THE STRESS GRADIENT HYPOTHESIS: PLANT FACILITATION AT THE FOREST-TUNDRA TRANSITION (MEALY MOUNTAINS, LABRADOR, CANADA)

BRITTANY CRANSTON

THE STRESS GRADIENT HYPOTHESIS:

PLANT FACILITATION AT THE FOREST-TUNDRA TRANSITION

(MEALY MOUNTAINS, LABRADOR, CANADA)

by:

Brittany Cranston

A thesis submitted to the
School of Graduate Studies
in partial fulfilment of the
requirements for the degree of
Master of Science

Department of Biology
Memorial University of Newfoundland

May 2009

St John's Newfoundland and Labrador

Abstract

Facilitative interactions (Stress Gradient Hypothesis) at the Forest-Tundra (F-T) Transition zone around the globe have yet to be evaluated. As climate warms, the balance between facilitation and competition shifts, allowing species to expand their ranges. This study investigates the role of nurse shrubs (*Betula glandulosa* [dwarf birch], and *Vaccinium uliginosum* [bilberry]) in facilitating recruitment of *Picea mariana* and *P. glauca* (black and white spruce) in the boreal F-T Transition zone. Treatments were designed to disentangle the roles of nurse height, shade, and nurse canopy removal by herbivores in determining recruitment success. Seeds germinated at temperatures 5°C below the minimum required for germination in the open and at significantly greater levels (20% ± 6.12 of *P. mariana* and 12% ± 2.8 of *P. Glauca*) compared with the non-shrub reference site (0.5%) and previous studies. Treatments generally had negligible effects on growth, but nurse litter increased levels of soil calcium, suggesting a weak net positive association. If viable seed is available, shrubs will facilitate recruitment. The balance between competition and facilitation is highly site dependent, and cannot be generalized over larger spatial scales. Understanding how species interactions are affected by regional climate change is critical for predicting range expansion of boreal trees.

Keywords: Stress Gradient Hypothesis, nurse effect, facilitation, *Picea mariana, P. glauca,* recruitment, Forest-Tundra Transition zone, dwarf shrubs, Labrador

Acknowledgements

I would first like to extend a huge thank you to my supervisor Luise Hermanutz for her invaluable assistance in every aspect of this project from initial planning, to field work, to analysis and writing. I would also like to thank the members of my committee, Paul Marino and Keith Lewis, not only for their guidance and support but also for all the laughs in the field. Thank you to John Jacobs for helping me with all things climate, and to Liz Sutton for so kindly sharing her equipment. Thank you to all colleagues who provided essential moral support in the field and lab, and particularly Julia Wheeler for so kindly sharing her data. I would have never made it through the 6AM slug trappings, the blackflies, the stouts, and the sub-zero swims without you all! Thanks to the Labrador Tree Nursery which not only provided me with seeds, but also a place to grow the seedlings.

Funding and logistical support was provided by IPY-PPS Arctic, IPY-CiCAT, Parks Canada, and the Northern Scientific Training Program (NSTP).

Finally, my most sincere thanks to the Innu Nation in whose traditional territory my research, and that of LHRG, took place. Thank you for providing us with such a beautiful and fascinating landscape in which to work.

Table of Contents

List of Tables

List of Figures

List of Plates

Glossary of Short Forms

B- *Betula* treatment

B_H- Herbivory treatment

B_R- Cleared treatment

B_S- Shaded treatment

B_T- Trimmed treatment

CiCAT- Climate influences on Canadian Arctic Tundra

df- degrees of freedom

F-T Transition- Forest-Tundra Transition

GDD -Growing Degree Days

GLM- General Linear Model

IPCC- Intergovernmental Panel on Climate Change

LHRG- Labrador Highlands Research Group

NSTP- Northern Scientific Training Program

PAR- Photosynthetic Active Radiation

PCA- Principle Components Analysis

PPS Arctic- Present day processes, Past changes, and Spatiotemporal variability of biotic, abiotic and socio-environmental conditions and resource components along and across the Arctic delimitation zone; an international IPY project lead by Dr. Karen Harper, Dalhousie University

PRS- Plant Root Simulator^{TM}

SGH- Stress Gradient Hypothesis

V- *Vaccinium* nurse treatment

Glossary of terms

Failure to Thrive: Seedling mortality due to natural causes (non-herbivory deaths) such as desiccation, drowning, failure to produce adequate rooting system, etc.

Forest-Tundra Transition Zone: Commonly referred to as 'treeline', it is the area where open canopy forest transitions to arctic-alpine shrubland

Growing Degree Days (GDD): a measure of the heat accumulated during a single growing season for a base temperature of 5°C

Positive interactions: All non-consumer interactions among two or more species in which at least one of the species involved is positively affected

Pseudoreplication: The use of inferential statistics to test for treatment effects with data from experiments where either treatments are not replicated (though samples may be) or replicates are not statistically independent (Hurlbert 1984)

List of Appendices

1.0 Introduction and Overview

1.1 Plant-Plant interactions: The balance between competition and facilitation

Facilitation, the positive effects of a plant on the growth and establishment of other plants, has long been part of successional theory, but only in the last few decades has the importance of facilitation and its abiotic drivers been recognized as an integral force structuring plant communities (Holmgren et al. 1997). The recoupling of facilitation and environmental factors has shed new light on patterns of species coexistence, particularly in harsh environments (Michalet 2006)

The relative importance of facilitation in structuring plant communities is thought to differ between environments having high and low net primary productivity. Competition structures plant communities in environments where net primary productivity is high (Grime 1979); whereas in habitats with low productivity, such as deserts, alpine or arctic environments, the importance of plant-plant interactions is unclear (Klanderud and Totland 2005). Three competing hypotheses have been suggested: firstly, interactions between plant species may be negative because individuals are competing for limiting resources (Chapin and Shaver 1985). Secondly, positive interactions between plant species may be most important because neighbouring vegetation ameliorates harsh environmental conditions improving survival, growth, and reproduction of individual plants (Bertness and Callaway 1994). Thirdly, both positive and negative plant-plant interactions may have negligible effects on community structure relative to direct impacts from abiotic conditions (Grime 1977). This thesis will focus on the second hypothesis, the *Stress Gradient Hypothesis*, and its importance in structuring communities at the point of transition between boreal forest and alpine tundra in the Mealy Mountains, Labrador.

1.2 The Stress Gradient Hypothesis

Bertness and Callaway (1994) proposed in the Stress Gradient Hypothesis (SGH) that the importance of positive interactions to the organization of plant communities increases with stress as competition decreases. The SGH suggests that in environments having high levels of biotic and/or abiotic stress, positive spatial associations, often between seedlings (beneficiaries) and sheltering adults (nurses) of another species are common, pervasive, and even predictable (Bertness and Callaway 1994; Callaway and Walker 1997). This facilitation of young seedlings by an adult neighbour is known as the "nurse effect". The SGH and associated nurse effects have been tested in environments including desert (McAuliffe 1984; Franco and Nobel 1988,1989; Flores-Martinez et al. 1995; Tewksbury and Lloyd 2001), Mediterranean (Smit et al. 2007; Gomez-Aparicio et al. 2004; Castro et al. 2002), salt marsh (Bertness 1984; Bertness and Hacker 1994; Bertness and Yeh 1994), tundra (Carlsson and Callaghan 1991), and alpine (Klanderud 2005; Choler et al. 2001). As a result, multiple nurse mechanisms have been revealed; nurse plants, particularly shrub species, alter and ameliorate the microenvironment for their beneficiaries in harsh environments by providing shelter from desiccating and damaging winds (Cavieres et al. 2002; Smith et al. 1983; Carlsson and Callaghan 1991), increased accumulation of snow (Akhalkatsi et al. 2006; Carlsson and Callaghan 1991), ameliorating physical and chemical soil properties (Akhalkatsi et al. 2006; Castro et al. 2002), protection from solar irradiance and extreme temperatures (Akhalkatsi et al. 2006; Smith et al. 1983; Kudo and Suzuki 2003; Castro et al. 2002; Sturm et al. 2001; Carlsson and Callaghan 1991), and protection from herbivory (McAuliffe 1984; Gomez et al. 2001). These nurse mechanisms can be further categorized as protection from environmental harshness, amelioration of soil structure and chemistry, and protection from herbivory and seed predation or any combination thereof. In this way, the SGH recouples the roles of abiotic and biotic effects in structuring plant communities (Bertness and Callaway 1994).

Protection from Environmental Harshness

Nurse plants can promote the establishment and growth of their beneficiaries by protecting them from damage from harsh winds (Squeo 1991; Carlsson and Callaghan 1991; Arroyo et al. 2003; Cavieres et al. 2002), extreme temperatures (Hastwell and Facelli 2003; Franco and Nobel 1989), and desiccation (Castro et al. 2002). Beneficiaries can be protected from harsh winds either directly, by growing underneath a nurse plant (Cavieres et al. 2002), or indirectly, through nurse snow-trapping (Baumeister and Callaway 2006; Carlsson and Callaghan 1991; Germino et al. 2002). In arid environments, where moisture is limiting, high winds can cause soil erosion, surface drying, suffocating dust clouds, denuded soil surfaces, and excess heat and water loss (Squeo 1991; Carlsson and Callaghan 1991; Arroyo et al. 2003; Cavieres et al. 2002). For example, nurse cushion plants growing at treeline in the Patagonian Andes of Chile reduce the cooling and desiccating effects of wind and facilitate the recruitment of other species (Cavieres et al. 2002). The proportion of species growing inside cushions was higher than those growing outside, and this effect increased with elevation. Field manipulations, of nurse shrubs (*Salix* [willow]) have also shown increased recruitment and overwinter survival of *Chamerion angustifolium* (fireweed) transplants in the alpine zone of the Rocky Mountains of Colorado. Extremely low survival in exposed sites lacking *Salix* nurse shrubs confirms the role of *Salix* shrubs in facilitating survival of *C. angustifolium* at the upper margin of its current range (Dona and Galen 2007).

Shrubs and trees can also trap snow (Walsh et al. 1997), which can decrease plant stress (Baumeister and Callaway 2006; Carlsson and Callaghan 1991; Germino et al. 2002) in Alpine, Arctic, and sub-Antarctic communities. By trapping snow, nurses insulate beneficiaries as well as protect them from mechanical damage by blowing snow and ice (Baumeister and Callaway 2006; Carlsson and Callaghan 1991; Germino et al. 2002). Callaway (1998) found that snow accumulation by mature *P. albicaulis* (whitebark pine) nurse plants decreased stress and facilitated growth of *Abies lasiocarpa* (subalpine fir) seedlings at high elevations along an exposed ridge near the treeline in the Rocky

Mountains. Strong facilitative effects were associated with mature trees that trap snow. Snow trapping by nurse shrubs has also been simulated using plywood shelters (Carlsson and Callaghan 1991) in the alpine-subarctic tundra of Swedish Lapland. Results suggest that snow accumulation provides above ground shelter, increasing height and number of shoots in beneficiaries (Carlsson and Callaghan 1991).

Shade provided by a nurse plant can both facilitate (in extreme temperatures through increased soil moisture, trapping and retaining heat in cool climates) and inhibit (when light is limiting by reducing incoming PAR and soil temperatures) beneficiaries beneath their canopies. Balances of net facilitative and inhibitory effects on photosynthetic ability and phenotypic plasticity are complex (Hastwell and Facelli 2003). For example, the Relative Growth Rate (RGR) and survival of the seedlings of an Australian perennial shrub *Enchylaena tomentosa* (ruby saltbush) have been positively associated with shading (Hastwell and Facelli 2003). Shading can also reduce daily variations in soil surface temperatures. This facilitative effect is particularly pronounced in environments where plants may become heat stressed. For example, seedlings of the cactus *Carnegiea gigantea* (saguaro) are only found in habitats sheltered by *Ambrosia deltoidea* (triangle-leaf bursage) or *Cercidium microphyllum* (yellow palloverde) nurse shrubs in desert communities (Franco and Nobel 1989). Maximum and minimum temperatures in exposed areas reached 70.1 and 17.5°C respectively where as under a nurse plant the soil was cooled by nearly 20°C from daytime maxima (Franco and Nobel 1989).

Amelioration of Soil Structure and Chemistry

A nurse plant can modify physical and chemical characteristics of the soil beneath its canopy, particularly through increased soil moisture and availability of soil nutrients where these are limiting factors. Soil moisture under a nurse plant is generally higher and transpiration of understory species consequently lower as a result of lower Photosynthetic Active Radiation (PAR) reaching the soil and reduced soil temperatures (Castro et al. 2002). Studies have shown that nurse plants may actively increase water availability via hydraulic lift (Richards and Caldwell 1987), however this is generally limited to arid environments (Castro et al. 2002). For example, soil moisture was 3% higher under the nurse shrub *Salvia lavandulifolia* (Spanish sage) than in the open in Mediterranean mountains and as a result survival of *Pinus* (pine) seedlings increased 30% under the nurse shrub (Castro et al. 2002).

Nurse plants can also increase available soil nutrients through increased litter fall and accelerated nutrient cycling as a result of higher soil moisture levels (Castro et al. 2002). Carlsson and Callaghan (1991) discussed the possibility that increases in growth of the sedge *Carex biglowlii* could be the result of pockets of high nutrient levels found beneath nurse shrubs in an alpine-subarctic tundra community. They further tested this by comparing the growth of *C. biglowlii* when provided with additional nutrients (fertilizers) with individuals receiving only water.

The results showed that those individuals provided with additional nutrients, similar to those associated with the nurse shrubs increased not only in height, but also in the number of shoots produced in the wet arctic tundra of Sweden.

Protection from Herbivores and Seed Predators

Protection from nurse plants can facilitate the establishment and growth of beneficiary seedlings by reducing herbivory and seed predation (McAuliffe 1984; Gomez et al. 2001). Seedlings associated with nurse shrubs are often found to be less influenced by herbivory than those in the open, as dense and/or thorny shrubs can act as effective mechanical barriers to small herbivores (Padilla and Pugnaire 2006; Valiente-Banuet and Ezcurra 1991; Holmgren et al. 1997). For *Pinus sylvestris* (Scots pine) saplings grown in the high mountain region of the Mediterranean, individuals which grew directly beneath a dense canopy of shrubs experienced less herbivore damage (goats, sheep, Spanish ibex) than individuals growing in the open (Zamora et al. 2001). Padilla and Pugnaire (2006) go so far as to suggest the utilization of thorny, unpalatable shrub species in the restoration of degraded environments which are limited by high rates of grazing and herbivory.

Net Facilitation-Inhibition Balance

The mechanisms driving the nurse effect are complex and vary in intensity depending on the environmental context and location (Table 1), as well as life histories and stress factors (Maestre et al. 2009). As the nurse effect is subject to such a range of mechanisms, the balance between competition and facilitation is highly site dependent, and hence cannot be generalized over larger spatial scales. Despite net positive associations, a nurse plant can have a simultaneous weakly competitive effect on the beneficiary through inhibition of seedling emergence from leaf litter (Wang and Kemball 2005; Jaderlund et al. 1996), the reduction in PAR beyond tolerable levels (Franco and Nobel 1988, 1989); the release allelopathic substances (Callaway et al. 1991), shortened growing seasons due to later snowmelt (Schönenberger 2001; Galen and Stanton 1991), and an increase in herbivory on seedlings by providing herbivore refugia (Bestelmeyer et al. 2007; Whittington-Jones et al. 2008).

The ability of a nurse to facilitate a beneficiary is also dependent on their own reactions to stress (i.e., stress-tolerator, or competitor), as well as whether the stress is resource (light, moisture, nutrients, etc) or non-resource (wind, temperature, etc) based (Table 2). In stress-tolerant beneficiaries (such as the conifer species used in this study), and competitive nurses, facilitation is expected to be prevalent under resource-driven stress (Maestre et al. 2009). When both the nurse and beneficiary are stress-tolerators, facilitation is most important at moderate stress levels. When stress is predominantly non-resource based both competitive and stress-tolerant nurses can facilitate beneficiaries with equal efficiency, as they provide a distinctly mechanical protection to the elements which can offset resource-based competition (Maestre et al. 2009). The strength of a facilitative effect is therefore dependent not only on environmental stress, but also the nature of that stress (i.e., resource or non resource based) as well as the response to stress by both the nurse and beneficiary.

Table 1: Benefits of the nurse effect and the environments in which they occur

Desiccating/damaging winds	Dry, moisture limited
Accumulation of snow	Arctic, Antarctic, alpine
Physical and chemical soil properties	All
Solar irradiance	Hot and dry
Extreme temperatures	Extreme hot and extreme cold
Protection from herbivory	All

Table 2: Strength of facilitation by competitive and stress-tolerant nurse plants for stress tolerant beneficiaries when stress is resource and non-resource based. + = net facilitation, ++ = strong net facilitation, - = net competition. *Adapted from Maestre et al. 2009.*

Stress levels	Competitive Nurse	Stress-Tolerant Nurse
Resource Based *(light, moisture, nutrients)*		
Low	-	-
Moderate	+	+
High	++	-
Non Resource Based *(wind, temperature)*		
Low	-	-
Moderate	+	0
High	++	+

1.3 Treeline and Alpine Environments

Alpine environments exist wherever a mountain reaches higher than the regional treeline. Separated by lowlands, alpine environments can be islands of isolated, treeless areas that usually experience a more extreme climate (i.e., colder temperatures, higher winds, and late lying snow packs) than the forested regions at lower elevations (Billings 1974). The boundary separating boreal forest from the treeless alpine vegetation (the "treeline") is very rarely a true line and can be exceedingly difficult to identify in field (Elliot-Fisk 1983). In most cases the 'line' is more imaginary than real and represents a zone where forest begins to thin and is more aptly referred to as the 'boreal Forest-Tundra (F-T) Transition zone'. Within this zone, trees can become stunted, gnarled, and shrub-like in form (i.e., krummholz) and are sometimes isolated into patches of single species stands (Elliott-Fisk 1983).

The F-T Transition is highly sensitive to climatic change (ACIA 2002; Gamache and Payette 2004). With tree species at their climatic threshold, they generally respond positively to increases in temperature (Payette 2007). As a consequence of global climate change, the balance between positive and negative plant-plant interactions is not at equilibrium and may shift easily in these environments, causing a radical restructuring of plant communities at or above the current tree limit as trees expand into alpine areas (Klanderud and Totland 2005). Though it is primarily climate controlled in North America (Payette 2007); factors such as probability of establishment, mechanical and physiological tissue damage, annual carbon balance, and growth limitations may also determine altitudinal limits of boreal species (Körner 1998). This complex suite of interactions makes predicting advance or retreat of trees, and the rates at which this might occur, exceedingly difficult and predictions of changes in range limits spatially variable. Because the upslope migration of trees could eliminate many alpine plant species, a detailed understanding of these processes is essential.

The potential for and impacts of facilitative interactions (SGH) at the F-T Transition zone around the globe has yet to be evaluated. To understand the role shrubs may play in facilitating the local advance of trees into the alpine zone, the role of biotic and abiotic interactions in present day plant communities must be better understood (Henry and Molau 1997; Price and Waser 1998; Klanderud and Totland 2005; Barber et al. 2008; Chapin et al. 1995; Walker et al. 2006;).

1.4 Study Site: Mealy Mountains, Labrador, Canada

This study was conducted at the F-T Transition in the Mealy Mountains of Labrador, Canada (N 53° 36.6' W58° 49.0'), 20 km SE of Lake Melville. Within the main study area, two sites above the regional altitudinal tree limit in an eastward tending valley, were used (Figure 1.4.1).

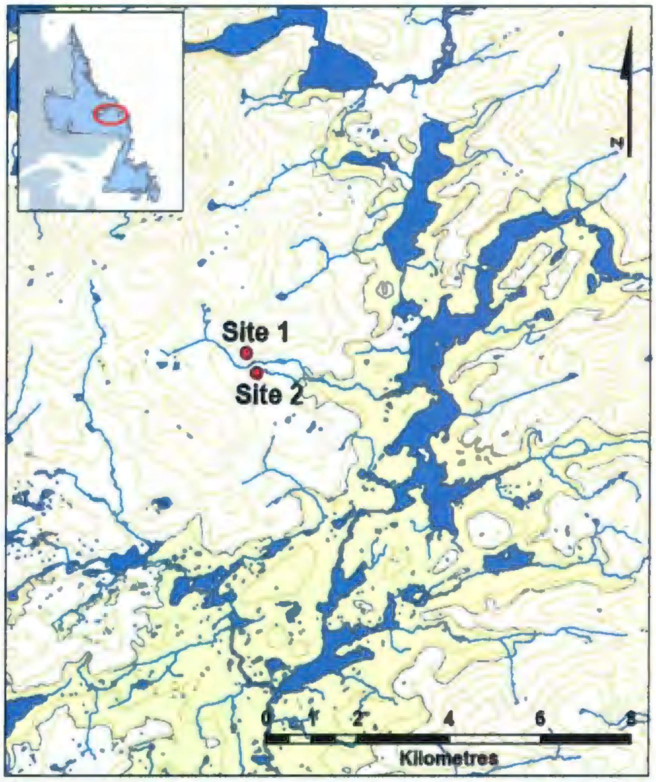

Figure 1.4.1 Location of the two study sites in the Mealy Mountains of south eastern Labrador, Canada

Climate

Climate monitoring has taken place in the Mealy Mountains since 2001. Three stations record climate data along an altitudinal gradient of 570m a.s.l. to 1000m a.s.l. Climate in the Mealy Mountains is characterized by cool summers (July average of 13.2°C at 570m a.s.l. and an annual maximum of 31°C) and cold winters (January average of -15.4°C at 570m a.s.l. and an absolute minimum of -40°C at 1000m a.s.l.). Temperatures are strongly influenced by elevation with a lapse rate of approximately -0.7°C/100m. Precipitation amounts average -3000mm annually. Permafrost is discontinuous, and soil temperatures range from -0.1°C at 570m a.s.l, to -0.7°C in alpine areas (LHRG 2007). Long-term monitoring indicates a summer warming trend from 1993 onwards with climate models predicting a warming of at least 2°C by 2090 in Labrador (LHRG 2007). Although this is moderate warming compared to the 5°C warming predicted for the rest of Canada (Environment Canada, 2002), a warming of 2°C would significantly increase the number of growing degree days (GDD) and hence primary productivity in Labrador. At the F-T Transition (600m a.s.l.) there were 616 GDD in 2007, and 683 GDD in 2008, as compared to 400 in 2007, and 97 in 2008 in the alpine zone (995m a.s.l.).

Vegetation

The growing period for vegetation in the Mealy Mountains is from mid May, when the snow melts, to mid- to late September. Vegetation communities follow an altitudinal gradient from closed canopy forest, to open canopy, to alpine tundra, the latter of which consists of only ~10% of land cover in the study site (LHRG 2007). F-T Transition zone species (*Picea mariana* [Mill.] B.S.P. [black spruce], *P. glauca* [Moench] Voss. [white spruce], *Abies balsamea* [L.] Mill. [balsam fir] and *Larix laricina* (Du Roi) K. Koch [eastern larch]) reach an altitudinal limit at ~600m a.s.l. (Figure 1.4.2). *Picea mariana* and *P. glauca* are the most abundant tree species in the F-T Transition zone in Labrador (Payette 2007; Elliott-Fisk 1983) and in the study valley (R. Jameson, pers. comm. 2009). Above the tree limit, the F-T Transition zone is dominated by shrubs to approximately 700m a.s.l, and consists predominantly of *Betula glandulosa* [Michx.] (Arctic dwarf birch), *Alnus* *crispa* [Chaix.] D. C., and *A.rugosa* [L.] *Moench.* (alder spp.), and *Salix spp.* (willow). Above the shrub zone, the alpine tundra 'crowns' the mountain tops at the highest elevations (above 1000m). The alpine zone is dominated by dwarf vegetation including *Vaccinium vitis-idaea* [L.] (partridge berry), *V. uliginosum* [L.] (tundra bilberry), *Phyllodoce caerulea* [L.] *Bab.* (mountain heather), *Rhododendron groenlandicum* [Oeder] Kron & Judd (Labrador tea), and *Empetrum nigrum* [L.] (black crowberry). All zones are characterized by a multitude of moss and lichen species.

Plate 1.4 Transition from open canopy woodland to shrub zone, Moraine Valley, Mealy Mountains, Labrador. September 2008

Consumers

Several species of vertebrate consumers may feed on conifer seedlings in the study area. Rodent herbivores appear to be the most prevalent include

Phenacomys intermedius (heather vole), *Zapus hudsonius* (meadow jumping mouse), and *Synaptomys borealis* (northern bog lemming, LHRG 2007). *Lagopus lagopus* (willow ptarmigan) and *Deroceras leave* (native slug species) also consume young *P. mariana* (Moss 1974; Moss 2004; Munier 2006) but they are not considered primary seedling herbivores in this area. Deciduous vegetation is subject to intense herbivory as well. Adult *B. glandulosa* for example is considered a food source for *P. intermedius*, and *Rangifer tarandus caribou* (Mealy Mountain Caribou) (de Groot and Wein 1997).

1.5 Study Species

Two nurse species were tested: the first, an erect shrub, *B. glandulosa* (height = 0.6m-1m), and the second, a more prostrate berry producing shrub, *V. uliginosum*(height = 0.07-0.15m). These two species will enable comparison of nurses with large and small canopies, thus affecting their ability to protect seedlings in this environment and determining their suitability as nurse shrubs. Two conifer species were used as 'beneficiary' seedlings: *P. mariana* and *P. glauca*. These species were chosen as they are the most abundant tree species in the F-T Transition zone and have slightly different temperature, moisture, and nutrient requirements for establishment and growth. They are also the species that are predicted to expand their ranges with climate change. Including both species in the study will allow for comparisons in their responses to neighbour interactions.

Picea mariana

Picea mariana is an evergreen coniferous tree commonly found in boreal and subalpine regions with a northernmost limit of 57°N (Mckay and Catling 1979; Ryan 1995). It is the most common tree species found in Labrador and is the dominant F-T Transition zone tree species in the Mealy Mountains (R. Jameson, pers. comm. 2009). It grows in wet, organic soils and, because of its shallow rooting system, is capable of growing over permafrost (Burns and Honkala 1990). Common seedbeds include *Sphagnum,* and *Pleurozium, Ptilium,* and *Hylocomium* feathermosses; however rapid growth rates of these seedbeds can impede seedling growth as the conifers are overgrown (Burns and Honkala 1990). *Picea mariana* generally require a minimum temperature of 15°C for germination, and experiences most rapid growth at 20°C (Black and Bliss 1980; Jobidon et al. 2003). It grows 0.5-2.5cm during the first season (Burns and Honkala 1990) and may eventually reach up to 18m (Ryan 1995) in height under ideal environmental conditions. *Picea mariana* is considered stress-tolerant (Baker 1949; Attree et al. 1995). Under the influence of intense, unrelenting winds, *P. mariana* may be slow growing, exhibiting a gnarled and dwarfed growth form known as krummholz, or elfinwood (Pielou 1988; Ryan 1995). *Picea mariana* produces cones but can also reproduce asexually through layering, creating tree islands and extensive krummholz patches (Burns and Honkala 1990; Bégin and Filion 1999).

Picea glauca

Picea glauca is an evergreen coniferous tree commonly found in boreal and subalpine regions with a northernmost limit of 60°N (Ryan 1995; Mckay and Catling 1979). It is a common F-T Transition zone species in northern Labrador and Québec and is the dominant species in coastal areas (Payette 2007). It can grow in a variety of soils including glacial, lacustrine, marine or alluvial. The most common seedbeds for *P. glauca* are *Pleurozium, Ptilium*, and *Hylocomium* feathermosses. Growth rates of seedlings can be limited in exposed areas where an absence of mosses results in soil moisture loss (Burns and Honkala 1990). It requires a temperature between 10-24°C for germination and experiences most rapid growth between 20-25°C (Burns and Honkala 1990). *Picea glauca* can grow 1-2cm during its first season, and sometimes reaches heights of 25m. *Picea glauca* is a stress-tolerator (Baker 1949; Attree et al. 1995), and its most common growth form in the alpine zone of the Mealy Mountains is tree islands (pers. observation 2007). *Picea glauca* produces cones but can also reproduce asexually through layering, creating tree islands (Burns and Honkala 1990; Bégin and Filion 1999).

Betula glandulosa

Betula glandulosa is a dioecious, deciduous shrub found in subarctic to subalpine areas (Chung 1989). It is variable in growth form and can be found as either completely prostrate, or 'ground hugging' in areas of high winds and low temperatures, to nearly 2m in height in more temperate zones, or sheltered areas

(Chung 1989; Hermanutz et al. 1989). It can expand its range rapidly via vegetative layering and can quickly dominate a landscape (Hermanutz et al. 1989). This tendency to rapidly disperse and dominate communities is characteristic of a competitive species (Cottrell 1996; de Groot and Wein 1999), which, as opposed to a stress-tolerator can withstand harsh conditions by out-competing neighbours for limited resources. *Betula glandulosa* is often associated with zones of high snow accumulation at its northernmost range and is often completely covered in snow through winter and early spring (Chung 1989). At the F-T Transition zone in Moraine Valley, *B. glandulosa* grows to heights of 1m with a dense canopy of leaves that reduce PAR reaching the soil by ~90%. This provides a high level of protection for beneficiaries growing beneath its canopy. An expansion of *B. glandulosa* populations into the arctic tundra is currently underway. Sturm et al (2001) have noted a North American expansion of *B. nana* of 320km^2 in the past 50 years alone. This is a closely related species to *B. glandulosa* and is prevalent in the western Arctic transition. This expansion has not only altered the structure of tundra plant communities, but has also changed the partitioning of energy in the summer, and trapping and distribution of snow in the winter (Barber et al. 2008). Range expansion is most pronounced in valley bottoms and hillsides and should it continue will have significant ramifications for ecosystem structure and function (Tape et al. 2006).

Vaccinium uliginosum

Vaccinium uliginosum is a deciduous, ericaceous, berry-producing dwarf shrub that can be found in a wide range of environments through the boreal, alpine, subarctic, and arctic zones (Trelawny 1993; Newcomb 1977). It is common in *Sphagnum* bogs, wet heathlands, open canopy coniferous forests and exposed alpine environments throughout Newfoundland and Labrador (Ryan 1995). *Vaccinium uliginosum* blooms from mid-June through July and produces edible fruits in early autumn (Ryan 1995; Newcomb 1977). In the study area it was found to grow to heights of 0.15m, significantly shorter than *B. glandulosa*, but still providing a moderate amount of protection to plants growing beneath its canopy. Because of high degrees of phenotypic plasticity, *V. uliginosum*, can easily withstand low resource levels and is considered to be a stress-tolerant species (Johnson 2005; Moola and Mallik 1998)

1.6 Thesis objective

Nurse shrubs can control the availability of resources to other organisms via physical changes in biotic or abiotic materials (Jones et al. 1997). They generally achieve this by casting shade, altering moisture regimes, reducing wind exposure, providing a mechanical barrier to herbivory, moderating extreme temperatures, as well as aerating and stabilizing the soil (Callaway and Walker 1997).

The objective of this study was to determine the principal mechanisms by which two dwarf shrubs, *B. glandulosa* and *V. uliginosum*, may facilitate, or nurse, establishment and growth of planted seeds and seedlings of *P. mariana* and *P. glauca* in the F-T Transition zone of the Mealy Mountains, Labrador. I predict, as suggested by Maestre et al. (2009) based on interactions between plant species along a rainfall gradient, that species with a competitive response to stress (*B. glandulosa)* will be more effective at protecting *Picea* seedlings from non-resource based stressors than the stress-tolerating *V. uliginosum.* Also, I predict there will be a strong temporal aspect to the nurse effect in this study. Initially nurse shrubs will have little effect on recruitment of *P. mariana* and *P. glauca*, but once seedlings grow above the seedbed into the severe boundary layer, the importance of shrub facilitation will increase.

Co-Authorship Statement

The Manuscript for this thesis was co-authored by Dr Luise Hermanutz of Memorial University of Newfoundland. I was the principle contributor for all aspects including initial proposal, experimental design, implementation of field methods, analysis of data, and the preparation of the manuscript.

References

ACIA (Arctic climate impact assessment). 2005. Arctic climate impact assessment: Scientific report, Cambridge: *University of Cambridge Press*, p. 1042

Akhalkatsii, M., Abdaladze, O., Nakhutsrishvili, G., Smith, W. K. 2006. Facilitation of seedling microsites by *Rhododendron Caucasicum* extends the *Betula Litwinowii* Alpine Treeline, Caucasus Mountains, Republic of Georgia. *Arctic, Antarctic and Alpine Research* **38**(4): 481-488

Arroyo, M., Cavieres, L., Penaloza, A., Arroyo-Kalin, M. 2003. Positive associations between the cushion plant *Azorella monantha* (Apiacaea) and alpine plant species in the Chilean Patagonian Andes. *Plant Ecology* **169**(1): 121-129

Attree, S. M., Pomeroy, M. K., Fowke L. C. 1995. Development of white spruce (*Picea glauca* (Moench.) Voss) somatic embryos during culture with abscisic acid and osmoticum, and their tolerance to drying and frozen storage. *Journal of Experimental Botany* **46**(285): 433-439

Baker, F. S. 1949 A revised tolerance table. *Journal of Forestry* **47**(3):179-181

Barber, D. B., Lukovich, J. V., Keogak, J., Baryluck, S., Fortier, L., and Henry, G. H. R. 2008. The changing climate of the arctic. *Arctic* **61** Suppl. 1: 7-26

Baumeister, D., Callaway, R. M. 2006. Facilitation by *Pinus flexilis* during succession: A hierarchy of mechanisms benefits other plant species. *Ecology* **87**(7): 1816-1830

Bégin, C., Filion, L. 1999. Black spruce (*Picea mariana*) architecture. Canadian *Journal of Botany* **77**(55): 664-672

Bertness, M. D. 1984. Ribbed mussels and *Spartina alterniflora* production in a New England salt marsh. *Ecology* **65**(66): 1794-1807

Bertness, M. D., Callaway, R. M. 1994. Positive interactions in communities. *Trends in Ecology and Evolution* **9**(5): 191-193

Bertness, M. D., Hacker, S. D. 1994. Physical stress and positive associations among marsh plants. *American Naturalist* **144**(3): 363-372

Bertness, M. D., Yeh, S. M. 1994. Cooperative and competitive interactions in the recruitment of marsh elders. *Ecology* **75**(8): 2416-2429

Bestelmeyer, B. T., Khalil, N. I., Peters, D. P. C. 2007. Does shrub invasion indirectly limit grass establishment via seedling herbivory: A test at grassland-shrubland ecotones. *Journal of Vegetation Science* **18**(3): 363-370

Billings, W. D., Bliss, L. C. 1959. An alpine snowbank environment and its effects on vegetation, plant development and productivity. *Ecology* **40**(33): 388-397

Black, A., R., Bliss, L. C. 1980. Reproductive ecology of *Picea mariana* (Mill.) B. S. P., at treeline near Inuvik, Northwest Territories, Canada [Forest Tundra]. *Ecological Monographs* **50**(3): 331-354

Burns, R. M., and Barbara H. Honkala, tech. coords. 1990. Silvics of North America: 1. Conifers; 2. Hardwoods. Agriculture Handbook 654. *U.S. Department of Agriculture*, Forest Service, Washington, DC. vol.2, 877 p.

Callaway, R. M. 1998. Competition and facilitation on elevation gradients in subalpine forests of the northern Rocky Mountains, USA. *Oikos* **82**(3): 561-573

Callaway, R. M., Nadkarmi, N. M., Mahall, B. E., 1991. Facilitation and interference of *Quercus douglasii* on understory productivity in central California. *Ecology* **72**(4): 1484-1499

Callaway, R. M., Walker, L. R. 1997. Competition and Facilitation: A synthetic approach to interactions in plant communities. *Ecology* **78**(7): 1958-1965

Carlsson, B. A., Callaghan, T. V. 1991. Positive plant interactions in tundra vegetation and the importance of shelter. *Journal of Ecology* **79**(4): 973-983

Castro, J., Zamora, R., Hódar, J. A., Gómez, J. M. 2002. Use of shrubs as nurse plants: A new technique for reforestation in Mediterranean Mountains. *Restoration Ecology* **10**(2): 297-305

Cavieres, L., Arroyo, M. T. K., Peñaloza, A., Molina-Montenegro, M., Torres, C. 2002. Nurse effect of *Bolax gummifera* cushion plants in the alpine vegetation of the Chilean Patagonian Andes. *Journal of Vegetation Science* **13**(4): 547-554

Chapin III, F. S., Shaver, G. R., 1985. Individualistic growth response of tundra plant species to environmental manipulations in the field. *Ecology* **66**(2): 564-576

Chapin III, F. S., Shaver, G. R., Giblin, A. E., Nadelhoffer, K. J., Laundre, J., A. 1995. Responses of arctic tundra to experimental and observed changes in climate. *Ecology* 76(3): 694-711

Choler, P., Michalet, R., Callaway, R. M. 2001. Facilitation and competition on gradients in alpine plant communities. *Ecology* **82**(12): 3295-3308

Chung, I. 1989. Eastern North America as seen by a botanist: pictorial. I. The arctic region. *Samhwa Printing Company*: Seoul, Korea

Cottrell, T. R. 1996. Use of plant strategy ordination, DCA and ANOVA to elucidate relationships among habitats of *Salix planifolia* and *Salix monticola*. *Journal of Vegetation Science* **7**(2): 237-246

Dona, A. J., Galen, C. 2007. Nurse effects of alpine willows (Salix) enhance overwinter survival at the upper range limit of fireweed, *Chamerion angustifolium*. *Arctic, Antarctic, and Alpine Research* **39**(1): 57-64

Elliott-Fisk, D. L. 1983. The stability of the Northern Canadian tree limit. *Annals of the Association of American Geographers* **73**(4): 560-576

Environment Canada, 2006. Climate trends and variations bulletin. Available from www.msc-smc.ec.gc.ca/ccrm/buletin/archive_e.cfm

Flores-Martinez, A., Ezcurra, E., Sanchez-Colon, S. 1994. Effect of *Neobuxbaumia tetetzo* on growth and fecundity of its nurse plant *Mimosa luisiana*. *Journal of Ecology* **82**(2): 325-330

Franco, A. C., Nobel, P. S. 1988. Interactions between seedlings of *Agave deserti* and the nurse plant *Hilaria rigida*. *Ecology* **96**(6): 1731-1740

Franco, A. C., Nobel, P. S. 1989. Effect of nurse plants on the microhabitat and growth of cacti. *Journal of Ecology* **77**(3): 870-886

Galen, C., Stanton, M. L. 1991. Consequences of emergence phenology for reproductive success in *Ranunculous adoneus* (Ranunculaceae). *American Journal of Botany* **78**(7): 978-988

Gamache, I., Payette, S. 2004. Height growth response of tree line black spruce to recent climate warming across the forest-tundra of eastern Canada. *Ecology* **92**(5): 835-845

Germino, M. J., Smith, K. S., Resor, A. C. 2002. Conifer seedling distribution and survival in an alpine-treeline ecotone. *Plant Ecology* **162**(2): 157-168

Gómez, J. M., Hódar, J. A., Zamora, R., Castro, J., Garcia, D. 2001. Ungulate damage on Scots pines in Mediterranean environments: Effects of association with shrubs. *Canadian Journal of Botany* **79**(6): 739-746

Gómez-Aparicio, L., Zamora, R., Gómez, J. M., Hódar, J. A., Castro, J., Baraza, E. 2004. Applying plant facilitation to forest restoration: A meta-analysis of the use of shrubs as nurse plants. *Ecological Applications* **14**(4): 1128-1138

Grime, J. P. 1977. Evidence for the existence of three primary strategies in plants and its relevance to ecological and evolutionary theory. *American Naturalist* **111**(982): 1169

Grime, J. P. 1979. Plant strategies and vegetation processes. *John Wiley and Sons*, Chichester, New York, NY

de Groot, W. J., Wein, R. W. 1997. *Betula nana* L. and *Betula glandulosa* Michx. *Journal of Ecology* **85**(2): 241-264

de Groot, W. J., Wein, R. W. 1999. *Betula glandulosa* Michx. response to burning and postfire growth temperature and implications of climate change. *International Journal of Wildland Fire* **9**(1): 51-64

Hastwell, G. T., Facellie, J. M. 2003. Differing effects of shade-induced facilitation on growth and survival during the establishment of a chenopod shrub. *Journal of Ecology* **91**(6): 941-950

Henry, G. H. H., Molau, U. 1997. Tundra plants and climate change: the InternationalTundra Experiment (ITEX). *Global Change Biology* **3**(1): 1-9

Hermanutz, L.A., D.J. Innes and I.M. Weis. 1989. Clonal structure of arctic dwarf birch (*Betula glandulosa*) at the northern limit of its range. *American Journal of Botany* **76**(55): 755-61.

Holmgren, M., Scheffer, M., Huston, M. 1997. The interplay of facilitation and competition in plant communities. *Ecology* **78**(7): 1966-1975

Jaderlund, A., Zackrisson, O., Nilsson, M.-C. 1996. Effects of bilberry (*Vaccinium myrtillus* L.) litter on seed germination and early seedling growth of four boreal tree species. *Journal of Chemical Ecology* **22**(5): 973-986

Jobidon, R., Roy, V., Cyr, G. 2003. Net effect of competing vegetation on selected environmental conditions and performance of four spruce seedling stock sizes after eight years in Québec (Canada). *Annals of Forest Science* **60**(7): 691-699

Johnson, G. 2005. Photosynthesis: Ecology. Encyclopaedia of Life Sciences. John Wiley and Sons, LTD:1-7

Jones, C. G., Lawton, J. H., Shachak, M. 1997. Positive and negative effects of organisms as physical ecosystem engineers. *Ecology* **78**(7): 1946-1957

Klanderud, K. 2005. Climate change effects on species interactions in an alpine plant community. *Journal of Ecology* **93**(1): 127-137

Klanderud, K., Totland, O. 2005. The relative importance of neighbours and abiotic environmental conditions for population dynamic parameters of two alpine plant species. *Journal of Ecology* **93**(3): 493-501

Körner, C., 1998. A re-assessment of high elevation treeline positions and their explanation. *Oecologia* **115**(4): 445-459

Kudo, G., Suzuki, S. 2003. Warming effects on growth, production, and vegetation structure of alpine shrubs: a five-year experiment in northern Japan. *Oecologia* **135**(2): 280-287

Labrador Highlands Research Group. 2007. Annual report of research 2006. Memorial University, Department of Geography. St John's NL, A1B 3X9. From www.mun.ca/geog/lhrg/Report_of_Research_2006.pdf

Liancourt, P., Callaway, R. M., Michalet, R. 2005. Stress tolerance and competitive-response ability determine the outcome of biotic interactions. *Ecology* **86**(6): 1611-1618

McAuliffe, J. R. 1984. Sahuaro-nurse tree associations in the Sonoran Desert: competitive effects of sahuaros. *Oecologia* **64**(3): 319-321

McKay, S. and Catling, P. 1979. Trees, shrubs, and flowers to know in Ontario. *J.M. Dent and Sons* (Canada) Ltd.: Oshawa.

Maestre, F. T., Callaway, R. M., Valladares, F., Lortie, C. J. 2009. Refining the stress- gradient hypothesis for competition and facilitation in plant communities. *Journal of Ecology* **97**(2): 199-205

Michalet, R. 2006. Is facilitation in arid environments the result of direct or complex interactions? *New Phytologist* **169**(1): 3-6

Moola, F. M., Mallik, A. U. 1998. Morphological plasticity and regeneration strategies of velvet leaf blueberry (*Vaccinium myrtilloides* Michx.) following canopy disturbance in boreal mixedwood forests. *Forest Ecology and Management* **111**(1): 35-50

Moss, M. 2004. The impact of slug herbivory on post-fire regeneration of black spruce in lichen woodland in Terra Nova National Park, NL. M.E.S. project, Environmental Science programme, Memorial University. St John's, NL

Moss, R., 1974. Winter diets, gut Lengths, and interspecific competition in Alaskan ptarmigan. *The Auk* **91**(4): 737-746Munier, A. 2006. Seedling establishment and climate change: The potential for forest displacement of alpine tundra (Mealy Mountains, Labrador). M.Sc. thesis, Dept of Biology, Memorial University, St. John's, NL

Munier, A. 2006. Seedling establishment and climate change: The potential for forest displacement of alpine tundra (Mealy Mountains, Labrador). M.Sc. thesis, Dept of Biology, Memorial University, St. John's, NL.

Newcomb, L. 1977. Newcomb's wildflower guide. *Little, Browne, and Co.*: Boston, MA

Padilla, F. M., Pugnaire, F. I. 2006. The role of nurse plants in the restoration of degraded environments. *Frontiers in Ecology and the Environment* **4**(4): 196-202

Payette, S. 2007. Contrasted dynamics of the Northern Labrador tree lines caused by climate change and migrational lag. *Ecology* **88**(3): 770-780

Pielou, E. C. 1988. The world of evergreens. *Comstock Publishing Associates*: Ithaca, NY

Price, M. V., Waser, N. M. 1998. Effects of experimental warming on plant reproductive phenology in a subalpine meadow. *Ecology* **79**(4): 1261-1271

Richards, J. H., Caldwell, M. M. 1987. Hydraulic lift: Substantial nocturnal water transport between soil layers by *Artemisia tridentata* roots. *Oecologia* **73**(4): 486-489

Ryan, A. G. 1995. Trees and shrubs of Newfoundland and Labrador. Parks division, Department of Environment and Lands, Province of Newfoundland

Schönenberger, W. 2001. Cluster afforestation for creating diverse mountain forest structures – a review. *Forest Ecology and Management* **145**(11): 121-128

Smit, C., Vanderbergh, C., den Ouden, J., Müller-Schärer, H. 2007. Nurse plants, tree saplings and grazing pressure: changes in facilitation along a biotic environmental gradient. *Oecologia* **152**(2): 265-273

Smith, W. K., Knapp, A. K., Pearson, J. A., Varman, J. H., Yavitt, J. B., Young, D. R. 1983. Influence of microclimate and growth form on plant temperatures of early spring species in a high elevation prairie. *American Midland Naturalist* **109**(2): 380-389

Smith, W. K., Germino, M. J., Hancock, T. E., Johnson, D. M. 2003. Another perspective on altitudinal limits of alpine timberlines. *Tree Physiology* **23**(1616): 1101-1112

Squeo, F. A., Rada, F., Azocar, A., Goldstein, G. 1991. Freezing tolerance and avoidance in high tropical Andean plants: Is it equally represented in species with different plant height? *Oecologia* **86**(3): 378-382

Sturm, M., Racine, C., Tape, K. 2001. Increasing shrub abundance in the Arctic. *Nature* **411**(6837): 546-547

Tape, K., Sturm, M., Racine, C. 2006. The evidence for shrub expansion in Northern Alaska and the Pan-Arctic. *Global Change Biology* **12**(4): 686-702

Tewksbury, J. J., Lloyd, J. D. 2001. Positive interactions under nurse-plants: spatial scale, stress gradients and benefactor size. *Oecologia* **127**(3): 425-434

Trelawny, J. G. 1993. Wildflowers of the Yukon and Northwestern Canada, including adjacent Alaska. *Sono Nis Press*: Victoria, BC.

Valiente-Banuet, A., Ezcurra, E. 1991. Shade as a cause of the association between the cactus *Neobuxbaumia tetetzo* and the nurse plant *Mimosa luisana* in the Tehuacan Valley, Mexico. *Journal of Ecology* **79**(4): 961-971

Walker, M. D., Wahren, C. H., Hollister, R. D., Henry, G. H. R., Alquist, L. E., Alatalo, J. M., Bret-Harte, M. S., Calef, M. P., Callaghan, T. V., Caroll, A. B., Epstein, H. E., Jonsdottir, I. S., Klein, J. A., Magnusson, B., Molau, U., Oberbauer, S. F., Rewa, S. P., Robinson, C. H., Shaver, G. R., Suding, K. N., Thomson, C. C., Tolvanen, A., Totland, O., Turner, P. L., Tweedie, C. E., Webber, P. J., Wookey, P. A. 2006. Plant community responses to experimental warming across the tundra biome. *Proceedings of the National Academy of Science* **103**(5):1342-1346

Walsh, N. E., McCabe, T. R., Welker, J. M., Parsons, A. N. 1997. Experimental manipulations of snow-depth: effects of nutrient content on caribou forage. *Global Change Biology* **1**(3): 158-164

Wang, G. G., Kemball, K. J. 2005. Balsam fir and white spruce seedling recruitment in response to understory release, seedbed type, and litter exclusion in trembling aspen stands. *Canadian Journal of Forest Research* **35**(3): 667-673

Whittington-Jones, G. M., Bernard, R. T. F., Parker, D. M. 2008. Bushclumps as refugia for small mammals in two Eastern Cape conservation areas. *African Zoology* **43**(2): 273-276

Zamora, R., Gomez, J. M., Hodar, J. A., Castro, J., Garcia, D. 2001. Effect of browsing by ungulates on sapling growth of Scots pine in a Mediterranean environment: consequences for forest regeneration. *Forest Ecology and Management* **144**(1-3): 33-42

2.0 Do Shrubs Act as Nurse Plants at the Treeline?

Abstract

Facilitative interactions (Stress Gradient Hypothesis) at the Forest-Tundra (F-T) Transition zone around the globe have yet to be evaluated. As climate warms, the balance between facilitation and competition shifts, allowing species to expand their ranges. This study investigates the role of nurse shrubs (*Betula glandulosa* [dwarf birch], and *Vaccinium uliginosum* [bilberry]) in facilitating recruitment of *Picea mariana* and *P. glauca* (black and white spruce) in the boreal F-T Transition zone. Treatments were designed to disentangle the roles of nurse height, shade, and nurse canopy removal by herbivores in determining recruitment success. Seeds germinated at temperatures 5°C below the minimum required for germination in the open and at significantly greater levels (20% ± 6.12 of *P. mariana* and 12% ± 2.8 of *P. glauca*) compared with the non-shrub reference site (0.5%) and previous studies. Treatments generally had negligible effects on growth, but nurse litter increased levels of soil calcium, suggesting a weak net positive association. If viable seed is available, shrubs will facilitate recruitment. The balance between competition and facilitation is highly site dependent, and cannot be generalized over larger spatial scales. Understanding how species interactions are affected by regional climate change is critical for predicting range expansion of boreal trees.

Keywords: Stress Gradient Hypothesis, nurse effect, facilitation, *Picea mariana, P. glauca,* recruitment, Forest-Tundra Transition zone, dwarf shrubs, Labrador

2.1 Introduction

The expansion of shrub-dominated habitat associated with climate change has the potential to alter alpine and tundra plant assemblages (Henry and Molau 1997; Price and Waser 1998; Klanderud and Totland 2005; Chapin et al. 1995; Walker et al. 2006). These alterations of the abiotic environment are driven by a strong positive feedback mechanism and can respond to changes in climate on a decadal time scale (Sturm et al. 2001; 2005). A shift to a shrub dominated landscape increases snow depth and persistence, insulating the soil which in turn increases microbial activity and thus nutrient availability, finally facilitating further expansion of shrubs (Sturm et al. 2005). This feedback often results in the local loss of shade-intolerant alpine species as well as significantly altering energy partitioning and nutrient cycling in summer, and snow accumulation and albedo in winter within a relatively short time span (Danby and Hik 2007; Barber et al. 2008; Sturm et al. 2001, 2005; Chapin et al. 1995).

Many studies have investigated shrub expansion into alpine and arctic tundra in North America, including Alaska (Sturm et al. 2001; Tape et al. 2006; Chapin et al. 2005), Northwest Territories (Lantz, 2008), Yukon (Myers-Smith 2007) and Quebec (Fortin and Pilote mountaincartobraphy.com). Since 1950, the cover of shrubs in Northern Alaska has increased at an average rate of 1.2% per decade either by infilling, increasing in size, or expansion to new areas (Chapin et al. 2005; Tape et al. 2006; Sturm et al. 2001). A series of photographs taken from 1948 to 2004 at Mount Jaques-Cartier, Quebec clearly show both expansion and infilling of shrubs in Southern Quebec at a rate of 0.28% per year (Fortin and Pilote mountaincartobraphy.com). With the predicted rise in temperatures associated with climate change, and the resultant invasion and facilitation of shrubs, a general trend of a northward and upward migration of boreal trees has been predicted (ACIA 2005; IPCC 2007).

The factors determining latitudinal and altitudinal tree limits vary globally, but the significance of plant-plant interactions is clear (Cannone et al. 2007). Here, forests transition to the shrub dominated Forest-Tundra (F-T) Transition zone, which is rapidly expanding its range as the climate warms, possibly acting as a conduit for the future migration of boreal trees (Barber et al 2008). These low, dense shrubs survive in harsh environments in part because of their ability to modify the microclimate beneath their canopies (Grace 2002). For understory plants, shrubs may facilitate survival and growth by providing shelter from desiccating and damaging winds (Carlsson and Callaghan 1991; Smith et al. 1983), protection from herbivores (Carlsson and Callaghan 1991), increased snow trapping (Carlsson and Callaghan 1991; Akhalkatsi et al. 2006), improved soil structure and nutrient content (Akhalkatsi et al. 2006; Castro et al. 2002), and protection from solar irradiance and extreme temperatures (Kudo and Suzuki 2003; Carlsson and Callaghan 1991; Akhalkatsi et al. 2006; Smith et al.1983; Castro et al. 2002). These modifications may facilitate the establishment and growth of other species, in particular trees, within the ecosystem.

The ability of a nurse to facilitate a beneficiary is also dependent on their own reactions to stress (i.e., stress-tolerator, or competitor), as well as whether the stress is resource (light, moisture, nutrients, etc) or non-resource (wind, temperature, etc) based. In stress-tolerant beneficiaries (such as the *Picea spp.* used in this study), and competitive nurses, facilitation is expected to be prevalent under resource-driven stress (Maestre et al. 2009). When both the nurse and beneficiary are stress-tolerators, facilitation is most important at moderate stress levels. However, when stress is predominantly non-resource based both competitive and stress-tolerant nurses can facilitate beneficiaries with equal efficiency, as they provide a distinctly mechanical protection to the elements which can offset resource-based competition (Maestre et al. 2009). The strength of a facilitative effect is therefore dependent not only on environmental stress, but also the nature of that stress (i.e., resource or non resource based) and the resultant response by both the nurse and beneficiary. By ignoring the role of these biotic interactions, a significant mechanism of potential advance, or retreat of boreal tree species, is overlooked. Because the nurse effect is subject to such a range of mechanisms, the balance between competition and facilitation is highly site dependent. Factors controlling the rate of forest expansion vary globally

Positive interactions, or facilitation, are important factors in structuring plant assemblages in a variety of ecosystems (Bertness and Callaway 1994; Callaway and Walker 1997). As predicted by the Stress Gradient Hypothesis [SGH], the relative importance of facilitation versus competition in structuring plant assemblages changes along a stress gradient such that facilitation becomes more important in harsher environments (Bertness and Callaway 1994). This hypothesis has not yet been tested at the boreal F-T Transition zone. The Northern Quebec and Labrador F-T Transition zone is a climatically stressed ecosystem for boreal tree species (Payette 2007). Already at their climatic thresholds, *Picea mariana* [Mill.] B.S.P. (black spruce) and *P. glauca* [Moench] Voss. (white spruce), which are the dominant trees of the F-T Transition zone, provide an opportunity to study whether shrubs in the F-T Transition zone exert a net positive or negative effect on the establishment and growth of these species in an environment where the climate is predicted to warm 2°C by 2090 (Environment Canada, 2002).

The SGH has historically been tested by searching for naturally established associations in which beneficiaries are growing beneath the canopy of a nurse. By focusing on already established associations, the effects of facilitation on beneficiaries during early life stages (i.e., germination, emergence, and early growth), a crucial stage for determining the success of seedling establishment is overlooked. This study investigates the potential role of facilitation in promoting the establishment from seeds and growth of purposefully planted seeds and seedlings of *P. mariana* and *P. glauca* in close association with shrubs in the F-T Transition zone. Although adults of both *Picea* species are present here, seedlings were planted as no naturally established *Picea* seedlings were found in the study site (Cranston 2009). An analysis of juvenile and adult conifer distribution (including *P. mariana* and *P. glauca*) in the lower F-T Transition zone of the Mealy Mountains, Labrador has revealed a positive spatial association within a 0.5m radius, suggesting that juvenile recruitment is limited by facilitation of larger adult congeners (D. DeFields, pers. comm. 2009). This lack of *Picea* seedlings in the F-T Transition zone is not limited to the Mealy Mountains. In the Yukon, it appears that *P. mariana* seedlings may require a disturbance, principally a recent burn, in order to successfully sexually reproduce at the F-T Transition zone (C. Brown, pers. comm. 2009).

Arctic-alpine shrubs have the ability to control the availability of resources to other organisms via physical changes in biotic or abiotic materials (Jones et al. 1997), making shrubs ideal candidates for investigating whether facilitation plays a role in structuring these plant communities. The objective of this study was to determine if *B. glandulosa* [Michx.] (arctic dwarf birch) and *V. uliginosum* [L.] (tundra bilberry) nurse, or facilitate recruitment of *P. mariana* and *P. glauca* in the upper F-T Transition zone of the Mealy Mountains during the crucial seed and seedling stage and, if they do, the principal mechanisms by which this occurs. To achieve this, abiotic and biotic factors that typically determine the balance between facilitation and competition were evaluated: the effect of nurse shade, height, and clearing of the nurse canopy by herbivores on the recruitment (emergence, growth and survival) of conifer seedlings. The following predictions were tested: 1) Both *B. glandulosa* and *V. uliginosum* will alter the abiotic environment (light, moisture, temperature, nutrients) beneath their canopies resulting in a net facilitative association with their beneficiaries; 2) Erect *B.*

glandulosa (~ 1m) will be a more effective nurse than the more prostrate, *V. uliginosum* (15cm) which has a less substantial canopy; 3) Recruitment will be higher in nurse treatments which allow moderate amounts of radiation to reach the light limited seedlings while still providing protection; 4) Herbivory on seedlings will be increased beneath nurse shrubs as both are palatable to small mammals and may provide shelter, attracting herbivores;

2.2 Methods

Study Area

This study was conducted in the F-T Transition zone of the Mealy Mountains, Labrador, Canada (N 53° 36.6' W58° 49.0'). Climate is characterized by cool summers (July average of 13.2°C at 570m a.s.l, annual maximum 31°C) and cold winters (January average of -15.4°C at 570m a.s.l, absolute minimum of -40°C at 1000m a.s.l) (LHRG 2007). Temperatures decline with increased elevation with a lapse rate of approximately -0.7°C/100m (LHGR 2007). Precipitation averages 2000-3000mm annually (LHRG 2007), with no summer moisture deficiency. The altitudinal limit is ~600m a.s.l. for *P. mariana, P. glauca, Abies balsamea* [L.] Mill. (balsam fir) and *Larix laricina* [Du Roi] K. Koch (eastern larch). The Shrub zone is dominated by dwarf vegetation including *B. glandulosa* and *V. uliginosum, Alnus crispa* [Chaix] D. C., *A. rugosa* [L.] Moench (alder spp.), *V. vitis-idea* [L.] (partridge berry), *Phyllodoce caerulea* [L.] Bab. (mountain heather), *Rhododendron groenlandicum* [Oeder] Kron & Judd (Labrador tea), and *Empetrum nigrum* [L.]

(black crowberry). Both zones also have a species-rich understory of mosses and lichens.

Experimental Design

To determine the principal mechanisms of facilitation in the association of nurse shrubs and seedlings of *P. mariana* and *P. glauca* in the F-T Transition zone, recruitment of *Picea spp*. in non-shrub reference plots dominated by a lichen seedbed was compared to plots dominated by nurse shrubs. The non-shrub reference site was not included in the statistical analysis due to confounding variables (location, microtopography, and slight differences in seedbed composition) but was included in graphical representations of the data for comparison. Two representative *B. glandulosa* stands were chosen to represent the range of densities found at the site. The *B. glandulosa* stand in Site 1 (~10 stems per m2 and 0.5m high) was less dense than Site 2 (~35 stems per m^2 and 1m high), which had a predominantly *Sphagnum* dominated seedbed, absent in Site 1.

A total of 36 plots were established; three treatment replicates were positioned randomly in each of two sites (Figure 2.2.1; Table 3). Because an extensive $300m^2$ search revealed no *Picea* seedlings growing naturally in the study area (Appendix I), an experimental approach was adopted in which seeds of both *P. mariana* and *P. glauca* and seedlings of *P. mariana* were planted in each $1m^2$ plot (Figure 2.2.1). Seed source for both 2007 and 2008 field seasons was from *P. mariana* and *P. glauca* from the Goose Bay (Labrador) area roughly 100 km west of the study site, and were obtained from the Department of Natural Resources Tree Nursery at Happy Valley-Goose Bay. Seedlings were reared at the nursery for two weeks prior to planting in the field.

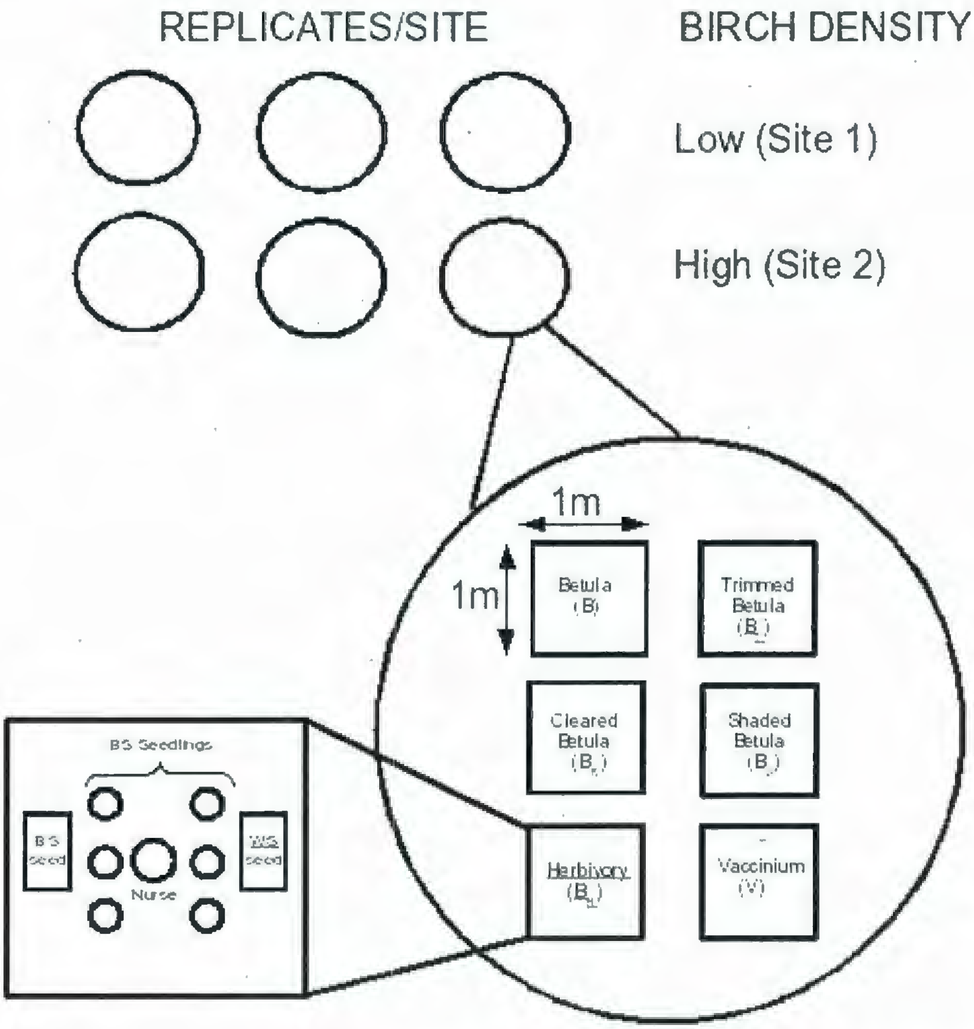

Figure 2.2.1 Experimental design depicting study plots and their allocation within sites. BS=*P. mariana*, WS=*P. glauca*. The nurse in the centre of each plot denotes the position of the stem. Nurse canopies extend to cover all seeds and seedlings within the plot. Data for the non-shrub reference site were collected adjacent to low density Site 1 and collected by Julia Wheeler (unpublished MSc. thesis)

Table 3: Summary of treatments involved in the experimental design. Treatments were designed to test nurse mechanisms including shade, herbivory, and height. PAR is measured as a percent of ambient levels.

Mechanism	Treatments	Nurse Species	PAR	Replicates	Notes
Reference	Lichen dominated, open tundra site used as a non-shrub reference in figures. Data provided by J. Wheeler (unpublished), not included in statistical analysis				
Site	Site 1	Low density *B. glandulosa* stand (~10 stems per m^2)			Sites contain half the replicates from all treatments
	Site 2	High density *B. glandulosa* stand (~35 stems per m^2)			
Shade	Unaltered canopy (**B**)	*B. glandulosa*	5-10%	6	$\mathbf{B_S}$ plots shaded using greenhouse shade cloth Height of *B. glandulosa* nurses ~1m
	Trimmed canopy ($\mathbf{B_T}$)	*B. glandulosa*	70-80%	6	
	Artificially cleared canopy	*B. glandulosa*	95-100%	6	
	artificially shaded canopy ($\mathbf{B_S}$)	*B. glandulosa*	0-5%	6	
Herbivory	Nurse cleared by herbivory ($\mathbf{B_H}$)	*B. glandulosa*	70-80%	6	$\mathbf{B_H}$ plots contain a central 'latrine' of rodent feces
Height	Unaltered canopy (V)	*V. uliginosum*	70-80%	6	Height of *V. uliginosum* *~15cm*

Abiotic Factors and Conifer Recruitment

To investigate the ability of nurse shrubs to alter the abiotic environment, soil moisture and temperature and nutrients were analysed. Soil moisture and temperature were measured on all dry, sunny days when the soil was not saturated with water between the dates of July 10th and July 16th of 2008 (n=180). Soil moisture (% saturation) was measured using a Delta T™ probe, and temperature (°C) using the Hannah™ probe. In both cases, measurements were taken approximately 8cm below the surface and direct contact with roots or rocks was avoided. During the summer of 2007 anion:cation exchange resin probes (Plant Root Simulator™ [PRS]) probes from Western Ag Innovation Inc. (Saskatoon, SK) were used to compare soil nutrients above detection levels (Total N, Ca, K, P, Fe, Zn, B, S, Pb, and Al) across select nurse treatments predicted to differ in nutrient levels. Two anion and two cation probes were inserted into the soil in each plot for *Betula* (**B**), Herbivory (**B_H**), *Vaccinium* (**V**), and Cleared (**B_R**) treatments for a sample size of n=48 pairs and left for 71 days (July 10th-September 19th, 2007, capturing a significant portion of the growing season which is an optimal time to measure soil nutrients).

On July 4th, 2007, 216 *P. mariana* seedlings were planted, 6 in each of the 36 plots. During the first growing season (2007) seedlings were monitored every three days beginning July 5th and ending July 11th for a total of 3 measurements, and once again September 18th to capture the end of the first growing season and the buds had set. Seedling recruitment success, measured as number of emerged seeds, growth (height and number of needles), and survival were recorded. During 2008, seedlings were remeasured June 23rd to determine overwintering success, July 18th, and September 13th to mark the end of the second growing season. Dead seedlings were collected throughout the second growing season, and cause of death determined. Mortalities were attributed either to herbivory (vertebrate, invertebrate), or failure to thrive (mortality due to natural causes [non-herbivory deaths]). Occasionally the presence of scat (ptarmigan or vole) or slug eggs provided evidence of the herbivores involved. Native slug species *Deroceras laeve* was evident on all plots (Appendix II; Appendix III).

In 2008, *P. mariana* and *P. glauca* seeds were planted in each plot (1080 seeds per species) to determine emergence success. Seeds were planted June 22 by being pressed into the soil and were marked with coloured toothpicks. Following the emergence of the first seeds July 13, plots were checked daily ending July 18 to determine the success of emergence. Seeds were checked again September 13th at the end of the growing season. All procedures were also completed in the non-shrub reference site by Julia Wheeler (unpublished MSc. thesis).

Data Analysis

Comparison of abiotic factors among treatments: All statistical analyses were performed using Minitab v. 15.0. Percent moisture saturation was analyzed using the General Linear Model (GLM) ANOVA with weighted means as the response variable and treatment, site, the interaction between the two, and plots nested within sites as the explanatory variables. A Box-Cox transformation was applied which corrected for a violation of the homogeneity of variance assumptions of the GLM. Means weighted per plot were added to the model to avoid pseudoreplication (Hulbert 1984; McArdle 1996). Soil temperature was analyzed using non-parametric Kruskal-Wallis test, when the error could not be normalized using a Box-Cox or natural log transformation. Tukey's HSD was applied to the soil moisture data as a Post Hoc analysis for comparisons among treatments. Principle Components Analysis (PCA) was used to compare levels of soil nutrients among treatment and to determine if there was an association to nurse shade, height, or herbivory on nurses. To test for significance of the treatment effect in the PCA, the rule set forth by Hair, Anderson, and Tatham (1987) for small sample sizes (n<100) was followed: For small sample sizes, loadings of ±0.3 are considered significant, ±0.4 are considered strongly significant, and ±0.5 are very strongly significant.

Seedling Recruitment: Emergence, growth, number of needles, and survival were analyzed using the GLM ANOVA with weighted means as the response variable and treatment, site, the interaction between the two, and plots nested within sites as the explanatory variables. A natural log transformation was applied to both *Picea* species to correct for a violation in homogeneity of variances as well as non-normal error. The height of *P. mariana* seedlings at planting varied greatly from 6mm to 20.8mm. For this reason height at planting was initially used as a covariate in the model, however, its effect on growth rate was not significant. This indicates that the size at planting did not affect subsequent measurements and so it was excluded from the analysis. A Box-Cox transformation was applied to the growth data to obtain a normal error distribution. Finally, a GLM Three-Way ANOVA was applied to determine cause of death using percent death caused by each factor (slug herbivory, vertebrate herbivory, or failure to thrive) as a response variable, with treatment, site, their interaction, and plots nested within sites as the explanatory variable. Tukey's HSD was applied as a Post Hoc analysis to the percent survival data when a significant interaction effect was uncovered to distinguish which interactions were significantly different.

2.3 Results

Data from the non-shrub reference site was not included in the statistical analysis due to potential confounding variables previously mentioned, but was included in graphical representations of the data for comparison. While this does not allow for the determination of the statistical significance of the nurse effect, it does reveal a biological trend which suggests a weak net-positive association between nurse shrubs and beneficiary seedlings.

Abiotic factors:

Soil moisture is lower in all nurse treatments than the non-shrub reference site (Figure 2.3.1). Treatment type ($F_{5,164}$= 3.83, p=0.003) but not site ($F_{1,164}$=4.51, p=0.591) significantly affects soil moisture levels (Figure 2.3.1), which supports general SGH predictions (Table 4). The *Vaccinium* (**V**) treatment had significantly higher (7.72%) soil moisture than the *Betula* (**B**) treatment in both study sites (Tukey HSD: $F_{5,174}$=3.46, p<0.005). High density Site 2 had significantly lower soil temperatures (9.58°C ± 1.7) than the low density Site 1 (13.45°C ± 1.21) (Kruskal-Wallis: H_1=116.77, p<0.001). Treatment types did not significantly affect temperature (Kruskal-Wallis: H_5=4.31, p=0.506) (Figure 2.3.1) indicating that nurse shade, height, and the removal of the nurse canopy by herbivory do not affect soil temperatures (Table 4).

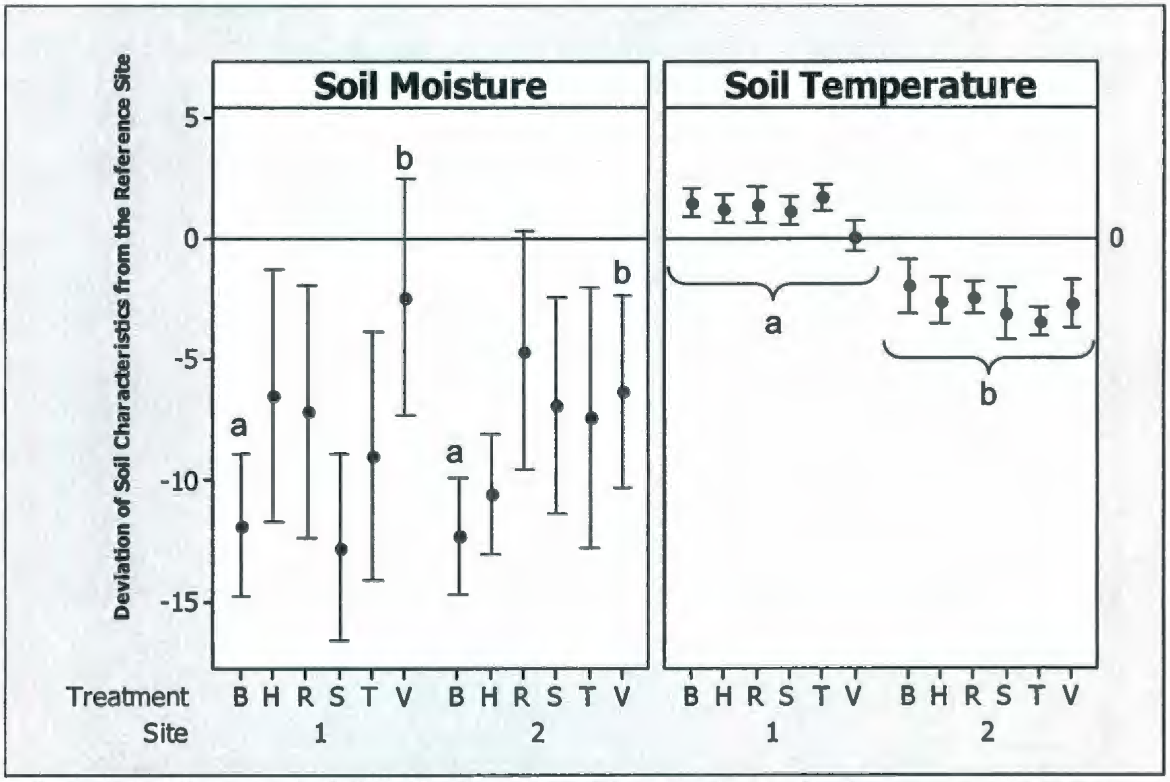

Figure 2.3.1 Average deviation of soil moisture (% saturation) and soil temperature (°C) from the non-shrub reference (zero line) for treatment and site in 2008. 95%confidence intervals are included. Treatments include *Betula* nurse (**B**), *Betula* nurses that have been cleared by rodent herbivory (**H**), artificially cleared *Betula* nurses (**R**), artificially shaded *Betula* nurses (**S**), artificially trimmed *Betula* (**T**), and *Vaccinium* nurses (**V**). Site 1 is a low density birch stand with feathermoss and lichen seedbed, and Site 2 is a high density birch stand with *Sphagnum* seedbed. The symbol 'a' denotes a significant difference from the symbol 'b' using a GLM ANOVA with weighted means.

Calcium levels in nurse treatments (450.79 µg/10cm^2 ±380.77) are consistently higher than in the non-shrub reference site (230.93 µg/10cm^2 ± 171.82) (examined *Betula* [**B**], *Betula* with Herbivory [**B$_H$**], Artificially Cleared *Betula* [**B$_R$**], and *Vaccinium* [**V**]) than in the non-shrub reference site (Figure 2.3.2). Among treatments, soil nutrients do not differ significantly: When Total N, P, and Fe are high, soil moisture is low, and when Ca, S, and soil temperature are high, *B. glandulosa* density and K are low in all treatments. The first two principal components explain 41.1% of the variation; however the absence of clear, defined clusters coded to nurse treatments in the biplot, suggests that there is no effect of treatment type on soil nutrients. (Appendix IV).

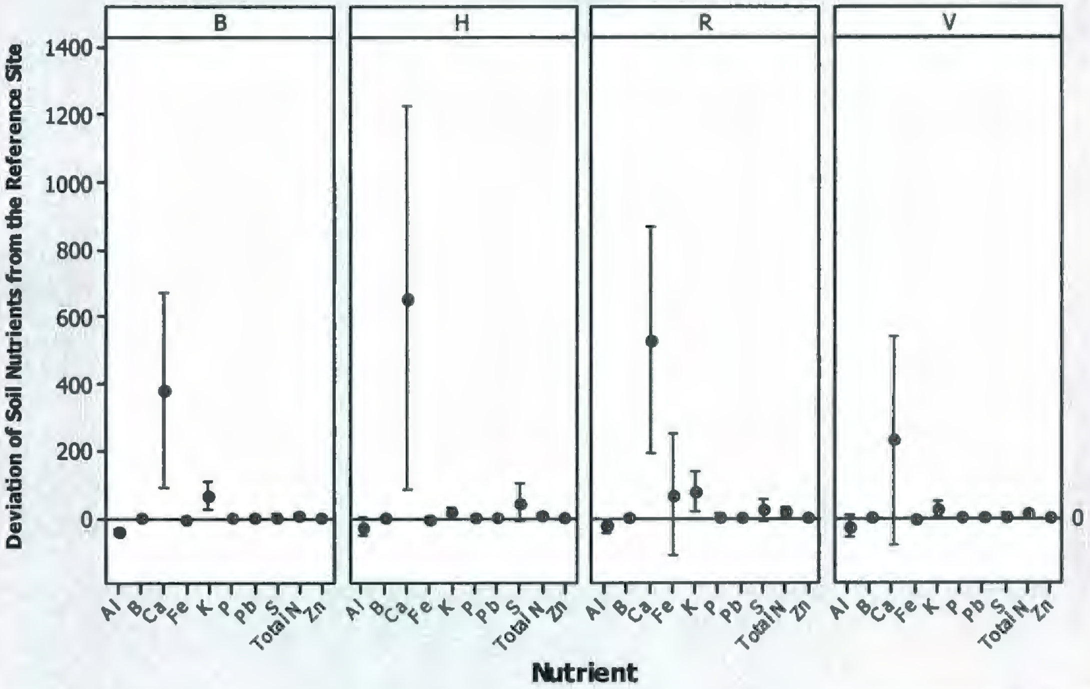

Table 2.3.2: Average deviation of soil nutrients from the non-shrub reference (zero line) in *Betula* (**B**), *Betula* Herbivory (**H**), Cleared *Betula* (**R**) and *Vaccinium* (**V**) nurse treatments with 95% confidence intervals. Soil nutrients include Aluminum (Al), Boron (B), Calcium (Ca), Iron (Fe), Potassium (K), Phosphorous (P), Lead (Pb), Sulphur (S), Total Nitrogen (Total N), and Zinc (Zn). Nutrients were measured using Plant-Root Simulator Probes during the summer of 2007.

Recruitment:

Emergence of both *P. mariana* and *P. glauca* is consistently higher in the experimental treatments than the non-shrub reference site (Figure 2.3.3). Of the 1080 seeds of each *P. mariana* and *P. glauca* seeds planted, a total of 217 *P. mariana* (20% ± 6.12) and 126 *P. glauca* (12% ± 2.8) seeds emerged compared with only 3 seeds total (0.5%) in the non-shrub reference site. Seed emergence for *P. mariana* was significantly higher in high density Site 2, which has soil temperatures 3.87°C lower than low density Site 1 ($F_{1,17}$=6.21, p=0.023), but there was no significant difference among treatments ($F_{5,17}$=0.83p=0.547). There was no significant effect of either treatment type or site on the emergence of *P. glauca* seeds (Treatment: $F_{5,20}$=1.85, p=0.148; Site: $F_{1,20}$=0.47 p=0.501).

Seedling growth in *P. mariana* was analyzed as a combination of height (Figure 2.3.4) and number of needles per individual (Figure 2.3.5). The height of *P. mariana* seedlings did not differ among treatments (2007: $F_{5,117}$=1.96, p=0.090; 2008: $F_{5,63}$=0.58, p=0.716) in either year, and heights for all treatments were similar to the non-shrub reference site for both growing seasons. Again, this is contrary to the predictions of the SGH (Table 4). During 2007, there was no effect of site on seedling height (Site: $F_{1,117}$=3.22, p=0.075), but by the second year of growth (2008), seedlings were significantly taller in low density Site 1 than high density Site 2 ($F_{1,63}$=4.65, p=0.035).

At planting, individual seedlings differed little in number of needles (4-5). However by the second growing season the number of needles per individual varied considerably (17-31) which in combination with needle removal by herbivores resulted in large confidence intervals in some instances (see Figure 2.3.5). Needle number was significantly higher in low density Site 1 than in high density Site 2 during 2007 and 2008 growing seasons (2007: $F_{1,128}$=6.15, p=0.014; 2008: $F_{1,73}$=7.74, p=0.007), but did not differ among treatment types (2007: $F_{5,128}$=0.99, p=0.429; 2008: $F_{5,73}$=1.76, p=0.133). However, by fall of 2008, seedlings beneath nurse canopies began to produce more needles than those in the non-shrub reference site.

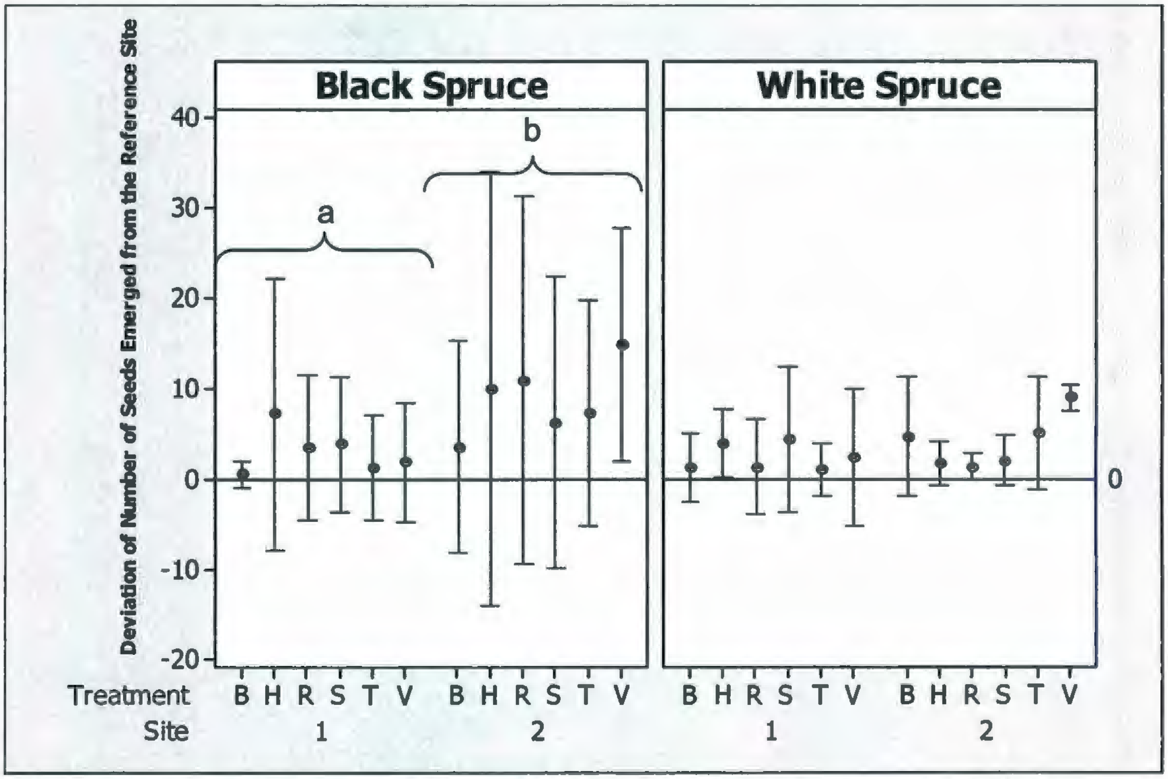

Figure 2.3.3 Deviation of the average number of *P. mariana* and *P. glauca* seeds which emerged during the first growing season (2008) from the non-shrub reference (zero line). 95% confidence intervals are included. Treatments include *Betula* nurse (**B**), *Betula* nurses that have been cleared by rodent herbivory (**H**), artificially cleared *Betula* nurses (**R**), artificially shaded *Betula* nurses (**S**), artificially trimmed *Betula* (**T**), and *Vaccinium* nurses (**V**). Site 1 is a low density birch stand with feathermoss and lichen seedbed, and Site 2 is a high density birch stand with *Sphagnum* seedbed. The symbol 'a' denotes a statistically significant difference from 'b' using a GLM ANOVA with weighted means.

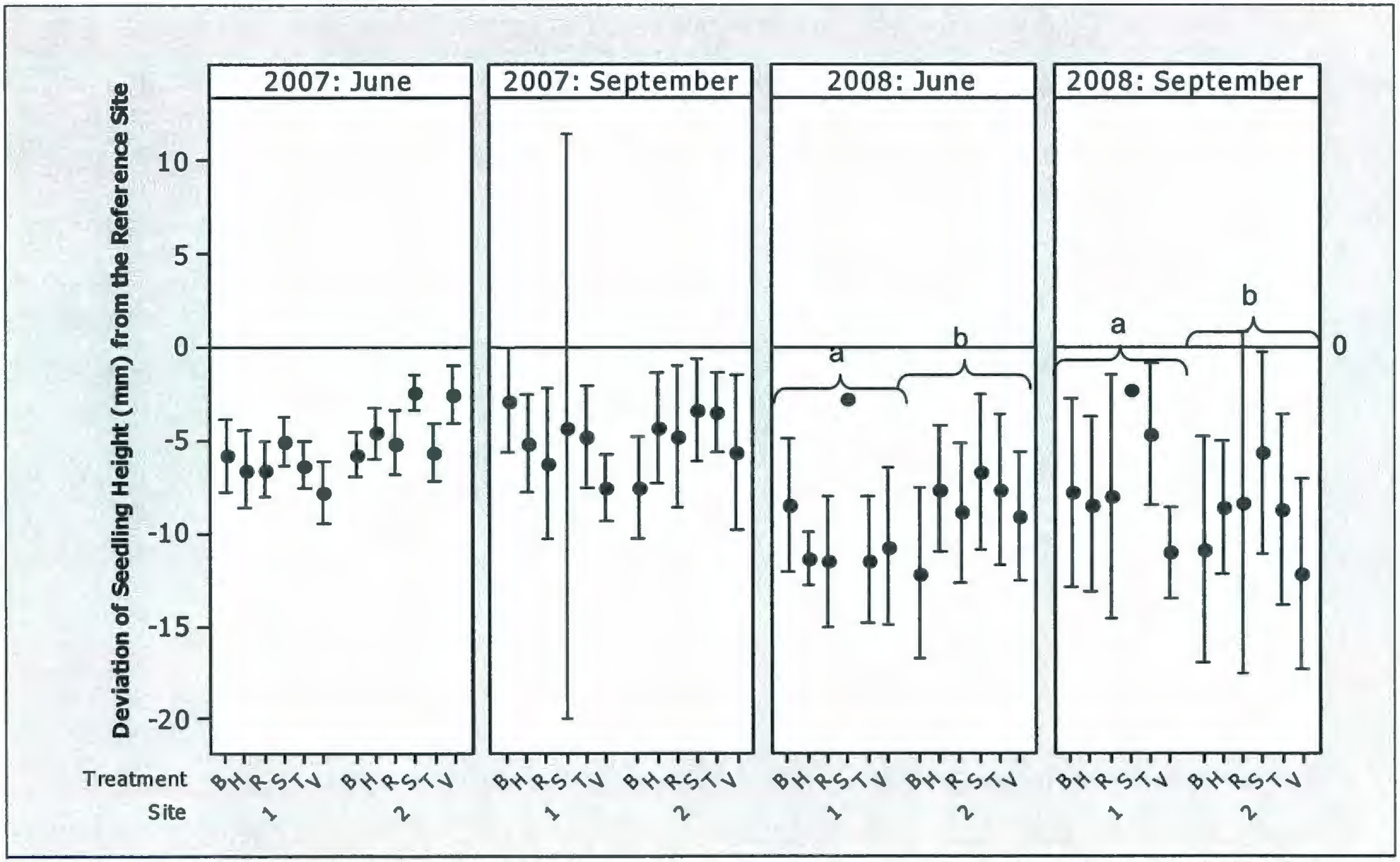

Figure 2.3.4: Deviation of seedling growth (mm) from the non-shrub reference (zero line) for the beginning and end of the first (2007) and second (2008) growing seasons. 95% confidence intervals are included. Treatments include *Betula* nurse (**B**), *Betula* nurses that have been cleared by rodent herbivory (**H**), artificially cleared *Betula* nurses (**R**), artificially shaded *Betula* nurses (**S**), artificially trimmed *Betula* (**T**), and *Vaccinium* nurses (**V**). Site 1 is a low density birch stand with feathermoss and lichen seedbed, and Site 2 is a high density birch stand with *Sphagnum* seedbed. The symbol 'a' denotes a significant difference from 'b' using a GLM ANOVA with weighted means.

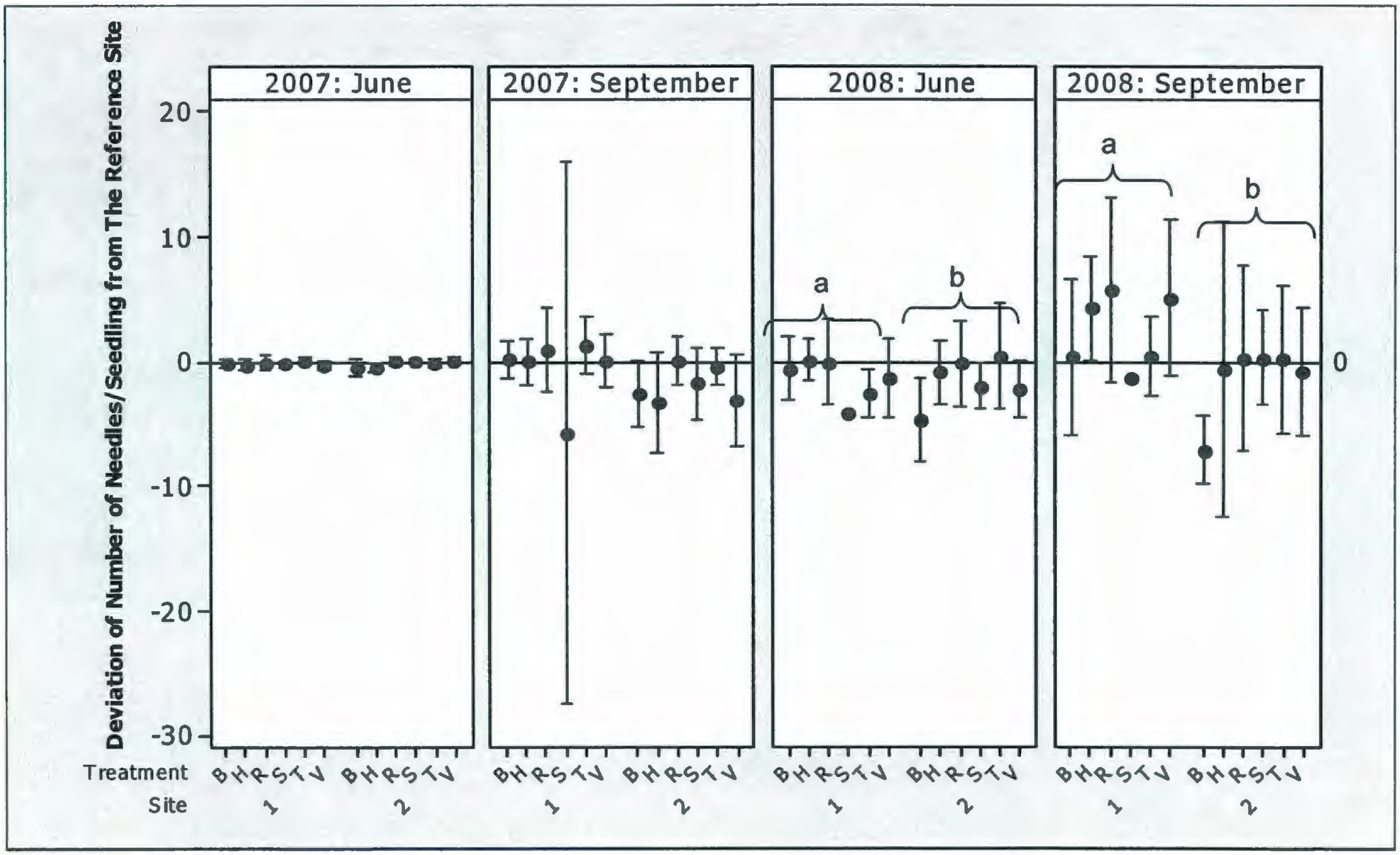

Figure 2.3.5: Deviation of average number of needles produced per seedling from the non-shrub reference (zero line) for the beginning and end of the first and second growing seasons. 95% confidence intervals are included. Treatments include *Betula* nurse (**B**), *Betula* nurses that have been cleared by rodent herbivory (**H**), artificially cleared *Betula* nurses (**R**), artificially shaded *Betula* nurses (**S**), artificially trimmed *Betula* (**T**), and *Vaccinium* nurses (**V**). Site 1 is a low density birch stand with feathermoss and lichen seedbed, and Site 2 is a high density birch stand with *Sphagnum* seedbed. The symbol 'a' denotes a significant difference from 'b' using a GLM ANOVA with weighted means

Analysis of the cause of death for each mortality event of *P. mariana* seedlings showed that the main causes of death were slug herbivory (25%), vertebrate herbivory (25%), and failure to thrive (50%). Voles were the main vertebrate herbivores at this site; however there were 3 instances of ptarmigan herbivory. Failure to thrive was mainly due to desiccation and in two instances appeared to result from competition with neighbouring plants (Appendix III). Neither nurse treatment (slug: $F_{5,24}$=0.91, p=0.538; vertebrate: $F_{5,24}$=0.15, p=0.977; failure to thrive: $F_{5,24}$=0.64, p=0.674) nor site (slug: $F_{1,24}$=0.39, p=0.538; vertebrate: $F_{1,24}$=1.29, p=0.264; failure to thrive: $F_{1,24}$=0.09, p=0.760) had a significant effect on the likelihood of herbivory on *P. mariana* seedlings during the first two growing seasons suggesting that nurses do not protect seedlings.

Seedling survival follows a Type I survivorship where the highest mortality occurs early in life and decreases with age (Figure 2.3.6). Over the first two growing seasons, survival was not significantly affected by treatment ($F_{5,8}$=1.07, p=0.443) or site ($F_{1,8}$=0.07, p=0.794). There was a significant interaction between treatment and site ($F_{5,8}$=6.49, p=0.011) as a consequence of higher survival in low density Site 1 in all treatments but Shaded ($\mathbf{B_S}$) and Trimmed ($\mathbf{B_T}$). The extraordinarily high error associated with the percent survival suggest that individual herbivory events observed in low density Site 1, is a result of entire plots being destroyed by herbivores. Despite the initial lack of significant differences among treatments with respect to herbivory on seedlings, by the end of the second growing season *P. mariana* seedlings in low density Site 1, *Betula* (**B**), Trimmed (**B_T**), and Herbivory (**B_H**) treatments had greater survival than the non-shrub reference site. This is suggestive of a nurse effect after two growing seasons.

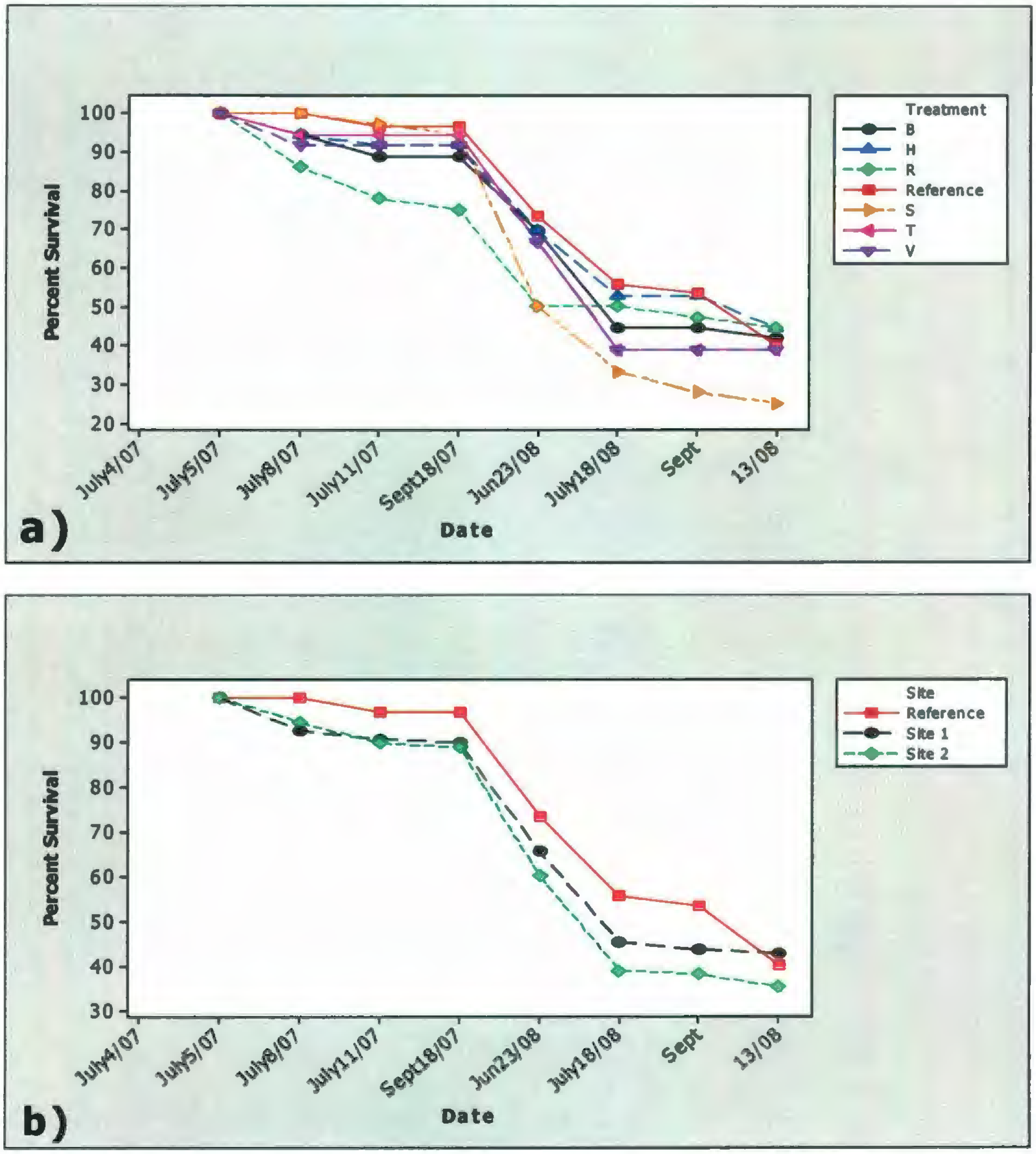

Figure 2.3.6 Survivorship curves of *P. mariana* seedlings for the first two growing seasons for treatment [a)] and site [b)]. Treatments include the non-shrub reference site (**Reference**), *Betula* nurse (**B**), *Betula* nurses that have been cleared by rodent herbivory (**H**), artificially cleared *Betula* nurses (**R**), artificially shaded *Betula* nurses (**S**), artificially trimmed *Betula* (**T**), and *Vaccinium* nurses (**V**). Site 1 is a low density birch stand with feathermoss and lichen seedbed, and Site 2 is a high density birch stand with *Sphagnum* seedbed.

Table 4: Summary of Stress Gradient Hypothesis predictions for nurse mechanisms at the Forest-Tundra Transition zone and the observed result

Nurse Mechanism	SGH Predictions	Observed Result
Nurse Shade	Recruitment will be highest beneath Trimmed *Betula* ($\mathbf{B_T}$) because of higher PAR, and lowest beneath Shaded *Betula* ($\mathbf{B_S}$)	Recruitment not affected
Nurse Height	Recruitment beneath *Betula* nurses will be higher than *Vaccinium* (**V**)	Soil moisture highest beneath Vaccinium nurses ($F_{5,174}$=3.46, p<0.005) Recruitment not affected
Protection from Herbivory	Herbivory will be highest beneath nurses with the most protective (dense) canopies (i.e., *Betula* [**B**], Shaded *Betula* [$\mathbf{B_S}$])	Herbivory not affected Recruitment not affected
Site Effect	Recruitment in Site 1 (low density birch) will be highest because of the benefits of protection and higher PAR of the nurse shrubs	Temperature highest in Site 1 (H_1=116.77, p<0.001) *P. mariana* Emergence highest in Site 2 ($F_{1,17}$=6.21, p=0.023) *P. mariana* Growth highest in Site 1 for 2008 ($F_{1,63}$=4.65, p=0.035) More Needles in Site 1 for 2007 ($F_{1,128}$=6.15, p=0.014) and 2008 ($F_{1,73}$=7.74, p=0.007) for *P. mariana*

2.4 Discussion

The SGH has been investigated in a wide range of environments but has not been studied at the boreal F-T Transition zone. This study suggests a net-positive facilitative association of *Picea* seeds/seedlings and nurse shrubs in the F-T Transition zone of the Mealy Mountains but after 2 years the positive benefit is mainly expressed in low density nurse shrubs, supporting the first prediction that

B. glandulosa and *V. uliginosum* can alter the abiotic environment resulting in a net facilitative nurse effect. Low shrub density Site 1 had the warmest soil and all nurse treatments had higher levels of calcium than the non-shrub reference site illustrating the variability associated with plant-plant interactions across even small spatial scales. Moreover, emergence success of both *P. mariana* (20% ± 6.12) and *P. glauca* (12% ± 2.8) seeds planted beneath nurse shrubs is higher than seeds planted in the non-shrub reference site, however, seedling height and number of needles were not facilitated by either nurse, which is inconsistent with the SGH. It was not clear that *B. glandulosa* created a significantly stronger nurse effect than *V. uliginosum* which provided the highest soil moisture levels observed, thus refuting the second prediction. The third prediction, that recruitment would be highest beneath nurses with moderate amounts of shade, was rejected as no single treatment provided putatively enhanced conditions for seedling establishment and growth. Finally, the fourth prediction that herbivory on seedlings would be highest beneath nurse shrubs was also rejected as it was relatively constant across all treatments.

Soil Properties and their Effects on Emergence

Soil moisture for all nurse treatments was lower than in the non-shrub reference site, suggesting that nurse plants reduce water availability to immediate neighbours without providing the benefit of hydraulic lift, frequently observed in arid environments (Callaway 1998; Pugnaire et al. 1996; Castro et al. 2002).

While *V. uliginosum* nurses were able to retain higher soil moisture than *B. glandulosa*, this amelioration was not reflected in higher *Picea* recruitment and so is not considered integral in seedling facilitation. This is likely because moisture is not limiting in the Mealy Mountains (Jacobs and Chan, unpublished data); all nurse treatments must retain sufficient soil moisture for *P. mariana* and *P. glauca* to germinate, emerge, and grow. Amelioration of soil moisture conditions may therefore be a characteristic of moisture limited environments only where hydraulic lift increases availability of limited soil water to beneficiaries (Franco and Nobel 1989).

Another benefit a nurse can provide to its beneficiary is buffering against extreme temperatures (Franco and Nobel 1988, 1989; Carlsson and Callaghan 1991; Kudo and Suzuki 2003; Castro et al. 2002). Nurse shade protects seedlings from temperature extremes by reducing PAR reaching the soil and hence evapotranspiration during the summer months (Sturm et al. 2001; Akhalkatsi et al. 2006). If shade was a driving mechanism behind the facilitation of *Picea spp.* in this study, lower temperatures would be expected in treatments with the most shade (i.e., *Betula* (**B**), and Shaded (**B$_S$**) treatments). For example, *P. mariana* seedlings which germinate in low intensity post-fire environments have highest survival in microclimates which provide shade (Moss 2004). In this study, there was no significant reduction in soil temperature among shade treatments and seed establishment did not differ. However, the high shrub density Site 2 with its greater canopy shading did have significantly cooler summer soil temperatures.

The ~4°C reduction in soil temperature under high density *B. glandulosa* is potentially strong enough to adversely affect recruitment success of *Picea spp.* that require a range of 10-24°C for germination (Burns and Honkala 1990; Black and Bliss 1980) because soil temperatures in this site drop below the 10°C threshold. However, 20% of *P. mariana*, and 12% *P. glauca* seeds emerged beneath nurse shrubs. This is high emergence success relative to previous studies in the Mealy Mountains which report emergence of <5% in alpine heath, tree island, and open canopy habitats (Munier 2006; Wheeler, unpublished MSc. thesis). Despite temperatures below the threshold for germination, emergence was not impeded beneath nurses and was in fact significantly facilitated by both *B. glandulosa* and *V. uliginosum*, a phenomenon likely resulting from a combination of facilitative nurse effect. If viable seed can reach these areas, it seems likely that establishment (i.e., germination and emergence) will be facilitated beneath these dwarf shrubs.

Shading not only ameliorates temperature extremes and reduces soil temperatures, but can also negatively affect beneficiaries by reducing available PAR to levels insufficient for growth. Although *Picea* seedlings are generally light limited beneath shrub canopies, they are stress-tolerant and can persist under low light conditions (Baker 1949; Attree et al. 1995). The negative effects of lower PAR availability are therefore limited and can sometimes be restricted to the edge of a nurse patch, or stand (Voicu and Comeau 2006). This suggests that if the nurse stands in Site 1 and Site 2 are of sufficient size, the negative effects of shade from the *B. glandulosa* may be negligible on the growth *Picea* seedlings, resulting in a net neutral association.

Nurse plots have consistently higher levels of calcium (450.79 µg/10cm^2± 380.77) than the non-shrub reference site (230.93 µg/10cm^2± 171.82); however within sites and among treatments, soil nutrient levels did not differ. This lack of a treatment effect suggests that herbivory on nurse plants (including latrines left behind by rodent herbivores), shade, and height, has no effect on soil nutrients.

Calcium plays a role in cell wall formation and stabilization, membrane stabilization, metabolic regulation, and even tolerance of cold, salinity, drought and shade in *Picea spp.* (Fink 1991, DeHayes et al. 1999). The typical needle 'yellowing' observed in declining conifers is characteristic of a calcium deficiency (Fink 1991). At the community level, calcium availability is associated with ecosystem productivity and vigour (Kobe et al. 2002). The high calcium input under nurse shrubs is likely from litter leachate and may facilitate growth and survival of *Picea* seedlings (Kobe et al. 2002; DeHynes et al. 1999). Unfortunately, no calibrated correlations can be made between PRS™ probe nutrient supply rate data and minimum calcium levels required for conifer growth determined using conventional extractions. PRS™ probes measure dynamic *in situ* nutrient flux over time to a constant surface area as opposed to available nutrient pools at a point in time. It is therefore impossible to compare the calcium levels observed to other studies.

Mechanical barriers such as dry, nutrient deficient lichen seedbeds and the accumulation of nurse leaf litter can impede seed emergence in *Picea spp.* Germination may be restricted by rapid fluctuations in moisture content characteristic of lichen seedbeds (Foster 1985; Richardson and Hall 1973), but successful establishment can be facilitated in desiccation fractures in the lichen layer where soil moisture is maintained near field capacity and so may not have reduced emergence success in the study area (Power 2005; Lafleur et al. 2002). Deciduous litter beds can also significantly reduce the ability of conifer seedlings to establish by creating a mechanical barrier between air and soil (Koroleff 1954, Parent et al. 2006). A broadleaf litter layer (including *B. papyrifera* [white birch], *Populus tremuloides* [trembling aspen], and *Prunus pensylvanica* [pin cherry] leaves) of only 2.5-3.0cm in thickness, for example, is enough to reduce emergence of *A. balsamea* seedlings by 57% (Parent et al. 2006). *Picea* seedlings that germinate in a litter layer 5-10cm thick rarely survive (Chapin et al. 2004). In this study, *P. mariana* and *P. glauca* seeds associated with a nurse shrub had consistently higher emergence success than the non-shrub reference site. Insulation from extreme temperatures, protection from post-dispersal seed predation, as well as calcium inputs from the litterfall from the time the seeds fall in autumn/winter to when they germinate in July most likely outweighs the negative mechanical effect of the deciduous litter in this study, but this hypothesis would need to be tested.

Seedling Growth and Survival

Low nurse density in this study provides preferable growing conditions to *P. mariana* by allowing more PAR to reach seedlings, increasing soil temperatures but still providing protection from harsh winds. Both seedling height and number of needles were greater in low density Site 1 during the second growing season (2008). This higher growth in the low density Site 1 may be a consequence of higher light availability, warmer temperatures and the absence of the *Sphagnum* mat present in high density Site 2. The growth and morphology of *Picea spp.* have been shown to respond positively to higher light levels (Claveau et al. 2002). Although *Picea* are relatively shade tolerant, under extreme low light conditions, vertical growth can be reduced or nearly cease in favour of resource storage (Messier et al. 1999). However, as previously discussed, the two *B. glandulosa* stands involved in this study are likely of sufficient size to reduce the effects of PAR limitations by shade (Voicu and Comeau 2006).

Slower growth rates in high density Site 2 are likely caused by seedbed interactions. High shrub density is associated with higher cover and thickness of *Sphagnum* seedbed. *Sphagnum* cells contain a secondary metabolite [*p*-hydroxy-β-(carboxymethyl)-cinnamic acid, also known as 'Sphagnum acid' (Rudolph 1972)], that is easily leached into the soil and inhibits tree growth (Verhoeven and Toth 1995). *Sphagnum* dominated seedbed can also be considerably cooler than in areas lacking *Sphagnum*. For example, *Sphagnum* dominated sites have been shown to thaw 22 days later than sites dominated by *Pleurozium* feathermoss

(Bisbee et al. 2001). This is likely the case in the Mealy Mountains, *Picea* seedlings could be subject to later snowmelt, a shortened growing season, and allelopathic chemicals, resulting in the slower growth rates observed. Any benefits accrued through the association with dense *B. glandulosa* in high density Site 2 are likely outweighed by the strong allelopathic influence of *Sphagnum* absent in low density Site 1

The role of nurse shrubs in inhibiting herbivory on seedlings varies among studies. Nurse shrubs, particularly unpalatable, thorny species, are generally believed to provide a mechanical barrier to herbivory on seedlings (Holmgren et al. 1997; Padilla and Pugnaire 2006). However they can also provide refugia to small herbivores, and thus increase herbivory (Whittington-Jones et al. 2008; Bestelmeyer et al. 2007). In this study, the nurse plants are palatable, and they appear to increase the abundance of herbivores (rodents and slugs). Yet seedling mortality caused by herbivory did not differ among treatments, suggesting that shrubs do not increase herbivory of seedlings in this study despite their high palatability.

The decreased survival of seedlings in the non-shrub reference site compared with those in the shrubs , as well as growth via needle production suggest a temporal aspect of the nurse effect in this study. In both growing seasons (2007 to 2008) survival of *P. mariana* seedlings is relatively constant in all treatments and both study sites relative to the non-shrub reference site. It is not until the end of the second growing season (September 2008) that survival in the non-shrub reference site decreases. Similarly, by the end of the second growing season, the number of needles per seedlings in the nurse treatments begins to surpass the non-shrub reference site. This suggests that early in the development of *P. mariana* seedlings, sheltering by some seedbed types (feathermosses, and lichen) is the predominant factor determining survival. Once the seedlings grow above the protection of the seedbed, facilitation provided by nurse shrubs becomes increasingly important (Smith et al. 2003). The boundary layer between seedbed and the open air is often severe and exposes new shoots and needles to wind, extreme temperatures, herbivory, and desiccation (Smith et al. 2003). This trend is not applicable to the high density Site 2 where *Sphagnum* is the dominant seedbed. The net effect of nurse shrubs on *P. mariana* and *P. glauca* seedlings in this study seems to depend on ontogeny, which is supported by an influential paper by Walker and Callaway (1997). Further tracking of the seedlings need to be done to determine if this trend would persist with seedlings becoming increasingly dependent on their nurse neighbours as they grow away from the seedbed.

Conclusion

Nurse shrubs have the ability to alter the biotic and abiotic environment beneath their canopies in such a way as to ameliorate conditions and facilitate recruitment of *P. mariana* and *P. glauca* that have grown above the seedbed. If temperatures continue to rise in this area as has been predicted, ameliorating environmental conditions for the conifers, the SGH predicts that the net interaction between *B. glandulosa* and *Picea spp.* will eventually shift to a predominantly competitive one, particularly as established seedlings grow, overcrowd and outcompete their nurses. The expansion of alpine shrubs (such as dwarf birch and willow), currently underway in North America, may facilitate conifer establishment and could therefore pave the way for a similar expansion of the boreal forest into the alpine zone (Payette 2007). The principal mechanisms by which this expansion is achieved varies: High density *B. glandulosa* stands increase emergence of *P. mariana* despite reducing summer temperatures below the threshold for successful germination in the open tundra, while the prostrate *V. uliginosum* takes up less water, allowing higher soil moisture availability to beneficiaries, and finally, both nurse species provide an increased supply of soil calcium which aids in many physiological processes.

A significant limitation to this research was the length of the study. Field work was limited to two field seasons. It was not until the second growing season ended (September 2008) that shrub facilitation began to reveal itself in seedling survival rates. Future research should focus on extending the time frame of seedling facilitation studies to include at least the first three growing seasons. *Picea* seedlings are small and may only grow 2cm or less in their first year. At this height, seedbed species are more likely to drive recruitment success. During the second and third year's growth, as seedling pass the seedbed boundary layer, the importance of shrub facilitation increases and may be easily detected.

Exploration of the SGH typically involves investigating naturally established nurse-beneficiary associations, but a systematic search of 300m^2 in the upper F-T Transition zone of the Mealy Mountains did not reveal any naturally established seedlings in the study site (Appendix I). This indicates a lack of viable seed production (R. Jameson, pers. comm. 2009). If viable seeds are produced in the future as the climate warms and are able to disperse to the shrub dominated F-T Transition zone, this study suggests *Picea spp*. will be facilitated by nurse shrubs and expand their range northward and upward in elevation. Future research should track seedling recruitment within this key landscape feature. The most prevalent interaction for conifers, significantly stressed beyond the F-T Transition zone, will be facilitation by neighbouring nurse shrubs (Callaway 1998) allowing for greater recruitment success beyond their traditional distributional limit. By ignoring the role of biotic interactions, a significant mechanism of potential advance, or retreat of boreal tree species, is overlooked. Knowledge of species interactions in the F-T Transition zone will allow us to understand how ranges will change with climate warming, therefore assisting in planning for impacts on other ecosystem users.

Acknowledgements

We thank Paul Marino and Keith Lewis for their comments and advice during this study and to all our colleagues for field support, and particularly Julia Wheeler and John Jacobs for providing unpublished data. Thanks to the Labrador Tree Nursery (Dept of Natural Resources) for use of their facility in Labrador, and providing seeds. Funding and logistical support was provided by Memorial University, IPY-PPS Arctic, IPY-CiCAT, Parks Canada, and the Northern Scientific Training Program (NSTP).

References

ACIA (Arctic climate impact assessment). 2005. Arctic climate impact assessment: Scientific report, Cambridge: University of Cambridge Press, p. 1042

Akhalkatsii, M., Abdaladze, O., Nakhutsrishvili, G., Smith, W. K. 2006. Facilitation of seedling microsites by *Rhododendron Caucasicum* extends the *Betula Litwinowii* alpine treeline, Caucasus Mountains, Republic of Georgia. *Arctic, Antarctic and Alpine Research* **38**(4): 481-488

Attree, S. M., Pomeroy, M. K., Fowke L. C. 1995. Development of white spruce (*Picea glauca* (*Moench.) Voss*) somatic embryos during culture with abscisic acid and osmoticum, and their tolerance to drying and frozen storage. *Journal of Experimental Botany* **46**(285): 433-439

Baker, F. S. 1949 A revised tolerance table. *Journal of Forestry* **47**(3):179-181

Barber, D. B., Lukovich, J. V., Keogak, J., Baryluck, S., Fortier, L., and Henry, G. H. R 2008. The changing climate of the Arctic. *Arctic* **61** Suppl. 1:7-26

Bertness, M. D., Callaway, R. M. 1994. Positive interactions in communities. *Trends in Ecology and Evolution* **9**(5): 191-193

Bestelmeyer, B. T., Khalil, N. I., Peters, D. P. C. 2007. Does shrub invasion indirectly limit grass establishment via seedling herbivory: A test at grassland-shrubland ecotones. *Journal of Vegetation Science* **18**(3): 363-370

Bisbee, K. E., Gower, S. T., Norman, J. M., Nordheim, E. V. 2001. Environmental controls on ground cover species composition and productivity in a boreal black spruce forest. *Oecologia* **29**(2): 261-270

Black, A., R., Bliss, L. C. 1980. Reproductive ecology of *Picea mariana* (Mill.) B. S. P., at treeline near Inuvik, Northwest Territories, Canada [Forest Tundra]. *Ecological Monographs* **50**(3): 331-354

Burns, R. M., and Barbara H. Honkala, tech. coords. 1990. Silvics of North America: 1. Conifers; 2. Hardwoods. Agriculture Handbook 654. *U.S. Department of Agriculture*, Forest Service, Washington, DC. vol.2, 877 p.

Callaway, R. M. 1998. Are positive interactions species-specific? *Oikos* **82**(11): 202-207

Callaway, R. M., Walker, L. R. 1997. Competition and facilitation: A synthetic approach to interactions in plant communities. *Ecology* **78**(7): 1958-1965

Canonne, N., Sgorbati, S., Guglielmin, M. 2007. Unexpected impacts of climate change on alpine vegetation. *Frontiers in Ecology and the Environment* **5**(7): 360-364

Carlsson, B. A., Callaghan, T. V. 1991. Positive plant interactions in tundra vegetation and the importance of shelter. *Journal of Ecology* **79**(4): 973-983

Castro, J., Zamora, R., Hódar, J. A., Gómez, J. M. 2002. Use of shrubs as nurse plants: A new technique for reforestation in Mediterranean Mountains. *Restoration Ecology* **10**(2): 297-305

Chapin III, F. S., Sturm, M., Serreze, M. C., McFadden, J. P., Key, J. R., Lloyd, A. H., McGuire, A. D., Rupp, T. S., Lynch, A. H., Schimel,J. P., Beringer,J., Chapman,W. L., Epstein,H. E., Euskirchen, E. S., Hinzman, L. D., Jia,G., Ping, C. –L., Tape, K. D., Thompson,C. D. C., Walker, D. A., Welker, J. M. 2005. Role of land-surface changes in arctic summer warming. *Science* **310**(2748): 657-660

Chapin III, F. S., Shaver, G. R., Giblin, A. E., Nadelhoffer, K. J., Laundre, J., A. 1995. Responses of Arctic tundra to experimental and observed changes in climate. *Ecology* **76**(3): 694-711

Chapin III, F. S., Vierek, L. A., Adams, P. C., Van Cleve, K., Fastie, C. L., Ott, R. A., Mann, D., Johnson, J. F. 2004. Successional processes in the Alaskan boreal forest. *In* Alaska's changing boreal forest. *Oxford University Press*: Oxford, UK

Claveau, Y., Messier, C., Commeau, P. G., Coates, K. D. 2002. Growth and crown morphological responses of boreal conifer seedlings and saplings with contrasting shade tolerance to a gradient of light and height. *Canadian Journal of Forest Research* **32**(3): 458-468

Danby, R. K., Hik, D. S. 2007. Variability, contingency and rapid change in recent subarctic alpine tree line dynamics. *Journal of Ecology* **95**(2): 352-363

DeHynes, D. H., Schaberg, P. G., Howley, G. J., Strimbeck, G. R. 1999. Acid rain impacts on calcium nutrition and forest health. *BioScience* **49**(10): 789-800

Environment Canada, 2006. Climate trends and variations bulletin. Available from www.msc-smc.ec.gc.ca/ccrm/buletin/archive_e.cfm

Fink, S. 1991. The micromorphological distribution of bound calcium in needles of Norway spruce [*Picea abies* (L.) *Karst*.]. *New Phytologist* **119**(1): 33-40

Fortin, G., Pilote, J –L. 2007. Multidate mapping approach to determine alpine and subalpine vegetation variations on Mount Jacques Cartier, Quebec, Canada (1973-2004). www.mountaincartography.org/publications/papers/papers_lenk_08/**fortin** pdf

Foster,D. R. 1985. Vegetation development following fire in Picea mariana (*black spruce*)- *Pleurozium* forests of southeastern Labrador, Canada. *Journal of Ecology* **73**(22): 517-534

Franco, A. C., Nobel, P. S. 1988. Interactions between seedlings of *Agave deserti* and the nurse plant *Hilaria rigida*. *Ecology* **96**(6): 1731-1740

Franco, A. C., Nobel, P. S. 1989. Effect of nurse plants on the microhabitat and growth of cacti. *Journal of Ecology* **77**(3): 870-886

Grace, J., Berninger, F., Nagy, L. 2002. Impacts of climate change on the treeline. *Annals of Botany* **90**(4): 537-544

Hair Jr, J. F., Anderson, R. E., Tatham, R. L. 1987. Multivariate analysis. *Macmillan Publishing Company*: New York, NY

Henry, G. H. H., Molau, U. 1997. Tundra plants and climate change: the International Tundra Experiment (ITEX). *Global Change Biology* **3**(1): 1-9

Holmgren, M., Scheffer, M., Huston, M. 1997. The interplay of facilitation and competition in plant communities. *Ecology* **78**(7): 1966-1975

Hurlbert, J. H. 1984. Pseudoreplication and the design of ecological field experiments. *Ecological Monographs* **54**(2): 187-211

IPCC. 2007. Climate change 2007 synthesis report. Contribution of working groups I, II, and III to the 4th assessment report of the Intergovernmental Panel on Climate Change. [Core writing team, Pachain, R. K., Reisinger, A. (eds.)]. IPCC, Geneva, Switzerland. p. 104

Jones, C. G., Lawton, J. H., Shachak, M. 1997. Positive and negative effects of organisms as physical ecosystem engineers. *Ecology* **78**(7): 1946-1957

Klanderud, K., Totland, O. 2005. The relative importance of neighbours and abiotic environmental conditions for population dynamic parameters of two alpine plant species. *Journal of Ecology* **93**(3): 493-501

Kobe, R. K., Likens, G. E., Eager, C. 2002. Tree seedling growth and mortality responses to manipulations of calcium and aluminum in a northern hardwood forest. *Canadian Journal of Forest Research* 32(**6**): 954-966

Koroleff, A. 1954. Leaf litter as a killer. *Journal of forestry* **52**(3): 178-182

Kudo, G., Suzuki, S. 2003. Warming effects on growth, production, and vegetation structure of alpine shrubs: a five-year experiment in northern Japan. *Oecologia* **135**(2): 280-287

Labrador Highlands Research Group. 2007. Annual report of research 2006. Memorial University of Newfoundland, Department of Geography: St John's NL, A1B 3X9

Lafleur, B., Bradley, R. L., Francoeur, A. 2002. Soil modifications created by ants along a post-fire chronosequence in lichen-spruce woodland. *Ecoscience* **9**(11): 63-73

Lantz, T. C. 2008. Relative influence of temperature and disturbance on vegetation dynamics in the low Arctic : an investigation at multiple scales. PhD. Thesis. Forestry, University of British Columbia:Vancouver, BC.

McArdle, B. H. 1996. Levels of evidence in studies of competition, predation, and disease. *New Zealand Journal of Ecology* **20**(1): 7-15

Maestre, F. T., Callaway, R. M., Valladares, F., Lortie, C. J. 2009. Refining the stress- gradient hypothesis for competition and facilitation in plant communities. *Journal of Ecology* **97**(2): 199-205

Messier, C., Doucet, R., Ruel, J. –C., Claveau, Y., Kelley, C., Lechowicz, M. J. 1999. Functional ecology of advance regeneration in relation to light in boreal forests. *Canadian Journal of Forest Research* **29**(6): 812-823

Moss, M. 2004. The impact of slug herbivory on post-fire regeneration of black spruce in lichen woodland in Terra Nova National Park, NL. M.E.S. project, Environmental Science programme, Memorial University. St John's, NL

Munier, A. 2006. Seedling establishment and climate change: The potential for forest displacement of alpine tundra (Mealy Mountains, Labrador). M.Sc. thesis, Dept of Biology, Memorial University, St. John's, NL.

Myers-Smith, I. 2007. Shrub line advance in alpine tundra of the Kluane region: mechanisms of expansion and ecosystem impacts. *Arctic* **60**(4): 447-451

Padilla, F. M., Pugnaire, F. I. 2006. The role of nurse plants in the restoration of degraded environments. *Frontiers in Ecology and the Environment* **4**(4): 196-202

Parent, S., Walsh, D., S, M. –J., Morin, H., Gagnon, R. 2006. Emergence of balsam fir seedlings under increasing broadleaf litter thickness. *Canadian Journal of Forest Research* **36**(8): 1976-1981

Payette, S. 2007. Contrasted dynamics of the Northern Labrador tree lines caused by climate change and migrational lag. *Ecology* **88**(3): 770-780

Power, R. G. 2005. Seed bed micro-sites and their role in post-fire succession of the lichen-black spruce woodland in Terra Nova National Park, Newfoundland. M.Sc. thesis, Dept of Biology, Memorial University, St. John's, NL.

Price, M. V., Waser, N. M. 1998. Effects of experimental warming on plant reproductive phenology in a subalpine meadow. *Ecology* **79**(4): 1261-1271

Pugnaire, F. I., Haase, P., Puigdefabregas, J. 1996. Facilitation between higher plant species in a semiarid environment. *Ecology* **77**(5): 1420-1426

Richardson, J., Hall, J. P. 1973. Natural regeneration after disturbance in the forests of Central Newfoundland. *Canadian Forest Service*, Information Report N-X-86. P 62

Rudolph, H. 1972. Identifikation der Czapekschen Sphagnolkristalle. *Biochemie and Phusiologie der Pflanzen* **163**: 110-112

Smit, C., Vanderbergh, C., den Ouden, J., Müller-Schärer, H. 2007. Nurse plants, tree saplings and grazing pressure: changes in facilitation along a biotic environmental gradient. *Oecologia* **152**(2): 265-273

Smith, W. K., Knapp, A. K., Pearson, J. A., Varman, J. H., Yavitt, J. B., Young, D. R. 1983. Influence of microclimate and growth form on plant temperatures of early spring species in a high elevation prairie. *American Midland Naturalist* **109**(2): 380-389

Smith, W. K., Germino, M. J., Hancock, T. E., Johnson, D. M. 2003. Another perspective on altitudinal limits of alpine timberlines. *Tree Physiology* **23**(16): 1101-1112

Sturm, M., Racine, C., Tape, K. 2001. Increasing shrub abundance in the Arctic. *Nature* **411**(6837): 546-547

Sturm, M., J. Schimel, G. Michaelson, J. M. Welker, S. F. Oberbauer, G. E. Liston, J. Fahnestock, and V. E. Romanovsky. 2005. Winter biological processes could help convert Arctic tundra to shrubland. *BioScience* **55**(1): 17- 26

Tape, K., Sturm, M., Racine, C. 2006. The evidence for shrub expansion in Northern Alaska and the Pan-Arctic. *Global Change Biology* **12**(4): 686-702

Verhoeven, J. T. A., Toth, E. 1995. Decomposition of Carex and Sphagnum litter in fens: Effect of litter quality and inhibition by living tissue homogenates. *Soil Biology and Biochemistry* **27**(3): 271-275

Voicu, M. F., Comeau, P. G. 2006. Microclimatic and spruce growth gradients adjacent to young aspen stands. *Forest Ecology and Management* **221**(1-3):13-26

Walker, M. D., Wahren, C. H., Hollister, R. D., Henry, G. H. R., Alquist, L. E., Alatalo, J. M., Bret-Harte, M. S., Calef, M. P., Callaghan, T. V., Caroll, A. B., Epstein, H. E., Jonsdottir, I. S., Klein, J. A., Magnusson, B., Molau, U., Oberbauer, S. F., Rewa, S. P., Robinson, C. H., Shaver, G. R., Suding, K. N., Thomson, C. C., Tolvanen, A., Totland, O., Turner, P. L., Tweedie, C. E., Webber, P. J., Wookey, P. A. 2006. Plant community responses to experimental warming across the tundra biome. *Proceedings of the National Academy of Science* **103**(5):1342-1346

Wheeler, J. Black spruce seedbed interactions: Facilitation and recruitment in the Boreal-Arctic ecotone (Mealy Mountains, Labrador). M.Sc. thesis, Dept of Biology, Memorial University, St. John's, NL. Unpublished

Whittington-Jones, G. M., Bernard, R. T. F., Parker, D. M. 2008. Bushclumps as refugia for small mammals in two Eastern Cape conservation areas. *African Zoology* **43**(2): 273-276

3.0 General Conclusions

Alpine and arctic ecosystems will be most affected by rising temperatures associated with climate change. In these environments there are invariably communities mixed with some species highly stressed while others are at optimal conditions (Franco and Nobel 1989). Understanding the mechanisms which regulate this balance is crucial for developing a predictive model of treeline advance. By ignoring the role of biotic interactions, a significant mechanism of potential advance, or retreat of boreal tree species, is overlooked. The SGH predicts that the most prevalent interaction for conifers, significantly stressed beyond the F-T Transition zone, will be facilitation by neighbouring nurse shrubs (Callaway 1998) allowing for greater recruitment success beyond their traditional distributional limit. The results of this study suggest that, if viable seed is available, nurse shrubs will facilitate the recruitment of *Picea spp.* above the current altitudinal and latitudinal limit of boreal tree species. However, this positive association between conifer seedlings and nurse shrubs is contingent on seedlings surviving the first growing season to surpass the often severe boundary layer above the seedbed. As the climate continues to warm, shrub expansion into the alpine tundra will accelerate, potentially acting as a conduit for local expansion of trees. The resultant forest displacement and loss of alpine biodiversity could be detrimental to the ecological integrity of the region.

The Labrador Highlands Research Group (LHRG) is an interdisciplinary association of researchers focused on expanding our understanding of a boreal-alpine environment in the highlands of Labrador, with particular focus on how the vegetation and wildlife of the region respond to climate change. As a part of LHRG, this research has shed new light on the dynamic nature of the F-T Transition zone and some of the mechanisms which determine local limits of boreal trees within it. The results of this study have highlighted the importance of considering not only the role of abiotic factors, such as climate, but also biotic associations in determining recruitment success of conifers above traditional boundaries. By investigating current plant-plant associations and how they mediate community structure at the F-T Transition zone today, we gain insight on the potential range of impacts that future climate change may have on this dynamic system, a key objective of LHRG (LHRG 2007).

A significant limitation to this research was the length of the study. Field work was limited to two years. It was not until the second growing season ended (September 2008) that shrub facilitation began to reveal itself in seedling survival rates. Had a third, or even fourth, field season been possible, I predict that the strength of shrub facilitation would only increase as seedlings grow away from the protection of the seedbed. Also, because the nurse effect is subject to such a range of effects, the balance between competition and facilitation is highly site dependent. Factors which control the rate of forest expansion will vary globally.

Future research should focus on extending the time frame of seedling facilitation studies to include at least the first three growing seasons. *Picea* seedlings are small and may only grow 2cm or less in their first year. At this height, seedbed species are more likely to drive recruitment success. During the second and third year's growth, as seedling pass the seedbed boundary layer, the importance of shrub facilitation increases and their influence may be detected.

The significant site effects also bear further investigation. Low density Site 1 has two critical differences from high density Site 2. Firstly, the *B. glandulosa* stand in low density Site 1 (~10 stems per m^2 which are 0.5m high) was less dense than high density Site 2 (~35 stems per m^2 which are 1m high). This greater density may have resulted in the reduced growth rates seen in high density Site 2 via reduced summer temperatures. Secondly, the seedbed in high density Site 2 was dominated by *Sphagnum* moss. The allelopathic 'Sphagnum acid' contained in its cell walls is easily leached, difficult to decompose, and can reduce growth of overstory trees by increasing soil moisture and pH, decreasing temperatures and prolonging soil thawing (Verhoeven and Toth 1995; Bisbee et al. 2001). While *Sphagnum* mosses can protect *Picea* seedlings from the effects of drought, their extremely rapid growth rate tends to overgrow and smother seedlings, resulting in as much as 100% mortality (Drobyshev 1999;Hörnberg et al. 1997).

Finally, a debate which has recently arisen in the literature concerning the SGH is the severity of the stress required to detect an effect. In a response to a study by Maestre et al. 2005, in which insufficient evidence was found to support the SGH, Lortie and Callaway (2006) questioned whether the stress gradient was sufficient. Nowhere in the original study (Bertness and Callaway 1994), or in the later refinement (Callaway and Walker 1997) is it stated that a certain degree of environmental stress is required for the predictions to hold true. There must be a minimum stress gradient for the SGH to be supported, but of all the studies investigating it, there is no explicit consideration of the severity of this gradient. While the study in the Mealy Mountains was conducted in close proximity to the altitudinal tree limit, *Picea spp*. and *A. balsamea* still grow in this area, albeit sparsely and only as gnarled, dwarfed, krummholz. These trees are certainly stressed and at their climatic threshold, however it is possible that the level of stress experienced at these early life history stages here is not strong enough to detect strong facilitative effects. Future research could focus on extending the stress gradient and investigating how the roles of facilitation change in Labrador.

References

Bertness, M. D., Callaway, R. M. 1994. Positive interactions in communities. *Trends in Ecology and Evolution* **9**(5): 191-193

Bisbee, K. E., Gower, S. T., Norman, J. M., Nordheim, E. V. 2001. Environmental controls on ground cover species composition and productivity in a boreal black spruce forest. *Oecologia* **29**(2): 261-270

Callaway, R. M., Walker, L. R. 1997. Competition and facilitation: A synthetic approach to interactions in plant communities. *Ecology* **78**(7): 1958-1965

Callaway, R. M. 1998. Competition and facilitation on elevation gradients in subalpine forests of the northern Rocky Mountains, USA. *Oikos* **82**(3): 561-573

Drobyshev, I. V., 1999. Regeneration of Norway spruce in canopy gaps in *Sphagnum-Myrtillus* old-growth forests. *Forest Ecology and Management* **115**(1): 71-83

Franco, A. C., Nobel, P. S. 1989. Effect of nurse plants on the microhabitat and growth of cacti. *Journal of Ecology* **77**(3): 870-886

Hornberg, G., Ohlson, M., Zackrisson, O. 1997. Influence of bryophytes and microrelief conditions on Picea abies seed regeneration patterns in boreal old-growth swamp forests. *Canadian Journal of Forest Research* **27**(7): 1015-1023

Labrador Highlands Research Group. 2007. Annual report of research 2006. Memorial University of Newfoundland, Department of Geography. St John's NL, A1B 3X9. From www.mun.ca/geog/lhrg/Report_of_Research_2006.pdf

Lortie, C. J., Callaway, R. M. 2006. Re-analysis of meta-analysis: support for the stress gradient hypothesis. *Journal of Ecology* **94**(1): 17-16

Maestre, F. T., Valladeres, F., Reynolds, J. F. 2005. Is the change of plant-plant interactions with abiotic stress predictable? A meta-analysis of field results in arid environments. *Journal of Ecology* **93**(4): 748-757

Verhoeven, J. T. A., Toth, E. 1995. Decomposition of *Carex* and *Sphagnum* litter in fens: Effect of litter quality and inhibition by living tissue homogenates. *Soil Biology and Biochemistry* **27**(3): 271-275

Appendix I: Community Structure

To investigate the effect of shrubs on community structure in closed vs open areas, a series of 25m transects were established: 3 transects were run through the *B. glandulosa* stand at each site (Site 1 [low density] and Site 2 [high density]), and 3 were run directly outside each stand for a total of 12 transects. For each $1m^2$ quadrat along the transect, all species were identified, percent cover estimated, and the number of *B. glandulosa* stems recorded. No naturally established *Picea* seedlings were found. In order to simplify statistical analyses, percent cover of species were categorized and are summarized in Table AI.

Table AI: Categories of percent cover in community structure analysis

Percent Cover	Category
0-5%	1
6-25%	2
26-50%	3
51-75%	4
76-100%	5

To determine if changes in community structure were associated with *B. glandulosa* vs. non-*B. glandulosa* sites, or high vs. low density *B. glandulosa* sites, a Principle Components Analysis (PCA) was run. The PCA conducted on percent cover and species composition data show a significant effect of *B. glandulosa* density on community composition (Figure a.I; Table AII).

The first principal component (PC1) shows a significant gradient from vegetation associated with open areas (*Cassiope tetragona, Salix herbacea,* *Carex biglowlii*) to that associated with shade (*Solidago spp., Kalmia polifolia, V. uliginosum, Trientalis borealis, Empetrum nigrum*) which accurately describes vegetation trends observed at the low density *B. glandulosa* site (Site 1). A second gradient (PC2) follows a trend similar to the first from lichen and moss spp. to *Betula glandulosa, Sphagnum spp., Salix spp.*, and *Cornus canadensis* which describe vegetation strands observed at the high *B. glandulosa* density Site 2.

This is an indication that the presence of *B. glandulosa* as well as its density influences community structure. The clusters in Figure a.III show that community structure is significantly different inside *B. glandulosa* stands versus outside however this trend is clearly stronger in relation to the high density *B. glandulosa* Site 2 as is evident by the separation of inside/outside clusters. Should shrubs continue to advance and increase in density into the alpine zone, plant community structure will certainly be affected.

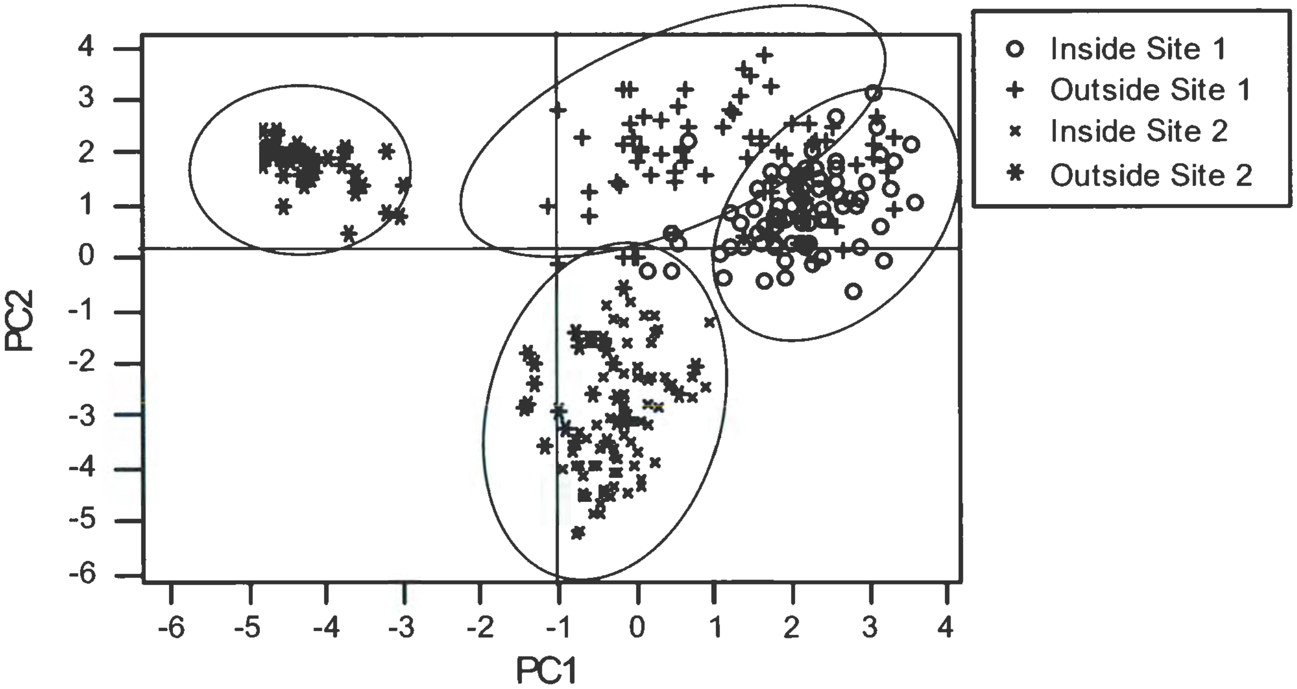

Figure a.I Biplot of Principle Component 1 (Eigen value = 5.42) vs Principal Component 2 (Eigen value = 5.28) showing clustering of transects inside vs outside *B. glandulosa* stands 1 and 2. The Eigen value for the third Principal Component was 2.77 with a variance of 0.08. PC1 accounted for 15 % of the variation, PC2 accounted for 15%; and PC 3 accounted for 7.7%

Table AII Loadings for the first two components of the PCA on community structure. Significant loadings are shown in bold

Variable	PC1	PC2
Tall Betula glandulosa	0.179	**-0.319**
Tall Vaccinium uliginosum	0.051	0.009
Tall Kalmia polifolia	0.085	0.031
Tall Rhododendron groenlandicum	0.037	-0.031
Low Betula glandulosa	0.004	0.083
Low Rhododendron groenlandicum	-0.013	-0.129
Low Kalmia polifolia	**0.223**	**0.205**
Empetrum nigrum	**0.200**	0.047
Salix spp.	-0.033	-0.290
Rubus chamaemorus	0.045	-0.230
Solidago spp.	**0.226**	0.117
Coptis trifolia	0.179	**0.201**
Comandra umbellata	-0.014	-0.199
Salix herbacea	**-0.324**	0.162
Linnaea borealis	0.029	-0.015
Diapensia lapponica	-0.174	0.078
Cornice canadensis	0.039	**-0.278**
Arctostaphylos uva-ursi	0.017	0.013
Cassiope tetragona	**-0.346**	0.157
Carex biglowii	**-0.276**	-0.090
Rush spp.	**0.273**	0.200
Low Vaccinium uliginosum	**0.204**	-0.027
Pedicularis spp.	0.025	-0.090
Phyllodocae caerulea	0.179	0.157
Maianthemum	0.152	0.097
Trientalis borealis	**0.233**	0.132
Lycopodium spp.	0.165	0.184
Vaccinium vitis-idaea	0.034	-0.006
Moss spp.	0.163	**0.245**
Lichen spp.	-0.127	**0.246**
Sphagnum	0.099	**-0.341**
Litter	-0.085	-0.018
Water	0.059	0.038
Bare ground	-0.079	0.029
Rock	**-0.288**	0.128

Appendix II: Slug Trapping

To determine the abundance of slugs in the study area, trapping began June 30th, 2008. 18 traps, made of $10cm^2$ corrugated cardboard were set at 6pm and collected at 6am the following morning. They were evenly spaced in two rows of three in each low density *B. glandulosa* Site 1, high density *B. glandulosa* Site 2, and in a non-shrub reference Site 3. Trapping continued at a rotation of three days on, three days off for 15 days and a total of 168 trapping days. At the time the traps were collected, relative humidity and temperature were recorded.

A total of three slugs (*Deroceras leave*) were caught (Figure a.II). This method of slug trapping has been previously tested (Moss 2004). Our low success rate was likely due to chance alone and not an artefact of the method. Because of the unexpectedly low sample size, statistical analysis was impossible. However, trapping did confirm the existence of slugs in the study area as suspected by the presence of slug trails and eggs found in the area of planting.

Plate a.II *Deroceras laeve* slug caught using corrugated cardboard slug trap

[1] Post-fire seedling recruitment at the southern limit of lichen woodland. Melissa Moss and Luise Hermanutz. MS Submitted

Appendix III: Seedling Mortality

To compare levels of vertebrate and invertebrate herbivory among the treatments, cause of seedling death was attributed either to herbivory (vertebrate, invertebrate), or failure to thrive (Figure a.III). Occasionally the presence of scat (ptarmigan or vole) or slug eggs provided evidence of the herbivores involved. Competition between neighbouring plants was clear when the peat pellet containing the dead seedling was colonized by another plant, often a bryophyte. In these cases, death of the seedling was often gradual as it was out-competed by its neighbour. Failure to thrive was assigned when no other cause of death could be attributed. Transplant death was limited in all plots; there was very low mortality immediately after planting (<5%), moisture was not limiting, and there was little evidence of peat pellets heaving out of the soil. Herbivory on seedlings was easily identifiable by the characteristic "clean nip" damage at the base of the stem or needles by vertebrate predators and the gradual "rasping" damage of slugs. Slug trails, eggs and adults themselves were observed on and around peat pellets containing young *P. mariana*.

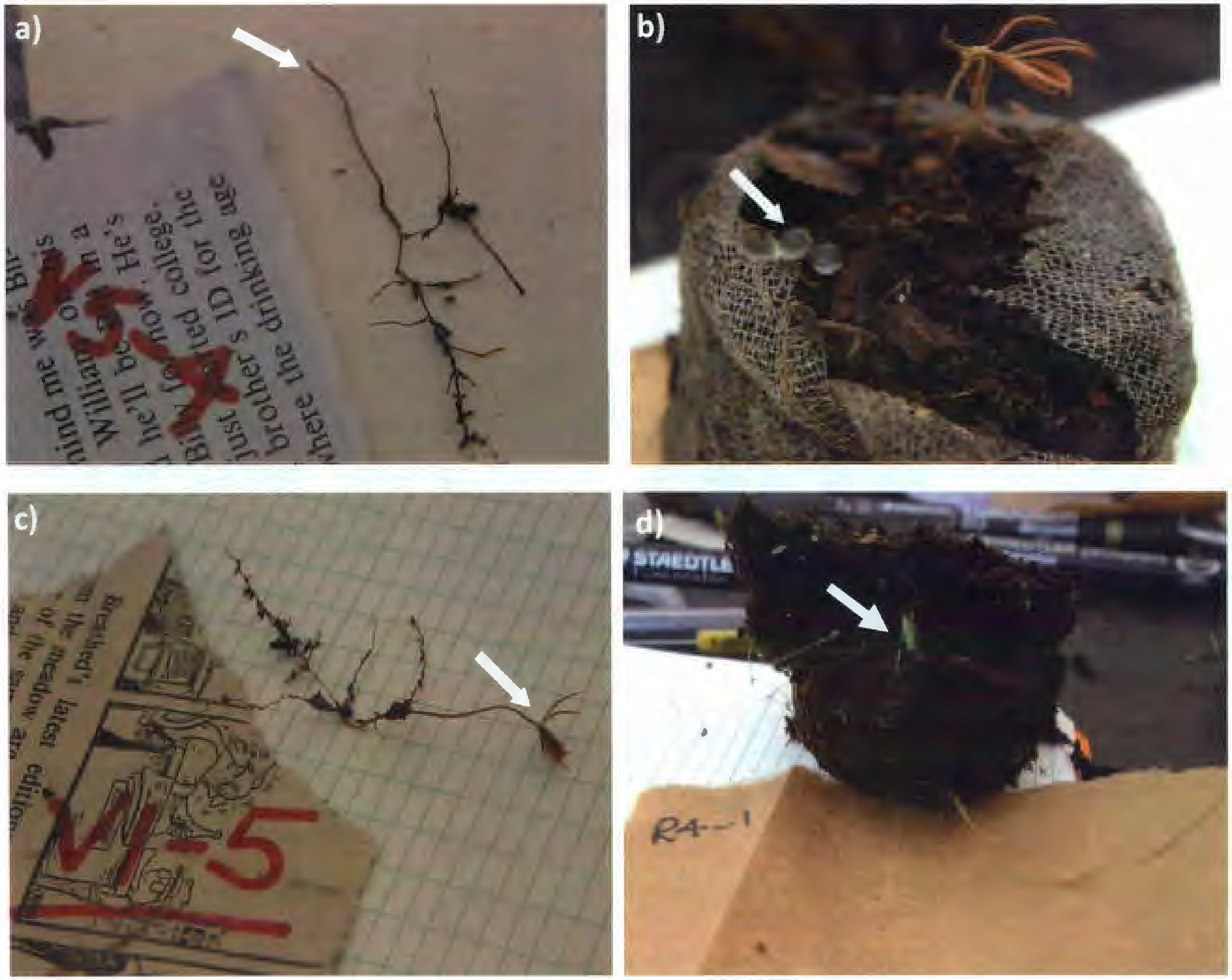

Plate a.III Seedling mortalities as a result of a) vertebrate herbivory *(note: clean bite removing needles)* b) slug herbivory *(note slug eggs)* c) failure to thrive *(note: needles still present, no sign of herbivory and substantial root system remaining)* d) competition *(note invading leader perhaps from Vaccinium neighbour)*

Appendix IV: Soil Nutrient Analysis

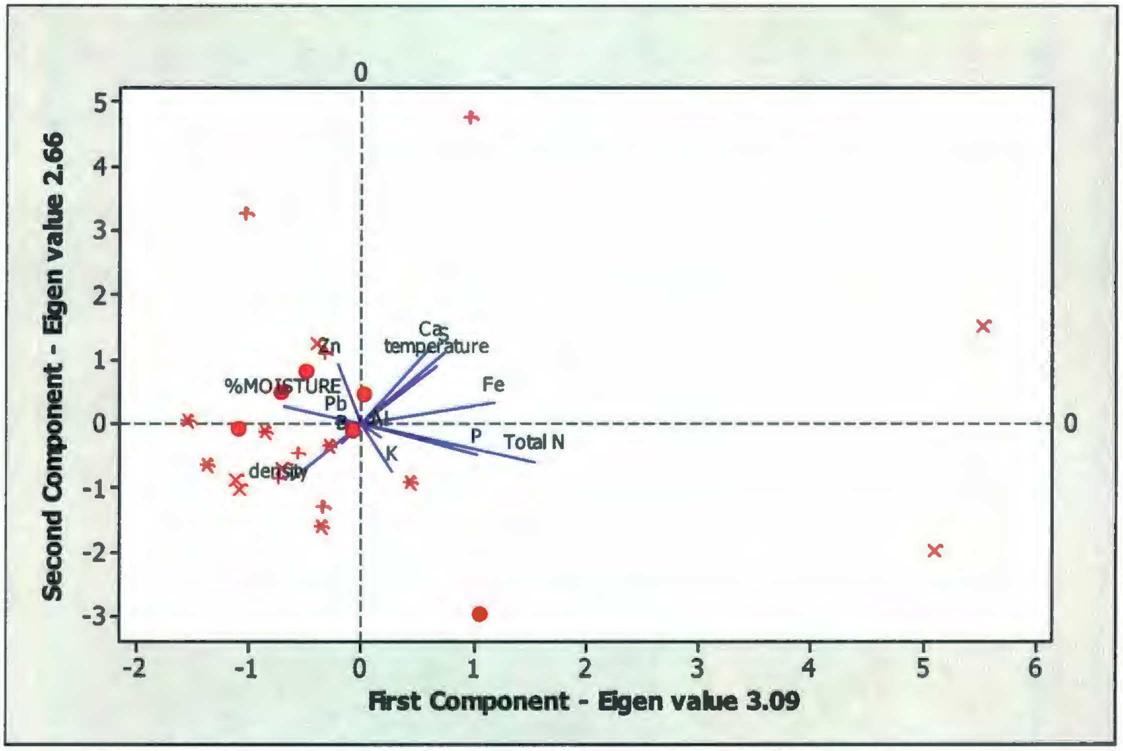

Figure a.IV Biplot of loadings for Principle Components 1 versus 2 for soil characteristics, over laying points coded for treatment type (•=Betula (**B**), Ӿ= Vaccinium (**V**), += Herbivory ($\mathbf{B_H}$), x= Removed ($\mathbf{B_R}$)). PC1 accounted for 22% of the variation, PC2 accounted for 19%, and PC3 accounted for 16%.

The biplot for soil nutrient data shown no clear clusters of points coded for nurse treatments (Figure a.IV; Table AIII). This indicates that any gradient for nutrient loadings is not strongly affected by treatment type. The first and second components explain 22.1% and 19% of the variation respectively, for a cumulative percentage of 41.1%.

Table AIII Loadings for the first two principal components for the PCA on soil nutrients: Significant loadings are shown in bold.

Variable	PC1	PC2
Total N	**0.503**	-0.224
Ca	0.203	**0.450**
K	0.091	-0.282
P	**0.335**	-0.187
Fe	**0.382**	0.123
Zn	-0.068	0.349
B	-0.056	-0.112
S	0.241	**0.412**
Pb	-0.070	0.008
Al	0.056	-0.083
Percent Moisture	-0.223	0.105
Temperature	0.217	0.339
Shrub Density	-0.201	**-0.327**