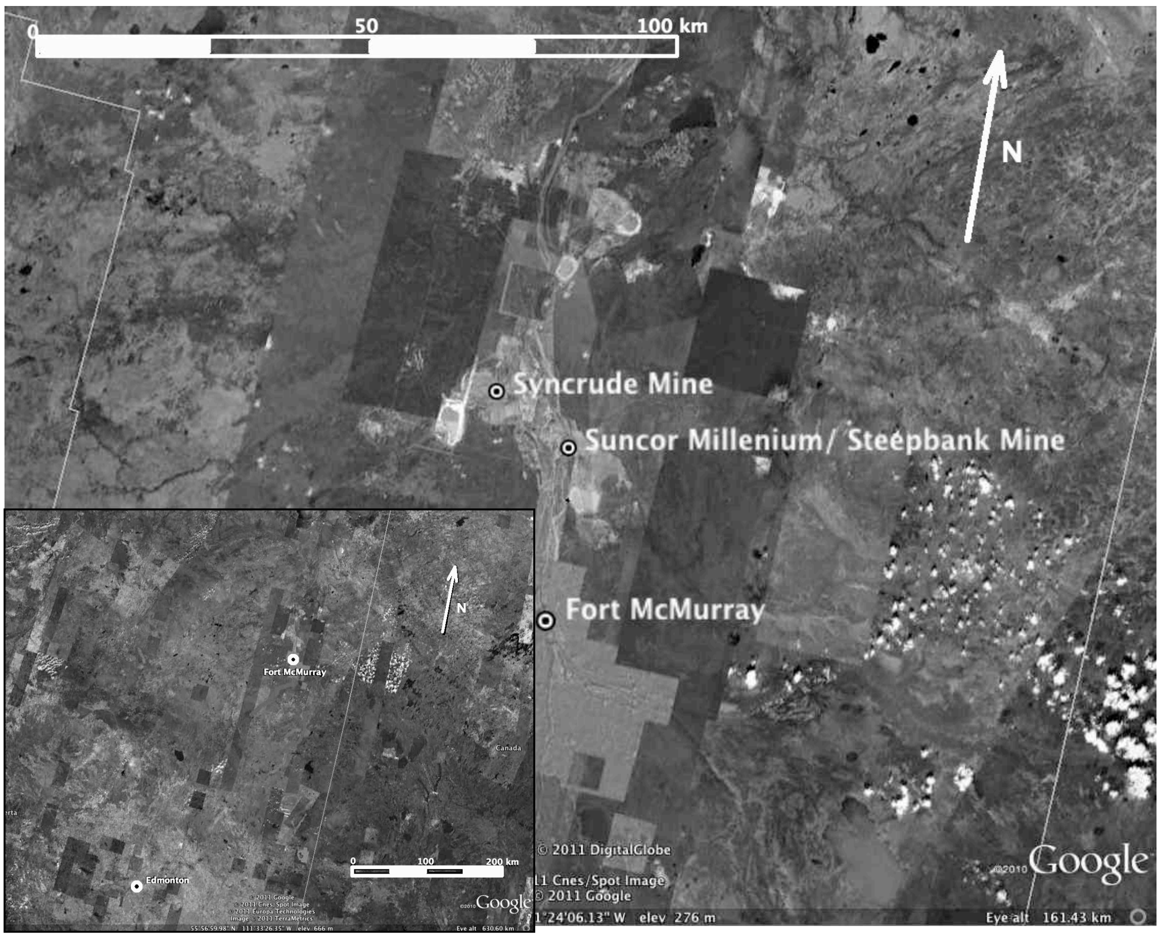
The indicators of lichen establishment chosen for assessment were:

- Fragment retention; defined as percentage of fragments remaining in the plot at each assessment,
- Vigour of the fragments placed in the trial plots using a qualitative rating scale,
- Change in two-dimensional lichen cover in the trial plots by photographic analysis, and
- Microscopic assessment of lichen growth (e.g. hyphal development, lateral branching).

Fragment retention is a good proxy for lichen survival, though it is not exactly the same – missing lichens may simply have moved, but are still alive elsewhere on the site. Reindeer lichens must also attach to a stationary substrate to continue to grow and form their characteristic mats, thus, for fragment dispersal to be a viable method of establishing lichen cover on the reclaimed site, fragments must remain in place after being dispersed onto a suitable substrate. Vigour is an indicator of likely future survival and growth of fragments that remain in plots. The assessment of lichen growth is difficult compared to larger vegetation species usually used in reclamation due to slow growth rate. Lichen mat growth is typically measured by change in area (e.g. Williston and Cichowski 2006), and change in area has been used for lichen fragment growth as well (e.g. Roturier et al. 2007; Roturier and Bergsten 2009). The presence of hyphal growth, annual branching, or lateral branching visible through microscopic assessment of the fragments may provide indication of lichen growth prior to measurable changes in lichen area.

2.2 Study Area Background

Suncor Energy Inc. and Shell Canada Energy funded this research as part of the Canadian Oil Sands Network for Research and Development (CONRAD) Environmental and Reclamation Research Group (ERRG) research program. The research trial was installed on Suncor's Millenium/Steepbank property (known colloquially as the Suncor mine, and hereafter referred to as Suncor), 25 km north of Fort McMurray, Alberta (Map 1).



Map 1 Suncor Mine location near Fort McMurray (inset map: NE Alberta).

2.2.1 Climate and Ecological Setting

<u>Climate</u>

The Suncor mine is situated in the Boreal Mixedwood ecological zone of Alberta, where the winters are long and cold, and the summers are warm, with marked differences between day and night temperatures (Beckingham and Archibald 1996). The growing season (annual frost free period) is usually 60 to 70 days from May to late August (Longley 1968 in Greenlee 1978). Mean summertime temperature (May to August) in Fort McMurray is 14.3°C, with a mean minimum of 7.5°C and mean maximum of 20.0 °C, and an average of 1755.3 Growing Degree Days (GDD) above 0°C (Environment Canada 2009). Winter (November to February) mean temperature is –14.4°C, with a mean minimum of –19.5°C and mean maximum of –9.3°C. The extreme low on record is –50.6°C in winter (recorded in 1947) and an extreme high of 37°C was recorded in summer (recorded in 1991).

Most precipitation falls in the summer months; the 30-year precipitation normal from 1971 – 2001 is 194 mm for the wet months (June through August), for a total annual precipitation of 455 mm (Environment Canada 2009). The dry season is from November to May, though up to 30 cm of snow can fall over the course of the winter months. This region experiences a net moisture deficit in most years, with an average potential evapotranspiration demand of 480 mm for the Fort McMurray region (Longley 1968 in Greenlee 1978).

The temperature and rainfall recorded at Suncor's property during the duration of the trial from 2009 to 2010 are provided in Figure 1 and Figure 2 with the 30-year rolling average obtained from Environment Canada (2009).

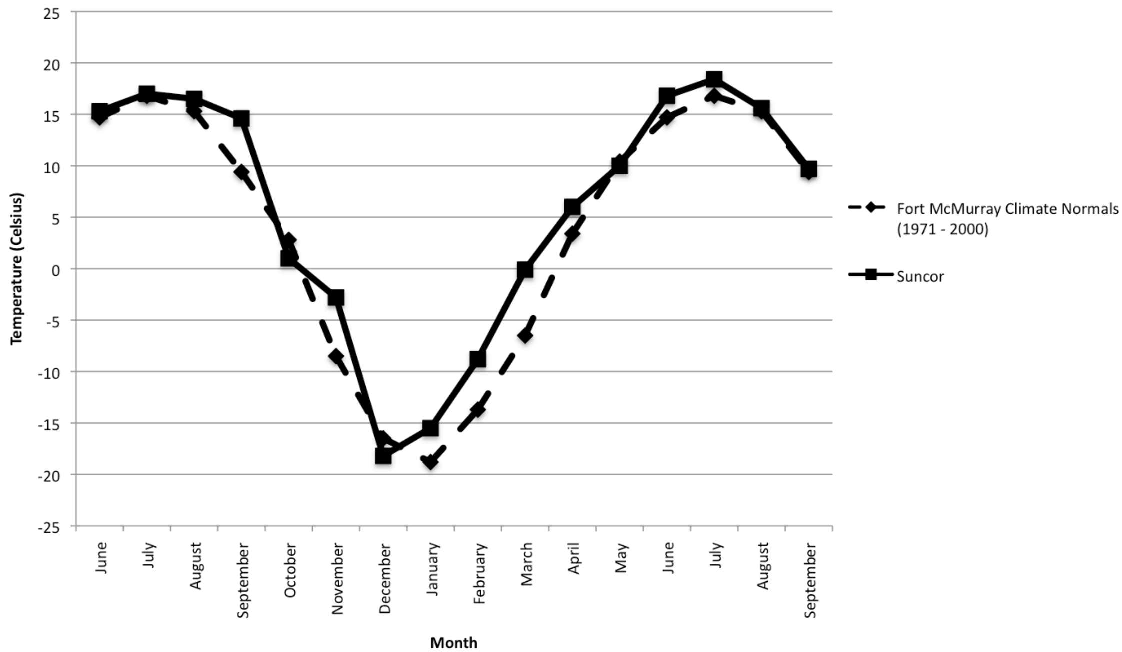


Figure 1 Study area average monthly temperature 2009-2010 versus Fort McMurray climate normal rolling average (1971-2000).

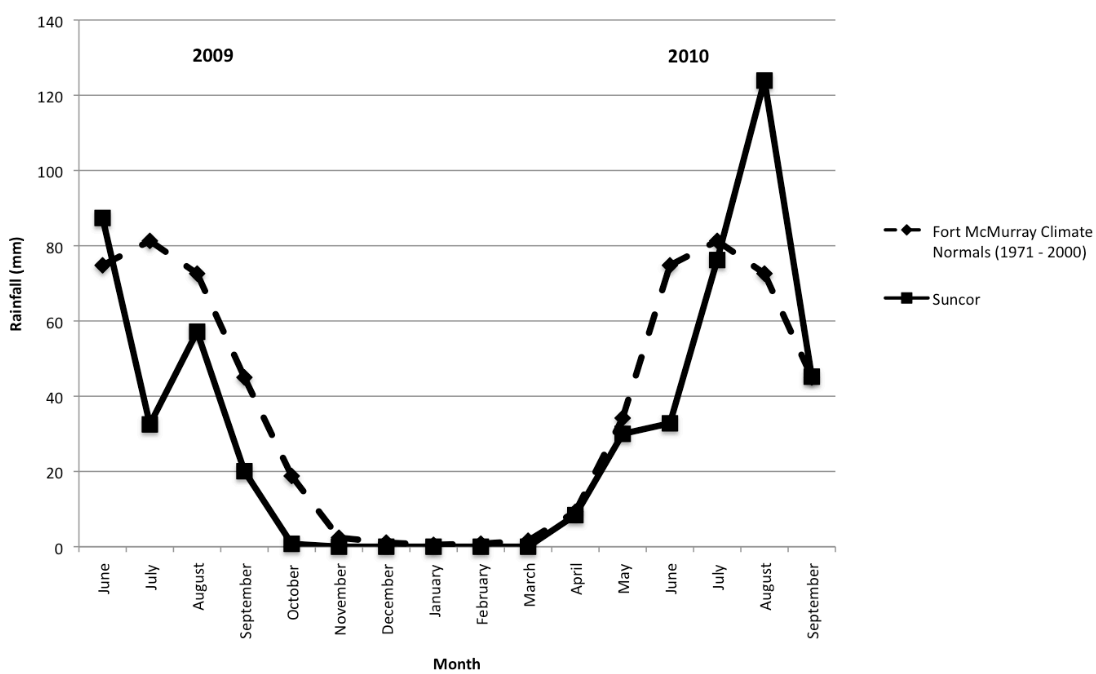


Figure 2 Study area average monthly rainfall 2009-2010 versus Fort McMurray climate normal rolling average (1971-2000).

The mean temperature for the duration of the trial did not deviate from normal, and the mean precipitation was slightly lower than normal during both growing seasons, except for the 70 percent higher than normal precipitation received during the month of August 2010.

Vegetation and Soils

The following description of the vegetation is taken from Beckingham and Archibald (1996), unless otherwise noted.

The study area is in the Mixedwood Section of the Boreal Forest Region. The forests in this region are dominated by stands of trembling aspen (*Populus tremuloides*), balsam poplar (*Populus balsamifera*), white birch (*Betula papyrifera*), white spruce (*Picea glauca*) and balsam fir (*Abies balsamea*), and mixed forests are common. Black spruce (*Picea mariana*) and tamarack (*Larix laricina*) dominate the lower areas with wetter edaphic conditions, and dry upland sites are dominated by jack pine (*Pinus banksiana*) with ericaceous shrubs in the understory.

The reindeer lichens are present on nutrient-poor ecosites with xeric to hygric moisture regimes throughout the Mixedwood Section. On well-drained upland sites, the lichens form a contiguous carpet beneath a jack pine canopy (ecosite phase a1: Lichen-Jack Pine). The dominant disturbance regime for jack pine forests in the boreal forest is forest fire; the mean fire return interval recorded for jack pine-lichen woodlands in northeastern Alberta and northern Saskatchewan (somewhat to the north of the study area) is approximately 38 years (Carroll and Bliss 1982). Jack pine colonizes sites immediately following fire, and the successional sequence for the vegetation community is similar to that recorded for lodgepole pine-lichen stands in British Columbia, with acrocarpus mosses arriving with the lodgepole pine, followed by the *Cladonia* cup lichens and then *Cladonia mitis* (see Coxson and Marsh 2001). However, in the Boreal Mixedwoods region, researchers found that *Cladonia mitis* colonizes burned sites immediately instead of with a 20 or more year delay (Dunford et al. 2006). In the absence of fire or with increasing soil moisture status, the Lichen-Jack Pine ecosite grades into the Labrador tea-mesic Jack Pine – Spruce (c1) ecosite phase, and the lichen cover is reduced in favour of the feathermosses. Bogs ('i' ecosites), poor fens ('j' ecosites), and Labrador tea – subhygric sites ('g' ecosites) all also have reindeer lichens present on localized patches of peat hummocks above the water table.

The trial locations are all on reclaimed sites intended to support dry upland jack pine sites, which in the undisturbed landscape have eolian, glaciofluvial, or fluvial-eolian sandy parent materials. The reclaimed sites will have soil profiles reconstructed using overburden material from the mining operation. These overburden materials are comprised mainly of the McMurray and Clearwater Formation materials; sodic shales and sandstones, overlain by lacustrine deposits of bedded silt, clay, and sand, and blankets of peat (Greenlee 1978).

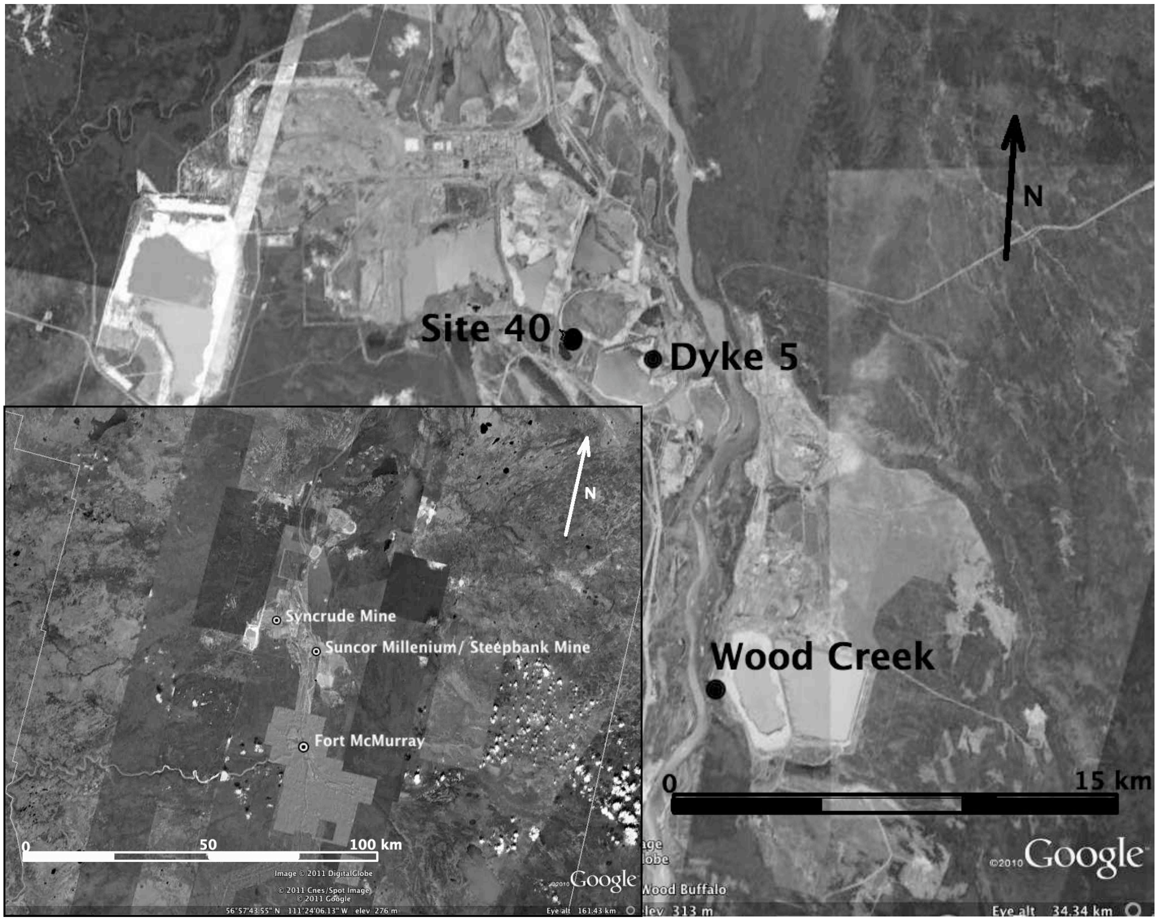
2.3 Trial Installation and Baseline Data Collection

2.3.1 Site Selection

During an initial site visit on May 27th 2009, four potential trial sites identified by the industry sponsors and a lichen source site at Wood Creek were located. In July 2009, two of the sites selected in May were dropped due to lack of suitability, but one site was added, for a total of three trial locations (Map 2). One dropped site was a newly planted area on level tailings sand capped with 30 cm of peat/mineral soil mix, and was free of herbaceous growth at the time of the first visit. This site was dropped from the trial installation in July since the site had come to support a dense growth of grasses and forbs after several weeks of rain. The second site selected in May but dropped from the trial was dropped at the request of the operators due to lodgepole pine (*Pinus contorta v. latifolia*) being the dominant tree species on the site. Industry sponsors believed that lodgepole pine stands should not be included in the trial, as the operators no longer plant lodgepole pine, and those sites may not be representative of typical reclaimed sites; lodgepole pine was previously used due to its ready availability from nurseries, but the more site-appropriate jack pine is now used instead.

Two of the trial sites that were selected were chosen to represent the oldest white spruce forests on the site, where a moss and lichen ground cover had already established (see Section 3.3 for a description of the cryptogam cover present). Both sites are located on Waste Area #19 (Lease 86/17) overburden dump, reclamation Site 40. Site 40 contains 15.2 ha of a hill covering north and west through south aspects, which was capped with 14 cm of direct-sourced muskeg soil, and planted with white spruce in 1985. One trial site is located on the north aspect (24 % slope), while the other is located on the south (14 % slope). The planting plan for north side of the site consisted of strips several metres across of alternating deciduous (hybrid poplar (*Populus deltoides x. nigra*), willow (*Salix* spp.), dogwood (*Cornus sericea*), prickly rose (*Rosa acicularis*) buffaloberry (*Shepherdia canadensis*), wolf willow (*Eleagnus commutata*), and saskatoon (*Amelanchier alnifolia*)) plantings and white spruce plantings, perpendicular to the slope. On the south side, the strips were planted parallel to the slope. Trees were planted approximately 2 metres apart in rows. The site was reportedly infill-planted with white spruce and lodgepole pine in 1994, though younger trees were not evident in the trial installation areas. The site was fertilized twice a year with N-P-K fertilizer in varying mixtures until 1989.

The third site is at Dyke 5, planted in 1997 on tailings sand capped with 36 cm of direct-sourced soil. The Dyke 5 site is 8.9 hectares, facing predominantly west (16 % slope), planted with white spruce, jack pine, balsam poplar, aspen, chokecherry (*Prunus virginiana*), and prickly rose. Jack pine dominated the site, with small roses found within the rows of conifers. Ground cover exclusion was not complete, as some alfalfa (*Medicago sativa*) (self-seeded) was still present, as well as some grass. The site was reportedly infill-planted with white spruce and aspen in 2001, but no young trees were evident in the trial area. The site was fertilized twice a year with N-P-K fertilizer at varying mixtures until 2001.



Map 2 Lichen transplant trial sites and Wood Creek source site on the Suncor Mine – Inset Map: Suncor Mine near Fort McMurray (see Maps 3 and 4 for detail).

2.3.2 Lichen Source Collection and Preparation

During reconnaissance to determine potential source sites and trial sites, a previously disturbed remnant white spruce/jack pine forest stand with abundant reindeer lichen was located on the Suncor Millenium/Steepbank property near Wood Creek (Map 2). Additional source sites in high deposition areas within the Voyageur South lease area and within the Joslyn North Mine lease area were also identified as potential lichen source sites, but Wood Creek was used since the lichen growing there should arguably be best adapted to the conditions on the mine site.

The most common reindeer lichen in the potential source sites was found to be *Cladonia mitis*, with lesser amounts of *Cladonia rangiferina* and *Cladonia stellaris* within the *C. mitis* mat. Thus, all source material collected was *C. mitis* to ensure adequate source material amounts, and to control for variation in establishment success that may exist between the species. *C. mitis* is also known to be the most tolerant of wind and direct sunlight (Ahti 1961; Brodo, Duran Sharnoff and Sharnoff 2001; Vitt, Marsh and Bovey 1988), and is frequently the first to colonize disturbed areas (Dunford et al. 2006; Helle, Aspi and Tarvainen 1983; Webb 1998), making it the most likely to be suited to the conditions of a reclaimed site[9].

Lichen source material was collected from three different locations within the Wood Creek forest. All locations likely receive full sun during parts of the day, and dappled to full shade at other times, so that the lichen source material should be adapted to the changing light conditions likely to be experienced on the trial sites. The lichen mats selected for harvesting were sprayed with distilled water from a laboratory wash bottle prior to harvest to prevent the lichens from being brittle and crumbling when handled. The uppermost 2 cm of each lichen thallus collected was separated out from the surrounding mat and placed into an opaque white plastic bag. Lichen thalli were collected in bags of 100 to 200 for ease of counting, and as each bag was completed, it was blown up with air and then tied.

The bags were stored in the shaded cab of the truck while being transported. The lichens were sprayed with a small amount of distilled water when they appeared to be drying out, and the bags were refilled with air when they began to deflate in an attempt to keep the lichens in a humid environment with enough air to prevent rotting, but enough moisture to prevent drying out and becoming brittle. Lichens that were stored overnight prior to placement in plots were kept in the researchers' hotel room in a dim corner, and checked for adequate moisture and air in the bags. On the last night, the first signs of rot were

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[9] *Cladonia arbuscula* is also described for this area, and has identical habitat requirements and morphology as *C. mitis*, though it is frequently said to be distinguishable in the field by the more prevalent 'wind-swept' appearance of the upper branches, which bend in one direction, versus the open, spreading branch form of *C. mitis* (Brodo, Duran Sharnoff and Sharnoff 2001; Goward 1999). Some authors treat *C. arbuscula* and *C. mitis* separately (e.g. Brodo, Duran Sharnoff and Sharnoff 2001; Goward 1999) while others treat them as subspecies, one species, or two species which intergrade to such a degree wherever they are conspecific, that there is little utility in trying to separate them (e.g. Ahti 1959; Kotelko, Doering and Piercy-Normore 2008; Osyczka 2006). Genetic analysis by (Stenroos et al. 2002) failed to separate *C. arbuscula* and *C. mitis*, thus I have chosen to refer to the lichen species I used as *C. mitis*.

discovered in lichens in a few of the bags, and these thalli were promptly disposed of, and the bags opened for better ventilation.

All of the thalli placed in the trials were firm and showed no signs of decay at the time of placement.

2.3.3 Trial Design

All trials were installed between July 6th and July 8th, 2009, with the assistance of two to three Suncor staff and an undergraduate assistant from the University of Victoria.

Layout

At each site, a total of 18 plots were laid out along a transect in 6 blocks of 3 plots each, one replicate of each treatment, and the order of the treatment plots in each block was randomized. The transect was installed between the tree rows, perpendicular to the slope, ie. along the same elevation to avoid changing edaphic conditions with slope position. Each block of three plots was placed at least 5 meters from the next (Figure 3), though on each of the trial locations, deciduous planting strips running perpendicular through the trial transect resulted in some treatment blocks being spaced up to 10 or 15 meters. Blocks were moved slightly up or down from the transect bearing to avoid trees. The degree of canopy cover varied along the transect, as no areas of the reclamation site were found to be homogeneously spaced enough to allow all plots to be in full canopy openings without moving to a very different position on the slope. Replicate plots within each block were separated by 1 m. Each replicate plot is a 0.70 x 0.70 m square (0.49 m^2), marked in the upper right and lower left corner with 45 cm red-flagged pigtail stakes pushed approximately 30 cm into the ground. An aluminum tree tag identifying each plot replicate number and treatment was affixed to the upper right stake.

In the center of the 0.49 m^2 plot, a 0.30 x 0.30 m square containing the treatment substrate was delineated using latex paint, then a strip of ground 0.2 m wide around it was cleared of all vegetation, duff, and large roots to prevent vegetation encroachment on the lichen treatments during the trials, and ensure that the treatment area boundary remained well-defined.

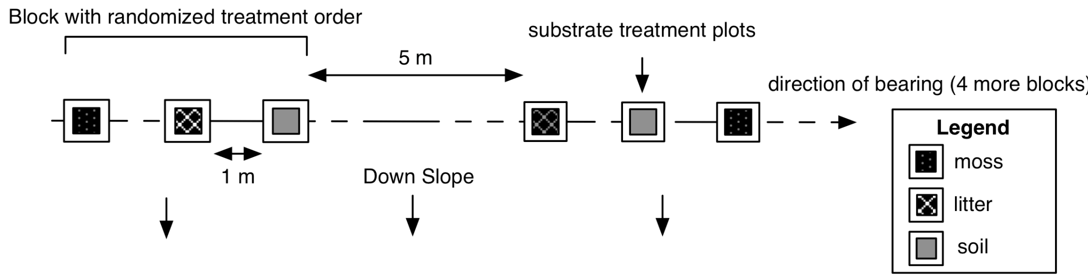
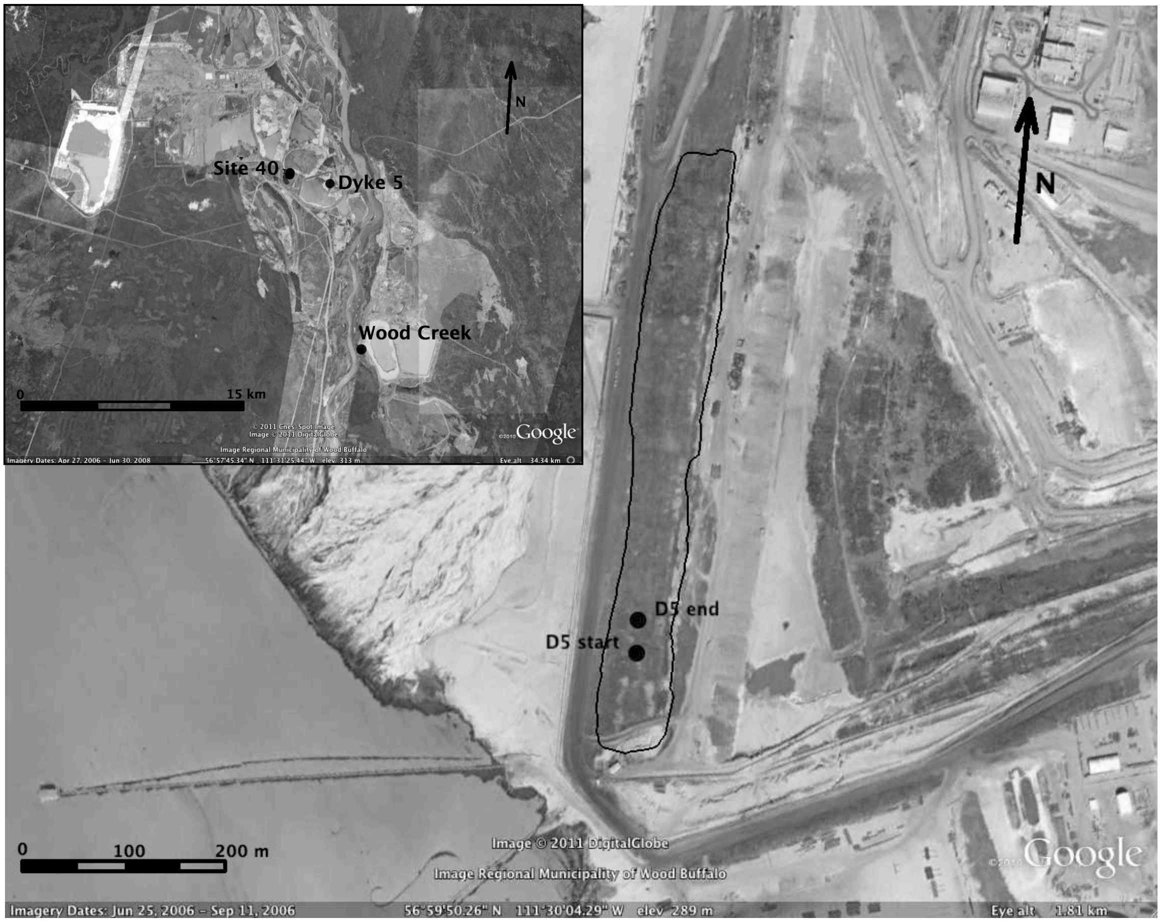


Figure 3 Lichen transplant transect layout illustration.

UTM coordinates were recovered at the upper right corner of each plot using a Garmin eTrex GPS, and plotted on GoogleEarth (Map 3 and Map 4).

Map 3 Site 40 transplant trial transect location - Inset Map: Trial locations on Suncor Mine (see Map 2 for Suncor Mine location in NE Alberta).



Map 4 Dyke 5 transplant trial transect location - Inset Map: Trial locations on Suncor Mine (see Map 2 for Suncor Mine location in NE Alberta).

Treatments

The three treatments within each block were bare surface soil, pine needle litter, or moss. The surface soil treatments were created by removing all litter, moss, vegetation, and humic layers from the plot until sandy mineral soil material was reached. At Site 40, the replaced muskeg soil was apparent at some surface locations as a mixture of sandy soil and peaty clumps, though sandy overburden materials were also likely to be at the surface, and, due to the substantial addition of organic matter to the surface soil that had occurred on the site, it was difficult to determine whether the mineral soil at the surface was muskeg or overburden beneath a developing LFH horizon. At the Dyke 5 site, the surface soil treatment was also sandy salvaged soil (see Section 2.3.5 for full descriptions of site soils). The litter treatments were created by removing all vegetation and humic material, then placing a thin layer (similar to the carpet beneath adjacent trees) of conifer need litter collected from outside the plot within the 0.09 m^2 square. The moss treatments were usually created by simply leaving the existing moss cover intact, and removing any grass or forbs poking through it. On the Site 40 trials, this moss cushion was usually thick and continuous, consisting almost entirely of *Hylcomium splendens*. Only one plot on Site 40S was created by creating a composite moss cushions from moss salvaged from outside the plot. On Dyke 5, the moss species present were all small acrocarpus mosses, and the cover was usually patchy; however, no sections of the moss were moved from outside the plot, as the moss crust was thin, sparse and friable.

On each treatment substrate, 25 2-cm long lichen fragments were laid out in an equal-spaced grid, and three squares of black fiberglass mesh were stapled securely to three corners of the plot, with a single thallus of lichen secured to each mesh square with waxed polyester outdoor thread (Figure 4).

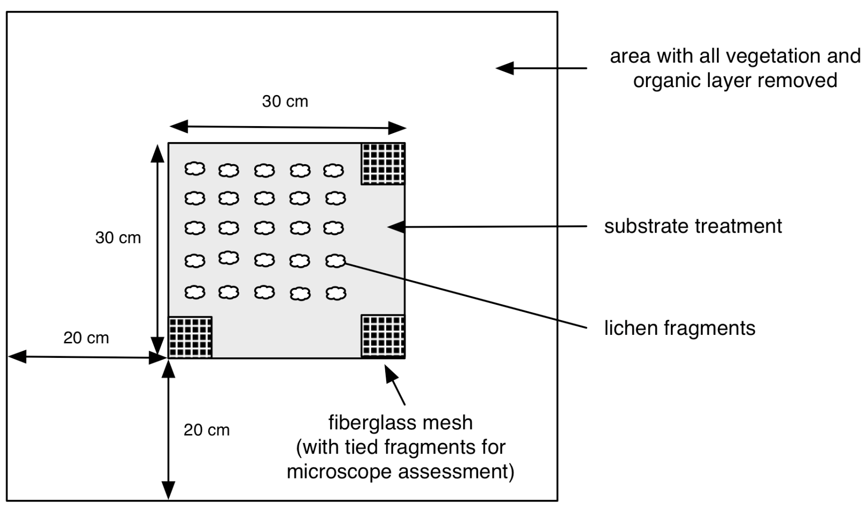


Figure 4 Lichen transplant plot layout illustration.

2.3.4 Baseline Lichen Growth Rate

Two samples of lichen were collected from open and forested positions in the source site in order to determine the baseline lichen growth rate on the study site for possible future comparison with lichen growth rate on the reclaimed sites. Each sample was collected in paper bags and transported back to the University of Victoria in a rigid, padded container to prevent fragmentation. The lichens were stored in the dark in unsealed paper bags then rehydrated with tap water and gently teased apart prior to measurement. Each thallus was separated out and the total length of live material was measured for 65 thalli from each cover type (130 thalli in total) using a length of string and a ruler; the string was laid along the thallus length, then removed and measured on the ruler. The number of annual branchings recorded was the number of branchings along the longest length of the thallus. The annual growth rate was then calculated for each podetium according to Equation 1:

Equation 1 Lichen annual growth rate calculation

$$\text{annual growth rate (mm/year)} = \frac{\text{total length (mm)}}{\text{number of annual branchings}}$$

The source site average growth rate was calculated for each cover category as an average of the 65 thalli measured from each class.

The average growth rate for C. mitis within the spruce forest at the Wood Creek site was 4.7 mm/year, and 3.8 mm/year in the open areas with Labrador tea cover (Table 1). While the only directly comparable measurement was Scotter's (1963) measurements in the black spruce forest in the Northwest Territories, where the average *C. mitis* growth rate was 3.6 mm, the *C. mitis* growth rate at Wood Creek is also within the range of annual growth measurements of other reindeer lichen species (which appear to have similar growth rates) in the boreal forest. The range of reported average growth rates for the reindeer lichens is from 3.4 mm/year (*C. stellaris* in northern Saskatchewan) to 5.9 mm/year (*C. rangiferina* in Russia) (see Table 2).

Table 1 *Cladonia mitis* growth rate at Wood Creek (under open and closed canopy cover)

Canopy Cover	Live Podetia Length (N = 65)		# Annual Divisions (N = 65)		Annual Growth Rate (N = 65)	
	Mean (mm)	**Range (mm)**	**Mean**	**Range**	**Mean (mm/yr)**	**Range (mm/yr)**
Open (0 % tree cover)	30	17 - 47	8	5 - 12	3.8	2.1 – 5.9
Forested (44 % tree cover)	42	16 - 74	9	5 - 14	4.7	2.2 – 7.4

Table 2 Reindeer lichen growth rates in the literature

Location	Species	Annual Growth Rate		Reference
		Mean (mm/yr)	**Range (mm/yr)**	
Suncor – Open	***C. mitis***	**3.8**	**2.1 – 5.9**	
Suncor - Forested	***C. mitis***	**4.7**	**2.2 – 7.4**	
Seward Peninsula, Alaska – Open	*C. rangiferina*	5.1	-	Pegau (1968)
	***C. arbuscula*[1]**	**5.4**	**-**	
	C. stellaris[2]	4.6	-	
Seward Peninsula, Alaska – Forested	*C. rangiferina*	5.6	-	
	C. arbuscula	**5.5**	**-**	
	C. stellaris	5.8	-	
Tahlston River, NWT - Forested	*C. rangiferina*	4.1	2.7 – 6.0	Scotter (1963)
	C. mitis	**3.6**	**2.7 – 4.9**	
	C. stellaris	3.4	2.5 – 4.8	
N. Saskatchewan – Forested	*C. rangiferina*	4.9	-	Scotter (1962 in 1963)
	C. stellaris	4.1	-	
Newfoundland - Forested	*C. rangiferina*	4.8	-	Ahti (1957 in Scotter,1963)
	C. stellaris	-	6 - 7	
Russia - Forested	***C. arbuscula***	**4.5**	**3.5 – 6.1**	Andreev (1954 in Ahti, 1959)
	C. rangiferina	5.9	4.2 – 5.1	
	C. stellaris	3.9	3.0 – 5.7	
Finland - Open	*C. rangiferina*	3.9	-	Helle et al. (1983)
	C. mitis	**3.2**	**-**	
Finland – Open Forest	*C. rangiferina*	3.9	-	
	C. mitis	**3.3**	**-**	
Finland – Dense Forest	*C. rangiferina*	4.3	-	
	C. mitis	**3.0**	**-**	

[1] authors listed used the synonym *Cladonia sylvatica* (L.) Hoffm. for *C. arbuscula* (Wallr.) Flotow. (Esslinger 2009)
[2] authors listed used the synonym *Cladonia alpestris* (L.) Rabenh. for *C. stellaris* (Opiz) Pouzar & Vězda (Esslinger 2009)

The growth rate of *C. mitis* at the Wood Creek site is within the range of annual growth recorded elsewhere in the circumpolar boreal forest.

2.3.5 Site Conditions

Canopy Cover

Light conditions at each site were characterized using 1) a spherical densiometer from the center of each block to record the canopy cover, and 2) a quantum meter calibrated to measure photosynthetic photon flux in sunlight (Apogee Instruments model BQM) to record the incident light at ground level in each plot. The time of day, sky conditions (clear or overcast), and shade status (ie. full sun, dappled shade, full shade) of the plot was recorded with each flux measurement. Where the flux changed constantly due to moving sunspots, the upper and lower range of the flux measured was recorded. The flux measurement was repeated at each site visit during the first year so that a record of the range of light incidences in the plots could be created. The flux data are not discussed further in this report, as the measurements did not add value to the description of site light conditions beyond what was provided by the canopy cover measurements. The flux varied widely in any given plot depending on the cloud cover and time of day; since the measurements were only taken during site visits, and not continuously over time, it was not possible to draw accurate conclusions about the true range of light conditions on the plots. For example, it was overcast and raining during the September 2009 site visit while taking measurements on Site 40 North (morning) and Dyke 5 (mid day), but was temporarily clear and sunny while at Site 40 South (afternoon).

The densiometer measurements were taken according to the manufacturer's directions (Lemmon RE, Forest Densiometer Instruction Sheet); holding the instrument at elbow level, held out from the body so that the researchers face is just outside the grid. Each of the 24 squares within the grid on the mirrored surface of the densiometer was divided into four imaginary squares. The number of canopy openings falling on each square equivalent in size to an imaginary quarter was then counted, for a total out of 96. This procedure was repeated facing each cardinal direction at each measurement point. The canopy cover for each block was later calculated as the averaged canopy opening score from each of the four directions, multiplied by 1.04.

The slope and aspect of each site was also recorded; only one slope and aspect measurement was needed for each site, as the slope was found to be constant along the length of the transects on each waste dump.

The average and range of canopy values encountered along the transects at each site, and in the sampling areas at Wood Creek, are provided in Table 3.

Table 3 Mean and range canopy covers on source and reclaimed sites

Site	Canopy Cover	
	Mean (%) (N)	Range (%)
Wood Creek	44 (2)	41 - 46
Site 40 -North	54 (6)	41 - 64
Site 40 - South	53 (6)	25 - 86
Dyke 5	22 (6)	10 - 43

The Wood Creek site had two distinct areas with lichen cover; one in the spruce forest, and another without any forest cover, dominated by Labrador tea. The mean canopy cover in the forested area was fairly constant throughout the small area, with the sample sites, separated by approximately 12 metres, having canopy cover values of 41 and 46 percent. The Dyke 5 site had a lower canopy cover than Wood Creek in all but one plot location (average 22 percent, ranging from 10 to 43 percent), which translated into higher average photosynthetic flux in the plots on this site on sunny days. The Site 40N and Site 40S trial locations had a higher average canopy cover than the Wood Creek site. Site 40N has the same minimum canopy cover as Wood Creek, but there are areas with much denser canopy cover. The Site 40S trial location has the most variable canopy cover, ranging from 25 percent cover in the more widely spaced sections, to 86 percent in the densest cover areas, where the tree spacing was very close and plots were close to tree boles.

<u>Substrate</u>

One composite soil, moss, and conifer litter sample was collected from each trial site and the Wood Creek source site at the time of plot installation. Soil samples were collected from the 0 – 5 cm mineral soil layer. Each sample is a composite of 6 subsamples taken from the area adjacent to each of trial plot blocks along the transect and combined in the field. Samples were frozen at the end of each sampling day, and remained frozen until prepared at ALS Laboratories, in Fort McMurray, Alberta.

Soil (and moss and litter) was collected as a composite sample across the transects, rather than as replicates for each plot or block, in order to assess coarse differences between the sites rather than to describe variability within the sites. Soil chemistry can vary substantially over distances of anywhere from a few millimeters to a few kilometers, so it is always difficult to determine what level of spatial replication counts as replication versus pseudo-replication, and to what spatial scale of soil chemistry variability the site vegetation is responding (e.g to chemistry differences over several meters, or in the few millimeters around the root-soil interface). The reindeer lichens, however, do not appear to derive nutrition through their hyphae from the substrates they are anchored on, meeting their needs instead from dry and wet deposition from the atmosphere (Crittenden, Katucka and Oliver 1994 in Crittenden 2000; Crittenden, Scrimgeour and Ellis 2004; Ellis et al. 2005; Hyvarinen and Crittenden 2000). Thus, there was no reason to assume that smaller spatial scale variability in the substrate chemistry between the plots would affect the lichens directly, whereas they may be affected by coarse differences in average site chemistry that results in differences in the vegetation community; e.g. nutrient rich, neutral soils on these reclaimed sites may indicate a potential long term competitive disadvantage for the reindeer lichens, which are successful in acidic, nutrient poor environments where there is less competition from higher plants.

Soil parameters assessed were pH (1:2 H_2O extract); Electrical Conductivity (EC) (1:2 H_2O extract); Sodium Adsorption Ratio (SAR); plant-available sulphate (SO_4), nitrogen (NO_3), ammonium (NH_4), phosphate (PO_4) and potassium (K); organic nitrogen (Total Kjeldahl Nitrogen or NH_3-N); strong acid extractable metals (by ICP-OES); and particle size (CSSS standard)[10].

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[10] All laboratory analysis methods and references are attached in the laboratory Chain of Analysis forms attached as Appendix E.

Tissue (litter and moss) parameters assessed were total Nitrogen (LECO combustible-N), total nutrients (by ICP-scan) strong acid extractable metals (by ICP-OES and ICP-MS) (see Appendices C and D for full results).

Soil

The reclaimed sites have similar soil textures, pH, sodium adsorption ratio (SAR), and electrical conductivity (EC) to each other (Table 4). The pH (H_2O) of the soil in the 0 – 5 cm depth on the reclaimed sites was 7.55 (Site 40 North) to 7.77 (Site 40 South), while the pH of the soil at the Wood Creek site was 4.22. Typical pH ($CaC1_2$) given for pine- and spruce-lichen forest soil in the region is in the 4.0 – 4.6 range (WBEA 2007)[11].

SAR, pH, and EC

The SAR and EC values are well within the limits for forest-supporting soil at all sites, though EC on reclaimed sites is slightly higher than surface soils in offsite areas, which, like Wood Creek, have typical values < 0.1 dS/m (WBEA 2007). The texture of the reclaimed sites is slightly coarser than at the Wood Creek site, which has approximately 20 percent clay and 10 percent silt fractions, compared to the reclaimed sites with 10 percent clay and 20 percent silt fractions (and 70 percent sand on all sites – see Appendix B for full soil analysis results). The difference in the soil textures between the Wood Creek and reclaimed sites is well within the range of soil textures associated with reindeer lichen-supporting sites (e.g. see Ahti 1977; Botting and Fredeen 2006; Brodo, Duran Sharnoff, and Sharnoff 2001; Brulisauer, Bradfield and Maze 1996; Sulyma 2000; Thomson 1967).

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[11] pH in water was chosen instead of pH in $CaCl_2$, as (Carter 1993) states this is the most accurate pH for soils with low EC and that are not being fertilized. PH in 0.1M $CaCl_2$ for most soils is approximately 0.5 pH units lower than in water.

Table 4 Sodium adsorption ratio, electrical conductivity, pH and texture of source and reclaimed sites

Site	pH	Sodium Adsorption Ratio (SAR)	Electrical Conductivity (EC) (dS/m)	Texture
Wood Creek	4.22	<0.1	<0.1	Sandy clay loam
Site 40 -North	7.55	0.19	0.29	Sandy loam
Site 40 - South	7.77	0.22	0.16	Sandy loam
Dyke 5	7.72	0.17	0.27	Sandy loam

Nutrients

Organic nitrogen was analyzed in the soil samples as Total Kjeldahl Nitrogen (TKN) and as the calculated value of TKN – NH_3. The values were identical within 0.001 percent, suggesting that the majority of the organic nitrogen in the 0 – 5 cm layer of soil exists bound within organic matter, not as free NH_3. TKN concentration was highest on the Site 40N site (0.199 %), and lowest at the Wood Creek site (0.067 %). TKN on Site 40S was 0.179 percent, and 0.122 percent on Dyke 5.

Dyke 5 had the highest concentration of plant-available nitrogen in the 0 – 5cm soil depth than all of the other sites (Figure 5); most of the available nitrogen on Dyke 5 was NO_3-N, while all the other reclaimed sites and Wood Creek had NO_3-N concentrations below detection limits. The remaining available nitrogen was found as NH_4-N, which was higher at the Wood Creek site than the reclaimed sites. Plant-available sulphate was much higher on Site 40, both aspects (68 and 45 mg/kg, respectively), than either Wood Creek or Dyke 5 (4 and 6 mg/kg, respectively); while Dyke 5 had much higher available phosphate levels than all the other sites (31 mg/kg versus less than 6 mg/kg on all other sites). The higher concentration of phosphate and nitrate on Dyke 5, the most readily leachable macronutrients on these sandy soils, may be due to the more recent history of fertilization (last fertilized 2001 versus 1989 for Site 40, and never for Wood Creek). The higher sulphate levels on Site 40 may be due to deposition from stack emissions; however in that case, nitrate should be elevated as well. Further nutrient cycling and deposition data would be required to determine the reasons for the different nutrient pool sizes on the reclaimed sites.

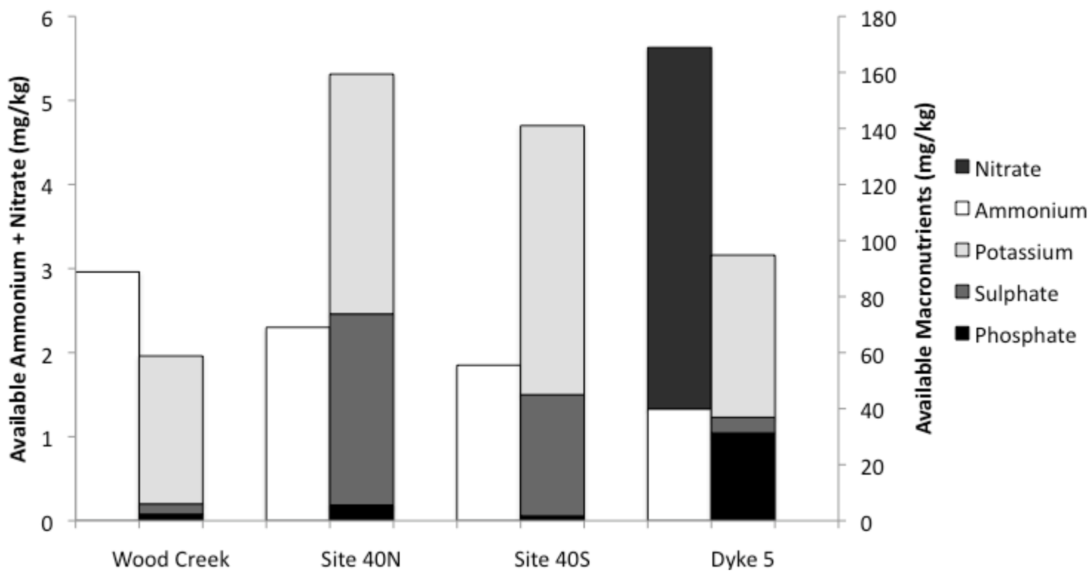

Figure 5 Soil macronutrient concentrations in the 0-5cm layer on Wood Creek and reclaimed transplant trial sites.

The exchangeable cation (Na^{2+}, Mg^{2+}, K^{+}, and Ca^{2+}) values are also higher in the 0 – 5 cm layer of soil on the reclaimed sites than in the Wood Creek soil (Figure 6).

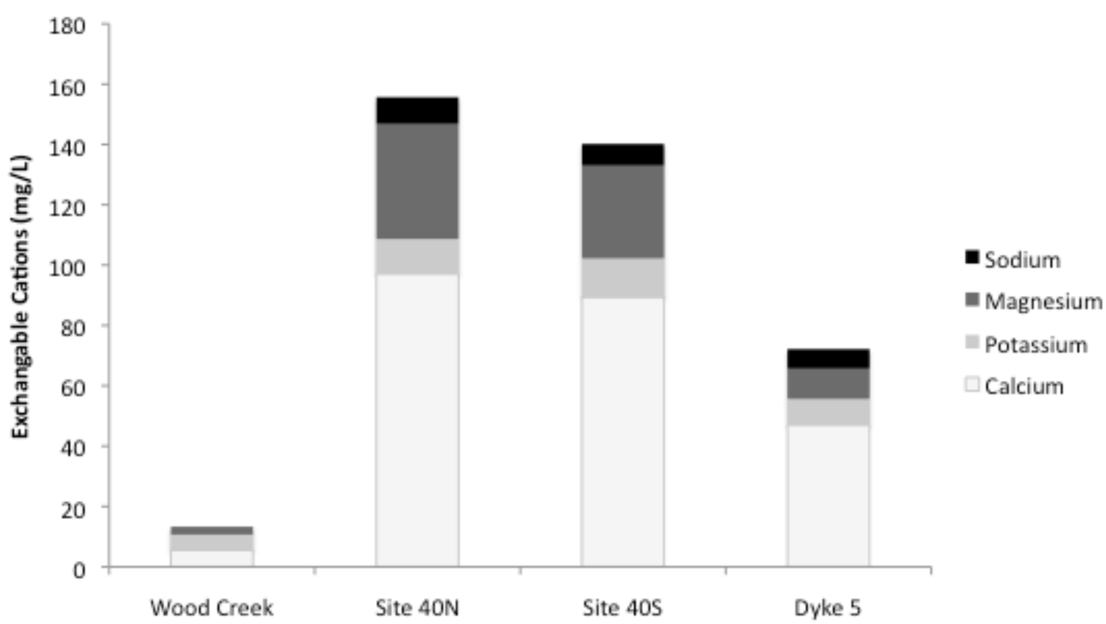

Figure 6 Soil exchangeable cation concentrations in the 0-5cm layer on Wood Creek and reclaimed transplant trial sites

Strong Acid Extractable Elements

The ICP-OES strong acid-extractable elements in the soil were compared to the Canadian Council of Ministers of the Environment guidelines (CCME 2007) and Alberta Contaminated Sites Regulation guidelines (Alberta Environment 2009) for Agricultural and Parkland soils (see Appendix B for full soil results). All soil metal parameters were below the most stringent CCME and Alberta CSR guidelines, with the exception of tin (Sn) in the Wood Creek soil sample, which was 5.6 mg/kg; above the 5 mg/kg guidelines for Agricultural soils under CCME guidelines, and Alberta CSR guidelines for both Agricultural and Parkland soils. The soil samples at each site are one aggregate sample from six subsamples along the transect and do not provide an estimate of the variation within the site. Thus, for the tin concentration in the Wood Creek soil, this might indicate a higher overall tin concentration in the soil, or one subsample with very high concentrations. Tin is not found to be a significant component of stack emissions in the oil sands region (Golder Associates Ltd. 2002 in WBEA 2007), or to have significant correlations with the health and vigour of arboreal fruticose lichens in acid deposition areas (WBEA 2007), thus, it is not likely that this elevated tin concentration comes from ongoing deposition that may affect the lichens.

The soil chemistry data indicate that the reclaimed site soils have higher pH and available nutrients than the Wood Creek source site and typical lichen-containing ecosites in the boreal forest; this may affect the development of the vegetation communities on these sites, and foster species that could outcompete the poorly competitive reindeer lichens for space.

<u>Litter and Moss</u>

Litter and moss composite sample data are not discussed within this report, because there is no reference for expected litter and moss chemistry values to compare to. The data were collected for completeness should future research on lichen substrate chemistry on the site indicate these values might be useful.

2.4 Methods

2.4.1 Fragment Retention and Vigour

At each monitoring visit (beginning and end of the growing season), the number of live thalli remaining in the plots, and the number of thalli in each vitality class was recorded. The vitality of the thalli was recorded using a 5-point scale adapted from Liden et al. (2004) shown in Table 5.

Table 5 Scale (adapted from Liden et al. 2004) used to assess lichen vigour on transplant trial plots

Category	Description
1	Necrotic, diseased
2	Bleached or melanic
3	Fragmented but otherwise vital
4	No infection or decay, some change in pigmentation
5	No parasitism, decay, further fragmentation, or change in pigmentation evident

The number of fragments visible outside of the plot, their distance from the 30 cm^2 inner plot, and their position relative to the plot on the slope (above, below, left, or right) was also recorded, but the data were not analyzed.

2.4.2 Growth and Establishment

At each visit one of the fragments tied to the fiberglass mesh in each plot was removed from the site for microscopic examination of hyphal growth (hyphal growth from branch ends and internodes), annual branching, and lateral branching. There was no published procedure for transporting storing lichen fragments treated in this way, so I developed my own; the mesh-anchored fragments were transported from the site and stored in clear plastic CD cases with the inserts removed. Each case had two pieces of mesh attached to the case with printer labels. The CD cases provided a rigid container that prevented crushing of the fragments, with an opening at the hinge to provide airflow. All samples were rinsed with distilled water to removed loose debris prior to microscopic examination and storage. All samples collected were stored for less than one week prior to microscopic examination, and were kept in a desiccated state in low light conditions at all times except during the rinse.

Photo Analysis

Photos of each plot were taken following the placement of the lichen in the plots using a Canon PowerShot A710 IS 7.1 megapixel camera mounted on a Manfrotto 190B tripod, which has an extendable camera mount arm which can be swiveled out and set horizontally at angles of 90 degrees or greater. This method was used by Roturier and Bergsten (2009) and Williston and Cichowski (2006) for plots on level ground; for the plots on the slope there was some difficulty in obtaining the appropriate angle with the boom arm to keep the camera at 90 degrees from the plot, and at a distance of 1 metre.

Photos taken at each visit were analysed using public domain image analysis software ImageJ version 1.41o (Rasband 2008). Each photograph was taken using a marked 30-cm ruler for scale, which was used to calibrate the scale of the photographs in ImageJ using the 'Analyze → Set Scale' function. The surface area of each lichen thallus was thus measured using the 'Analyze →Analyze Particles' function, and summed to give the total area of lichen in cm^2 for each plot.

Prior to area measurement, each photograph was converted from full colour to 8-bit (black and white), and occasionally the brightness or contrast of the photo was then adjusted to provide a starker relief between the lichen and the substrate. The threshold of each photograph was then manually adjusted to select the lichen pixels prior to running 'Analyze Particles'.

Plots with low contrast between the lichen and substrate (usually Litter plots, but also Soil plots where fragments were melanic and similar in colour to the soil) required erasure of other light-coloured objects in the plots using the 'Eraser' function in the 'Macro Tool Sets' prior to running 'Analyze Particles'.

In photographs with high contrast between the lichen and the substrate, a threshold value that selected pixels with a white value of approximately 190 to 255 (maximum range 0-

255, with pure black pixels receiving a value of 0, and pure white pixels receiving a value of 255) was usually adequate to capture the lichens. At subsequent visits, lower thresholds were frequently selected due to the change in colour of the lichens, or low contrast on overcast days. The process was subjective, so most plots were analyzed using Analyze Particles several times with slight adjustments to the threshold value to check the accuracy of the calculated outline against the original photograph. Once a threshold value provided an outline closest to the visible outline of the lichen in the original photo, that measurement was selected to provide the baseline lichen area for the plot. The total area per plot was recorded, and the average area per lichen fragment was calculated based on the recorded number of fragments in each plot.

Microscopic Examination

A Nikon SMZ800 10x to 63x dissecting microscope was used to view the lichen samples removed from the plots at each monitoring visit. Photos were taken of some fragments as exemplars of the features being described using a Wild M420 macrozoom system with 5.8x to 38x digital camera.

Prior to viewing with the microscopes, each fragment was sprayed with distilled water to rehydrate.

2.4.3 Statistical Analysis

The tests used for this statistical analysis were conservative and, coupled with the variability between the replicates and the low replication (of treatments on each site, and of sites), conclusions about the hypotheses of this trial based on these tests are preliminary. The conclusions were meant to be preliminary, as this was a pilot project aimed at opening avenues of investigation for further reindeer lichen research by reclamation practitioners, and was not meant to be a definitive study on lichen transplant techniques.

Statistical analysis of the effect of substrate treatment was completed for the assessment parameters lichen retention and lichen vigour, but not fragment area. Fragment area was excluded from statistical analysis when it became apparent that change in lichen fragment area was in large part a second metric to measure lichen vigour: the change in lichen fragment size was predominantly a decrease as fragments fragmented further, and fragmentation was already captured in vigour. Thus, by using both the vigour ratings and fragment area measurements to compare treatments, the likelihood of committing a Type I error, erroneously finding a significant effect on lichen vigour due to treatment when no treatment effect exists, was increased. Fragment area also turned out to be a poor assessment parameter due to the choice of measurement technique: recording the change in area of the lichen fragments with ImageJ was not useful, as the amount of fragmentation and burial of the fragments made the change in visible size of the fragments a poor indication of actual condition. In addition, many of the lichen fragments darkened as the trial progressed, decreasing the contrast between the lichens and the substrate and making distinguishing the lichens from the substrate increasingly difficult for the software.

Non-Parametric Test Selection Rationale

All statistical analyses of lichen retention and vigour were completed using Kruskal-Wallis Tests followed by Wilcoxon Rank Sum Tests for multiple comparisons. Comparisons of fragment retention and vigour were completed separately for each site and for each assessment date. Non-parametric tests make no assumptions about the distribution or variance of the data, and are thus less powerful relative to parametric methods; i.e. they are more likely to result in a Type II error - the acceptance of the null hypothesis (that there is no significant difference between treatment means) when the null hypothesis is actually false - than parametric tools such as ANOVA or MANOVA. However, the assumptions underlying the use of parametric statistics are i) that the data have normal error distributions, and ii) that the variance of the treatment groups is the same (Leach 1979; Ostle and Malone 1988; Snedecor and Cochrane 1980; Underwood 1997). The fragment retention data from September 2009, and vigour data from all monitoring visits were non-normally distributed. The variances for each parameter were also frequently not equal among treatment groups. In addition, some plots had fragment retention rates of zero in 2010, resulting in uneven numbers of replicates for analysis of the vigour data, The use of ANOVA/MANOVA on non-normal data increases the likelihood of Type I error, or concluding that the treatments are significantly different when they are not, while the use of ANOVA/MANOVA on data with unequal variances increases the likelihood of a Type II error, or concluding that the treatments are not significantly different when they are. Since the use of ANOVA/MANOVA could result in a greater chance of a Type I error (as well as a loss of power due to assumption violations), non-parametric tests were used for all analyses.

All analyses and figures were completed using open-source statistical computing software *R* (R Development Core Team 2008).

The family-wise error rate for fragment retention and vigour comparisons between treatments in each site-by-assessment date family of inferences was controlled at α = 0.10. Thus, differences in each of fragment retention or vigour due to treatment on each site during each assessment period are significant at an α of 0.05. Each site-by-assessment date was treated as a separate family of errors since inferences were not aggregated across sites (i.e. the statistical difference between treatments was evaluated separately for each site when deciding whether to reject the experimental null hypothesis that there is no difference in the success of establishing lichen fragment transplants between treatments), and the comparisons between treatments at each assessment date were planned to provide an indication of the time since transplant required to see difference between treatments. Treatment comparisons were completed within sites, rather than across sites using site as a factor, due to the rationale described in Factor Selection Rationale, below.

When there was a finding of significant difference between treatments with a Kruskal-Wallis Test (α = 0.05), the per-comparison error rate for multiple comparisons between treatments was adjusted using the Dunn-Sidak equation so that each treatment comparison (moss-litter, moss-soil, and litter-soil) is significant at α = 0.0169 with the Wilcoxon Rank Sum Test.

Factor Selection Rationale

Both trial areas on Site 40 were 24/25 years old, and the site at Dyke 5 was 12/13 years old; Site 40N and Dyke 5 both faced moist aspects (north and east, respectively) while Site 40S faced a dry aspect. Site age was not included as a factor in the statistical analysis of the trial data, as any differences in lichen survival or vigour correlated with differences in site age would likely be due to changes in other site factors which vary with site age, such as canopy cover or understory development. Understory varied between the sites - more grass and legumes, less moss, and more bare soil on Dyke 5; well-developed feathermoss cover on Site 40N; and low moss cover on Site 40S – which may have had direct (e.g. competition for light with feathermoss) or indirect (e.g. runoff interception by moss mats) effect on lichen survival or vigour. According to Beckingham and Archibald (1996), *C. mitis* occurs on north, south and west aspects in the region, though typically at shallower slopes than reclaimed dumps, and most commonly on flats. *C. mitis* would likely have a competitive advantage on drier aspects, since the terrestrial lichens dominate in dry and poor edaphic conditions, but transplants may have better initial survival on moist sites. However, since none of the aforementioned factors which varied by site could be teased out and assessed separately with only three trial sites (except canopy cover, but see discussion below), they were not included in statistical analysis of treatment effects by a method such as GLM.

Canopy cover in the treatment plots varied from a low of 10 percent on Dyke 5 to a high of 86 percent on Site 40S. The relationship between canopy closure and lichen cover within sites has been described for a number of forested lichen sites (Lechowicz and Adams 1974; Sedia and Ehrenfeld 2003; Sulyma and Coxson 2001; Williston and Cichowski 2006), and positions with higher canopy cover on a forested lichen site typically have higher moss cover, while lichens dominate in the canopy openings (Pharo and Vitt 2000). However, Goward's (2000) observations were that lichen cover was not influenced by tree spacing - which affects canopy closure - when tree spacing was between 0.5 and 4 m in mature spruce forests in B.C., (when trees were greater than 4 m apart, increased lichen cover was promoted by the foraging activity of caribou).

Replication under different canopy covers on these trial plots was too low to use a linear regression to determine whether there was a relationship between canopy cover and lichen survival or vigour. Thus, I attempted to determine whether there was a rationale for grouping plots into canopy cover classes (e.g. open, partial, closed) during statistical analysis, and to determine what the cover class brackets should be using published data. The only published paper with complete percent canopy cover and lichen cover data I could find was by Carroll and Bliss (1982), who examined the relationship between vegetation community and fire return interval in jack pine-lichen stands in northern Alberta and Saskatchewan. *C mitis* cover was positively correlated with canopy cover on sites less than 50 years old, where *C. mitis* cover was also positively correlated with time since disturbance; as sites aged and self-thinned, there was a slight negative correlation between *C. mitis* cover and age (likely as *C. mitis* was replaced by *C. stellaris*) and hence *C. mitis* cover and canopy cover (see Appendix E for my statistical analysis of Carroll and Bliss's (1982) data). Thus, the data from Carroll and Bliss (1982) failed to indicate that canopy cover had a large enough effect on lichen establishment or survival to warrant including cover as a factor in my statistical analyses.

Kruskal-Wallis Rank Sum Test

The Kruskal-Wallis Rank Sum Test is used to test for statistical differences among three or more groups where the assumptions for an ANOVA are violated (Ostle and Malone 1988). Because the Kruskal-Wallis Rank Sum Test involves assigning ranks to the values in each group and comparing the distribution of the ranks rather than comparing variances about the mean (as in ANOVA), the null hypothesis for the Kruskal-Wallis Rank Sum Test is that the probability distributions of the ranks in each group are identical, and the alternative hypothesis is that the groups do not have the same median (Ostle and Malone 1988).

Wilcoxon Rank Sum Test

The Wilcoxon Rank Sum Test is used to test for statistical differences between two groups where the variances are unequal and the errors are not normally distributed (Crawley 2005); this test was used to make pairwise comparisons between treatments where a significant difference was found using the Kruskal-Wallis Rank Sum Test, as well as to compare initial and final vigour for each treatment on each site.

2.5 Results and Discussion

The results presented here are preliminary due to the limitations for statistical interpretation discussed in Section 2.4.3, i.e. low replication and sample size, and large variability between replicates within treatments necessitating the use of non-parametric statistics, which limits the statistical power of the comparisons and the ability to make inferences about the existence of treatment effects. The low number of sites which were eligible to be included in the study also reduced the ability to draw statistical inferences on the potential contribution of site factors aside from substrate on the establishment of the lichens on the trials.

2.5.1 Fragment Retention

Fragment movement out of the plots was likely due to water, wind, and animals. All three sites were on slopes (ranging from 14-26 %), thus, most fragment movement was expected to be down slope, and primarily due to water transport during heavy rainfall events.

Animal activity was evident at the first monitoring visit in the plots in the Site 40N and Site 40S trial locations: most of the plot tags had been chewed or clawed, and clods of moss and litter had rolled or been kicked into some of the plots. Coincident with the animal activity in the older forested plots, fragment movement out of the plots was frequently to positions upslope of the plots or to either side, rather than predominantly below the plots as was seen on the Dyke 5 plot.

Lichen retention for all sites and assessment dates are shown in Figure 7. The box-and-whisker plots show the median (the horizontal bar), and the top and bottom of the boxes show the 75th and 25th percentiles, respectively. The whiskers extend to the minimum and maximum values in the dataset, except where the values are smaller or greater than 1.5 times the interquartile range (IQR); where this occurs, the whiskers show the 1.5

minimum or maximum IQR, with outliers denoted as hollow circles. Where no upper or lower whiskers are shown, all values, except any outliers, are contained within the upper and lower quartiles, respectively.

Substrate treatments within Dyke 5 joined by a line with an asterisk have significantly different median fragment retention following multiple pairwise Wilcoxon Rank Sum Tests with $\alpha = 0.0169$ for each comparison (to maintain a family-wise error rate of 0.05); there was no significant difference between substrates on Sites 40N or 40S at any assessment date using the Kruskal-Wallis Rank Sum Test.

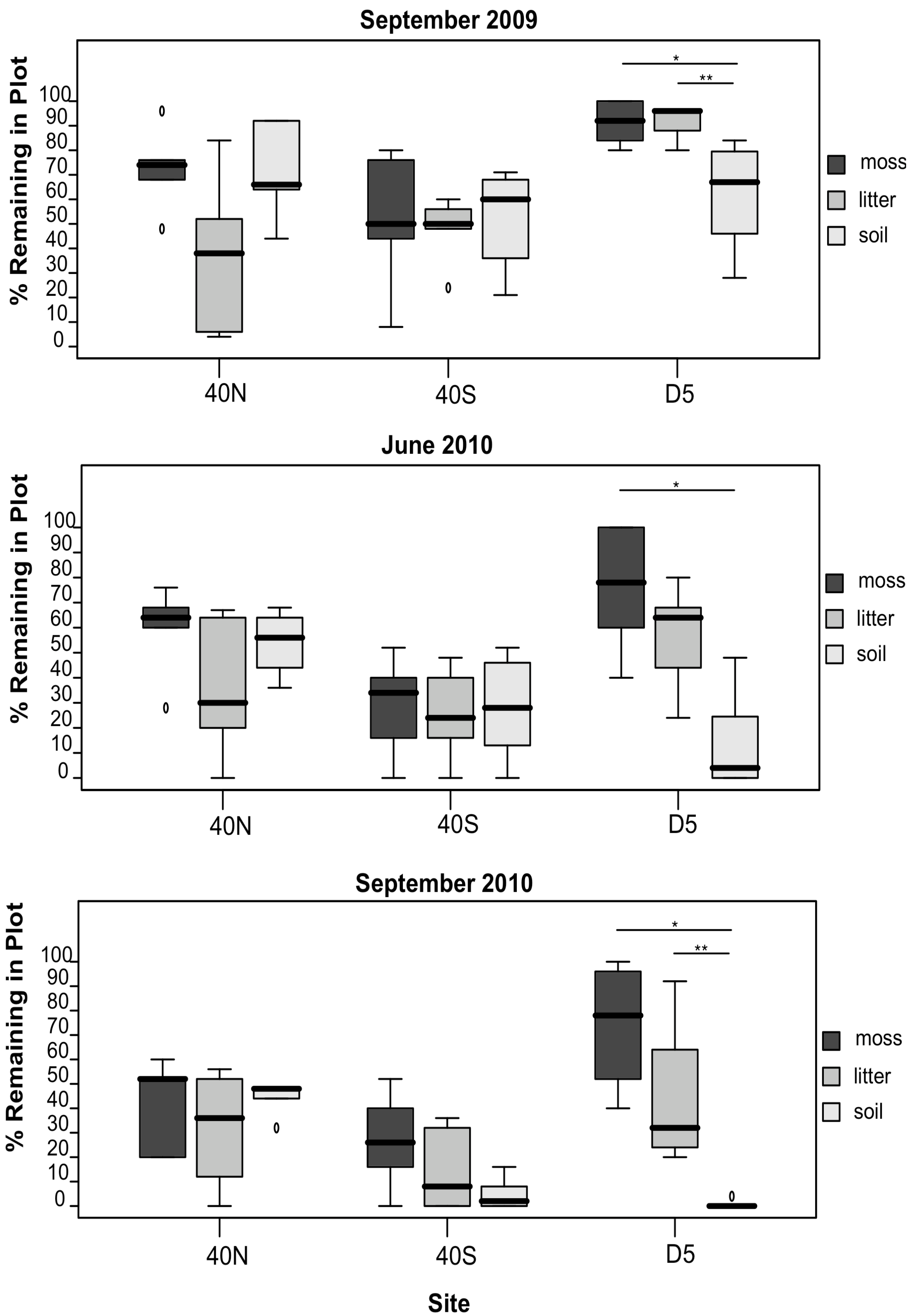

Figure 7 Fragment retention (% remaining in plot) on different substrates by site for all assessment dates.

Variance in fragment retention within substrate treatments was high, and the only site for which substrate made a significant difference in fragment retention was Dyke 5 (see Table 6 for Kruskal-Wallis Rank Sum Test statistics for all sites on all assessment dates).

Table 6 Kruskal-Wallis Rank Sum Test results for comparison of fragment retention by substrate (moss, litter, soil) on all sites for all assessment dates. Numbers in bold show statistically significant differences at α = 0.05

Assessment Date	**Site**	χ^2	df	*p*
September 2009	Site 40N	2.399	2	0.301
	Site 40S	0.2478	2	0.883
	Dyke 5	9.649	2	**0.008**
June 2010	Site 40N	3.435	2	0.180
	Site 40S	0.169	2	0.919
	Dyke 5	10.679	2	**0.005**
September 2010	Site 40N	1.403	2	0.496
	Site 40S	4.503	2	0.105
	Dyke 5	13.695	2	**0.001**

On Dyke 5, median fragnment retention in September 2009 was significantly higher on moss than soil (Wilcoxon Rank Sum, W=39.5, p=0.010), and litter than soil (Wilcoxon Rank Sum, W=33, p=0.014), with no significant difference between fragment retention on moss and litter (Wilcoxon Rank Sum, W=16.5, p=0.8507). In June 2010, median fragment retention was significantly higher on moss than soil (Wilcoxon Rank Sum, W=41, p=0.005), with no significant difference between moss and litter or litter and soil (Wilcoxon Rank Sum, W=22, p=0.234, and W= 32, p=0.022, respectively). At the end of the trial in September 2010, median fragment retention was significantly higher on the moss (78 %) and litter (38 %) treatments than the soil treatment (0 %) (Wilcoxon Rank Sum, W=42, df=2. p=0.002, and W=35, df=2, p=0.003, respectively), with no significant difference between median fragment retention on moss and litter (Wilcoxon Rank Sum, W=24, p=0.121).

Overall fragment retention was low after two growing seasons; the median fragment retention was less than 50 percent for all treatments except moss on Site 40N (52 %) and Dyke 5 (78 %).

2.5.2 Lichen Vigour

Vigour was rated on a scale from 1 through 5, with 5 being the highest score (healthy and unchanged from initial condition). Categories 3, 4, and 5 were healthy thalli; category 3 denoted a healthy thallus that had fragmented (obvious loss of branches, or very small fragment remaining), and category 4 denoted thalli that were healthy but had changed colouration (fragments placed in plots with more shade than source location darkened, and those in plots with more sun exposure frequently bleached). Categories 2 and 1 were unhealthy thalli showing signs of tissue necrosis or disease.

For statistical comparisons of the fragment vigour on each substrate, the weighted mean[12] of the fragment vigour of each replicate plot was used. Plot vigour for all sites and assessment dates are shown in Figure 8.

Substrate treatments within Dyke 5 and Site 40S joined by a line with an asterisk have significantly different distributions following multiple pairwise Wilcoxon Rank Sum Tests with $\alpha = 0.0169$ for each comparison (to maintain a family-wise error rate of 0.05); there was no significant difference between substrates on Site 40N using the Kruskal-Wallis Rank Sum Test (see Table 7 for Kruskal-Wallis Rank Sum Test statistics for fragment vigour for all sites on all assessment dates).

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[12] The sum of the number of fragments in each fragment category multiplied by the vigour category value, divided by the total number of fragments in the plot.

Table 7 Kruskal-Wallis Rank Sum Test results for comparison of fragment vigour by substrate (moss, litter, soil) on all sites for all assessment dates. Numbers in bold show statistically significant differences at α = 0.05

Assessment Date	Site	χ^2	df	*p*
September 2009	Site 40N	1.679	2	0.432
	Site 40S	1.213	2	0.545
	Dyke 5	10.680	2	**0.005**
June 2010	Site 40N	5.443	2	0.066
	Site 40S	11.67	2	**0.003**
	Dyke 5	11.121	2	**0.004**
September 2010	Site 40N	4.924	2	0.085
	Site 40S	6.290	2	**0.043**
	Dyke 5	2.213	2	0.331

There was no significant difference in fragment vigour among the different substrates on Site 40S in September 2009 or September 2010, but there were significant differences in fragment vigour between substrates in June 2010. In June 2010, median fragment vigour was significantly higher on moss than soil (Wilcoxon Rank Sum, W=36, p=0.0.005) and on litter than soil (Wilcoxon Rank Sum, W=36, p=0.005) on Site 40S; there was still no significant difference between median fragment vigour on moss and litter (Wilcoxon Rank Sum, W=20.5, p=0.742).

On Dyke 5, there were significant differences in median fragment vigour among substrates in September 2009 and June 2010, but not in September 2010. In September 2009, median fragment vigour was significantly higher on moss than soil (Wilcoxon Rank Sum, W=39, p=0.012), and on litter than soil (Wilcoxon Rank Sum, W=35, p=0.006) but there was no significant difference between fragment vigour on moss and litter (Wilcoxon Rank Sum, W=21, p=0.311). In June 2010, median fragment vigour was higher on moss than soil (Wilcoxon Rank Sum, W=42, p=0.001), but there was no significant difference between moss and litter (Wilcoxon Rank Sum, W=26, p=0.049) or litter and soil (Wilcoxon Rank Sum, W=24.5, p=0.208).

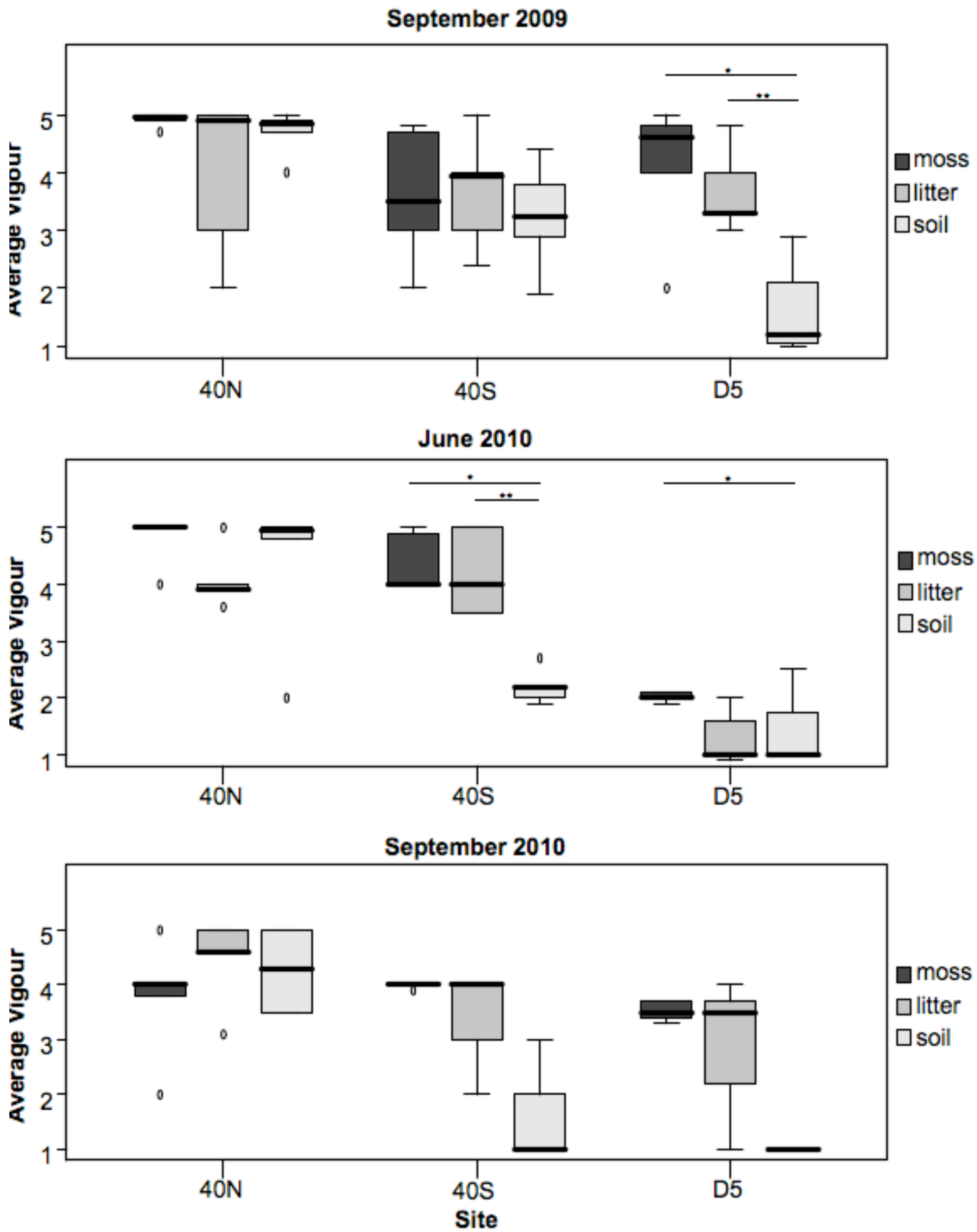


Figure 8 Fragment vigour (% remaining in plot) on different substrates by site for all assessment dates.

Even though fragment retention for plots on Site 40N was low (see Section 2.5.1, above), the remaining lichen fragments remaining were healthy on all treatments. On Site 40S, median fragment vigour was also high on moss and litter plots in June 2010 and September 2010, with an initial drop in median vigour during the first growing season due primarily to fragmentation of the transplanted thalli. The median fragment vigour appeared to increase on the plots after September 2009, as the thalli that had fragmented were no longer recorded in the plots. On Dyke 5, median fragment vigour was high on moss and litter plots in September 2009 and September 2010, but was low in June 2010. Since the vigour data presented includes only remaining fragments (missing fragments cannot have vigour), in some cases it appears that some treatments have thalli that improve in vigour (e.g. Dyke 5 moss and litter treatments); however, the vigour of the plots 'improved' due to the death of diseased thalli, and only healthy thalli remained and were recorded until the end of the trial (i.e. all missing fragments were removed from the weighted mean vigour for the plots).

The median vigour on the soil substrate on Dyke 5 was low by the end of the first growing season and remained low, while the median vigour on soil substrate on Site 40 was high in September 2009, but declined by June 2010, remaining low through the second growing season. The proportion of remaining thalli in each vigour category in each plot at each data collection point is shown in Figure 9 through Figure 11 below.

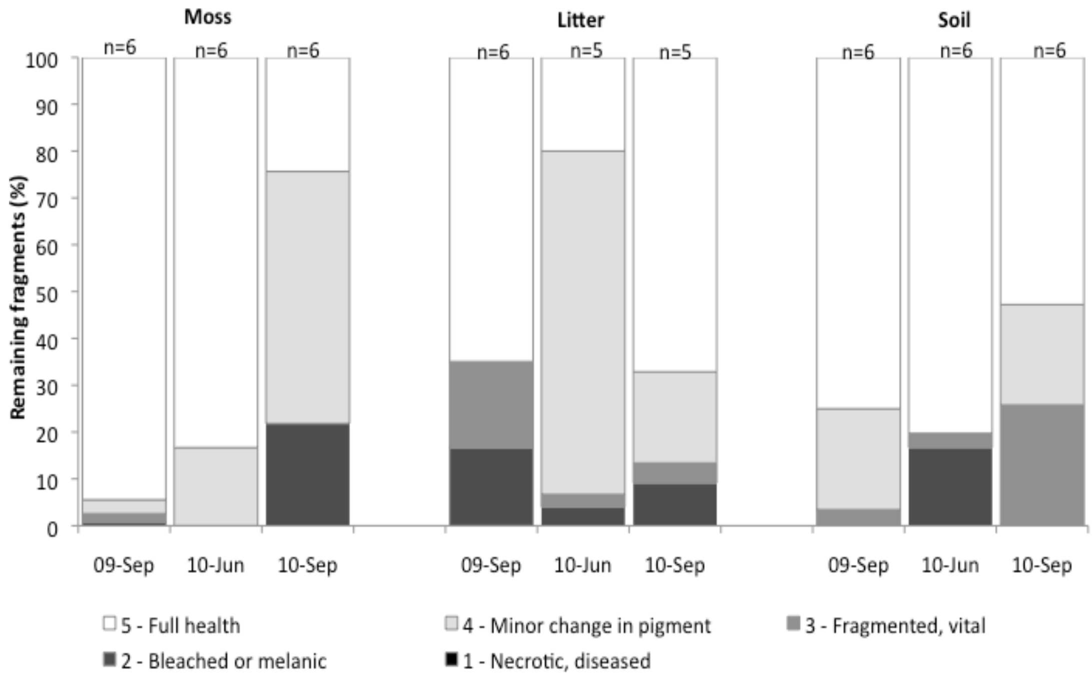


Figure 9 Site 40N lichen fragment vigour by treatment and assessment date.

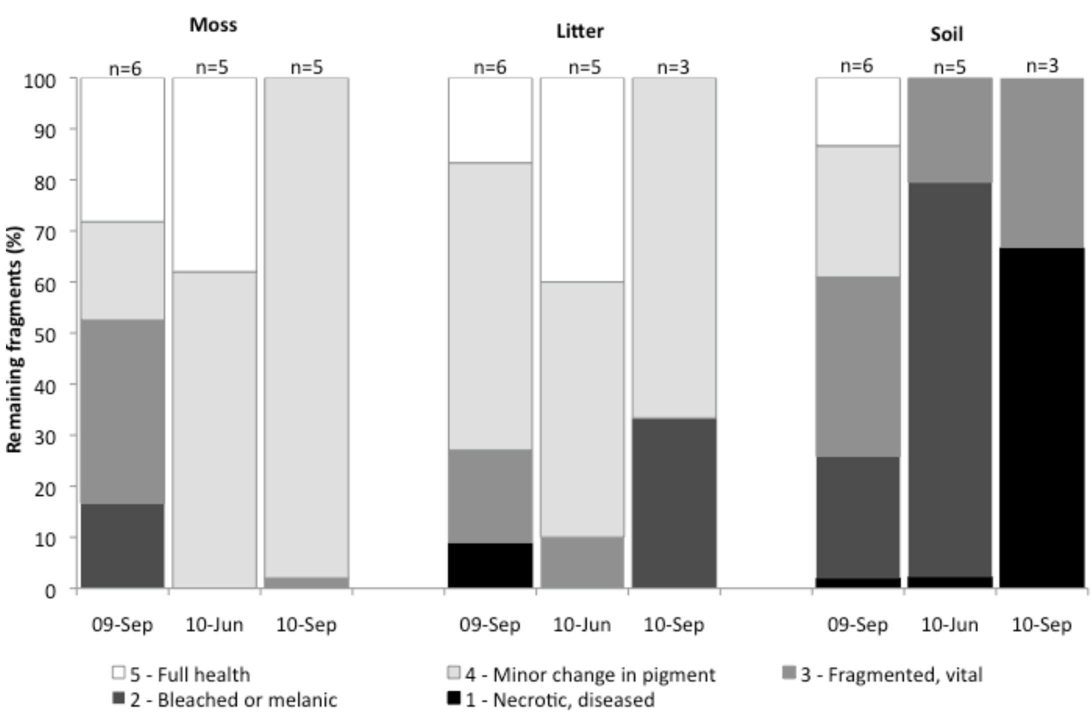


Figure 10 Site 40S lichen fragment vigour by treatment and assessment date.

Site 40S had a higher proportion of melanic thalli due to increased algae production, suggesting that the plots likely received more shade than at the source site. Thalli on the soil plots on Site 40S were frequently in poor condition due to being scoured and coated with sand transported by water. Thalli on Dyke 5 were frequently bleached, suggesting the death of algae due to increased sun exposure on Dyke 5 relative to the source site. Thalli on the soil treatment on Dyke 5 were in very poor condition due to scouring and coating with sand.

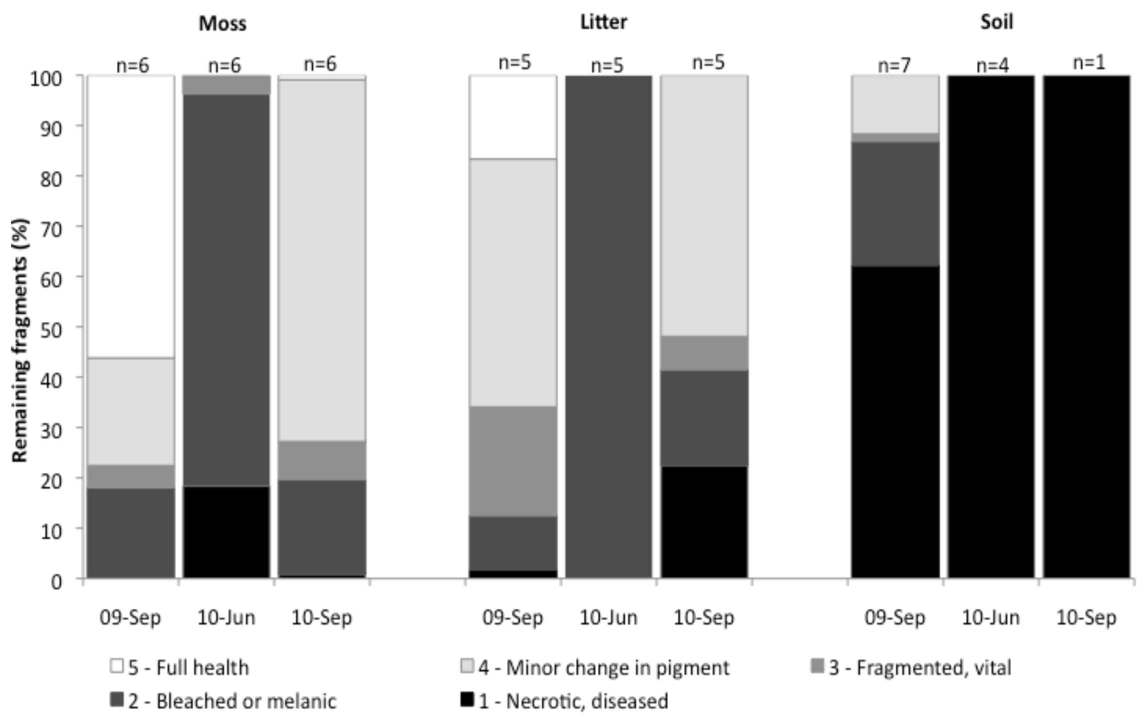


Figure 11 Dyke 5 lichen fragment vigour by treatment and assessment date.

The fragments found outside the plots were usually as vigorous as the fragments remaining within the plot, except where the transport appeared to be by water, as this seemed to result in fragments becoming coated or buried in soil.

2.5.3 Lichen Growth – Photo Assessment

The mean lichen cover values and mean fragment sizes for each set of substrate treatments on each site was not statistically different at the time of installation (Figure 12), but the lichen cover on each replicate varied widely; individual fragment sizes ranged from 0.6 to 1.4 cm^2, and total cover ranged from 13.2 cm^2 to 36.3 cm^2.

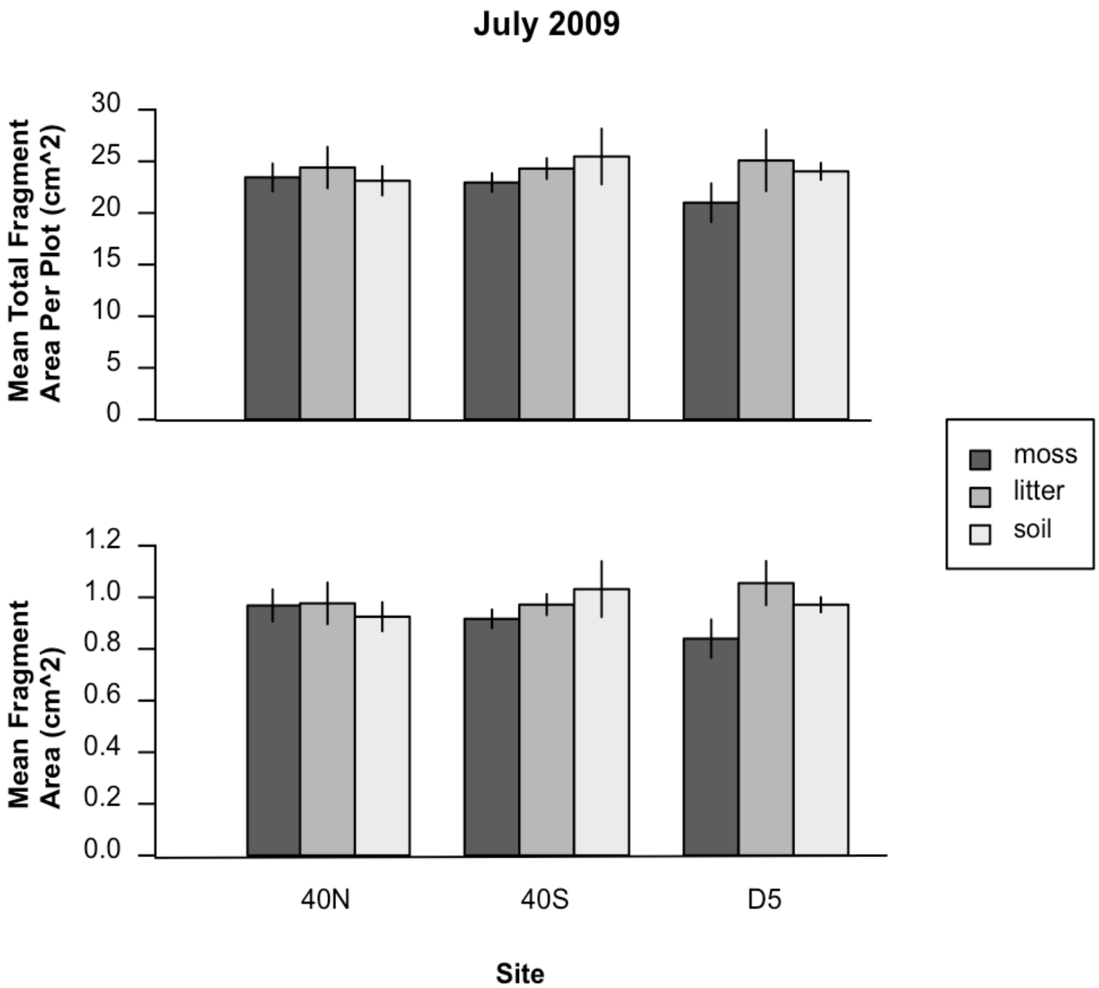


Figure 12 Initial mean total fragment area and mean fragment area by site (bars denote 1 standard error).

Statistical analysis of the change in total lichen fragment area or area per fragment throughout the trial was not completed, since the area analysis captured metrics of the trial plot establishment that were already better captured using either fragment retention or the vigour scale. Changes in total lichen cover in each plot were primarily due to differing rates of fragment retention, and changes in mean area per fragment did not capture the growth of fragments - where average fragment size was found to be increasing, it merely represented the loss of smaller fragments. Change in average size per fragment was instead a measure of the degree of fragmentation experienced by the thalli in each plot, which was also captured in the vigour assessment.

2.5.4 Lichen Growth – Microscopic Assessment

Cladonia lichens adhere to their substrates via a microscopic hyphal network or rhizines that can penetrate up to a few millimeters into their substrate. While reindeer lichens can continue to grow without this connection (in fact, must be able to do so in order to establish in new sites via fragmentation of the thallus), the fragments must eventually adhere to the substrate in order to start forming a mat. The more quickly the fragments can fix themselves to a new substrate, the less likely they will be to be moved to less suitable areas or fragmented further in transport. Field transplant studies which evaluated the vigour and survival of arboreal fruticose lichens, did not assess the transplants for evidence of adherence to the substrate; the transplanted thalli were either tied to the new locations (ie, twigs) or transplanted between sites on their original substrates (i.e. entire branches were moved with the lichens intact) (Hilmo and Såstad 2001; Hilmo 2002; Liden et al. 2004). The Roturier et al. (2001) study did not directly observe new hyphal connections, as these were inferred from the nascent mat formation around the original fragments after two years.

In Webb's (1998) paper, she describes new growth from fragments occurring as new branch formation from internodes, and undifferentiated thallus (primary thallus) growth on organic materials. I removed a set of thallus fragments secured to mesh (for ease of retrieval) at each monitoring visit to assess microscopically for signs of hyphal growth from the newly broken stem ends of the fragments (only growth from the new breaks was included, as hyphal growth from older breaks may have occurred prior to transplant).

By the end of the two-year trial, 21 percent of the total fragments collected from the trials had been found with new hyphae, and 41 percent of the fragments collected in September 2009 had grown hyphae.

Approximately 90 percent of the fragments assessed in each monitoring period were also found to have few to abundant hapters growing from the apical tips or latter two years of new growth, which would also assist in attaching the lichens to their substrate (Photo 1).

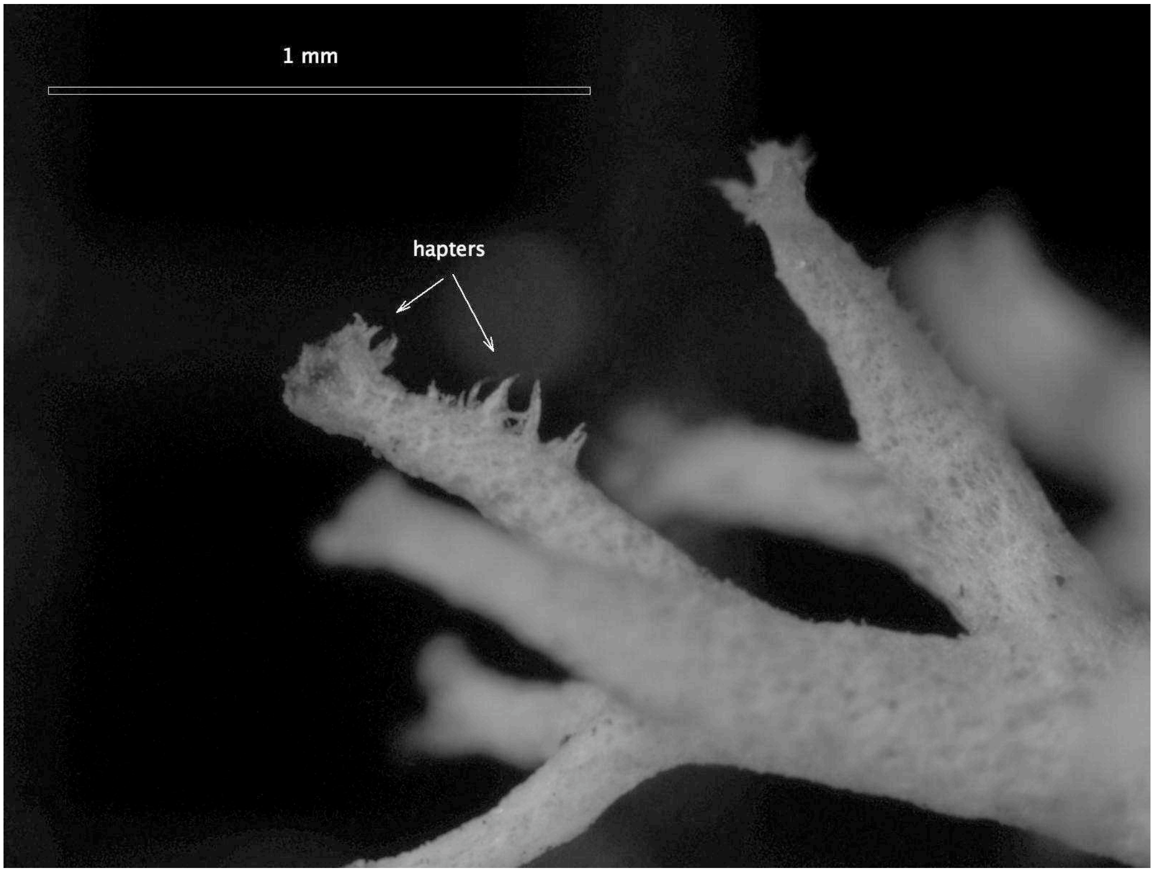


Photo 1 Microscope photo of hapters on transplanted lichen fragment.

Hapters are specialized organs, frequently described in parasitic or climbing higher plants but also seen in lichens, which puncture tissue and act as anchors and occasionally transport nutrients from a host to a parasite. Smith (1921) was the only author in the literature reviewed to mention this feature on the mat-forming lichens, though she claims that these structures are “very frequent” on *C. rangiferina* and *C. sylvatica* [sic (syn. *C. mitis*)], and are “important organs” for these lichens on tundra and heath habitats, where they facilitate the cohesion of the lichen mats, and allow them to “climb” with the surrounding higher vegetation as it grows, detaching from the ground altogether. Attaching to the surrounding shrubs (usually *Calluna* spp.) prevents the lichen from being overgrown, and allows the lichens to raise their photosynthesizing apices toward the light (Smith 1921). The baseline growth assessment samples also had abundant hapter formation, so it is likely that many of the trial fragments had these hapters at installation; however, the presence of these organs suggests that *C. mitis* (or other reindeer lichen species) fragments may be able to rapidly establish attachments to new substrates using hapters instead of needing to develop new hyphae.

Many of the fragments (23 %) collected during September 2009 and September 2010 were also observed to have red or brown apothecia on apical tips, and these were particularly abundant on the most vigorous fragments (Photo 2).

Photo 2 Microscope photo of mature apothecia on transplanted lichen fragment.

The finding of apothecia was unexpected, as McCune and Geiser (1997) stated that apothecia are entirely lacking on *C. mitis*, and other authors have stated apothecia formation is rare to uncommon on this species (Brodo, Duran Sharnoff and Sharnoff 2001; Pope 2005; Vitt, Marsh and Bovey 1988). However, Kotelko, Doering and Piercy-Normore (2008) also found apothecia on approximately 21 percent of the *C. arbuscula* in their study area. Apothecia are reportedly found on lichens growing under favourable site conditions, in particular, where there is abundant incident light and moisture (Smith

1921). The baseline growth assessment samples were found to have apothecia on approximately 25 percent of the fragments.

Apothecia recorded in 2010 may have been formed under trial site conditions, so the presence of apothecia is included in the summary of microscopic growth indicators in Table 8, along with the proportion of lichen fragments in each treatment observed to have new hyphal development, lateral branching (e.g. Photo 3) or recent branch division (e.g Photo 4). Recent branch divisions were only observable at the end of the growing seasons). At the last assessment in September 2010, 31 percent of the total number of lichen fragments collected had lateral branches.

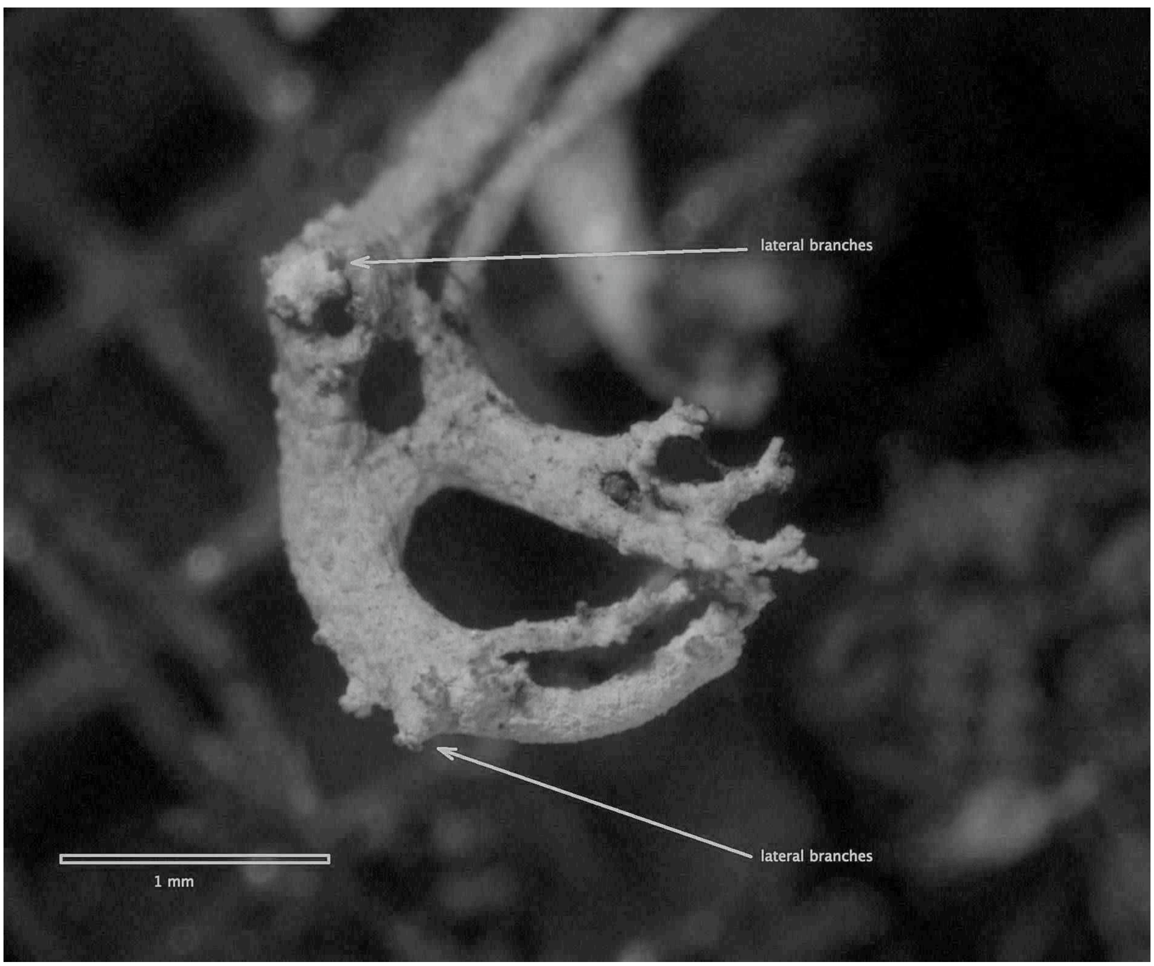


Photo 3 Microscope photo of lateral branching on transplanted lichen fragment.

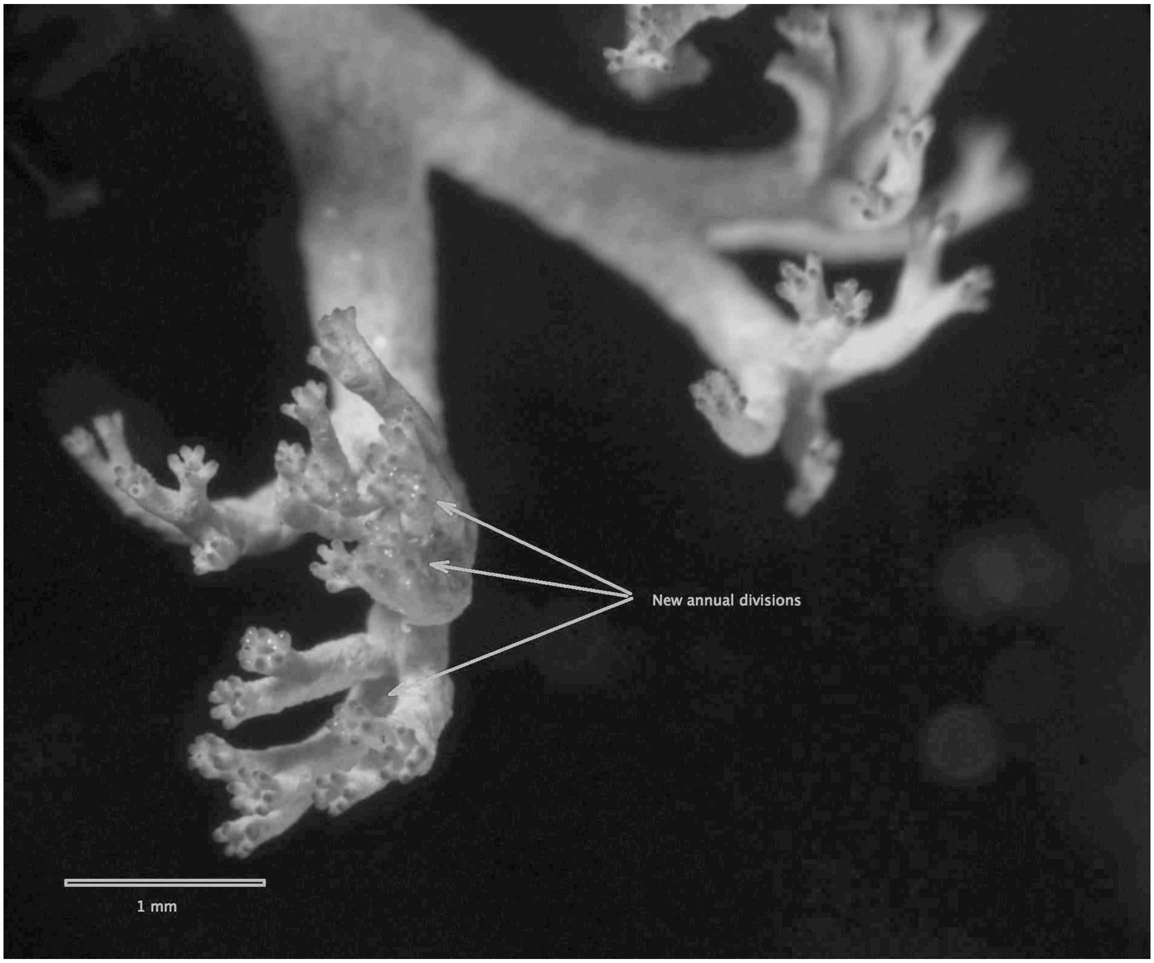


Photo 4 Microscope photo of new branch division on transplanted lichen fragment.

The fragments secured to mesh in the trials had very low vigour compared to the other fragments in the plots, possibly due to tissue damage from being secured, or unfavourable moisture or heat conditions created by the mesh; by September 2010 many of the fragments were necrotic to varying degrees, if not missing entirely. No statistical analyses were performed to compare the microscopic differences between treatments, as microscopic observations were merely meant to determine whether any microscopic changes in the lichens could be observed within the timeframe of the trial.

Table 8 Microscopic growth assessment results

Site	Substrate	Number of fragments retrieved from treatment			% Fragments with Apothecia			% Fragments with Hyphae			% Fragments with New Branch Divisions		% Fragments with Lateral Branching		
		Sept-09	Jun-10	Sept-10	Sept-09	Jun-10	Sept-10	Sept-09	Jun-10	Sept-10	Sept-09	Sept-10	Sept-09	Jun-10	Sept-10
40N	moss	6	6	5	0	17	20	50	17	40	83	100	0	0	40
	litter	6	6	4	50	17	25	67	17	25	83	100	0	0	25
	soil	6	6	4	0	17	25	50	17	25	33	100	0	0	75
40S	moss	6	6	2	33	50	0	67	33	0	100	50	0	0	0
	litter	6	6	3	0	33	33	33	33	33	50	66	0	0	33
	soil	6	5	2	0	33	50	33	0	0	83	0	0	0	50
D5	moss	6	6	6	83	33	0	17	33	17	100	50	0	0	33
	litter	5	5	3	20	20	0	50	17	0	100	33	0	0	0
	soil	7	7	3	14	14	0	0	0	0	57	33	0	0	0

3.0 Cryptogam Diversity Assessment

3.1 Introduction

There are no descriptions of the terrestrial lichen diversity or succession patterns on reclaimed oil sands sites. A small biodiversity assessment of the other terrestrial lichens in the trial sites was undertaken in order to provide some background data on existing lichen diversity, and compare that with the successional communities described by (Ahti 1959; Ahti 1977) for spruce- and pine-lichen forests.

Ahti (1959, 1977) provides a rough succession pattern from bare soil (usually following a burn) for spruce- and pine- lichen forests in the hemi-boreal forests:

1. Bare soil (1-3 years following burn)
2. Crustose lichens stage (3 – 10 years)
3. Cup lichen stage (10 – 30(-50) years) including *Cladonia crispata*, *C. cristatella*, *C. deformis*, *C. pyxidata*, *C. cornuta*, *C. gracilis*, etc.
4. First reindeer lichen stage (30(50-) – 80(-120) years) *C. mitis*, *C. rangiferina*, *C. arbuscula*
5. Second reindeer lichen stage (80(120-) – years) *C. stellaris*. This stage may never be reached in areas with frequent disturbance.

At the time of trial installation, the Site 40S and Site 40N trial sites were both found to have abundant cup lichens present. These lichens are found in habitats with the same characteristics as the reindeer lichens in terms of soil type and forest cover, only with less time since disturbance. Cup lichens arrive prior to the reindeer lichens after a disturbance due to their reproductive characteristics; these species produce abundant soredia, as well as fungal sexual spores, that can be spread long distances by wind and animals. The reindeer lichens do not produce soredia, and less commonly produce fungal sexual spores, colonizing new areas primarily by thallus fragments that have a smaller dispersal range than spores. Once the reindeer lichens arrive on a site, they slowly out-compete the cup lichens due to their longer lifespan and larger size (Ahti 1959).

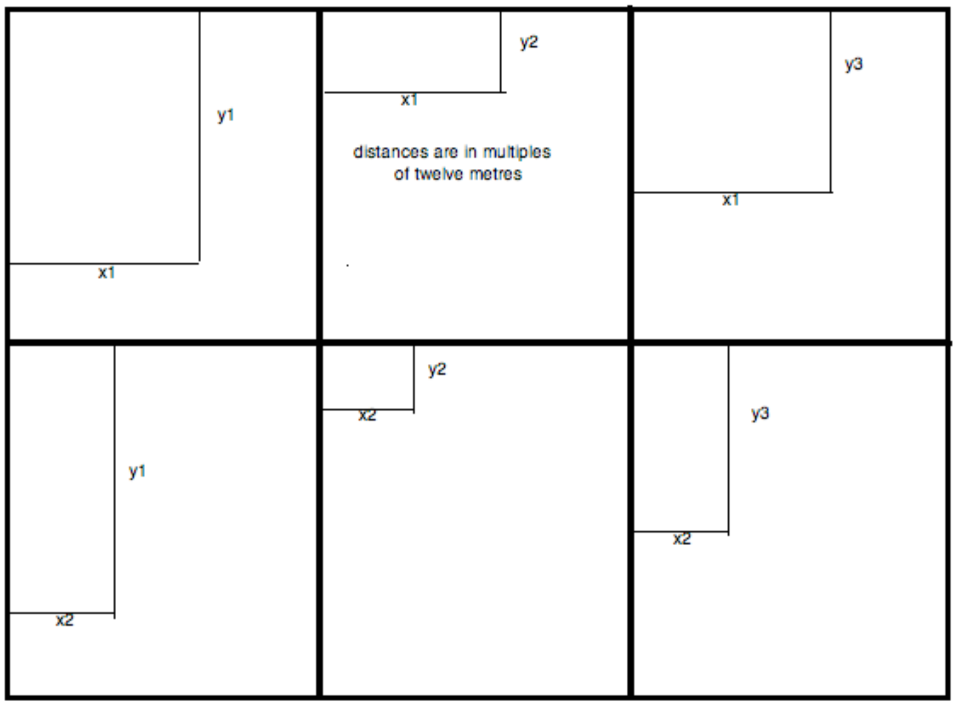
Both sites also had abundant *Hylcomium splendens* (step moss), which is a co-dominant species with the reindeer lichens in the boreal forests. This species tends to dominate in the moister microsites, while the lichens will predominate in the drier areas.

3.2 Methods

Field collection

The type of plot design used to evaluate species diversity of terrestrial lichens was found to be highly variable throughout the literature. Plot sizes ranging from 20 cm x 20 cm to 1 m^2, and were placed at intervals along transects (Botting and Fredeen 2006; Gignac and Dale 2005; Kotelko, Doering and Piercy-Normore 2008; McCune and Lesica 1992; McIntosh 2003) at regular (e.g. Gabriel and Bates 2005; Pharo and Beattie 1997) or random (e.g. Carleton 1990; Coppins and Shimwell 1971) positions within larger vegetation survey plots. McCune and Lesica (1992) found that the number of species found in a given replicate area using small (1 m^2) plots along transects was highly variable, and many plots were required (up to 45 in a 0.081 ha area, or 556 1 m^2 plots/ha) in order to approach the 'true' number of species in the study area (estimated by the total number of species found in all replicates); though a minimum of 25 plots (or 250 m^2 of sampled area per hectare) was sufficient to capture the true number of species in younger forests with fewer species. Based on McCune and Lesica's (1992) work, I installed three 100 m^2 plots (circular plots with radius 5.64 m) per hectare on the reclaimed areas, and five 100 m^2 plots per hectare on the Wood Creek area, for a minimum of 300 m^2 of sampled area per hectare (500 m^2 for Wood Creek, where higher species abundances were expected).

A stratified unaligned systemic sampling design was used to distribute the plots on the reclaimed sites (as described by Grieg-Smith 1983). On a scaled map (1:50,000), the sites were loosely stratified into square or rectangular sections according to vegetation type (deciduous or coniferous), and planting direction (sections of Site 40 were planted in rows perpendicular to the slope, et al. were planted parallel). Each stratified section was then divided into equal smaller sections, three to each hectare. One corner of each small section was designated as the origin (0,0), and plot centres were placed on randomly selected coordinates on an imaginary 12 m grid (to ensure that all areas within the plot had an approximately equal chance of being selected using a 11.3 m diameter plot with plot centres on grid intersections). The layout of this scheme is shown in Figure 13.

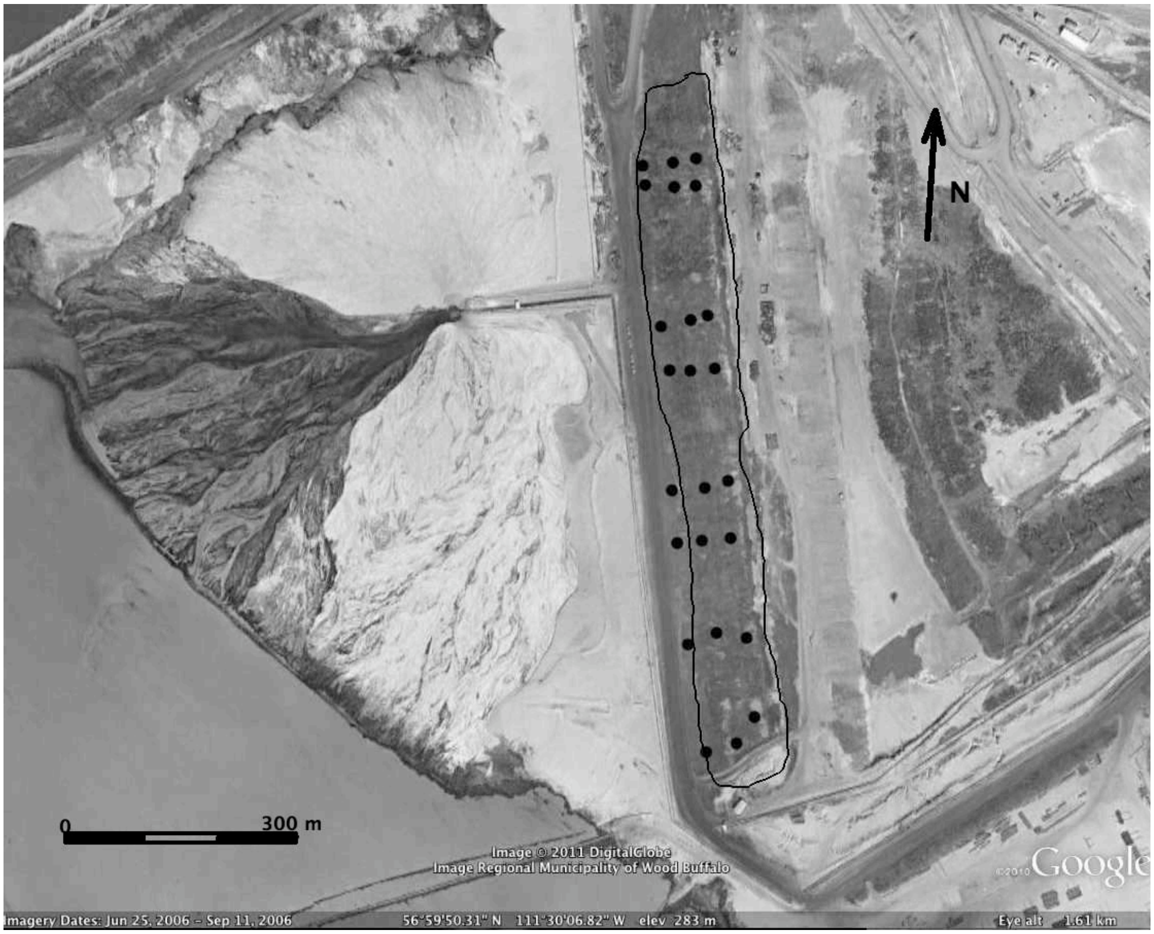


Note: x coordinate distances from the upper left corner of each box (0,0), in each row are the same across columns, and y coordinate distances from the upper left corner of each box (0,0) in each column are the same across rows – as denoted by coordinates with the same numerals.

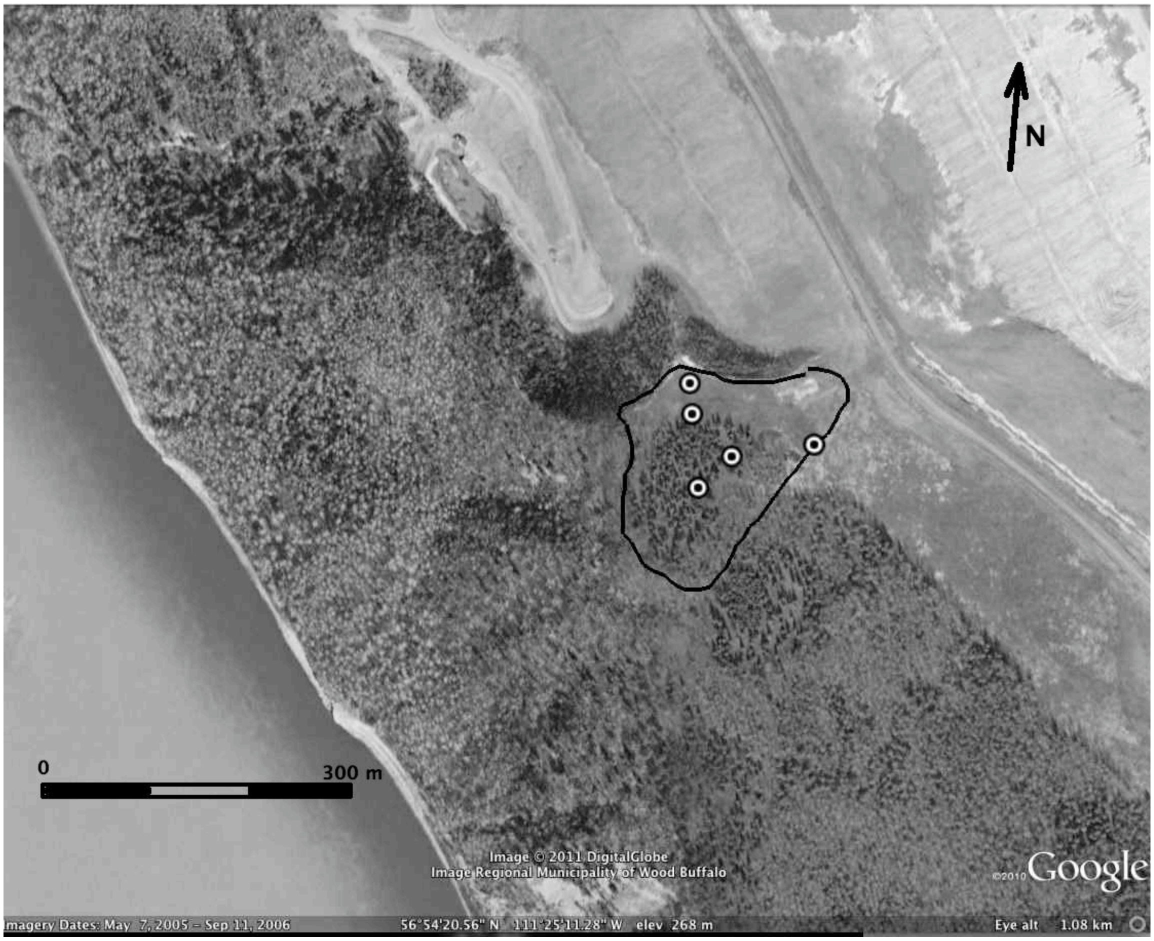
Figure 13 Cryptogam diversity assessment sampling layout.

In the field, plots were navigated to using a Garmin eTrex GPS, so plot locations are accurate within 5 to 8 m. Navigation points were marked in each 0,0 corner, and plots were found by navigating the pre-determined distance from the origin in metres along the x and y axes. A total of 70 plots were installed on three sites (41 on Site 40, 24 on Dyke 5, and 5 on Wood Creek) from June 3 to 9, 2010. Plot locations are show in Map 5 to Map 7.

Map 5 GPS locations of Site 40 cryptogam diversity assessment plots (see Map 2 for site location).



Map 6 GPS locations of Dyke 5 cryptogam diversity assessment plots (see Map 2 for site location).



Map 7 GPS locations of Wood Creek cryptogam diversity assessment plots (see Map 2 for site location).

Two people searched plots for lichen and moss species occurrences, and voucher specimens of each species found were collected. Specimens were placed in numbered paper envelopes to be transported to Victoria for verification, and information on adjacent species, substrate, and vegetation community were also collected. For common, easily recognizable species, such as *H. splendens*, a new specimen was not collected in each plot. A minimum of 20 minutes was spent in each plot, or until no new species were found after 15 minutes of searching, to a maximum of 45 minutes per plot.

Reconnaissance in 2009 indicated that the lichen species present on the reclaimed areas were likely to have very low cover values (<5%) and very patchy distributions; making accurate estimates of species cover difficult with small plots. In addition, identifying lichens to the species level frequently requires microscopic analysis and chemical testing. The most accurate methods of determining terrestrial lichen abundance (particularly for the *Cladonia*s) involve removing sods and determining the species present by weight in the laboratory (as in McIntosh 2003).

Instead, abundance for moss and lichen species found was estimated using a scale adapted from Holt, McCune and Neitlich (2009), who used it to record changes in lichen and bryophyte diversity and abundance in Montana forests over time (Table 9).

Table 9 Cryptogam species abundance scale (adapted from Holt et al. 2009) used to record lichen and moss abundance on the cryptogram diversity plots

Abundance value	Classification	Description
1	Rare	<3 thalli
2	Uncommon	4 – 10 thalli
3	Common	>10 thalli, <1 % cover
4	Abundant	1 – 5% cover
5	Prolific	6 – 25% cover
6	Dominant	> 26% cover

Laboratory Identification

A total of 86 moss and lichen vouchers were collected. Voucher specimens of mosses were identified to the family level, and in some cases the species level, with the aid of a compound microscope, and using Sargent and Lucas (2006), Schofield (1992), and Vitt, Marsh and Bovey (1988) as references. Lichens were identified to the genus level, and to the species level for *Cladonia* lichens with the aid of dissecting and compound microscopes, chemical reagent spot tests (calcium hypochlorite, potassium hydroxide, and paraphenalenediamine (PPD)), and long wave UV fluorescence. References included Brodo, Duran Sharnoff and Sharnoff (2001), Goward, McCune and Meidinger (1994), Goward (1999), and Vitt, Marsh and Bovey (1988).

3.3 Results and Discussion

Twelve relatively distinct vegetation communities were identified for Site 40 based on aspect (north, south, west and southwest) and dominant vegetation (aspen/grass; aspen/ woody; spruce; spruce/aspen/wolf willow; wolf willow; dogwood; spruce/willow), and cryptogammic data is summarized according to each community type (Table 10). Representative plot photos of each vegetation type can be found in Appendix G. Planting density, grass and alfalfa cover, and relative cover of deciduous versus coniferous species varied widely over the Dyke 5 site but there were no identifiably distinct vegetation communities, so cryptogammic data is summarized for the entire site. Cryptogammic data for three vegetation communities sampled on the Wood Creek reference site are also shown for comparison.

No lichen species were discovered on the Dyke 5 site, and only small acrocarpus mosses were found (Photo 5). Lichens, primarily *Cladonia* (non-reindeer lichen species) and *Peltigera* spp., were present in all reclaimed vegetation types on Site 40, and *Stereocaulon tomentosum* was found in spruce-dominated vegetation communities. *Cladonia mitis* was rare to uncommon in many spruce and aspen-dominated plots throughout Site 40; though this species was abundant at approximately 5 percent cover on the open plots at Wood Creek[13], and covered up to 25 percent of the ground in the forested plots. *Peltigera* spp. were common to abundant (covering <1% and 1 to 5% of plots, respectively) on Site 40's north aspect, and rare to uncommon (<10 occurrences per plot) on other aspects and at Wood Creek. A granular crust lichen associated with calcareous soils, *Lepraria neglecta*, was occasionally abundant in plots on the south aspect of Site 40 (but uncommon overall).

[13] Though *C. mitis* is a dominant component of the cryptogammic cover, coarse woody debris and Labrador tea cover most of ground in this area.

Photo 5 Acrocarpus mosses on Dyke 5.

Hylocomium splendens, the most common moss in the boreal forest, is also a common to dominant component of the cryptogammic cover on Site 40, accounting for up to 50 percent of the ground cover in the spruce community on the north aspect of Site 40, though it was absent in the aspen/woody understory and wolf willow plots. *H. splendens* was less common on the other aspects, accounting for less than 1 percent of the ground cover in tree-dominated plots, and absent from plots where deciduous shrub or grass cover were dominant.

The Polytricheae (hair cap) mosses, which are noted early colonizers on disturbed peat mining sites (Campeau and Blanchard 2010; Groeneveld and Rochefort 2002), were absent from all but one reclaimed site plot (on Site 40 West in the spruce/aspen/wolf willow community); but were common on forested sites at Wood Creek (<1 percent cover), and abundant (1 to 5 percent cover) on the open sites. *Pleurozium schreberi*, a common co-dominant moss with *H. splendens* in the boreal forest, was also absent from reclaimed sites, but was common on the open plots at Wood Creek (1-5 percent cover), and dominated the forested plots at Wood Creek, covering greater than 50 percent of the area. *Cladonia stygia* and *Cladonia stellaris*, which were common to prolific on the Wood Creek site on both forested and open areas, were also absent from all reclaimed areas.

Table 10 Moss and lichen abundance by vegetation community on the reclaimed and source sites

		Dyke 5	Site 40 North				Site 40 South			
		overall	At/grass	At/ woody	Sw	Eleagnus	At/ woody	At/grass	Cornus	Sw/At/ Eleagnus
		N = 24	N = 1	N = 1	N = 4	N = 1	N = 5	N = 5	N = 3	N = 9
		% cover	% cover	% cover	% cover	% cover	% cover	% cover	% cover	% cover
Feathermosses (stairstep)	*Hylocomium splendens*		26-50		26-50		<1	<1		<1
	Pleurozium schreberi									
	Rhytidiadelphus triquetrus		<1	<1						
Feathermosses (other)	Brachytheciaceae							<1	1-5	<1
	Drepanocladus aduncus		<1		1-5	<1		<1		<1
	Hypnaceae		<1	<1	<1	<1	<1	<1	<1	<1
	Thuidium recognitum				1-5				<1	<1
Haircap Mosses	Polytrichaceae									
Acrocarpus Mosses	Aulacomniaceae, Bryaceae, Dicranaceae, Ditrichaceae, Pottiaceae	6-25	<1	6-25	1-5	6-25	<1	1-5	1-5	1-5
Reindeer lichens (Cladonia spp.)	*Cladonia mitis*				uncommon		rare	rare		rare
	Cladonia stellaris									
	Cladonia stygia									
Other Lichens	*Cladonia* spp.		uncommon	uncommon	1-5	uncommon	uncommon	uncommon	uncommon	<1
	Lepraria neglecta						<1	<1	<1	<1
	Peltigera spp		<1	1-5	1-5	uncommon	rare		uncommon	uncommon
	Stereocaulon tomentosum				rare					rare

Table 10 Moss and lichen abundance by vegetation community on the reclaimed and source sites (cont'd)

		Site 40 Southwest		Site 40 West		Wood Creek		
		Sw/ Salix	Sw/ Cornus	At/ woody	Sw/At/ Eleagnus	At	open	Sw
		N = 1	N = 1	N = 1	N = 7	N = 1	N = 2	N = 2
		% cover	% cover	% cover	% cover	% cover	% cover	% cover
Feathermosses (stairstep)	*Hylocomium splendens*		6-25		<1		<1	1-5
	Pleurozium schreberi					<1	<1	>50
	Rhytidiadelphus triquetrus							
Feathermosses (other)	Brachytheciaceae		26-50	<1	1-5			
	Drepanocladus aduncus		1-5	<1	<1			
	Hypnaceae	>50	<1	<1	<1	26-50	1-5	
	Thuidium recognitum				<1			
Haircap mosses	Polytrichaceae				<1	<1	1-5	<1
Acrocarpus mosses	Aulacomniaceae, Bryaceae, Dicranaceae, Ditrichaceae, Pottiaceae	6-25	1-5	<1	1-5	<1	1-5	1-5
Reindeer lichens (Cladonia spp.)	*Cladonia mitis*				uncommon		1-5	6-25
	Cladonia stellaris						1-5	6-25
	Cladonia stygia						<1	1-5
Other Lichens	*Cladonia* spp.	rare	rare	rare	1-5		1-5	1-5
Other Lichens	*Lepraria neglecta*		1-5					
	Peltigera spp		rare	uncommon	uncommon	rare	rare	uncommon
	Stereocaulon tomentosum				rare			

The cryptogammic community summary for the community types with lichen trials are shown in Table 11, with the Wood Creek source site (open, disturbed site, and forested site) for comparison.

Table 11 Moss and lichen abundance summary for lichen trial sites

		Dyke 5	**Site 40**		**Wood Creek**	
			South	**North**		
			Sw/At/ Eleagnus	**Sw**	**open**	**Sw**
		% cover	**% cover**	**% cover**	**% cover**	**% cover**
Feathermosses (stairstep)	*Hylocomium splendens*		<1	26-50	<1	1-5
	Pleurozium schreberi				<1	>50
	Rhytidiadelphus triquetrus					
Feathermosses (other)	Brachytheciaceae		<1			
	Drepanocladus aduncus		<1	1-5		
	Hypnaceae		<1	<1	1-5	
	Thuidium recognitum		<1	1-5		
Haircap mosses	Polytrichaceae				1-5	<1
Acrocarpus mosses	Aulacomniaceae, Bryaceae, Dicranaceae, Ditrichaceae, Pottiaceae	6-25	1-5	1-5	1-5	1-5
Reindeer lichens (Cladonia spp.)	*Cladonia mitis*		rare	uncommon	1-5	6-25
	Cladonia stellaris				1-5	6-25
	Cladonia stygia				<1	1-5
Other Lichens	*Cladonia* spp.		<1	1-5	1-5	1-5
	Lepraria neglecta		uncommon			
	Peltigera spp		uncommon	1-5	rare	uncommon
	Stereocaulon tomentosum		rare	rare		

The list of *Cladonia* species found on the reclaimed site (Site 40) versus Wood Creek is given in Table12 below[14].

[14] Identification of species for the genus *Cladonia* provided in Table 10 has not been confirmed by an expert; the identifications are provisionary, pending confirmation.

Table 12 Cladonia species on Site 40 and Wood Creek

Species	Site 40	Wood Creek	
	combined	open	forested
Cladonia acuminata		X	
Cladonia botrydes	X	X	X
Cladonia cariosa	X		
Cladonia chlorophaea group	X		
Cladonia ciliata		X	
Cladonia coccifera		X	
Cladonia cornuta	X	X	X
Cladonia cristatella	X	X	X
Cladonia deformis			X
Cladonia ecmocyna s. *intermedia*	X	X	
Cladonia gracilis s. *turbinata*	X		
Cladonia macilenta	X	X	X
Cladonia multiformis	X	X	X
Cladonia pleurota		X	X
Cladonia pyxidata	X		
Cladonia singularis	X		
Cladonia squamosa	X		
Cladonia sulphurina		X	X
Cladonia uncialis		X	X

4.0 Summary and Conclusions

The primary objective of this research was to determine whether artificial introduction of reindeer lichen fragments could be a feasible part of the revegetation plan for reclaimed sites where these lichens form a significant part of the vegetation. There were two components to this research: a) a lichen transplant trial, where I investigated which commonly available substrates found in reclaimed forest sites would promote the best lichen fragment survival and vigour for a lichen 'seeding' program; and b) a diversity assessment of the reclaimed site to compare the existing cryptogam community with the expected community for the target ecosite.

4.1 Lichen Transplant Success

A caveat for the interpretation of this transplant trial is the low statistical power for the analysis. The tests used for this statistical analysis were conservative, as the data had large within-treatment variances and the number of sites included in the trial was small due to a low number of sites identified as eligible for the trial (i.e. with low forage cover, planted to jack pine or spruce, permanently reclaimed, and without other research trials present). The study was designed based on a similar trial on a domestic reindeer range in Sweden (Roturier et al. 2007) where the natural variation within the sites, and thus the variance within treatments, was much lower. Increasing the replication may have helped to increase the statistical power of the design, however, due to logistical constraints there was not enough time to install more replicates. Thus, while the conclusions based on these statistical analyses are preliminary, this was a pilot project aimed at opening avenues of investigation for further reindeer lichen research by reclamation practitioners, and they provide fodder for the creation of new hypotheses.

The effect of substrate on lichen transplant survival varied by site. On Site 40 North, there was no significant difference in the number of lichen fragments retained in a plot between the substrate types, and overall fragment retention on the trial plots was low; the median fragment retention was less than 50 percent for all treatments except moss on Site 40N (52 %) and Dyke 5 (78 %) after two growing seasons. However, the fragments that remained were relatively healthy and unfragmented; the average vigour of the lichens on the moss plots was statistically higher than those on the soil plots on Site 40 North and Dyke 5. The median retention of the lichen fragments on Dyke 5 was highest on the moss plots, at 78 percent, which was statistically higher than the retention on the litter and soil plots. The vigour of the transplants on the Dyke 5 site was not statistically higher on the moss plots than on the litter plots due to the high fragmentation rate of the lichen transplants on the site; however, the fragments, were not discoloured or diseased.

The difference between the substrate types was likely not as pronounced on the North aspect of Site 40 as on the South aspect due to the greater precipitation and runoff interception by the surrounding feathermoss mat. On Site 40 South and Dyke 5, the low survival of the lichens on the soil plots was possibly due to being washed away in the rains, as evidenced by the runnels in the plots; in plots where the lichens were not washed away, they were coated in sand and necrotic or buried in place. Removing moss or graminoid vegetation cover to reduce competition is therefore not a recommended site preparation for lichen fragment transplants without additional treatment to reduce overland water flow on the lichen transplant site. The greater retention on the Dyke 5 moss treatment compared to the moss treatments on Site 40 may be anomalous - there was no replication of the site types – though it may also be due to a difference is a site factor such as aspect or the moss species present. The mosses on the Dyke 5 site were acrocarpous species[15]; they provided stabilization of the soil surface to reduce soil movement and scouring, as well as an anchoring substrate for the lichens, but they were small and low growing, and did not compete with the lichens for light or overtop them like the feathermosses on Site 40 North.

The finding that substrate differences had significant effects on lichen retention on the youngest and most open site, Dyke 5, but not on the forested sites, is consistent with Roturier et al.'s (2007) findings that substrate differences made no difference to lichen

[15] Having the spore-bearing body at the end of a main stem or from the apical cell of a well-developed branch; as opposed to pleurocarpous species, which produce spore-bearing bodies from lateral branch or specialized leaf bud (Vitt, Marsh, Bovey 1988). Acrocarpus mosses and typically smaller and more compact than pleurocarpus mosses, which include species that have multiple-branching and stair-step growth forms.

retention in second-growth forests, but that moss and twig substrates improved lichen retention on clearcuts.

It was expected that the litter treatment would provide the stabilizing benefits of the moss substrates without the possibility of growing over and competing with the lichens, however, the litter treatment did not provide a good substrate. On Site 40 South, the litter treatments were washed away as easily as the soil substrate (while also adding material to bury the lichens with), while the lichens were partially covered with the continuing litter fall from the conifers on both the soil and litter plots on Site 40N (the moss appeared to overgrow the litterfall on the moss treatment). On Dyke 5, the continuing litterfall did partially bury the remaining lichens on the moss and litter plots, but the effect was most pronounced on the litter plots compared to the moss plots, since the lichens seemed to get worked down into the thickening litter layers.

Recording the change in area of the lichen fragments with ImageJ was not useful for the assessment of these lichen fragment trials, as the amount of fragmentation and burial of the fragments made the change in visible size of the fragments a poor indication of actual condition. In addition, many of the lichen fragments darkened as the trial progressed, decreasing the contrast between the lichens and the substrate and making distinguishing the lichens from the substrate increasingly difficult for the software. There was no statistically significant difference in the mean size of the lichen fragments on the different treatments by the end of the two year trial, though the mean measured size of the lichen fragments did decrease significantly since installation for the moss plots on Site 40N, and on the litter plots on Site 40S and Dyke 5. The decrease in size was due in part to the fragmentation of the lichens, as well as the partial burial of the lichens by the moss and litter resulting in the lower surface area captured by the camera. The use of area measurement by ImageJ would likely be a more suitable indicator of lichen establishment on a trial using larger lichen mats.

One of the most interesting findings of this trial was the discovery of lateral branching after two growing seasons on the microscopic assessment fragments. Hyphal growth from fragments of *Cladonia arbuscula* thalli has been observed in the laboratory after 7 to 14 weeks (Jahns 1993), and the growth rate of the reindeer lichens has been fairly well studied using annual branching (e.g. Ahti 1957; Ahti 1959; Helle, Aspi and Tarvainen 1983; Pegau 1968; Scotter 1963), but only one author had previously noted lateral branching in lichen fragments as a regrowth mechanism following lichen fragmentation due to logging (Webb 1998). In her study, Webb noted lateral branching on all logged or previously burned sites three years following disturbance, but not on sites two years since disturbance. This trial provides evidence for lateral branch formation as early as two years following fragmentation and transplantation. Direct evidence of continuing lichen growth as evidenced by new divisions in the tips of transplanted lichens was also observed.

The presence of apothecia on the lichen fragments was also unexpected, as McCune and Geiser (1997) stated that apothecia are entirely lacking on *C. mitis*, and other authors have stated apothecia formation is rare to uncommon on this species (Brodo, Duran Sharnoff and Sharnoff 2001; Pope 2005; Vitt, Marsh and Bovey 1988). However, Kotelko, Doering, and Piercy-Normore (2008) also found apothecia on approximately 21 percent of the *C. arbuscula* in their study area.

4.2 Cryptogam Diversity on Reclaimed Sites

There are no published studies of cryptogammic diversity on reclaimed oil sands sites, though forest establishment and soil development on these sites is being studied (Canadian Oil Sands Network for Research and Development, Environmental and Reclamation Research Group (CONRAD ERRG) 2009; Rowland et al. 2009). A small diversity assessment of the terrestrial lichens in the trial sites was undertaken to provide some background data on existing lichen diversity, and compare the species with what might be expected from the descriptions of seral communities described (Ahti 1959; Ahti 1977) for spruce- and pine-lichen forests.

The cup lichens were common to abundant on Site 40, which is consistent with Ahti's seral sequence, where the cup lichens dominate the terrestrial lichen community within 10 years of disturbance, and for up to 30 to 50 years. The early reindeer lichen stage of pine- and spruce-lichen forest types typically occurs 30 years after fire disturbance, and seldom later than 50 years, when *C. mitis* gains dominance over the cup lichens (*Cladonia* spp.) (Ahti 1959; Ahti 1977). The Site 40 reclaimed site is over 25 years old, and while *C. mitis*, is present, it is rare to uncommon; if this species is to become a dominant component of the reclaimed vegetation, it may take longer than in the typical fire recovery sequence. The later colonizers *Cladonia stellaris*, *Cladonia rangiferina* and *Cladonia stygia* that, together with *C. mitis*, are indicative of the al and c1 ecosites of the Central Mixedwood Boreal forest, were not present on the reclaimed sites.

Pioneer species of mosses such as *Ceratodon purpureus*, *Tortula ruralis* and *Pohlia nutans*, which are ubiquitous on disturbed areas in North America (Vitt, Marsh and Bovey 1988), were expected on the reclaimed sites. Other pioneering species of the boreal forest such as the hair cap mosses (*Polytrichum spp*.), and common later seral components such as the many species of feathermosses (e.g. *Hylocomium splendens*, *Pleurozium scheberi*, *Ptilium crista-castrensis*) could also be expected on an older forested site like Site 40.

Pleurozium schreberi (Schreber's moss or big red-stem feathermoss) is an indicator moss of the a1 and c1 ecosites (Beckingham and Archibald 1996), and was only present on the Wood Creek site. *Polytrichum piliferum* (awned hair cap moss), which is indicative of the a1 ecosite (ibid.), and is an early colonizer in disturbed lowland habitats (Campeau and Blanchard 2010), was also only present on the Wood Creek site.

Hylocomium splendens, or stair-step moss, indicative of the c1 ecosite, was found throughout Site 40, and was prolific in the spruce-dominated communities within that site.

In pine- and spruce-lichen forests, feathermosses such as *H. splendens* and the reindeer lichens frequently co-dominate on the forest floor, with mosses appearing to dominate in the moister microsites, and lichens in the drier canopy gaps (Kershaw 1977; Lechowicz and Adams 1974; Sulyma and Coxson 2001; Williston and Cichowski 2006). In the spruce-dominated community on the reclaimed Site 40 North aspect, *H. splendens* grew rapidly and overtopped the transplanted lichens in the trial area; however, during the diversity assessment areas with larger canopy gaps were found where cup lichens formed continuous mats. Reindeer lichens transplanted into these gaps may survive and replace the cup lichens.

On the South aspect of Site 40, the range in canopy covers was wider, and areas with low canopy cover were more frequent in the conifer-dominated sections. *H. splendens* was not as abundant as on the north aspect, but the lichens were also not as abundant; the ground cover was typically grass or bare ground. Thus, while feathermosses and lichens typically compete for microsites within suitable forests, the overall site conditions that are conducive to one group establishing on reclaimed sites appear to be conducive to the establishment of the other group as well.

4.3 Future Research

At the last sampling visit in September 2010, Francis Salifu, the Suncor Reclamation Research and Development Coordinator, pointed out an east-facing reclaimed area on the Tar Island overburden dump near the plant site, above the Athabasca River, reclaimed approximately 30 years ago (compared to 25 years ago for Site 40). The tree spacing and grass cover was similar to that of Site 40's southern aspect; but, on this site, reindeer lichens and *Polytrichum* moss species were a dominant portion of the understory (Photo 6).

Photo 6 Lichen-rich reclaimed site on Tar Island overburden dump

Unfortunately, the vegetation composition of this site was not known prior to the diversity assessment or trial installation, otherwise it would have been an important site to include in the study. This lichen-rich site is approximately half a kilometer from the nearest undisturbed spruce forest and possible propagule source across the river, while the southern aspect of Site 40 is nearly three times that distance from the nearest lichen source site in a patch of intact forest near the Suncor Main Security Gate.

The existence of this site raises many questions for future study: are there other sites on the Suncor property that have developed a reindeer lichen cover? What site conditions appear to control the development of a reindeer lichen mat on a reclaimed site – initial grass cover? Proximity to source sites? Aspect? Organic amendment source and type?

It would also be informative to measure the annual growth increments of the lichen on this site. First, to determine how old the lichens are, and thus get an idea of when they began establishing on the site, and second, to determine how their growth rate compares to that measured for the Wood Creek site.

A further avenue for future study would be to use the moss and lichen fragment transplant method used by (Campeau and Blanchard 2010), who spread harvested peatland surface materials, moss and lichens together, over a soil surface and then covered the plots in a straw mulch. I had decided against using a mulch or other covering for my lichen transplants due to a concern with keeping the lichens too moist and therefore destroying the symbiosis (Jahns 1973; Pyatt 1973), and because burial by litter in conifer forests has been observed to cause lichen mat mortality that takes years to recover (Kauppi 1990 in Williston and Cichowski 2006). Campeau and Blanchard's (2010) results, however, were that both mosses and lichens thrived under the light straw mulch cover, even though the lichens were not a target species for the restoration on those sites.

4.4 Conclusions and Recommendations

Lichen species are not routinely included in reclamation on the Athabasca oil sands mines, though the reindeer lichens are included in the planting lists for the a1 ecosite, and are components of other target reclamation ecosites such as c1, i, g, and j (Beckingham and Archibald 1996). The inclusion of reindeer lichens (and other terrestrial lichens) would be beneficial from two perspectives: wildlife habitat reclamation, particularly in the northern Athabasca region where caribou are still present; and to fulfill commitments to creating biodiverse reclaimed sites with similar community structure and composition to the surrounding natural ecosystems.

Lichens are a difficult species to transplant for land reclamation purposes, since they grow very slowly and are very small relative to the more commonly used tree, shrub, and graminoid species. The lichen transplant trials reviewed for this thesis usually followed the growth of lichen transplants for one to three years (with the exception of Gilbert (2002), who followed his transplants for ten years), so there was very little in the way of literature to support my hypothesis that lichens could be successfully introduced for reclamation. In order to demonstrate that reindeer lichens were a viable reclamation species, they must be shown to survive and continue to grow on the reclaimed site.

The transplant trial conducted for this study did demonstrate that transplanted lichen retention is possible for two growing seasons on reclaimed sites, but it is not certain what the long-term survival trend of the lichen transplants would be. Feathermosses on Site 40 were shown to overgrow lichen transplants within two years, but the acrocarpus mosses present on Dyke 5 resulted in higher retention of lichen fragments without the competition.

The soil pH and nutrient regime on the reclaimed sites was not similar to the lichen source site or other documented soils of reindeer lichen-containing sites in the surrounding boreal forest. Still, the reclaimed sites supported a number of the terrestrial mosses and lichens indicative of the boreal forest ecotypes, and the lichen community composition showed similarities to the seral stage communities that appear after a forest fire; however, they also supported moss species that are not typically associated with local ecosites, such as *Rhytidiadelphus triquetrus*, which may indicate that reclaimed community on these sites will support novel species assemblages (Hobbs, Higgs and Harris 2009).

The total cover and diversity of the lichens and mosses present was not compared statistically across the sites or with a range of natural sites in different seral stages. Future research could focus on better quantifying the composition of the cryptogammic community relative to natural stands at different stages of succession as an indicator of the successional trajectory of the reclaimed sites.

If I were to repeat this study, I would seek to reduce the limitations on the statistical interpretations inherent to this small trial. Since the effect of the treatment on retention and vigour varied between the sites, there are likely to be interaction effects between the substrate and other factors on the site; in addition, the presence of the lichen cover on the Tar Island overburden dump suggests that there may be some combination of site factors, such as distance to source locations, reclamation treatment (i.e. soil placement), or edaphic factors, that are suitable for reindeer lichen establishment on reclaimed sites on the Suncor mine, and by extension, other oil sands surface mines. I would focus on elucidating which of those factors were associated with lichen transplant success through a multiple regression or MANOVA, after increasing the number of sites included in the trial to encompass a wider range of planting ages, percent vegetation cover (not just tree canopy cover), soil replacement treatments, slope and aspect. Some of these factors could be incorporated into trial design through planned blocking within large reclamation areas, as well as through increasing the number of reclamation areas included in the trial.

This trial also only focused on one method of lichen transplant, small widely scattered fragments. For a repeat trial, I would compare the use of transplanting clumps of lichen versus larger transplant plots with a higher density of lichen fragments. Both alternative transplant methods could reduce the potential for transplanted fragments to be washed away by water moving down slopes or blown away, thereby reducing the importance of creating or finding suitable substrates for lichen establishment. Instead of using litter as a substrate treatment on field trials, I would use humus, which I had cleared away from the soil and litter treatments in this trial, and bare soil. I believe the effect of moss substrates on the establishment of reindeer lichen would need to be investigated separately, perhaps as a greenhouse trial, to evaluate the effect of the different moss growth forms (e.g. stairstep moss, other feathermosses, low cushion acrocarps, large acrocarps like the Polytricheae, etc.) on the ability of the lichen thalli to compete with the moss for space and/or secure themselves to the moss.

Recommendations:

1. Continue monitoring of lichen transplant sites to determine long-term transplant survival and growth trends. It would also be useful to expand the trial to include additional trial sites selected to allow analysis of the influence of site factors (aspect, canopy closure, ground cover type, etc.) on transplant success.

2. Explore the effect of different moss substrate species on lichen fragment establishment success. The variable growth forms of different mosses – forming stepped fronds or compact mats - may help or hinder lichen establishment. Choosing lichen transplant locations based on moss species present, or

influencing the moss community (e.g. by choosing moss species to be introduced using a technique like that recommended in point 3), could increase lichen transplant success.

3. Explore lichen and bryophyte transplant methods based on peat moss restoration research. Fragment introduction techniques designed to include a variety of species from the boreal forest, not just lichens, could be used to increase the species diversity on reclaimed sites. Spreading lichen and bryophyte fragments harvested from natural areas, as opposed to whole forest floor transplants, could exploit many species' natural reproduction methods to increase the amount of area reclaimed per unit of source area.

4. Perform a reconnaissance to select study sites from reclaimed areas of several ages and planting treatments across the Suncor mine property (or other ERRG member properties) to a) determine whether reindeer lichens have established on other reclaimed sites in addition to the Tar Island overburden dump, and b) assess the cryptogam diversity on other reclaimed sites.

5. Conduct a study to compare the composition of the cryptogam community on reclaimed sites with natural communities of the reclamation target ecosites at different stages of succession. The composition of the reclaimed site cryptogam communities may be an indicator of the potential successional trajectory of the reclaimed sites, whether to communities similar to those in the surrounding boreal forest, or novel species assemblages.

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Appendix A Ecosite Descriptions

Ecosite label	Ecosite Name	Edaphic Conditions		Tree Species	Characteristic Understory Components	Minor Understory Components
		Moisture	Nutrients			
a1	Lichen- Jack Pine	Xeric - submesic	v. poor – medium	Jack Pine	Blueberry Bearberry Bog cranberry Lily-of-the-valley Schreber's moss Awned hair cap Reindeer lichens	
c1	Labrador tea mesic Jack Pine- Black Spruce	Subxeric-subhygric	poor	Jack Pine Black Spruce	Labrador tea Bog cranberry Blueberry Green alder Twinflower Schreber's moss	Bunchberry Stair-step moss Knight's plume moss Reindeer lichen
g1	Labrador tea subhygric Jack Pine- Black Spruce	Subhygric - subhydric	poor	Black Spruce Jack Pine	Labrador tea Schreber's moss Stair-step moss	Bog cranberry Blueberry Prickly rose Knight's plume moss Peat moss Tufted moss Bunchberry Reindeer lichen

Ecosite label	Ecosite Name	Edaphic Conditions		Tree Species	Characteristic Understory Components	Minor Understory Components
		Moisture	Nutrients			
i(1,2)	Bog (Treed and Shrubby phases)	hygric - hydric	V. poor - poor	(Black spruce)	Labrador tea Bog cranberry Small bog cranberry Cloudberry Three-leaved Solomon's seal Peat moss Schreber's moss Stair-step moss Knight's plume moss Slender hair cap moss Reindeer lichen	
j(1,2)	Poor Fen (Treed and Shrubby phases)	Subhydric - hygric	v. poor-medium	(Tamarack) (Black Spruce)	Labrador tea Bog Birch Bog cranberry Small bog cranberry Cloudberry Three-leaved Solomon's seal Peat moss Golden moss Schreber's moss Tufted moss Slender hair cap moss Reindeer lichen	

Adapted from: Beckingham JD and Archibald JH. 1996. Field guide to ecosites of northern Alberta. Natural Resources Canada, Canadian Forest Service, Northwest Region, Northern Forestry Centre. Special Report No. 5. Edmonton, Alberta.

Appendix B
Soil Laboratory Analysis Results

	pH	SAR	EC	Saturation
	pH	SAR	dS m-1	%
Wood Creek	4.22	<0.1	<0.1	46
Site 40N	7.55	0.19	0.29	77
Dyke 5	7.77	0.22	0.16	44
Site 40S	7.72	0.17	0.27	36
DL	0.1	0.4	0.1	1

	Tot. Org. N	Total Kjeldahl N	NH3
	%	%	%
Wood Creek	0.066	0.067	0.001
Site 40N	0.198	0.199	0.001
Dyke 5	0.121	0.122	0.001
Site 40S	0.179	0.179	0
DL	0.02	0.02	

	Available Nutrients				
	NH4	NO3	PO4	K	SO4
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	2.96	<2.0	2.3	52.8	3.7
Site 40N	2.3	<2.0	5.6	85.6	68.2
Dyke 5	1.33	4.3	31.3	57.9	5.6
Site 40S	1.85	<2.0	1.7	96	43.3
DL	0.8	2	1	2	2

	Exchangeable Cations			
	Ca	K	Mg	Na
	mg/L	mg/L	mg/L	mg/L
Wood Creek	5.5	5.2	2.6	<4.0
Site 40N	97.1	11.6	38.1	8.9
Dyke 5	47	8.7	10	6.5
Site 40S	89.4	12.9	30.7	7.2
DL	1	2	1	4

	ICP-OES Elements				
	Ag	**Ba**	**Be**	**Cd**	**Co**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	<1.0	15.9	<1.0	<0.50	<1.0
Site 40N	<1.0	35.9	<1.0	<0.50	4.6
Dyke 5	<1.0	27	<1.0	<0.50	3
Site 40S	<1.0	34.8	<1.0	<0.50	4.2
DL	1	5	1	0.5	1

	ICP-OES Elements				
	Cr	**Cu**	**Mo**	**Ni**	**Pb**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	1.44	10.1	<1.0	<2.0	<5.0
Site 40N	8.3	14.3	<1.0	11.9	5
Dyke 5	4.85	4.3	1.1	10	<5.0
Site 40S	7.35	9.5	<1.0	11.1	<5.0
DL	0.5	2	1	2	5

	ICP-OES Elements				
	Sn	**Sr**	**Tl**	**V**	**Zn**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	5.6	2.7	<1.0	5.1	<10
Site 40N	<5.0	34	<1.0	16.1	29
Dyke 5	<5.0	17.5	<1.0	21.4	14
Site 40S	<5.0	31.2	<1.0	14.8	28
DL	5	1	1	1	10

	Sand 2mm - 0.05mm	**% Silt 0.05mm - 2um**	**% Clay <2um**	**SK-Texture**
	%	%	%	
Wood Creek	70	10	20	SL/SLC
Site 40N	68	20	11	SL
Dyke 5	71	18	11	SL
Site 40S	69	19	12	SL
DL	1	1	1	

Appendix C
Moss Laboratory Analysis Results

	ICP-MS				
	Sb	**As**	**Ba**	**Bi**	**Pb**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	0.058	0.668	80.8	<0.30	3.08
Site 40N	0.426	1.45	49.5	<0.30	3.42
Dyke 5	<0.050	1.84	51.0	<0.30	6.17
Site 40S	<0.050	3.08	57.1	<0.30	4.66
DL	0.05	0.05	0.1	0.3	0.1

	ICP-MS				
	Li	**Mo**	**Ni**	**Se**	**Tl**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	1.60	1.22	36.6	<1.0	0.038
Site 40N	4.11	2.29	27.4	<1.0	0.057
Dyke 5	3.65	1.54	44.5	<1.0	0.085
Site 40S	7.79	0.792	24.2	<1.0	0.112
DL	0.5	0.05	0.5	1	0.03

	ICP-MS		
	Sn	**U**	**V**
	mg/kg	mg/kg	mg/kg
Wood Creek	0.21	0.447	11.4
Site 40N	0.30	0.510	35.2
Dyke 5	<0.20	1.39	41.4
Site 40S	<0.20	0.754	21.5
DL	0.2	0.01	0.5

	ICP-OES				
	Al	**Be**	**Cd**	**Ca**	**Cr**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	2760	0.55	<0.50	3580	69.8
Site 40N	3040	0.96	<0.50	9870	32.7
Dyke 5	1900	1.83	<0.50	3950	64.5
Site 40S	5340	1.88	<0.50	8900	34.8
DL	10	0.2	0.5	10	0.5

	ICP-OES				
	Co	**Cu**	**Fe**	**Mg**	**Mn**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	2.14	5.73	2790	685	111
Site 40N	3.29	6.75	5730	2180	158
Dyke 5	5.62	5.91	11400	1090	232
Site 40S	4.81	8.42	10400	3620	158
DL	0.5	0.5	5	5	0.2

	ICP-OES				
	P	**K**	**Na**	**Sr**	**Ti**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	429	1520	<100	15.7	26.9
Site 40N	704	2430	<100	41.2	33.8
Dyke 5	315	660	<100	21.9	111
Site 40S	544	1650	<100	39.2	43.5
DL	20	100	100	0.3	0.5

	ICP-OES
	Zn
	mg/kg
Wood Creek	13.1
Site 40N	52.2
Dyke 5	26.3
Site 40S	35.3
DL	0.5

	ICP				
	B	**Ca**	**Cu**	**Fe**	**K**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	5.0	6470	42.8	4980	1830
Site 40N	21.7	10400	28.2	5740	1870
Dyke 5	13.0	6090	86.8	11500	1060
Site 40S	12.4	9450	51.1	8400	950
DL	1	100	1	10	100

	ICP				
	B	**Ca**	**Cu**	**Fe**	**K**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	5.0	6470	42.8	4980	1830
Site 40N	21.7	10400	28.2	5740	1870
Dyke 5	13.0	6090	86.8	11500	1060
Site 40S	12.4	9450	51.1	8400	950
DL	1	100	1	10	100

	ICP				
	Mg	**Na**	**P**	**S**	**Zn**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	1180	40.7	610	990	34.1
Site 40N	2020	49.1	790	1870	76.4
Dyke 5	1030	25.0	410	1970	43.7
Site 40S	2260	37.7	610	2330	43.4
DL	100	1	100	100	1

	ICP	
	Mn	**Leco N**
	mg/kg	%
Wood Creek	242	0.665
Site 40N	144	0.942
Dyke 5	192	0.409
Site 40S	151	1.10
DL	1	0.02

Appendix D
Litter Laboratory Analysis Results

	MET-DRY-MS				
	Sb	**As**	**Ba**	**Bi**	**Pb**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	0.073	0.994	91.1	<0.30	3.81
Site 40N	0.068	1.12	95.7	<0.30	2.36
Dyke 5	<0.050	0.629	16.7	<0.30	1.49
Site 40S	0.061	0.601	106	<0.30	1.13
DL	0.05	0.05	0.1	0.3	0.1

	MET-DRY-MS				
	Li	**Mo**	**Ni**	**Se**	**Tl**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	3.43	1.79	16.9	<1.0	0.103
Site 40N	3.38	1.59	19.0	<1.0	0.045
Dyke 5	1.06	0.680	9.79	<1.0	<0.030
Site 40S	2.71	0.749	8.41	<1.0	0.031
DL	0.5	0.05	0.5	1	0.03

	MET-DRY-MS		
	Sn	**U**	**V**
	mg/kg	mg/kg	mg/kg
Wood Creek	0.21	0.208	21.3
Site 40N	0.23	0.279	23.2
Dyke 5	<0.20	0.234	8.56
Site 40S	<0.20	0.117	9.11
DL	0.2	0.01	0.5

	MET-DRY-MS				
	Al	**Be**	**Cd**	**Ca**	**Cr**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	2860	0.67	<0.50	14400	18.5
Site 40N	2460	0.70	<0.50	23600	22.5
Dyke 5	726	0.41	<0.50	10800	13.7
Site 40S	1440	0.34	<0.50	32400	10.2
DL	10	0.2	0.5	10	0.5

	MET-DRY-MS				
	Co	**Cu**	**Fe**	**Mg**	**Mn**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	2.94	8.42	3970	1450	785
Site 40N	2.78	6.26	3970	2150	148
Dyke 5	1.63	3.49	2700	1150	66.2
Site 40S	1.21	4.49	2220	1440	125
DL	0.5	0.5	5	5	0.2

	MET-DRY-MS				
	P	**K**	**Na**	**Sr**	**Ti**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	801	1510	110	29.8	34.5
Site 40N	841	1920	210	126	29.2
Dyke 5	314	500	<100	22.5	40.2
Site 40S	647	1650	130	276	18.9
DL	20	100	100	0.3	0.5

	MET-DRY-MS
	Zn
	mg/kg
Wood Creek	74.6
Site 40N	190
Dyke 5	80.3
Site 40S	261
DL	0.5

	ICP-Nutrients				
	B	**Ca**	**Cu**	**Fe**	**K**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	14.2	12500	10.6	3510	1100
Site 40N	21.2	27600	12.8	3500	1250
Dyke 5	15.0	8830	40.6	7660	390
Site 40S	36.4	24700	7.8	1970	1270
DL	1	100	1	10	100

	ICP-Nutrients				
	Mg	**Na**	**P**	**S**	**Zn**
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg
Wood Creek	1280	78.4	690	1560	67.8
Site 40N	1930	113	720	1940	205
Dyke 5	1070	26.3	310	1740	76.9
Site 40S	1250	87.3	560	1440	199
DL	100	1	100	100	1

	ICP-Nutrients	
	Mn	**Leco N**
	mg/kg	%
Wood Creek	663	1.08
Site 40N	137	0.856
Dyke 5	137	0.636
Site 40S	94.5	0.833
DL	1	0.02

Appendix E
Laboratory Chain of Custody Forms

ALS Laboratory Group
ANALYTICAL CHEMISTRY & TESTING SERVICES

Environmental Division

Certificate of Analysis

SUNCOR ENERGY INC
ATTN: SARA DUNCAN
310-128 CROFT STREET
VICTORIA BC V8V 2E6

Report Date: 14-SEP-09 12:09 (MT)
Version: FINAL

Lab Work Order #: L806210 **Date Received:** 17-AUG-09

Project P.O. #: PO # 10000948024
Job Reference:
Legal Site Desc:
CofC Numbers:

Other Information:

Comments:

RAECHELLE JANZEN
Account Manager

ALS Canada Ltd.
Part of the **ALS Laboratory Group**
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Phone: +1 306 668 8370 Fax: +1 306 668 8383 www.alsglobal.com
A Campbell Brothers Limited Company

ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-1 WOOD CREEK 0-5CM SOIL							
Sampled By: SD on 07-JUL-09							
Matrix: SOIL							
Miscellaneous Parameters							
Conductivity (1:2)	<0.10		0.10	dS m-1	24-AUG-09	24-AUG-09	R919637
pH (1:2 soil:water)	4.22		0.10	pH	24-AUG-09	24-AUG-09	R919637
Particle Size Analysis: Hydrometer							
% Sand (2.0mm - 0.05mm)	70.0		1.0	%	21-AUG-09	24-AUG-09	R919009
% Silt (0.05mm - 2um)	10.0		1.0	%	21-AUG-09	24-AUG-09	R919009
% Clay (<2um)	20.0		1.0	%	21-AUG-09	24-AUG-09	R919009
Texture	Sandy loam / Sandy clay loam				21-AUG-09	24-AUG-09	R919009
Metals							
Silver (Ag)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Barium (Ba)	15.9		5.0	mg/kg		19-AUG-09	R913264
Beryllium (Be)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Cadmium (Cd)	<0.50		0.50	mg/kg		19-AUG-09	R913264
Cobalt (Co)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Chromium (Cr)	1.44		0.50	mg/kg		19-AUG-09	R913264
Copper (Cu)	10.1		2.0	mg/kg		19-AUG-09	R913264
Molybdenum (Mo)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Nickel (Ni)	<2.0		2.0	mg/kg		19-AUG-09	R913264
Lead (Pb)	<5.0		5.0	mg/kg		19-AUG-09	R913264
Tin (Sn)	5.6		5.0	mg/kg		19-AUG-09	R913264
Strontium (Sr)	2.7		1.0	mg/kg		19-AUG-09	R913264
Thallium (Tl)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Vanadium (V)	5.1		1.0	mg/kg		19-AUG-09	R913264
Zinc (Zn)	<10		10	mg/kg		19-AUG-09	R913264
Total Organic Nitrogen - Soil							
Available Ammonium-N							
Available Ammonium-N	2.96		0.80	mg/kg	22-AUG-09	22-AUG-09	R918365
Nitrogen, Total Organic							
Total Organic Nitrogen	0.066		0.020	%		26-AUG-09	
Total Kjeldahl Nitrogen (Organic N)							
Total Kjeldahl Nitrogen	0.067		0.020	%	21-AUG-09	24-AUG-09	R923115
Available N, P, K and S							
Available Nitrate-N							
Available Nitrate-N	<2.0		2.0	mg/kg	21-AUG-09	21-AUG-09	R917403
Available Phosphate & Potassium							
Available Phosphate-P	2.3		1.0	mg/kg	24-AUG-09	24-AUG-09	R918633
Available Potassium	52.8		2.0	mg/kg	24-AUG-09	24-AUG-09	R918633
Available Sulfate-S							
Available Sulfate-S	3.7		2.0	mg/kg	21-AUG-09	21-AUG-09	R917646
Sodium Adsorption Ratio							
SAR and Cations in saturated soil							
Calcium (Ca)	5.5		1.0	mg/L	24-AUG-09	24-AUG-09	R919030
Potassium (K)	5.2		2.0	mg/L	24-AUG-09	24-AUG-09	R919030
Magnesium (Mg)	2.6		1.0	mg/L	24-AUG-09	24-AUG-09	R919030
Sodium (Na)	<4.0		4.0	mg/L	24-AUG-09	24-AUG-09	R919030
SAR	<0.40	SAR:DL	0.40	SAR	24-AUG-09	24-AUG-09	R919030
Saturated Paste							
% Saturation	46.0		1.0	%	24-AUG-09	24-AUG-09	R919118
L806210-2 WOOD CREEK LITTER							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
ICPOES & ICPMS in Tissue (Dry Weight)							

* Refer to Referenced Information for Qualifiers (if any) and Methodology.

ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-2 WOOD CREEK LITTER							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
Metals in Tissue by ICPMS							
Antimony (Sb)-Total	0.073		0.050	mg/kg		05-SEP-09	R937105
Arsenic (As)-Total	0.994		0.050	mg/kg		05-SEP-09	R937105
Barium (Ba)-Total	91.1		0.10	mg/kg		05-SEP-09	R937105
Bismuth (Bi)-Total	<0.30		0.30	mg/kg		05-SEP-09	R937105
Lead (Pb)-Total	3.81		0.10	mg/kg		05-SEP-09	R937105
Lithium (Li)-Total	3.43		0.50	mg/kg		05-SEP-09	R937105
Molybdenum (Mo)-Total	1.79		0.050	mg/kg		05-SEP-09	R937105
Nickel (Ni)-Total	16.9		0.50	mg/kg		05-SEP-09	R937105
Selenium (Se)-Total	<1.0		1.0	mg/kg		05-SEP-09	R937105
Thallium (Tl)-Total	0.103		0.030	mg/kg		05-SEP-09	R937105
Tin (Sn)-Total	0.21		0.20	mg/kg		05-SEP-09	R937105
Uranium (U)-Total	0.208		0.010	mg/kg		05-SEP-09	R937105
Vanadium (V)-Total	21.3		0.50	mg/kg		05-SEP-09	R937105
Metals in Tissue by ICPOES							
Aluminum (Al)-Total	2860		10	mg/kg		31-AUG-09	R937236
Beryllium (Be)-Total	0.67		0.20	mg/kg		31-AUG-09	R937236
Cadmium (Cd)-Total	<0.50		0.50	mg/kg		31-AUG-09	R937236
Calcium (Ca)-Total	14400		10	mg/kg		31-AUG-09	R937236
Chromium (Cr)-Total	18.5		0.50	mg/kg		31-AUG-09	R937236
Cobalt (Co)-Total	2.94		0.50	mg/kg		31-AUG-09	R937236
Copper (Cu)-Total	8.42	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Iron (Fe)-Total	3970		5.0	mg/kg		31-AUG-09	R937236
Magnesium (Mg)-Total	1450		5.0	mg/kg		31-AUG-09	R937236
Manganese (Mn)-Total	785		0.20	mg/kg		31-AUG-09	R937236
Phosphorus (P)-Total	801		20	mg/kg		31-AUG-09	R937236
Potassium (K)-Total	1510	RRVAP	100	mg/kg		31-AUG-09	R937236
Sodium (Na)-Total	110		100	mg/kg		31-AUG-09	R937236
Strontium (Sr)-Total	29.8		0.30	mg/kg		31-AUG-09	R937236
Titanium (Ti)-Total	34.5		0.50	mg/kg		31-AUG-09	R937236
Zinc (Zn)-Total	74.6		0.50	mg/kg		31-AUG-09	R937236
Total N and Minerals							
Total ICP scan -Nutrients							
Boron (B)-Total	14.2		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Calcium (Ca)-Total	12500		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Copper (Cu)-Total	10.6		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Iron (Fe)-Total	3510		10	mg/kg	24-AUG-09	24-AUG-09	R918323
Potassium (K)-Total	1100		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Magnesium (Mg)-Total	1280		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sodium (Na)-Total	78.4		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Phosphorus (P)-Total	690		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sulfur (S)-Total	1560		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Zinc (Zn)-Total	67.8		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Manganese (Mn)-Total	663		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Total Nitrogen by Combustion							
Total Nitrogen by Leco	1.08		0.020	%	24-AUG-09	24-AUG-09	R918924
L806210-3 WOOD CREEK MOSS							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
ICPOES & ICPMS in Tissue (Dry Weight)							
Metals in Tissue by ICPMS							

* Refer to Referenced Information for Qualifiers (if any) and Methodology.

ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-3 WOOD CREEK MOSS							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
Metals in Tissue by ICPMS							
Antimony (Sb)-Total	0.058		0.050	mg/kg		05-SEP-09	R937105
Arsenic (As)-Total	0.668		0.050	mg/kg		05-SEP-09	R937105
Barium (Ba)-Total	80.8		0.10	mg/kg		05-SEP-09	R937105
Bismuth (Bi)-Total	<0.30		0.30	mg/kg		05-SEP-09	R937105
Lead (Pb)-Total	3.08		0.10	mg/kg		05-SEP-09	R937105
Lithium (Li)-Total	1.60		0.50	mg/kg		05-SEP-09	R937105
Molybdenum (Mo)-Total	1.22		0.050	mg/kg		05-SEP-09	R937105
Nickel (Ni)-Total	36.6		0.50	mg/kg		05-SEP-09	R937105
Selenium (Se)-Total	<1.0		1.0	mg/kg		05-SEP-09	R937105
Thallium (Tl)-Total	0.038		0.030	mg/kg		05-SEP-09	R937105
Tin (Sn)-Total	0.21		0.20	mg/kg		05-SEP-09	R937105
Uranium (U)-Total	0.447		0.010	mg/kg		05-SEP-09	R937105
Vanadium (V)-Total	11.4		0.50	mg/kg		05-SEP-09	R937105
Metals in Tissue by ICPOES							
Aluminum (Al)-Total	2760		10	mg/kg		31-AUG-09	R937236
Beryllium (Be)-Total	0.55		0.20	mg/kg		31-AUG-09	R937236
Cadmium (Cd)-Total	<0.50		0.50	mg/kg		31-AUG-09	R937236
Calcium (Ca)-Total	3580	RRVAP	10	mg/kg		31-AUG-09	R937236
Chromium (Cr)-Total	69.8		0.50	mg/kg		31-AUG-09	R937236
Cobalt (Co)-Total	2.14		0.50	mg/kg		31-AUG-09	R937236
Copper (Cu)-Total	5.73	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Iron (Fe)-Total	2790	RRVAP	5.0	mg/kg		31-AUG-09	R937236
Magnesium (Mg)-Total	685	RRVAP	5.0	mg/kg		31-AUG-09	R937236
Manganese (Mn)-Total	111	RRVAP	0.20	mg/kg		31-AUG-09	R937236
Phosphorus (P)-Total	429		20	mg/kg		31-AUG-09	R937236
Potassium (K)-Total	1520		100	mg/kg		31-AUG-09	R937236
Sodium (Na)-Total	<100		100	mg/kg		31-AUG-09	R937236
Strontium (Sr)-Total	15.7		0.30	mg/kg		31-AUG-09	R937236
Titanium (Ti)-Total	26.9		0.50	mg/kg		31-AUG-09	R937236
Zinc (Zn)-Total	13.1	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Total N and Minerals							
Total ICP scan -Nutrients							
Boron (B)-Total	5.0		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Calcium (Ca)-Total	6470		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Copper (Cu)-Total	42.8		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Iron (Fe)-Total	4980		10	mg/kg	24-AUG-09	24-AUG-09	R918323
Potassium (K)-Total	1830		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Magnesium (Mg)-Total	1180		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sodium (Na)-Total	40.7		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Phosphorus (P)-Total	610		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sulfur (S)-Total	990		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Zinc (Zn)-Total	34.1		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Manganese (Mn)-Total	242		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Total Nitrogen by Combustion							
Total Nitrogen by Leco	0.665		0.020	%	24-AUG-09	24-AUG-09	R918924
L806210-4 TH SOIL 0-5CM							
Sampled By: SD on 07-JUL-09							
Matrix: SOIL							
Miscellaneous Parameters							
Conductivity (1:2)	0.29		0.10	dS m-1	24-AUG-09	24-AUG-09	R919637

* Refer to Referenced Information for Qualifiers (if any) and Methodology.

ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-4 TH SOIL 0-5CM							
Sampled By: SD on 07-JUL-09							
Matrix: SOIL							
pH (1:2 soil:water)	7.55		0.10	pH	24-AUG-09	24-AUG-09	R919637
Particle Size Analysis: Hydrometer							
% Sand (2.0mm - 0.05mm)	68.0		1.0	%	21-AUG-09	24-AUG-09	R919009
% Silt (0.05mm - 2um)	20.0		1.0	%	21-AUG-09	24-AUG-09	R919009
% Clay (<2um)	11.0		1.0	%	21-AUG-09	24-AUG-09	R919009
Texture	Sandy loam				21-AUG-09	24-AUG-09	R919009
Metals							
Silver (Ag)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Barium (Ba)	35.9		5.0	mg/kg		19-AUG-09	R913264
Beryllium (Be)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Cadmium (Cd)	<0.50		0.50	mg/kg		19-AUG-09	R913264
Cobalt (Co)	4.6		1.0	mg/kg		19-AUG-09	R913264
Chromium (Cr)	8.30		0.50	mg/kg		19-AUG-09	R913264
Copper (Cu)	14.3		2.0	mg/kg		19-AUG-09	R913264
Molybdenum (Mo)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Nickel (Ni)	11.9		2.0	mg/kg		19-AUG-09	R913264
Lead (Pb)	5.0		5.0	mg/kg		19-AUG-09	R913264
Tin (Sn)	<5.0		5.0	mg/kg		19-AUG-09	R913264
Strontium (Sr)	34.0		1.0	mg/kg		19-AUG-09	R913264
Thallium (Tl)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Vanadium (V)	16.1		1.0	mg/kg		19-AUG-09	R913264
Zinc (Zn)	29		10	mg/kg		19-AUG-09	R913264
Total Organic Nitrogen - Soil							
Available Ammonium-N							
Available Ammonium-N	2.30		0.80	mg/kg	22-AUG-09	22-AUG-09	R918365
Nitrogen, Total Organic							
Total Organic Nitrogen	0.198		0.020	%		26-AUG-09	
Total Kjeldahl Nitrogen (Organic N)							
Total Kjeldahl Nitrogen	0.199		0.020	%	21-AUG-09	24-AUG-09	R923115
Available N, P, K and S							
Available Nitrate-N							
Available Nitrate-N	<2.0		2.0	mg/kg	21-AUG-09	21-AUG-09	R917403
Available Phosphate & Potassium							
Available Phosphate-P	5.6		1.0	mg/kg	24-AUG-09	24-AUG-09	R918633
Available Potassium	85.6		2.0	mg/kg	24-AUG-09	24-AUG-09	R918633
Available Sulfate-S							
Available Sulfate-S	68.2		2.0	mg/kg	21-AUG-09	21-AUG-09	R917646
Sodium Adsorption Ratio							
SAR and Cations in saturated soil							
Calcium (Ca)	97.1		1.0	mg/L	24-AUG-09	24-AUG-09	R919030
Potassium (K)	11.6		2.0	mg/L	24-AUG-09	24-AUG-09	R919030
Magnesium (Mg)	38.1		1.0	mg/L	24-AUG-09	24-AUG-09	R919030
Sodium (Na)	8.9		4.0	mg/L	24-AUG-09	24-AUG-09	R919030
SAR	0.19		0.10	SAR	24-AUG-09	24-AUG-09	R919030
Saturated Paste							
% Saturation	77.0		1.0	%	24-AUG-09	24-AUG-09	R919118
L806210-5 TH LITTER							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
ICPOES & ICPMS in Tissue (Dry Weight)							
Metals in Tissue by ICPMS							
Antimony (Sb)-Total	0.068		0.050	mg/kg		05-SEP-09	R937105

* Refer to Referenced Information for Qualifiers (if any) and Methodology

ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-5 TH LITTER							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
Metals in Tissue by ICPMS							
Arsenic (As)-Total	1.12		0.050	mg/kg		05-SEP-09	R937105
Barium (Ba)-Total	95.7		0.10	mg/kg		05-SEP-09	R937105
Bismuth (Bi)-Total	<0.30		0.30	mg/kg		05-SEP-09	R937105
Lead (Pb)-Total	2.36		0.10	mg/kg		05-SEP-09	R937105
Lithium (Li)-Total	3.38		0.50	mg/kg		05-SEP-09	R937105
Molybdenum (Mo)-Total	1.59		0.050	mg/kg		05-SEP-09	R937105
Nickel (Ni)-Total	19.0		0.50	mg/kg		05-SEP-09	R937105
Selenium (Se)-Total	<1.0		1.0	mg/kg		05-SEP-09	R937105
Thallium (Tl)-Total	0.045		0.030	mg/kg		05-SEP-09	R937105
Tin (Sn)-Total	0.23		0.20	mg/kg		05-SEP-09	R937105
Uranium (U)-Total	0.279		0.010	mg/kg		05-SEP-09	R937105
Vanadium (V)-Total	23.2		0.50	mg/kg		05-SEP-09	R937105
Metals in Tissue by ICPOES							
Aluminum (Al)-Total	2460		10	mg/kg		31-AUG-09	R937236
Beryllium (Be)-Total	0.70		0.20	mg/kg		31-AUG-09	R937236
Cadmium (Cd)-Total	<0.50		0.50	mg/kg		31-AUG-09	R937236
Calcium (Ca)-Total	23600		10	mg/kg		31-AUG-09	R937236
Chromium (Cr)-Total	22.5		0.50	mg/kg		31-AUG-09	R937236
Cobalt (Co)-Total	2.78		0.50	mg/kg		31-AUG-09	R937236
Copper (Cu)-Total	6.26		0.50	mg/kg		31-AUG-09	R937236
Iron (Fe)-Total	3970		5.0	mg/kg		31-AUG-09	R937236
Magnesium (Mg)-Total	2150		5.0	mg/kg		31-AUG-09	R937236
Manganese (Mn)-Total	148		0.20	mg/kg		31-AUG-09	R937236
Phosphorus (P)-Total	841		20	mg/kg		31-AUG-09	R937236
Potassium (K)-Total	1920		100	mg/kg		31-AUG-09	R937236
Sodium (Na)-Total	210		100	mg/kg		31-AUG-09	R937236
Strontium (Sr)-Total	126		0.30	mg/kg		31-AUG-09	R937236
Titanium (Ti)-Total	29.2		0.50	mg/kg		31-AUG-09	R937236
Zinc (Zn)-Total	190		0.50	mg/kg		31-AUG-09	R937236
Total N and Minerals							
Total ICP scan -Nutrients							
Boron (B)-Total	21.2		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Calcium (Ca)-Total	27600		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Copper (Cu)-Total	12.8		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Iron (Fe)-Total	3500		10	mg/kg	24-AUG-09	24-AUG-09	R918323
Potassium (K)-Total	1250		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Magnesium (Mg)-Total	1930		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sodium (Na)-Total	113		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Phosphorus (P)-Total	720		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sulfur (S)-Total	1940		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Zinc (Zn)-Total	205		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Manganese (Mn)-Total	137		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Total Nitrogen by Combustion							
Total Nitrogen by Leco	0.856		0.020	%	24-AUG-09	24-AUG-09	R918924
L806210-6 TH MOSS							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
ICPOES & ICPMS in Tissue (Dry Weight)							
Metals in Tissue by ICPMS							
Antimony (Sb)-Total	0.426		0.050	mg/kg		05-SEP-09	R937105

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ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-6 TH MOSS							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
Metals in Tissue by ICPMS							
Arsenic (As)-Total	1.45		0.050	mg/kg		05-SEP-09	R937105
Barium (Ba)-Total	49.5		0.10	mg/kg		05-SEP-09	R937105
Bismuth (Bi)-Total	<0.30		0.30	mg/kg		05-SEP-09	R937105
Lead (Pb)-Total	3.42		0.10	mg/kg		05-SEP-09	R937105
Lithium (Li)-Total	4.11		0.50	mg/kg		05-SEP-09	R937105
Molybdenum (Mo)-Total	2.29		0.050	mg/kg		05-SEP-09	R937105
Nickel (Ni)-Total	27.4		0.50	mg/kg		05-SEP-09	R937105
Selenium (Se)-Total	<1.0		1.0	mg/kg		05-SEP-09	R937105
Thallium (Tl)-Total	0.057		0.030	mg/kg		05-SEP-09	R937105
Tin (Sn)-Total	0.30		0.20	mg/kg		05-SEP-09	R937105
Uranium (U)-Total	0.510		0.010	mg/kg		05-SEP-09	R937105
Vanadium (V)-Total	35.2		0.50	mg/kg		05-SEP-09	R937105
Metals in Tissue by ICPOES							
Aluminum (Al)-Total	3040		10	mg/kg		31-AUG-09	R937236
Beryllium (Be)-Total	0.96		0.20	mg/kg		31-AUG-09	R937236
Cadmium (Cd)-Total	<0.50		0.50	mg/kg		31-AUG-09	R937236
Calcium (Ca)-Total	9870		10	mg/kg		31-AUG-09	R937236
Chromium (Cr)-Total	32.7		0.50	mg/kg		31-AUG-09	R937236
Cobalt (Co)-Total	3.29		0.50	mg/kg		31-AUG-09	R937236
Copper (Cu)-Total	6.75	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Iron (Fe)-Total	5730		5.0	mg/kg		31-AUG-09	R937236
Magnesium (Mg)-Total	2180		5.0	mg/kg		31-AUG-09	R937236
Manganese (Mn)-Total	158		0.20	mg/kg		31-AUG-09	R937236
Phosphorus (P)-Total	704		20	mg/kg		31-AUG-09	R937236
Potassium (K)-Total	2430	RRVAP	100	mg/kg		31-AUG-09	R937236
Sodium (Na)-Total	<100		100	mg/kg		31-AUG-09	R937236
Strontium (Sr)-Total	41.2		0.30	mg/kg		31-AUG-09	R937236
Titanium (Ti)-Total	33.8		0.50	mg/kg		31-AUG-09	R937236
Zinc (Zn)-Total	52.2	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Total N and Minerals							
Total ICP scan -Nutrients							
Boron (B)-Total	21.7		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Calcium (Ca)-Total	10400		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Copper (Cu)-Total	28.2		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Iron (Fe)-Total	5740		10	mg/kg	24-AUG-09	24-AUG-09	R918323
Potassium (K)-Total	1870		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Magnesium (Mg)-Total	2020		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sodium (Na)-Total	49.1		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Phosphorus (P)-Total	790		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sulfur (S)-Total	1870		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Zinc (Zn)-Total	76.4		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Manganese (Mn)-Total	144		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Total Nitrogen by Combustion							
Total Nitrogen by Leco	0.942		0.020	%	24-AUG-09	24-AUG-09	R918924
L806210-7 D5 SOIL 0-5CM							
Sampled By: SD on 07-JUL-09							
Matrix: SOIL							
Miscellaneous Parameters							
Conductivity (1:2)	0.16		0.10	dS m-1	24-AUG-09	24-AUG-09	R919637
pH (1:2 soil:water)	7.77		0.10	pH	24-AUG-09	24-AUG-09	R919637

* Refer to Referenced Information for Qualifiers (if any) and Methodology.

ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-7 D5 SOIL 0-5CM							
Sampled By: SD on 07-JUL-09							
Matrix: SOIL							
Particle Size Analysis: Hydrometer							
% Sand (2.0mm - 0.05mm)	71.0		1.0	%	21-AUG-09	24-AUG-09	R919009
% Silt (0.05mm - 2um)	18.0		1.0	%	21-AUG-09	24-AUG-09	R919009
% Clay (<2um)	11.0		1.0	%	21-AUG-09	24-AUG-09	R919009
Texture	Sandy loam				21-AUG-09	24-AUG-09	R919009
Metals							
Silver (Ag)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Barium (Ba)	27.0		5.0	mg/kg		19-AUG-09	R913264
Beryllium (Be)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Cadmium (Cd)	<0.50		0.50	mg/kg		19-AUG-09	R913264
Cobalt (Co)	3.0		1.0	mg/kg		19-AUG-09	R913264
Chromium (Cr)	4.85		0.50	mg/kg		19-AUG-09	R913264
Copper (Cu)	4.3		2.0	mg/kg		19-AUG-09	R913264
Molybdenum (Mo)	1.1		1.0	mg/kg		19-AUG-09	R913264
Nickel (Ni)	10.0		2.0	mg/kg		19-AUG-09	R913264
Lead (Pb)	<5.0		5.0	mg/kg		19-AUG-09	R913264
Tin (Sn)	<5.0		5.0	mg/kg		19-AUG-09	R913264
Strontium (Sr)	17.5		1.0	mg/kg		19-AUG-09	R913264
Thallium (Tl)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Vanadium (V)	21.4		1.0	mg/kg		19-AUG-09	R913264
Zinc (Zn)	14		10	mg/kg		19-AUG-09	R913264
Total Organic Nitrogen - Soil							
Available Ammonium-N							
Available Ammonium-N	1.33		0.80	mg/kg	22-AUG-09	22-AUG-09	R918365
Nitrogen, Total Organic							
Total Organic Nitrogen	0.121		0.020	%		26-AUG-09	
Total Kjeldahl Nitrogen (Organic N)							
Total Kjeldahl Nitrogen	0.122		0.020	%	21-AUG-09	24-AUG-09	R923115
Available N, P, K and S							
Available Nitrate-N							
Available Nitrate-N	4.3		2.0	mg/kg	21-AUG-09	21-AUG-09	R917403
Available Phosphate & Potassium							
Available Phosphate-P	31.3		1.0	mg/kg	24-AUG-09	24-AUG-09	R918633
Available Potassium	57.9		2.0	mg/kg	24-AUG-09	24-AUG-09	R918633
Available Sulfate-S							
Available Sulfate-S	5.6		2.0	mg/kg	21-AUG-09	21-AUG-09	R917646
Sodium Adsorption Ratio							
SAR and Cations in saturated soil							
Calcium (Ca)	47.0		1.0	mg/L	24-AUG-09	24-AUG-09	R919030
Potassium (K)	8.7		2.0	mg/L	24-AUG-09	24-AUG-09	R919030
Magnesium (Mg)	10.0		1.0	mg/L	24-AUG-09	24-AUG-09	R919030
Sodium (Na)	6.5		4.0	mg/L	24-AUG-09	24-AUG-09	R919030
SAR	0.22		0.10	SAR	24-AUG-09	24-AUG-09	R919030
Saturated Paste							
% Saturation	44.0		1.0	%	24-AUG-09	24-AUG-09	R919118
L806210-8 D5 LITTER							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
ICPOES & ICPMS in Tissue (Dry Weight)							
Metals in Tissue by ICPMS							
Antimony (Sb)-Total	<0.050		0.050	mg/kg		05-SEP-09	R937105
Arsenic (As)-Total	0.629		0.050	mg/kg		05-SEP-09	R937105

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ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-8 D5 LITTER							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
Metals in Tissue by ICPMS							
Barium (Ba)-Total	16.7		0.10	mg/kg		05-SEP-09	R937105
Bismuth (Bi)-Total	<0.30		0.30	mg/kg		05-SEP-09	R937105
Lead (Pb)-Total	1.49		0.10	mg/kg		05-SEP-09	R937105
Lithium (Li)-Total	1.06		0.50	mg/kg		05-SEP-09	R937105
Molybdenum (Mo)-Total	0.680		0.050	mg/kg		05-SEP-09	R937105
Nickel (Ni)-Total	9.79		0.50	mg/kg		05-SEP-09	R937105
Selenium (Se)-Total	<1.0		1.0	mg/kg		05-SEP-09	R937105
Thallium (Tl)-Total	<0.030		0.030	mg/kg		05-SEP-09	R937105
Tin (Sn)-Total	<0.20		0.20	mg/kg		05-SEP-09	R937105
Uranium (U)-Total	0.234		0.010	mg/kg		05-SEP-09	R937105
Vanadium (V)-Total	8.56		0.50	mg/kg		05-SEP-09	R937105
Metals in Tissue by ICPOES							
Aluminum (Al)-Total	726		10	mg/kg		31-AUG-09	R937236
Beryllium (Be)-Total	0.41		0.20	mg/kg		31-AUG-09	R937236
Cadmium (Cd)-Total	<0.50		0.50	mg/kg		31-AUG-09	R937236
Calcium (Ca)-Total	10800	RRVAP	10	mg/kg		31-AUG-09	R937236
Chromium (Cr)-Total	13.7		0.50	mg/kg		31-AUG-09	R937236
Cobalt (Co)-Total	1.63		0.50	mg/kg		31-AUG-09	R937236
Copper (Cu)-Total	3.49	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Iron (Fe)-Total	2700	RRVAP	5.0	mg/kg		31-AUG-09	R937236
Magnesium (Mg)-Total	1150		5.0	mg/kg		31-AUG-09	R937236
Manganese (Mn)-Total	66.2	RRVAP	0.20	mg/kg		31-AUG-09	R937236
Phosphorus (P)-Total	314		20	mg/kg		31-AUG-09	R937236
Potassium (K)-Total	500		100	mg/kg		31-AUG-09	R937236
Sodium (Na)-Total	<100		100	mg/kg		31-AUG-09	R937236
Strontium (Sr)-Total	22.5		0.30	mg/kg		31-AUG-09	R937236
Titanium (Ti)-Total	40.2		0.50	mg/kg		31-AUG-09	R937236
Zinc (Zn)-Total	80.3		0.50	mg/kg		31-AUG-09	R937236
Total N and Minerals							
Total ICP scan -Nutrients							
Boron (B)-Total	15.0		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Calcium (Ca)-Total	8830		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Copper (Cu)-Total	40.6		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Iron (Fe)-Total	7660		10	mg/kg	24-AUG-09	24-AUG-09	R918323
Potassium (K)-Total	390		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Magnesium (Mg)-Total	1070		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sodium (Na)-Total	26.3		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Phosphorus (P)-Total	310		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sulfur (S)-Total	1740		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Zinc (Zn)-Total	76.9		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Manganese (Mn)-Total	137		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Total Nitrogen by Combustion							
Total Nitrogen by Leco	0.636		0.020	%	24-AUG-09	24-AUG-09	R918924
L806210-9 D5 MOSS							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
ICPOES & ICPMS in Tissue (Dry Weight)							
Metals in Tissue by ICPMS							
Antimony (Sb)-Total	<0.050		0.050	mg/kg		05-SEP-09	R937105
Arsenic (As)-Total	1.84		0.050	mg/kg		05-SEP-09	R937105

* Refer to Referenced Information for Qualifiers (if any) and Methodology.

ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-9 D5 MOSS							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
Metals in Tissue by ICPMS							
Barium (Ba)-Total	51.0		0.10	mg/kg		05-SEP-09	R937105
Bismuth (Bi)-Total	<0.30		0.30	mg/kg		05-SEP-09	R937105
Lead (Pb)-Total	6.17		0.10	mg/kg		05-SEP-09	R937105
Lithium (Li)-Total	3.65		0.50	mg/kg		05-SEP-09	R937105
Molybdenum (Mo)-Total	1.54		0.050	mg/kg		05-SEP-09	R937105
Nickel (Ni)-Total	44.5		0.50	mg/kg		05-SEP-09	R937105
Selenium (Se)-Total	<1.0		1.0	mg/kg		05-SEP-09	R937105
Thallium (Tl)-Total	0.085		0.030	mg/kg		05-SEP-09	R937105
Tin (Sn)-Total	<0.20		0.20	mg/kg		05-SEP-09	R937105
Uranium (U)-Total	1.39		0.010	mg/kg		05-SEP-09	R937105
Vanadium (V)-Total	41.4		0.50	mg/kg		05-SEP-09	R937105
Metals in Tissue by ICPOES							
Aluminum (Al)-Total	1900		10	mg/kg		31-AUG-09	R937236
Beryllium (Be)-Total	1.83		0.20	mg/kg		31-AUG-09	R937236
Cadmium (Cd)-Total	<0.50		0.50	mg/kg		31-AUG-09	R937236
Calcium (Ca)-Total	3950	RRVAP	10	mg/kg		31-AUG-09	R937236
Chromium (Cr)-Total	64.5		0.50	mg/kg		31-AUG-09	R937236
Cobalt (Co)-Total	5.62		0.50	mg/kg		31-AUG-09	R937236
Copper (Cu)-Total	5.91	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Iron (Fe)-Total	11400		5.0	mg/kg		31-AUG-09	R937236
Magnesium (Mg)-Total	1090		5.0	mg/kg		31-AUG-09	R937236
Manganese (Mn)-Total	232		0.20	mg/kg		31-AUG-09	R937236
Phosphorus (P)-Total	315		20	mg/kg		31-AUG-09	R937236
Potassium (K)-Total	660	RRVAP	100	mg/kg		31-AUG-09	R937236
Sodium (Na)-Total	<100		100	mg/kg		31-AUG-09	R937236
Strontium (Sr)-Total	21.9		0.30	mg/kg		31-AUG-09	R937236
Titanium (Ti)-Total	111		0.50	mg/kg		31-AUG-09	R937236
Zinc (Zn)-Total	26.3	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Total N and Minerals							
Total ICP scan -Nutrients							
Boron (B)-Total	13.0		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Calcium (Ca)-Total	6090		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Copper (Cu)-Total	86.8		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Iron (Fe)-Total	11500		10	mg/kg	24-AUG-09	24-AUG-09	R918323
Potassium (K)-Total	1060		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Magnesium (Mg)-Total	1030		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sodium (Na)-Total	25.0		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Phosphorus (P)-Total	410		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sulfur (S)-Total	1970		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Zinc (Zn)-Total	43.7		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Manganese (Mn)-Total	192		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Total Nitrogen by Combustion							
Total Nitrogen by Leco	0.409		0.020	%	24-AUG-09	24-AUG-09	R918924
L806210-10 18 SOIL 0-5CM							
Sampled By: SD on 07-JUL-09							
Matrix: SOIL							
Miscellaneous Parameters							
Conductivity (1:2)	0.27		0.10	dS m-1	24-AUG-09	24-AUG-09	R919637
pH (1:2 soil:water)	7.72		0.10	pH	24-AUG-09	24-AUG-09	R919637
Particle Size Analysis: Hydrometer							

* Refer to Referenced Information for Qualifiers (if any) and Methodology.

ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-10 18 SOIL 0-5CM							
Sampled By: SD on 07-JUL-09							
Matrix: SOIL							
Particle Size Analysis: Hydrometer							
% Sand (2.0mm - 0.05mm)	69.0		1.0	%	21-AUG-09	24-AUG-09	R919009
% Silt (0.05mm - 2um)	19.0		1.0	%	21-AUG-09	24-AUG-09	R919009
% Clay (<2um)	12.0		1.0	%	21-AUG-09	24-AUG-09	R919009
Texture	Sandy loam				21-AUG-09	24-AUG-09	R919009
Metals							
Silver (Ag)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Barium (Ba)	34.8		5.0	mg/kg		19-AUG-09	R913264
Beryllium (Be)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Cadmium (Cd)	<0.50		0.50	mg/kg		19-AUG-09	R913264
Cobalt (Co)	4.2		1.0	mg/kg		19-AUG-09	R913264
Chromium (Cr)	7.35		0.50	mg/kg		19-AUG-09	R913264
Copper (Cu)	9.5		2.0	mg/kg		19-AUG-09	R913264
Molybdenum (Mo)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Nickel (Ni)	11.1		2.0	mg/kg		19-AUG-09	R913264
Lead (Pb)	<5.0		5.0	mg/kg		19-AUG-09	R913264
Tin (Sn)	<5.0		5.0	mg/kg		19-AUG-09	R913264
Strontium (Sr)	31.2		1.0	mg/kg		19-AUG-09	R913264
Thallium (Tl)	<1.0		1.0	mg/kg		19-AUG-09	R913264
Vanadium (V)	14.8		1.0	mg/kg		19-AUG-09	R913264
Zinc (Zn)	28		10	mg/kg		19-AUG-09	R913264
Total Organic Nitrogen - Soil							
Available Ammonium-N							
Available Ammonium-N	1.85		0.80	mg/kg	22-AUG-09	22-AUG-09	R918365
Nitrogen, Total Organic							
Total Organic Nitrogen	0.179		0.020	%		26-AUG-09	
Total Kjeldahl Nitrogen (Organic N)							
Total Kjeldahl Nitrogen	0.179		0.020	%	21-AUG-09	24-AUG-09	R923115
Available N, P, K and S							
Available Nitrate-N							
Available Nitrate-N	<2.0		2.0	mg/kg	21-AUG-09	21-AUG-09	R917403
Available Phosphate & Potassium							
Available Phosphate-P	1.7		1.0	mg/kg	24-AUG-09	24-AUG-09	R918633
Available Potassium	96.0		2.0	mg/kg	24-AUG-09	24-AUG-09	R918633
Available Sulfate-S							
Available Sulfate-S	43.3		2.0	mg/kg	21-AUG-09	21-AUG-09	R917646
Sodium Adsorption Ratio							
SAR and Cations in saturated soil							
Calcium (Ca)	89.4		1.0	mg/L	24-AUG-09	24-AUG-09	R919030
Potassium (K)	12.9		2.0	mg/L	24-AUG-09	24-AUG-09	R919030
Magnesium (Mg)	30.7		1.0	mg/L	24-AUG-09	24-AUG-09	R919030
Sodium (Na)	7.2		4.0	mg/L	24-AUG-09	24-AUG-09	R919030
SAR	0.17		0.10	SAR	24-AUG-09	24-AUG-09	R919030
Saturated Paste							
% Saturation	36.0		1.0	%	24-AUG-09	24-AUG-09	R919118
L806210-11 18 LITTER							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
ICPOES & ICPMS in Tissue (Dry Weight)							
Metals in Tissue by ICPMS							
Antimony (Sb)-Total	0.061		0.050	mg/kg		05-SEP-09	R937105
Arsenic (As)-Total	0.601		0.050	mg/kg		05-SEP-09	R937105

* Refer to Referenced Information for Qualifiers (if any) and Methodology.

ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-11 18 LITTER							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
Metals in Tissue by ICPMS							
Barium (Ba)-Total	106		0.10	mg/kg		05-SEP-09	R937105
Bismuth (Bi)-Total	<0.30		0.30	mg/kg		05-SEP-09	R937105
Lead (Pb)-Total	1.13		0.10	mg/kg		05-SEP-09	R937105
Lithium (Li)-Total	2.71		0.50	mg/kg		05-SEP-09	R937105
Molybdenum (Mo)-Total	0.749		0.050	mg/kg		05-SEP-09	R937105
Nickel (Ni)-Total	8.41		0.50	mg/kg		05-SEP-09	R937105
Selenium (Se)-Total	<1.0		1.0	mg/kg		05-SEP-09	R937105
Thallium (Tl)-Total	0.031		0.030	mg/kg		05-SEP-09	R937105
Tin (Sn)-Total	<0.20		0.20	mg/kg		05-SEP-09	R937105
Uranium (U)-Total	0.117		0.010	mg/kg		05-SEP-09	R937105
Vanadium (V)-Total	9.11		0.50	mg/kg		05-SEP-09	R937105
Metals in Tissue by ICPOES							
Aluminum (Al)-Total	1440		10	mg/kg		31-AUG-09	R937236
Beryllium (Be)-Total	0.34		0.20	mg/kg		31-AUG-09	R937236
Cadmium (Cd)-Total	<0.50		0.50	mg/kg		31-AUG-09	R937236
Calcium (Ca)-Total	32400		10	mg/kg		31-AUG-09	R937236
Chromium (Cr)-Total	10.2		0.50	mg/kg		31-AUG-09	R937236
Cobalt (Co)-Total	1.21		0.50	mg/kg		31-AUG-09	R937236
Copper (Cu)-Total	4.49	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Iron (Fe)-Total	2220		5.0	mg/kg		31-AUG-09	R937236
Magnesium (Mg)-Total	1440		5.0	mg/kg		31-AUG-09	R937236
Manganese (Mn)-Total	125	RRVAP	0.20	mg/kg		31-AUG-09	R937236
Phosphorus (P)-Total	647		20	mg/kg		31-AUG-09	R937236
Potassium (K)-Total	1650	RRVAP	100	mg/kg		31-AUG-09	R937236
Sodium (Na)-Total	130		100	mg/kg		31-AUG-09	R937236
Strontium (Sr)-Total	276		0.30	mg/kg		31-AUG-09	R937236
Titanium (Ti)-Total	18.9		0.50	mg/kg		31-AUG-09	R937236
Zinc (Zn)-Total	261	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Total N and Minerals							
Total ICP scan -Nutrients							
Boron (B)-Total	36.4		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Calcium (Ca)-Total	24700		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Copper (Cu)-Total	7.8		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Iron (Fe)-Total	1970		10	mg/kg	24-AUG-09	24-AUG-09	R918323
Potassium (K)-Total	1270		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Magnesium (Mg)-Total	1250		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sodium (Na)-Total	87.3		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Phosphorus (P)-Total	560		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sulfur (S)-Total	1440		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Zinc (Zn)-Total	199		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Manganese (Mn)-Total	94.5		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Total Nitrogen by Combustion							
Total Nitrogen by Leco	0.833		0.020	%	24-AUG-09	24-AUG-09	R918924
L806210-12 18 MOSS							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
ICPOES & ICPMS in Tissue (Dry Weight)							
Metals in Tissue by ICPMS							
Antimony (Sb)-Total	<0.050		0.050	mg/kg		05-SEP-09	R937105
Arsenic (As)-Total	3.08		0.050	mg/kg		05-SEP-09	R937105

* Refer to Referenced Information for Qualifiers (if any) and Methodology.

ALS LABORATORY GROUP ANALYTICAL REPORT

Sample Details/Parameters	Result	Qualifier*	D.L.	Units	Extracted	Analyzed	Batch
L806210-12 18 MOSS							
Sampled By: SD on 07-JUL-09							
Matrix: VEG							
Metals in Tissue by ICPMS							
Barium (Ba)-Total	57.1		0.10	mg/kg		05-SEP-09	R937105
Bismuth (Bi)-Total	<0.30		0.30	mg/kg		05-SEP-09	R937105
Lead (Pb)-Total	4.66		0.10	mg/kg		05-SEP-09	R937105
Lithium (Li)-Total	7.79		0.50	mg/kg		05-SEP-09	R937105
Molybdenum (Mo)-Total	0.792		0.050	mg/kg		05-SEP-09	R937105
Nickel (Ni)-Total	24.2		0.50	mg/kg		05-SEP-09	R937105
Selenium (Se)-Total	<1.0		1.0	mg/kg		05-SEP-09	R937105
Thallium (Tl)-Total	0.112		0.030	mg/kg		05-SEP-09	R937105
Tin (Sn)-Total	<0.20		0.20	mg/kg		05-SEP-09	R937105
Uranium (U)-Total	0.754		0.010	mg/kg		05-SEP-09	R937105
Vanadium (V)-Total	21.5		0.50	mg/kg		05-SEP-09	R937105
Metals in Tissue by ICPOES							
Aluminum (Al)-Total	5340		10	mg/kg		31-AUG-09	R937236
Beryllium (Be)-Total	1.88		0.20	mg/kg		31-AUG-09	R937236
Cadmium (Cd)-Total	<0.50		0.50	mg/kg		31-AUG-09	R937236
Calcium (Ca)-Total	8900		10	mg/kg		31-AUG-09	R937236
Chromium (Cr)-Total	34.8		0.50	mg/kg		31-AUG-09	R937236
Cobalt (Co)-Total	4.81		0.50	mg/kg		31-AUG-09	R937236
Copper (Cu)-Total	8.42	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Iron (Fe)-Total	10400		5.0	mg/kg		31-AUG-09	R937236
Magnesium (Mg)-Total	3620		5.0	mg/kg		31-AUG-09	R937236
Manganese (Mn)-Total	158	RRVAP	0.20	mg/kg		31-AUG-09	R937236
Phosphorus (P)-Total	544		20	mg/kg		31-AUG-09	R937236
Potassium (K)-Total	1650	RRVAP	100	mg/kg		31-AUG-09	R937236
Sodium (Na)-Total	<100		100	mg/kg		31-AUG-09	R937236
Strontium (Sr)-Total	39.2		0.30	mg/kg		31-AUG-09	R937236
Titanium (Ti)-Total	43.5		0.50	mg/kg		31-AUG-09	R937236
Zinc (Zn)-Total	35.3	RRVAP	0.50	mg/kg		31-AUG-09	R937236
Total N and Minerals							
Total ICP scan -Nutrients							
Boron (B)-Total	12.4		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Calcium (Ca)-Total	9450		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Copper (Cu)-Total	51.1		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Iron (Fe)-Total	8400		10	mg/kg	24-AUG-09	24-AUG-09	R918323
Potassium (K)-Total	950		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Magnesium (Mg)-Total	2260		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sodium (Na)-Total	37.7		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Phosphorus (P)-Total	610		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Sulfur (S)-Total	2330		100	mg/kg	24-AUG-09	24-AUG-09	R918323
Zinc (Zn)-Total	43.4		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Manganese (Mn)-Total	151		1.0	mg/kg	24-AUG-09	24-AUG-09	R918323
Total Nitrogen by Combustion							
Total Nitrogen by Leco	1.10		0.020	%	24-AUG-09	24-AUG-09	R918924

* Refer to Referenced Information for Qualifiers (if any) and Methodology.

Reference Information

QC Samples with Qualifiers & Comments:

QC Type Description	Parameter	Qualifier	Applies to Sample Number(s)
Internal Reference Material	Calcium (Ca)	DLA	L806210-1, -10, -4, -7
Internal Reference Material	Magnesium (Mg)	DLA	L806210-1, -10, -4, -7
Internal Reference Material	Potassium (K)	DLA	L806210-1, -10, -4, -7
Internal Reference Material	Sodium (Na)	DLA	L806210-1, -10, -4, -7
Matrix Spike	Aluminum (Al)-Total	E	L806210-11, -12, -2, -3, -5, -6, -8, -9
Matrix Spike	Iron (Fe)-Total	E	L806210-11, -12, -2, -3, -5, -6, -8, -9
Duplicate	Boron (B)-Total	G	L806210-11, -12, -2, -3, -5, -6, -8, -9
Comments: sample is organic material removed from soil and therefore atypical smf 11-sep-09			
Duplicate	Calcium (Ca)-Total	G	L806210-11, -12, -2, -3, -5, -6, -8, -9
Comments: sample is organic material removed from soil and therefore atypical smf 11-sep-09			
Duplicate	Copper (Cu)-Total	G	L806210-11, -12, -2, -3, -5, -6, -8, -9
Comments: sample is organic material removed from soil and therefore atypical smf 11-sep-09			
Duplicate	Sodium (Na)-Total	G	L806210-11, -12, -2, -3, -5, -6, -8, -9
Comments: sample is organic material removed from soil and therefore atypical smf 11-sep-09			
Duplicate	Sulfur (S)-Total	G	L806210-11, -12, -2, -3, -5, -6, -8, -9
Comments: sample is organic material removed from soil and therefore atypical smf 11-sep-09			
Duplicate	Zinc (Zn)-Total	G	L806210-11, -12, -2, -3, -5, -6, -8, -9
Comments: sample is organic material removed from soil and therefore atypical smf 11-sep-09			

Sample Parameter Qualifier Key:

Qualifier	Description
DLA	Detection Limit Adjusted For required dilution
E	Matrix Spike recovery outside ALS DQO due to analyte background in sample.
G	QC result did not meet ALS DQO. Refer to narrative comments for further information.
RRVAP	Reported Result Verified by Alternate Process
SAR:DL	SAR cannot be calculated due to undetectable Na. Detection Limit represents the maximum possible value.

Test Method References:

ALS Test Code	Matrix	Test Description	Method Reference**
EC-1:2-SK	Soil	EC 1:2 Soil to Water Extraction	CSSS (1993) 16.2.2, 18.3.1

1 part dry soil and 2 parts de-ionized water (by volume) is mixed. The slurry is allowed to stand with occasional stirring for 30 - 60 minutes. After equilibration, an extract is obtained by gravity filtration. Conductivity of the extract is measured by a conductivity meter.

Reference:
Carter, Martin R., Soil Sampling and Methods of Analysis, Can Soc. Soil Sci. method 16.2.2 and 18.3.1

ALS Test Code	Matrix	Test Description	Method Reference**
ETL-N-TOTORG-CALC-SK	Soil	Nitrogen, Total Organic	APHA 4500 Norg-Calculated as TKN - NH3-N
MET-DRY-ICP-ED	Tissue	Metals in Tissue by ICPOES	EPA 200.3/200.7-ICPOES
MET-DRY-MS-ED	Tissue	Metals in Tissue by ICPMS	EPA 200.3/200.8-ICPMS
METAL-ED	Soil	Metals	SW 846 - 3051/6010-ICP-OES
N-TOT-LECO-SK	Tissue	Total Nitrogen by Combustion	SSSA (1996) P.9773-974
N-TOTKJ-SK	Soil	Total Kjeldahl Nitrogen (Organic N)	FORESTRY CANADA (1991) P. 57-59

Organic Nitrogen in soil is converted to ammonia nitrogen using sulfuric acid with CuSO4 and K2SO4 as catalysts. The ammonia is determined by distillation into boric acid and titration with standard acid.

Reference:
Y.P. Kalra, and D.G. Maynard, 1991. Methods Manual For Forest Soil and Plant Analysis, Northwest Region. Forestry Canada p. 57-59

ALS Test Code	Matrix	Test Description	Method Reference**
NH4-AVAIL-SK	Soil	Available Ammonium-N	CSSS (1993) 4.2, 4.4

Ammonium (NH4-N) is extracted from the soil using 2 N KCl. Ammonium in the extract is mixed with hypochlorite to form indophenol blue and determined colorimetrically by auto analysis at 660 nm.

Reference:
Carter, Martin R., Soil Sampling and Methods of Analysis, Can Soc. Soil Sci. methodS 4.2 and 4.4

Reference Information

NO3-AVAIL-SK	Soil	Available Nitrate-N	CSSS (1993) 4.3

Available Nitrate and Nitrite are extracted from the soil using a dilute calcium chloride solution.
Nitrate is quantitatively reduced to nitrite by passage of the sample through a copperized cadmium column. The nitrite (reduced nitrate plus original nitrite) is then determined by diazotizing with sulfanilamide followed by coupling with N-(1-naphthyl) ethylenediamine dihydrochloride. The resulting water soluble dye has a magenta color which is measured at colorimetrically at 520nm.

Reference:
Carter, Martin. Soil Sampling and Methods of Analysis. Can. Soc. Soil Sci.(1993) method 4.3

NUTR-COMP-SK	Tissue	Total ICP scan -Nutrients	Comm Soil Sci 16:943/APHA 3120B
PH-SK	Soil	pH 1:2 H20 Extract	CSSS 16.2.2 - PH OF 1:2 WATER EXTRACT

1 part dry soil and 2 parts de-ionized water (by volume) is mixed. The slurry is allowed to stand with occasional stirring for 30 - 60 minutes. pH of the soil slurry is then measured using a pH meter.

Reference:
Carter, Martin R., Soil Sampling and Methods of Analysis, Can Soc. Soil Sci. method 16.2.2

PO4/K-AVAIL-SK	Soil	Available Phosphate & Potassium	Comm. Soil Sci. Plant Anal, 25 (5&6)
PSA-1-SK	Soil	Particle Size Analysis: Hydrometer	CSSS (1993) P.508-509

The hydrometer method is based on Stokes' Law which relates the radius of soil particles to the velocity of their sedimentation.
Air-dried soil is wetted with a dispersing agent and then mixed with water in a sedimentation cylinder. The soil is allowed to settle and particle density readings(g/L) are taken after 40 seconds and 6 hours. These readings correspond to silt + clay and clay content respectively. Sand content is calculated by difference.

Reference:
Carter, M.R., 1993. Soil sampling and methods of analysis. Can. Soc. Soil Sci. Ottawa Ont. 508-509

Kalra, Y.P., Maynard, D.G. 1991. Methods manual for forest soil and plant analysis. Forestry Canada. p. 42-45.

SAR-CALC-SK	Soil	SAR and Cations in saturated soil	CSSS 18.4-Calculation
SAT-PCNT-SK	Soil	Saturated Paste	CSSS (1993) 18.2.2
SO4-AVAIL-SK	Soil	Available Sulfate-S	NCR-13 (1998) p. 35-39

The soil is extracted with a weak calcium chloride solution. The calium chloride serves to reduce the extraction of organic materials and increases flocculation of the soil in the extract. Total S in the extract is then determined by ICP-AES, which is considered to be equivalent to the plant available S for mineral soils from the prairies.

Reference:
Recommended Methods of Soil Analysis for Canadian Praire Agricultural Soils.Alberta Agriculture(1988), p. 28

** ALS test methods may incorporate modifications from specified reference methods to improve performance.

The last two letters of the above test code(s) indicate the laboratory that performed analytical analysis for that test. Refer to the list below:

Laboratory Definition Code	Laboratory Location
ED	ALS LABORATORY GROUP - EDMONTON, ALBERTA, CANADA
SK	ALS LABORATORY GROUP - SASKATOON, SASKATCHEWAN, CANADA

Chain of Custody Numbers:

Reference Information

GLOSSARY OF REPORT TERMS

Surrogates are compounds that are similar in behaviour to target analyte(s), but that do not normally occur in environmental samples. For applicable tests, surrogates are added to samples prior to analysis as a check on recovery. In reports that display the D.L. column, laboratory objectives for surrogates are listed there.
mg/kg - milligrams per kilogram based on dry weight of sample
mk/kg wwt - milligrams per kilogram based on wet weight of sample
mg/kg lwt - milligrams per kilogram based on lipid-adjusted weight
mg/L - unit of concentration based on volume, parts per million.
< - Less than.
D.L. - The reporting limit.
N/A - Result not available. Refer to qualifier code and definition for explanation.

Test results reported relate only to the samples as received by the laboratory.
UNLESS OTHERWISE STATED, ALL SAMPLES WERE RECEIVED IN ACCEPTABLE CONDITION.
Analytical results in unsigned test reports with the DRAFT watermark are subject to change, pending final QC review.

Appendix F
Statistical Analysis of Carroll and Bliss (1982) Data

Pharo and Vitt (2000) investigated the relationship between reindeer lichen and canopy cover in their study of local variation in lichen and bryophyte cover in the eastern slopes region of Alberta. They found that reindeer lichen cover was statistically higher on the most open 10 percent of jack pine-lichen sites compared to the densest 10 percent of jack pine-lichen sites using ANOVA (R^2=0.30, p <0.001); however, they also found that the open sites were significantly older (p<0.001), and that stand age (R^2 = 0.30, p = <0.001) was the second-strongest explanatory factor for the PCA ordination differences between the open and dense sites after elevation. The most open 10 percent of sites had a mean canopy cover of 45.3 to 52.3 percent, while the densest 10 percent of sites had a mean canopy cover of 72.8 to 87 percent. The mean lichen cover on the open sites was 29.2 percent, and on the densest site was 16.4 percent. Pharo and Vitt (2000) conclude that there is a real, though weak, correlation between canopy cover and stand age on jack pine-lichen sites.

Carroll and Bliss (1982) also collected data on reindeer lichen cover, canopy cover, and stand age in jack pine-lichen forests in northeast Alberta and Saskatchewan. The complete plot data were included in their report, so I used their data to better tease out the relationship between reindeer lichen cover, canopy cover, and stand age.

Cladonia mitis typically becomes a dominant component of the ground cover of jack pine-lichen forests between 30 and 50 years following fire; in areas that escape burning for more than 80 years, *C. mitis* may be replaced by *Cladonia stellaris* (Ahti 1959, 1977). Carroll and Bliss (1982) found that the mean fire return interval in their study area was 40.4 years. Thus, the vegetation community in a jack pine-lichen forest over 50 years old can be considered a mature forest.

Canopy cover on the mature sites ranged from 14 to 54 percent, and *C. mitis* cover ranged from 15 to 70 percent (mean 40 percent). No *C. mitis* cover was present in stands less than 7 years old, but in stands between 14 and 49 years of age, *C. mitis* cover ranged from 4.2 to 53.5 percent (mean 34 percent), while canopy cover ranged from 10 to 74 percent.

Statistical analysis of data was performed using linear regression modeling in R. Canopy cover, stand age, and the interaction between canopy cover and site age were examined as explanatory variables for *Cladonia mitis* cover in the study area forests, and canopy cover was found to have a statistically significant effect ($p = 0.00$) on the model ($R^2 = 0.4068$). When the data were divided into seral stands (less than 50 years old) and mature stands (71 to 181 years old), the correlation between *C. mitis* cover and canopy cover was only slight in mature forests ($R^2 = 0.1680$) and not statistically significant ($p = 0.091$), while *Cladonia mitis* cover was significantly positively correlated with canopy cover ($R^2 = 0.5993$, $p = 0.01$) in stands less than 50 years old (see Figure 1).

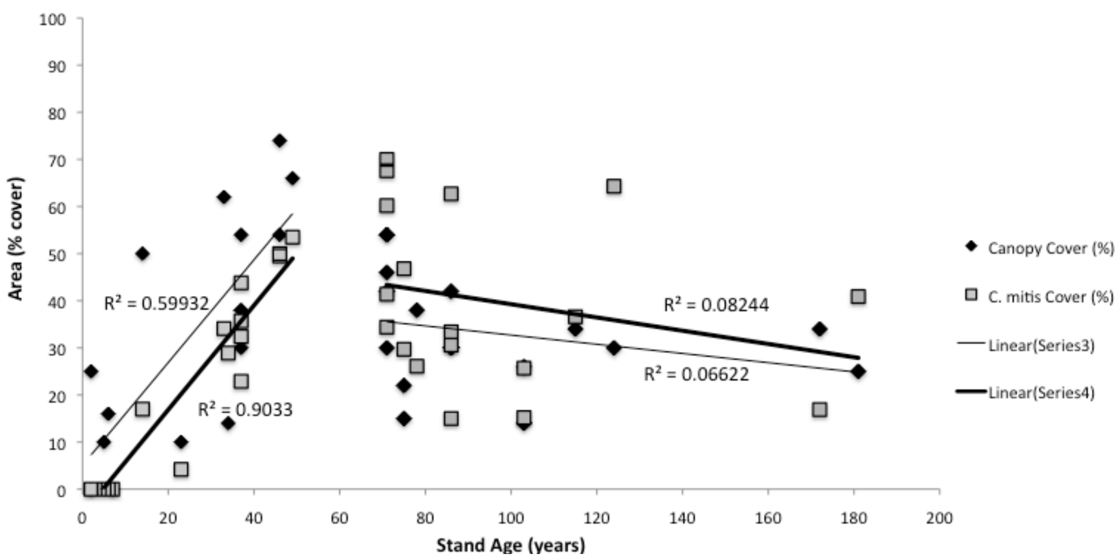

Figure 1 Relationship between canopy cover, Cladonia mitis cover, and site age

C. mitis cover was also very strongly and significantly positively correlated with site age on seral sites ($R^2 = 0.9033$, $p = 0.00$). On mature sites, there was no correlation between *C. mitis* cover and site age ($R^2 = 0.0824$), or canopy cover and site age ($R^2 = 0.0663$).

Thus, it appears that stand age is the strongest explanatory factor for *C. mitis* cover in jack pine-lichen forests, and that any correlation between canopy cover and *C. mitis* cover is due to the effect of stand age on canopy cover.

Appendix G
Representative Plot Photos of Cryptogam Diversity Assessment Community Types

Dyke 5

Site 40N wolf willow

Site 40N Aspen – grass understory

Site 40N Aspen - woody understory

Site 40N Spruce

Site 40S Dogwood

Site 40S Aspen-grass understory

Site 40S Aspen-woody understory

Site 40SW willow

Site 40SW Spruce-dogwood missing

Site 40W Aspen – woody understory

Site 40W Spruce - aspen