Fungal Assemblages in Moss & Lichen Mats of Boreal Manitoba

By

Jason Scott Robertson

A thesis submitted to the faculty of Graduate Studies in partial fulfillment of the Requirements for the degree of Master of Science

Department of Botany
University of Manitoba
Winnipeg, Manitoba

August, 2005

Fungal Assemblages in Moss & Lichen Mats of Boreal Manitoba

BY

Jason Scott Robertson

A Thesis/Practicum submitted to the Faculty of Graduate Studies of The University of Manitoba in partial fulfillment of the requirement of the degree

Of

Master of Science

Acknowledgements:

I would like to thank both my advisor and co-advisor, Dr. Tom Booth and Dr. Piercey-Normore for all their guidance and support, my committee members Dr. Sparling and Dr. Sumner for further guidance and constructive criticisms, and Dr. Kenkel for his guidance and suggestions in the statistical analyses. Special thanks to Narinder for his help with laboratory facilities and equipment, to Sara Beigi for assistance in molecular trouble shooting, to Ira Morriseau and Tiago Booth for their help as field assistants, to Tim Tebbe for being there when I needed him, and to my family for their support in my pursuit of this venture.

Abstract

Fungal assemblages in mats of *Cladonia mitis*, *C. rangiferina*, and *Pleurozium schreberi* in Manitoba were isolated and compared in July and September. Temporal changes in these fungal assemblages were seen over the two sampling periods. The fungal assemblages of *C. mitis* and *C. rangiferina* were significantly different from those found in *P. schreberi* during both sampling periods. Isolation of fungi from *C. mitis* and *C. rangiferina* involved subsampling the lichens into upper and lower strata, which were also compared, revealing significant differences between the upper canopies and the lower bases of the lichen mats. *Alternaria*, *Cladosporium*, and *Epicoccum*, common epiphytes, were found to be associated with the upper canopies of *C. mitis* and *C. rangiferina*, while *Mucor* and *Trichoderma*, common soil fungi, were found associated with the bases of *C. mitis* and *C. rangiferina*. Soil litter sampling beneath mats of *C. mitis* and *C. rangiferina* revealed 11 fungal taxa found in the lichen mats that were not in the soil, 6 fungal taxa common to both lichen mats and soil litter beneath them, and 5 fungal taxa present only in the soil litter layers. Fungi isolated from *C. amaurocraea*, *C. arbuscula*, *C. rangiferina*, *C. stellaris*, *C. uncialis, Evernia mesomorpha*, *Vulpicida pinastri, Stereocaulon alpinum*, and *Peltigera* spp., revealed significant differences in fungal assemblages across the various lichens. Microhabitat determines fungal assemblages present on lichens. The most important factor of microhabitat seen was moisture.

Amplification of a portion of nuclear small subunit ribosomal DNA (SSU rDNA) from the fungal partner of *C. arbuscula* revealed fragments of two different size classes in one region. One size class was suspected to contain an intron, while the other had no intron. Amplification of a separate region of SSU rDNA from the fungal partner of *C. arbuscula* revealed fingerprint banding patterns. Variation in banding patterns, as well as the presence or absence of the intron, was examined among samples both within and among 10 transects laid out on islands and lake shores over a 2 km area. Cluster analysis revealed no difference in variation within and among transects, indicating efficient gene flow over the area.

Table of Contents

	Page
Acknowledgements	i
Abstract	ii
Table of Contents	iii
List of Tables	vi
List of Figures	vii
Chapter 1: Introduction	1
Chapter 2: Comparisons of fungal assemblages found in forest mats of *Cladonia mitis*, *C. rangiferina,* and *Pleurozium schreberi* in southern Manitoba, with temporal aspects examined	8
Chapter 3: Spatial comparisons of the fungal assemblages found in mats of *Cladonia mitis* and *C. rangiferina* in southern Manitoba	30
Chapter 4: Intraspecific Variation Within the nuclear SSU rDNA for Populations of *Cladonia arbuscula* in Northern Manitoba	47
Chapter 5: Fungal assemblages associated with selected ground dwelling and epiphytic lichens	61
Chapter 6: Discussion	71
Literature Cited	77
Appendix 1a: July fungal assemblage frequency data pooled by sample	86
Appendix 2a: September fungal assemblage frequency data pooled by sample	89

Appendix 3a: July fungal assemblage frequency data 93

Appendix 3b: Principal component analysis summary of the July fungal assemblage frequency data 96

Appendix 3c: Multiple discriminate analysis summary of the July fungal assemblage frequency data, separating groups by mat species 101

Appendix 4a: September fungal assemblage frequency data 105

Appendix 4b: Principal component analysis summary of the September fungal assemblage frequency data 111

Appendix 4c: Multiple discriminate analysis summary of the September fungal assemblage frequency data, groups separated by mat species 118

Appendix 5a: Pooled July fungal assemblage frequency data by collection type group 123

Appendix 6a: Pooled September fungal assemblage frequency data by collection type group 125

Appendix 7a: Temperature data for La Broquerie, prior to July & September collections 127

Appendix 8a: Precipitation data from St. Labre, prior to July & September collections 129

Appendix 9a: Fungal assemblage frequency data for *Cladonia mitis* from the September data set 131

Appendix 9b: Principal component analysis summary of fungal assemblage frequency data for *Cladonia mitis* from the September data set 134

Appendix 9c: Multiple discriminate analysis of fungal assemblage frequency data for the *Cladonia mitis* September data set, groups separated by canopy & base 139

Appendix 10a: Fungal assemblage frequency data for *Cladonia rangiferina* from the September data set 143

Appendix 10b: Principal component analysis summary of fungal assemblage frequency data for *Cladonia rangiferina* from the September data set 146

Appendix 10c: Multiple discriminate analysis of fungal assemblage frequency data for the *Cladonia rangiferina* September data set, groups separated by canopy & base 152

Appendix 11a: Soil fungal assemblage frequency data for samples collected beneath mats of *Cladonia mitis* & *C. rangiferina* 156

Appendix 12a: Molecular data for *Cladonia arbuscula* 158

Appendix 13a: Fungal assemblage frequency data for the August data set 161

Appendix 13b: Principal component analysis summary of the fungal assemblage frequency data for the August data set 164

Appendix 13c: Multiple discriminate analysis summary of the fungal assemblage frequency data for the August data set, groups separated by lichen taxa 170

Appendix 14a: TLC data from the July collections 175

Appendix 14b: TLC data from the September collections 178

Appendix 14c: TLC data from the August collections for the molecular data sets 181

Appendix 14d: TLC data from the August collections for lichen samples surveyed for fungi 184

Appendix 15a: Photographic plates of fungi 186

List of Tables

Page

Table 2.1. Species Richness & Shannon-Wiener evenness diversity indices for the pooled July & September fungal assemblage collection type groups. 22

Table 2.2. List of fungal assemblages and occurrences for collection type groups for the July and September collections. 24

Table 2.3. Mean occurrences of the fungal assemblages found for each collection type group for the July and September data sets. 25

Table 2.4. Meteorological data for the three weeks proceeding sampling of the July and September collections. 27

Table 3.1. Species Richness & Shannon-Wiener diversity indices for fungal assemblages from the September data set, broken into canopy and base groups. 37

Table 3.2. List of fungal assemblages and occurrences for canopy and base groups from the September collections. 38

Table 3.3. List of fungi and their occurrences on washed litter from soil samples taken beneath *Cladonia mitis* and *C. rangiferina* mats. 40

Table 4.1. Site descriptions of transects from the Payuk lake region. 51

Table 5.1. Occurrences of fungal taxa isolated from lichen taxa sampled in northern Manitoba. 66

Table 5.2. List of the secondary compounds present in the lichen taxa sampled from northern Manitoba. 68

List of Figures

Page

Figure 2.1. Simplified sketch of a *Cladonia mitis* podetium, with canopy and base strata labelled 11

Figure 2.2. Dendrogram from a sums of squares cluster analysis of the fungal assemblages for the July data set. 18

Figure 2.3. Dendrogram from a sums of squares cluster analysis of the fungal assemblages for the Sepember data set. 19

Figure 2.4. Graph of ordinations from a multiple discriminate analysis separating the fungal assemblages found in *Cladonia mitis*, *C. rangiferina*, & *Pleurozium schreberi* for the July data set. 20

Figure 2.5. Graph of ordinations from a multiple discriminate analysis separating the fungal assemblages found in *Cladonia mitis*, *C. rangiferina*, & *Pleurozium schreberi* for the September data set. 21

Figure 2.6. Coefficients of association for fungal assemblages in the collection type group pairings from the July collections. 26

Figure 2.7. Coefficients of association for fungal assemblages in the collection type group pairings from the September collections. 26

Figure 3.1. Dendrogram from a sums of squares cluster analysis of the fungal assemblages of *Cladonia mitis* samples from the September data set. 35

Figure 3.2. Dendrogram from a sums of squares cluster analysis of the fungal assemblages of *Cladonia rangiferina* samples from the September data set. 36

Figure 3.3. Coefficients of association for fungal assemblages in canopy and base group pairings for the September collections. 39

Figure 4.1. Topographic map of Payuk lake, northern Manitoba, labelled with transect sites. 50

Figure 4.2. Diagram of a portion of the nuclear ribosomal DNA repeat unit, showing location and direction of primers used. 53

Figure 4.3. Gel photo of electrophoresis of PCR product from region A, showing the presence and absence of introns. 54

Figure 4.4. Gel photo of electrophoresis of PCR product from region B, showing banding pattern variation. 55

Figure 4.5. Dendrogram from a sums of squares cluster analysis of the data set from region B, with intron data from region A mapped on below the samples. 56

Figure 4.6. Topographic map of Payuk lake, northern Manitoba, labelled with transect sites, and showing textured dispersal patterns. 56

Figure 5.1. Graph of ordinations from a multiple discriminate analysis separating the fungal assemblages found in *Cladonia amaurocraea*, *C. arbuscula*, *C. rangiferina*, *C. stellaris*, *C. uncialis*, *Evernia mesomorpha*, *Peltigera* spp., *Stereocaulon alpinum*, & *Vulpicida pinastri*. 65

Chapter 1

Introduction

Fungal Assemblages in Moss & Lichen Mats of Boreal Manitoba

Introduction:

Lichens - nutrients, ground cover & decomposition

Lichens are prevalent on the floor of boreal forests (Knops *et al.*, 1991) and act as substratum filters. Highly branched fruticose lichens are particularly effective as filters, as the branching structure creates abundant traps for particulates. Particulates are trapped in considerable quantity on the external surface, and accumulate in the internal spaces of fruticose lichen thalli (Longton, 1988). This prevents nutrients in these particulates from reaching the soil.

Lichens decompose at a slower rate than vascular plants (Moore, 1984). This may be due to inhibitory substances present in lichens, affecting microbial activity (Greenfield, 1993). For example, some lichen substances, such as lichen acids, have antibiotic properties (Hale, 1983). As well, some secondary metabolites in lichens can inhibit the mineralization process (Parinkina *et al.*, 1995). Thus, lichens can also play a major role in nutrient cycling by immobilizing nutrients over large surface areas, and slowing the rate of decomposition (Longton, 1988), which may impact the accessibility of nutrients to the surrounding vegetation.

Dominant trees in boreal woodlands are *Picea mariana* (Mill.) BSP., *P. glauca* (Moench.) Voss, *Larix laricina* (Du Roi) K. Koch, *Pinus banksiana* Lamb., or *P. contorta* Dougl. (Ahti, 1997). Lichens such as *Cladonia* spp. can form as much as 50% of the above ground biomass in these forests (Ahti, 1997). Lichens may represent a potentially valuable source of nutrients, such as nitrogen in cyanobacterial lichens, a limiting element in boreal ecosystems (Greenfield, 1999).

Mosses - nutrients, ground cover & decomposition

Mosses also act as highly effective filters of particulates such as pollen, rain water, and litter (Linskens *et al.*, 1991; Oechel & Van Cleve, 1986), preventing nutrients from contacting the soil. As is the case with reindeer lichens, this is a result of the high degree of vegetative branching in the moss mat, offering abundant traps for particulates. Nutrients from these trapped particulates are directly accessible to the mosses, but not to

the surrounding vascular plant life, as many of the particulate nutrients are directly incorporated into the moss tissues, and prevented from reaching the soil. Where mosses make up a large portion of the ground cover (eg. 67-100% in a black spruce stand, and 42-70% in a white spruce stand) (La Roi & Stringer, 1976; Oechel & Van Cleve, 1986), incoming nutrients will likely be trapped.

In addition to acting as filters, many mosses also have a slow rate of decomposition, approximately 10% that of vascular plants (Oechel & Van Cleve, 1986). Decomposition in mosses is slowed by a low pH and a chemistry which prevents survival of decomposing organisms. As a result, these mosses can play a major role in nutrient cycling (Oechel & Van Cleve, 1986), by immobilizing nutrients over large areas and slowing decomposition (Longton, 1988).

Moss & Lichen Habitat Overlap

Considerable habitat overlap exists between mosses and lichens (Topham, 1997). Both mosses and lichens dominate the understory of black and white spruce forests (Ahti, 1997; La Roi & Stringer, 1976; Oechel & Van Cleve, 1986). Mosses tend to be found in the more mesic (moderately moist) sites, while lichens tend to be found in more xeric (very dry) sites (Ahti, 1997). Ground dwelling fruticose lichens generally succeed establishment of crustose and foliose lichens, and once established, remain the dominant lichen growth form throughout the lichen phase of forest succession. Mesic conditions and forest canopy closure, results in fruticose lichens being largely replaced by weft-forming mosses, such as *Pleurozium schreberi* (Brid.) Mitt. and *Hylocomium splendens* (Hedw.) B. S. G. (Johnson, 1981; Longton, 1988). Competition therefore occurs between mosses and lichens, with lichens tending to be better competitors than mosses in direct sunlight conditions. It is common for *Peltigera* lichens growing in mixed mats with feather mosses, such as *Pleurozium schreberi* and *Hylocomium splendens,* to overgrow portions of the mosses, shading them (Johnson, 1981).

Fungal Assemblages

As moss and lichen mats can play a major role in nutrient cycling (Greenfield, 1999; Oechel & Van Cleve, 1986; Longton, 1988), some work has been done on surveying

lichen and moss mats for fungi. For example, within lichen mats of *Cladonia* and *Stereocaulon,* several species of the fungal genera *Fusarium*, *Heteroconium*, and *Diplodina,* as well as *Mortierella vinacea* Dixon-Stewart and others, have been found present (Petrini *et al.*, 1990). *Fusarium* species, as well as *Mortierella vinacea*, are assumed to be living off the leachates and secondary metabolites of plant materials, such as pollen and leaf litter, trapped within lichen mats. Species of *Diplodina* and *Heteroconium* have been considered specific lichen-inhabiting fungi (Petrini *et al.*, 1990), though the exact nature of their interaction with the lichen host has not been examined.

Mosses as well, have been shown to provide a good microenvironment for supporting fungal growth (Felix, 1988). For example, *Cantharellus tubaeformis* Fr., *Cortinarius impennis* Fr., and *Paxillus involutus* (Batsch ex. Fr.) Fries have all been shown to have a close association with *Pleurozium schreberi*, though again the exact nature of these associations have yet to be determined. As well, some bryophilous ascomycetes are obligate parasites, growing exclusively on living mosses (Dobbler, 1997). Certain fungi also support moss growth. Growth promotion of moss protonema by compounds, eg. gibberellic acid, of some fungi, such as *Aspergillus flavus* Link ex. Gray, *Penicillium martensii* Biourge, *Mucor racemosus* Fresenius, & *Fusarium equiseti* (Corda) Sacc., is well known (Maltzahn & McQuarrie, 1958; Vaarama & Taren, 1959).

A wide variety of fungal assemblages can be found in lichen and moss mats. These include representatives of the Myxomycetes, Ascomycetes, Basidiomycetes, Zygomycetes, and Chytridiomycetes (Azmi & Seppelt, 1998; Carleton & Read, 1991; Ing, 1994; Moller & Dreyfuss, 1996; Thormann *et al.*, 2004 ; Thormann *et al.*, 2001). Some fungi associated with lichen and moss thalli live freely within the mats and others are biotrophic, ranging from mutualism to parasitism (Azmi & Seppelt, 1998; Carleton & Read, 1991; Ing, 1994; Moller & Dreyfuss, 1996).

Despite all the previous work on fungi from lichen and moss thalli, in past studies very often only the upper portions of the lichen and moss mats were surveyed for fungi, leaving the base of the mats unexplored to avoid results confounded by soil fungal communities (Petrini *et al.*, 1990; Pocock *et al.*, 1984). As well, no literature on temporal changes or seasonal effects on the fungi in lichen and moss mats have been reported. Finally, the literature on lichen and moss fungal assemblages, is often based around

bryophilous and lichenicolous fungi (Doebbeler, 1997 & 2001; Cole & Hawksworth, 2001). These fungi do not attack the substratum trapped within the mats, only the mats themselves. To date only two studies in the literature have set out to investigate the fungal communities that can be isolated from lichen thalli, which are not obligately lichenicolous - both of which were conducted in Europe (Girlanda *et al.*, 1997; Petrini *et al.*, 1990). Thus, literature relating to fungal assemblages within lichen and moss mats is, in general, lacking, and virtually non-existent in North-America.

Manitoba, being situated in the center of Canada, is a vegetative transition zone not only from east to west across the country, but also from south to north. Precambrian granitic rock (Canadian shield) runs from southeast to northwest through the province (Ritchie, 1956a). Manitoba is geologically very diverse (Lauhn-Jensen, 1987). Glacial lake Agassiz covered most of Manitoba at one time, creating numerous sediment deposits (Ritchie, 1956b). Due to large vegetation and habitat diversity (Ritchie, 1956a, 1956b, 1960a, & 1960b; Shay, 1984), as well as the large tracts of boreal forest which moss and lichen mats dominate (Ahti, 1997; La Roi & Stringer, 1976; Oechel & Van Cleve, 1986), Manitoba is thought to be an excellent place to study crypogams.

Given the overlap in habitat between lichen and moss mats, the slow decomposition rates of both, and the similar roles played by each as filters of particulates, fungal assemblages within moss and lichen mats ought to be similar. Hence any heterogeneity in the fungal assemblages seen between moss and lichen mats would be more likely due to the properties of the lichens, such as the presence of secondary compounds, than to environmental variables. Lichens produce a variety of secondary compounds which vary between species of lichen forming fungi (Brodo *et al.*, 2001). Thus, moss mats are thought to be an ideal control for the examination of fungi in lichen mats.

Objectives

General topics in the thesis focus on the biodiversity and biology of fungi in lichen mats, using moss mats as a control. Temporal, spatial, and environmental factors affecting the presence or absence, and species composition of fungal assemblages is explored, including potential genetic variation of the lichen mat itself.

The objectives of my studies were:

1) to determine if differences exist in the fungal assemblages found in forest floor mats of *Cladonia mitis*, *C. rangiferina*, and *Pleurozium schreberi*;

2) to determine if the fungal assemblages in forest floor mats change from the beginning to end of season;

3) to determine whether spatial differences (canopies & bases) in the fungal assemblages exist within forest floor mats of *Cladonia mitis* and *C. rangiferina*;

4) to compare the fungal assemblages in soil beneath the lichen to those in the lichen mat itself;

5) to compare population structure of fungal assemblages with that of the lichen fungus;

6) to determine fungal assemblages on species from 5 genera of lichen forming fungi.

The Thesis

Objectives listed above are sequentially addressed in the body of the thesis, running from chapters 2 through 6. Chapter 2 examines the fungal assemblages found in mats of *Cladonia mitis* Sandst., *C. rangiferina* (L.) Wigg., and *Pleurozium schreberi* in southern Manitoba and temporal changes in these assemblages (objectives 1 & 2). Chapter 3 further expands on the preliminary findings of Chapter 2 by focusing on the vertical strata within the forest floor mats of *C. mitis* and *C. rangiferina* and examining the fungal assemblages found at the soil surface beneath the lichen mats in southern Manitoba (objectives 3 & 4). Chapter 4 focuses on the fungal genetic variation within *C. arbuscula* mats to compare population structure of the lichen fungus with population structure of fungal assemblages in the lichen mat, and thus infer co-dispersal of fungal assemblages and lichen thalli (objective 5). Chapter 5 expands the number of lichen species sampled for fungi to include *Evernia mesomorpha* Nyl., *Peltigera* Willd. spp., *Stereocaulon alpinum* Laurer *ex* Funck, and *Vulpicida pinastri* (Scop.) J. –E. Mattsson & M. J. Lai, as well as increases the number of species of the genus *Cladonia* represented to

include: *C. amaurocraea* (Florke) Schaerer, *C. arbuscula* (Wall.) Rabenh., *C. rangiferina*, and *C. stellaris* (Opiz) Brodo, this time in northern Manitoba (objective 6).

Cladonia amaurocraea, *C. mitis*, *C. rangiferina*, and *C. stellaris* are among the most widely represented species of the genus *Cladonia* found in Manitoba (Piercey-Normore, 2003), and hence were chosen for study as they form highly branched fruticose lichen mats. *Stereocaulon* and *Peltigera* were chosen for study due to their nitrogen fixing abilities, *Evernia* for its epiphytic habitat, and *Vulpicida* for its foliose growth form as comparison with *Peltigera*.

Chapter 2

Comparisons of fungal assemblages found in forest mats of *Cladonia mitis*, *C. rangiferina,* and *Pleurozium schreberi* in southern Manitoba, with temporal aspects examined.

Introduction

Lichen and moss mats, particularly those of highly branched fruticose lichens, have been shown to provide a good microenvironment for fungal growth (Dobbler, 1997; Felix, 1988; Petrini *et al.*, 1990; Thormann *et al.*, 2004; Thormann *et al.*, 2001). Moss and highly branched fruticose lichens act as effective filters and trappers of particulates such as pollen and litter (Linskens *et al.*, 1991; Oechel & Van Cleve, 1986), due to their high degree of vegetative branching. Additionally, the highly branched thallus creates an environment with ameliorated temperatures and light intensities (Kershaw & Field, 1975; Kershaw & Harris, 1971). Generally, high humidity levels are maintained within the mats (Carleton & Dunham, 2003; Kershaw & Field, 1975). The microhabitat thus provides an ideal environment for fungal inhabitants.

Large overlap in habitat exists between lichen and moss mats (Topham, 1997). Given the similar environment, architecture, and roles played by each as filters of particulates, fungal assemblages within moss and lichen mats should be similar. Thus heterogeneity in assemblages seen across mats are likely due to physiology of the mat species, such as the presence of secondary compounds in lichens (Brodo *et al.*, 2001), making for interesting comparisons.

It is known that fungal assemblages are not necessarily consistent and very often change in composition over time. Keller & Bidochka (1998) examined temporal differences among soil fungal assemblages in deciduous forest, cedar forest, old field and alfalfa field. Soil type consisted of various loam series. They found that 53-85% of the species in the assemblages changed over the summer (Keller & Bidochka, 1998). Zak & Parkinson (1984) examined fungi growing on the roots of the grass *Agropyron trachycaulum* and noted species turnover rates ranging from 61-91% over a four week period. Bhat & Kaveriappa (1998) examined the aerial surface mycoflora of two endangered tree species: *Myristica fatua* var. *magnifica* and *Myristica malabarica*, in March, June, September and December, noting up to a 50% increase in species richness depending on the time substrata was sampled.

This study examines fungal assemblages on two species of reindeer lichens (fruticose lichens), i.e. *Cladonia mitis* and *C. rangiferina*. Though others have studied fungi on *C. mitis* and *C. rangiferina* (Petrini *et al.*, 1990), direct comparisons with mat

forming bryophyte biota in immediate proximity, as well as temporal surveys of fungal species compilations in mats of reindeer lichens have not been undertaken. *Pleurozium schreberi*, and its attendant fungal assemblages serves as a comparison for potential mat effects such as moisture and pH.

Materials and Methods

Collection Sites:

Four sites containing lichen mats were selected within an 8 x 8 kilometer region of the Sandilands Provincial forest. Site I was located 49° 23' N and 96° 15' W, approximately 1.8 km west from hwy. 210 along forestry trail 16, near Marchand provincial wayside park. Site II was located 49° 20' N and 96° 14' W, approximately 7 km south of hwy. 210 along forestry trail 20, near the town of Sandilands. Site III was located 49° 20' N and 96° 14' W, approximately 7.5 km south of hwy. 210 along forestry trail 20, near the town of Sandilands. Site IV was located 49° 20' N and 96° 14' W, approximately 8 km south of hwy. 210 down forestry trail 20, and off an ATV trail running northeast off trail 20.

Sites I and II contained lichen stands of *Cladonia rangiferina* and *C. mitis* mixed with the moss *Pleurozium schreberi*. Sites III and IV contained stands of *C. rangiferina* and *C. mitis* minus the presence of *P. schreberi*. Five permanent 5 x 5 meter plots were set up at each site. Plots containing *C. rangiferina*, *C. mitis*, and *P. schreberi* are hereafter referred to as mixed moss-lichen plots. Plots containing *C. rangiferina* and *C. mitis*, but no *P. schreberi*, are hereafter referred to as pure lichen plots.

Surveying Fungal Assemblages:

Collection of substratum

Prior to sampling, five of the ten plots (5x5m) in the mixed moss-lichen sites, and five of the ten plots in the pure lichen sites were selected using a random numbers generator. Within the selected plots, a grid of 25 ($1m^2$) quadrats was established and three quadrats were randomly selected (numbers generator) for sampling. The central portions of *Cladonia rangiferina* and *C. mitis* mats were sampled within each of the three $1m^2$ quadrats selected for sampling, using a 5cm diameter, 4cm deep bulk density ring. Lichen

mat samples were taken to the depth of the soil litter layer (sometimes to a depth greater than 4cm). With five plots, three samples / plot, and two species, the collecting protocol generated a total of 30 samples from the mixed moss-lichen sites, and 30 samples from the pure lichen sites (i.e. for each of the two lichen species there were 15 samples collected for each of the two site types). *Pleurozium schreberi* mats were also sampled for the moss thallus using the same procedures as for the lichens, except three randomly chosen plots were sampled at three random quadrats for a total of nine samples. All samples were placed in labelled plastic bags and taken back to the laboratory for further processing. Samples were collected on July 14th, 2003, and collections repeated again on September 28th, 2003.

Isolation of fungi

On return to the University of Manitoba Department of Botany laboratories, samples were separated into mat canopy and mat base portions (base portions being identified by their distinct lighter coloration, reduced branching, and representing almost half of the thallus height) (fig. 2.1).

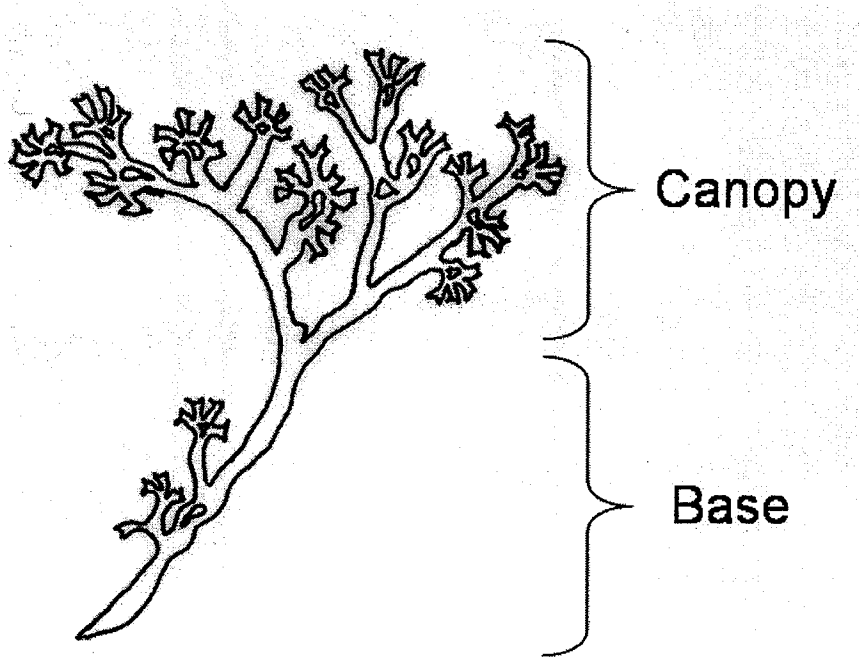


Fig. 2.1 – Simplified sketch of a *Cladonia mitis* podetium, with canopy and base strata labelled

Thallus fractions were then cut into 4-6mm lengths and 20 cut pieces of each fraction were placed into separate sterile Sartorius filter apparatuses and washed in 20 changes of sterile water to remove fungal spores (Muhsin & Booth, 1987). The washed thallus pieces were subsequently removed from the filter apparatus and equidistantly arranged on rose Bengal agar (Malloch, 1981) in lots of five per petri plate. The eight resultant petri plates required per sample, four with five plated pieces for the canopy derived pieces of the lichen, and four with five plated pieces for the 20 pieces from the base of the lichen, were incubated at ~ 20°C and checked daily for developing fungi (examination of assemblages associated with canopy and base thalli is the subject of the next chapter). Developing fungal colonies were observed and hyphae transferred to separate petri plates of potato dextrose agar (Malloch, 1981). Thus, with two collection dates, ten plots sampled, two lichen species/plot, three samples/species, eight plates/sample, and five thallus pieces per plate, ~ 4800 lichen thalli fragments were examined for fungi. Fungi from *Pleurozium schreberi* were also isolated using the same procedures except the samples were not separated into canopy and bases. With two collection dates, three plots sampled, three samples/plot, four plates/sample, and five thallus pieces per plate, ~ 360 moss thalli pieces were surveyed.

Identifications

In addition to identification of the lichens in the field, verification was done in the laboratory by detection of secondary compounds using thin-layer chromatography (TLC) (appendices 14a & 14b) according to methods described by Culberson *et al.* (1972; 1974) (vouchers of scanned images for the TLC plates are available in molecular laboratory of Dr. Piercey-Normore, U of M).

Fungi were observed for both macroscopic and microscopic morphological characteristics, and grown on several different media, including rose bengal agar, potato dextrose agar, czapek dox agar, and modified leonian's agar (Malloch, 1981), to stimulate the production of spores. Fungal identifications were based on Ainsworth (1973), Barnett & Hunter (1972), Dennis (1968), Ellis (1971; 1976), Hesseltine (1955), Hesseltine & Ellis (1973), Malloch (1981), Martin & Alexopoulos (1969), Miller (1984), Singer

(1986), and von Arx (1982). Where fungi could not be identified to species, the lowest taxonomic level of identification determined was stated, with a no. proceeding the taxon. Isolates were then grouped by morphotype into the appropriate numbered taxon.

Meteorological Data

Daily precipitation, maximum temperature, and minimum temperature for three weeks prior to the July and September collection periods was retrieved from Environmental Canada (2003), from the St. Labre and La Broquerie recording stations.

Data Analysis:

Fungal assemblage frequency data

Frequencies of individual fungal taxa on the moss and lichen thalli were taken as the number of finds divided by the number of starts (in this case 20) from each of the mat samples (canopy and base fractions). Fungal assemblage frequency data was subjected to three multivariate analysis methods: 1) Sums of squares cluster analysis; 2) Principle component analysis; and 3) Multiple discriminate analysis. All multivariate analyses were conducted using the program Syn-tax 2000 – Hierarchical Classification (Podani, 2001).

Sums of squares cluster analyses

Prior to analysis, fungal assemblage frequency data was pooled by sample, combining canopy and base fractions. Assemblage frequency data was then subjected to a sums of squares cluster analysis using a chord distance (scaled Euclidean distance) association coefficient for both the July and September collections (Kenkel & Booth, 1992).

Principle component analyses

Subsequent to the sums of squares cluster analyses, the unpooled fungal assemblage frequency data was subjected to a logarithmic transformation, followed by a principle component analysis for both the July and September data sets (Kenkel & Booth, 1992).

Multiple discriminate analyses

Component scores for the first three component axes from the principle component analysis were then subjected to a multiple discriminate analysis, attempting to separate mat samples by species (i.e. *Pleurozium schreberi*, *Cladonia mitis* & *C. rangiferina*) for both the July and September collections (Kenkel & Booth, 1992). Group centroids constructed were then tested to determine if they were significantly different using a χ^2 test (Moore, 2000).

Data Pooling

Fungal assemblage frequency data was also processed by pooling mat samples together, constructing five 'collection type groups' for both the July and September data sets: 1) *Cladonia mitis* samples from pure lichen plots (LM); 2) *C. mitis* samples from mixed moss-lichen plots (MM); 3) *C. rangiferina* samples from pure lichen plots (LR); 4) *C. rangiferina* samples from mixed moss-lichen plots (MR); and 5) *Pleurozium schreberi* samples from mixed moss-lichen plots (P).

Average species richness

Average species richness was calculated for each of the collection type groups for both the pooled July and September data sets (Krebs, 1972). Average species richness was calculated by summing the total number of fungal taxa in each plot of a collection type group (five plots are in a collection type group), and dividing it by the total number of plots in the collection type group (Krebs, 1972).

Shannon-Wiener index

Both diversity and taxa evenness were determined using the Shannon-Wiener index, sometimes called Shannon's index. These parameters were calculated for each mat type collection from both the pooled July and September data sets using the following formulae:

$$H = -\sum_{i=1}^{s} p_i \ln p_i$$

where H equals the index of species diversity, s equals the number of fungal species and p_i equals the proportion of the total sample that belongs to the ith species, and

$$E = \frac{H}{H_{\max}}$$

where E equals the evenness or equitability of the distribution (range = 0-1), H equals the observed species diversity, and H_{max} equals the maximum species diversity(i.e. ln(s)) (Krebs, 1972).

Occurrence

Occurrence was calculated for individual fungal taxa in the fungal assemblages from *Cladonia rangiferina*, *C. mitis*, and *Pleurozium schreberi*, by plot type (pure lichen or mixed moss-lichen plots), for both July and September data sets. Occurrence is the number of observations for a fungal taxon, over the collections for each collection type group. Occurrences for each fungal taxon were expressed as a percentage of the number of times a taxon was observed divided by 15 in the case of the lichens sampled, and by nine in the case of the fungal taxa from *P. schreberi* mat samples.

Coefficients of association

Coefficients of association (Krebs, 1972) for *Cladonia rangiferina*, *C. mitis*, and *P. schreberi* were calculated using collection type group, and the fungal assemblages isolated. Coefficients of association were calculated for collection type groups using the following formula:

$$V = \frac{ad - bc}{\sqrt{(a+b)(c+d)(a+c)(b+d)}}$$

where *a* equals the number of fungal taxa common to both groups, *b* equals the number of fungal taxa exclusive to group 1, *c* equals the number of fungal taxa exclusive to group 2, and *d* equals the number of fungal taxa excluded from both groups (Krebs, 1972).

Meteorological data

Data was broken up into three and one week periods, and mean max., min., and average temperatures, and respective standard deviations calculated, as well as number of days with precipitation, range of precipitation, total precipitation, last rainfall ≥ 2cm/24hrs. and mean precipitation with standard deviation determined.

Results

Fungi of the genera *Alternaria*, *Aureobasidium*, *Cladosporium*, *Epicoccum*, *Mortierella*, *Mucor*, *Penicillium*, and *Trichoderma* were isolated from lichen and moss mat samples taken during the July collections. As well, three taxa from the Sphaeropsidales were found and four different taxa of mycelia sterilia encountered (appendix 1a). Isolates of *Aureobasidium pullulans*, Sphaeropsidales, mycelia sterilia, and two taxa of *Mortierella* (# 3 & # 4) were unique to the July collections.

In the September collections, fungi of the genera *Absidia*, *Alternaria*, *Aspergillus*, *Cladosporium*, *Cunninghamella*, *Epicoccum*, *Mortierella*, *Mucor*, *Penicillium*, *Rhizopus*, and *Trichoderma* were isolated (appendix 2a). No Sphaeropsidales, mycelia sterilia, or *Aureobasidium pullulans*, were found in the September collections. Isolation of taxa of the genera *Absidia*, *Aspergillus*, *Cunninghamella*, and *Rhizopus* were unique to the September collections. Among the genera isolated in both the July and September lichen and moss mat collections, one taxon of *Mucor* (#5) and one taxon of *Cladosporium* (#2) were also unique to the fall collection.

Aureobasidium pullulans, *Epicoccum purpurascens*, *Mortierella* # 4, mycelia sterilia # 38, Sphaeropsidales # 36 and # 37, and a sclerotia forming taxon of *Penicillium*

were isolated only once during the July survey. All of the September fungal taxa were isolated multiple times.

In the July collections, the sums of squares analysis on the matrix of frequencies of fungal taxa (appendix 1a) over all mat collections, revealed clusters of *Pleurozium schreberi* samples (fig. 2.2, cluster B & C). Similarly, the sums of squares analysis on the matrix of frequencies from the September collections (appendix 2a), also revealed a cluster of *P. schreberi* samples (fig. 2.3, cluster C).

Fig. 2.2 – Sums of squares cluster analysis of July samples using chord distance as the association measure. Underlined samples are *Pleurozium schreberi*. Numbers represent samples in order of appearance in appendix 1a.

Fig. 2.3 – Sums of squares cluster analysis of September samples using chord distance as the association measure. Underlined samples are *Pleurozium schreberi*. Numbers represent samples in order of appearance in appendix 2a.

Multiple discriminate analyses for both the July and September matrix of frequencies of fungal taxa in mat collections, yielded significant results ($p = 0.05$) (appendices 3a-3c and 4a-4c). The chi squared values were 13.75, degrees of freedom (d.f.) = 6, and 35.97, d.f. = 6, for the July and September collections respectively. Figures 2.4 & 2.5 clearly indicate that fungal assemblages found in *Pleurozium schreberi* are significantly different from those found in *Cladonia rangiferina* and *C. mitis*. Also, figures 2.4 & 2.5 indicate that the fungal assemblages in *C. rangiferina* and *C. mitis* are not significantly different from one another.

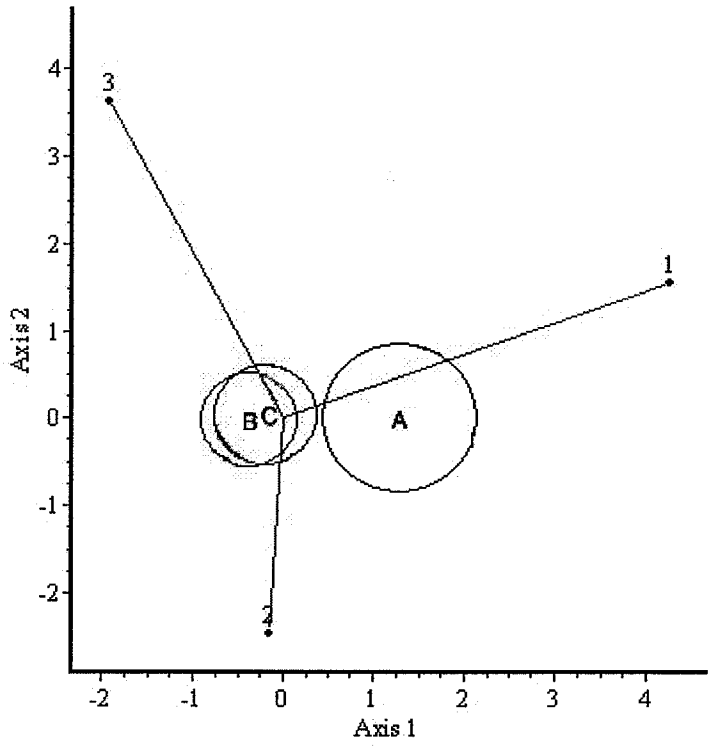


Fig. 2.4 - Graph of ordinations from a multiple discriminate analysis of the object scores from a principle component analysis for the fungal assemblages found in *Cladonia mitis, C. rangiferina, & Pleurozium schreberi* for the July collections. Circle A represents the 95% confidence interval (C.I.) for samples of *P. schreberi*, circle B represents the 95% C. I. for samples of *C. mitis*, and circle C represents the 95% C.I for samples of *C. rangiferina*.

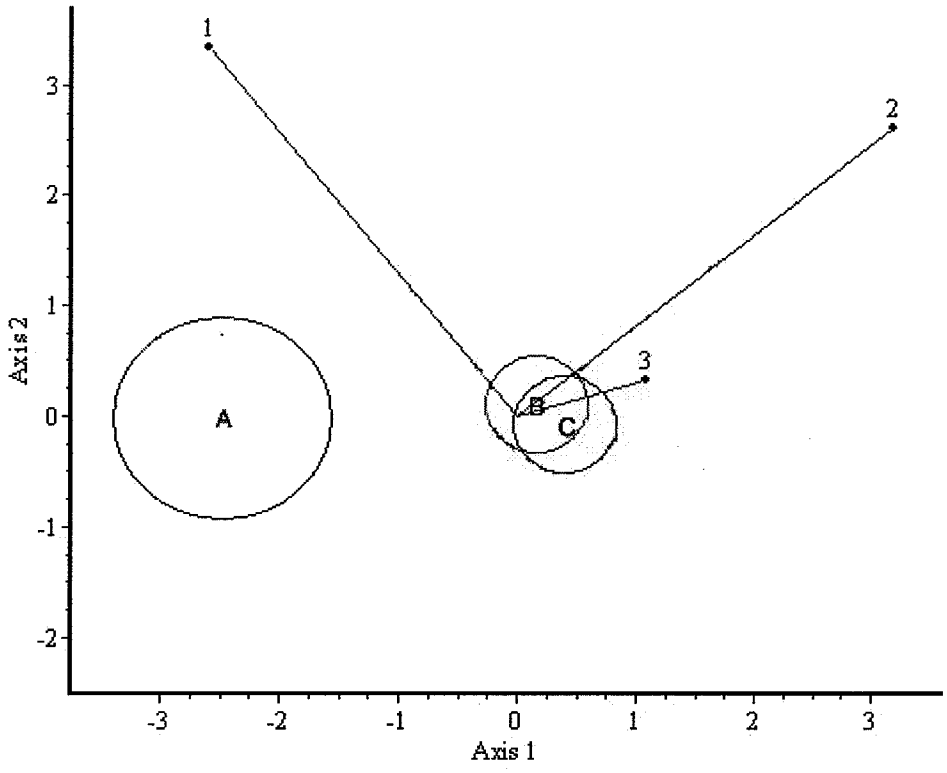


Fig. 2.5 - Graph of ordinations from a multiple discriminate analysis of the object scores from a principle component analysis for the fungal assemblages found in *Cladonia mitis, C. rangiferina, & Pleurozium schreberi* for the September collections. Circle A represents the 95% confidence interval (C.I.) for samples of *P. schreberi*, circle B represents the 95% C. I. for samples of *C. mitis*, and circle C represents the 95% C.I for samples of *C. rangiferina*.

Diversity indices for pooled matrix data (canopy and base fractions combined) for the July collections (appendix 5a) indicated that the highest fungal assemblage species richness was in mats of *Pleurozium schreberi* (P). Species richness in the mats declined as follows: *Cladonia rangiferina* in the pure lichen plots (LR), *C. mitis* in pure lichen plots (LM), *C. mitis* in mixed-moss lichen plots (MM), followed by *C. rangiferina* in mixed moss-lichen plots (MR) (table 2.1).

Table 2.1 - Species Richness & Shannon-Wiener evenness diversity indices for the pooled July & September fungal assemblage collection type groups.

	July		September	
Pooled Samples	**Species Richness**	**Evenness**	**Species Richness**	**Evenness**
LM:	3.8	0.39	9.6	0.53
MM:	2.8	0.25	9.6	0.42
LR:	4.4	0.31	10.4	0.55
MR:	1.8	0.11	9.6	0.33
P:	5	0.27	10.33	0.31

LM = *Cladonia mitis* samples from pure lichen plots
MM = *Cladonia mitis* samples from mixed moss-lichen plots
LR = *Cladonia rangiferina* samples from pure lichen plots
MR = *Cladonia rangiferina* samples from mixed moss-lichen plots
P = *Pleurozium schreberi* samples from mixed moss-lichen plots

Evenness within the fungal assemblages over the collection types (P, LR, LM, MM, MR) was highest in samples from *C. mitis* in pure lichen plots (LM). Evenness declined in order as follows: LR > P > MM followed by MR (table 2.1). All the fungal taxa within collection type groups appear to be uneven (< 0.4) and relatively low in species richness.

In the September mat collections (appendix 6a), the highest fungal assemblage species richness was found in mats of *Cladonia rangiferina* when sampled in the pure lichen plots (LR). Species richness in the mats declined as follows: *Pleurozium schreberi* (P), followed by the remaining three collection type groups being equal in their species richness (table 2.1). The evenness of the distribution of fungal assemblages within each group was highest in samples of LR. Evenness declined in order as follows: LM > MM > MR followed by P (table 2.1). Mixed moss-lichen collection type groups are low in their evenness of distributions (< 0.4) of fungal assemblages, while the pure lichen collection type groups are moderate (> 0.4 & < 0.6) in the evenness of their fungal assemblages.

Species richness in the September collections was considerably higher ($\bar{x} = 9.9$) than in the July collections ($\bar{x} = 3.6$) (table 2.1). Evenness of the fungal taxa on lichen thalli increased moderately (14-24%) in the September collections from the July collections. In *Pleurozium schreberi* samples, evenness of the mycota remained essentially the same (3% increase) (table 2.1).

On examination of occurrences from the July collections *Alternaria* was only found in pure lichen plots. *Trichoderma* was not found in mats of *Pleurozium schreberi.*

Mucor # 1 and mycelia sterilia # 14 were the only cosmopolitan fungal taxa across the collection type groups. Mycelia sterilia # 14, on average, had the highest occurrence in the fungal assemblages across the collection type groups (table 2.2).

Table 2.2 – List of fungal assemblages and occurrences for collection type groups for the July & September collections.

	LM		MM		LR		MR		P	
	July	Sept.	July	Sept.	July	Sept.	July	Sept.	July	Sept.
Absidia coerulea Bain.	-	1	-	0.8	-	0.8	-	0.8	-	1
Alternaria spp.	0.4	1	-	1	0.4	1	-	1	-	1
Aspergillus alutaceus group	-	-	-	-	-	-	-	0.2	-	-
Aureobasidium pullulans (de Bary) Arnaud	-	-	-	-	-	-	0.2	-	-	-
Cladosporium # 1	0.2	0.2	-	0.4	-	0.6	0.2	1	0.67	0.33
Cladosporium # 2	-	1	-	1	-	1	-	-	-	0.67
Cunninghamella elegans Lendner	-	-	-	-	-	0.2	-	-	-	-
Epicoccum purpurascens Ehrenb. Ex. Schlecht	0.2	1	-	0.6	-	1	-	0.8	-	0.67
Mortierella isabellina/vinacea complex	-	0.8	0.4	1	0.4	0.8	0.4	0.8	0.67	1
Mortierella # 3	-	-	-	-	0.2	-	-	-	0.33	-
Mortierella # 4	0.2	-	-	-	-	-	-	-	-	-
Mucor # 1	0.4	0.8	0.4	1	0.6	1	0.2	1	0.67	1
Mucor # 4	0.4	0.2	-	0.2	0.4	0.4	-	-	0.67	0.33
Mucor # 5	-	-	-	-	-	0.2	-	-	-	-
Mucor # 10	-	0.8	-	1	0.2	1	-	1	0.33	1
Penicillium (non-sclerotial)	0.2	1	0.2	1	0.6	0.8	-	0.8	0.33	1
Penicillium (sclerlotial)	0.2	0.2	-	-	-	0.4	-	0.8	-	0.67
Rhizopus oryzae Went & Prinsen Geerlings	-	0.6	-	0.4	-	0.2	-	0.2	-	0.67
Rhizopus # 2	-	-	-	0.2	-	-	-	0.2	-	-
Sphaeopsidales # 23	-	-	0.2	-	-	-	-	-	0.33	-
Sphaeropsidales # 36	-	-	-	-	0.2	-	-	-	-	-
Sphaeropsidales # 37	0.2	-	-	-	-	-	-	-	-	-
Sterile # 1	0.2	-	0.2	-	0.4	-	-	-	0.33	-
Sterile # 14	0.6	-	0.6	-	0.4	-	0.4	-	0.67	-
Sterile # 24	0.2	-	-	-	0.2	-	0.2	-	-	-
Sterile # 38	-	-	0.2	-	-	-	-	-	-	-
Trichoderma spp.	0.4	1	0.4	1	0.4	1	0.2	1	-	1

LM = *Cladonia mitis* samples from pure lichen plots
LR = *Cladonia rangiferina* samples from pure lichen plots
MM = *Cladonia mitis* samples from mixed moss-lichen plots
MR = *Cladonia rangiferina* samples from mixed moss-lichen plots
P = *Pleurozium schreberi* samples from mixed moss-lichen plots

Although *Alternaria* and *Trichoderma* were not found in mats of *Pleurozium schreberi* in the July collections, they were found in mats of *Pleurozium schreberi* in the September collections (table 2.2).

Only two of the twenty fungal taxa isolated in the July collections were cosmopolitan, i.e. in all of the collection type groups. In the September collections, ten out of the 17 fungal taxa: *Absidia coerulea*, *Alternaria* spp., *Cladosporium* #1, *Epicoccum purpurascens*, *Mortierella isabellina/vinaceae* complex, *Mucor* # 1 and # 4, *Penicillium* spp. (non-sclerotial), *Rhizopus oryzae*, and *Trichoderma* spp., were cosmopolitan (table 2.2) (photographs of select fungal taxa can be seen in appendix 15a).

September collection type groups contained more fungal taxa then the July collection type groups. The mean occurrences of fungal taxa were also considerably higher for the September collections (table 2.3). *Aspergillis* (*alutaceus* group) was only isolated from *C. rangiferina* in mixed moss-lichen plots, while *Cunninghamella elegans* was only isolated from *C. rangiferina* in pure lichen plots (table 2.2).

Table 2.3 - Mean occurrences of the fungal assemblages found for each collection type group for the July and September collections.

	July	September
P:	0.5	0.80
LM:	0.29	0.74
LR:	0.37	0.69
MM:	0.33	0.74
MR:	0.26	0.74

LM = *Cladonia mitis* samples from pure lichen plots
MM = *Cladonia mitis* samples from mixed moss-lichen plots
LR = *Cladonia rangiferina* samples from pure lichen plots
MR = *Cladonia rangiferina* samples from mixed moss-lichen plots
P = *Pleurozium schreberi* samples from mixed moss-lichen plots

In the July collections there were no strong associations (< 0.5) across any of the pairings of the five collection type groups (fig. 2.6).

	MM	LR	MR	P
LM	-0.04	0.04	0.10	-0.03
	MM	0.25	0.26	0.41
		LR	0.17	0.41
			MR	0.11

Fig. 2.6 - Coefficients of association for fungal assemblages in the collection type group pairings from the July collections. See table 2.3 for acronym explanations.

In contrast, the coefficients of association data for the September collections revealed strong positive associations (< 0.6) between LM and MM, LM and LR, LM and P, MM and P, and LR and P (fig.2.7).

	MM	LR	MR	P
LM	0.67	0.66	0.35	1
	MM	0.23	0.35	0.67
		LR	-0.20	0.66
			MR	0.35

Fig. 2.7 - Coefficients of association for fungal assemblages in the collection type group pairings from September collections. See table 2.3 for acronym explanations.

Meteorological data (appendices 7a & 8a) revealed approximately twice the number of days with precipitation both one and three weeks preceding the September collections as compared to the July collections (table 2.4). Total precipitation one week prior to collections was approximately the same for July and September. Total precipitation over the three weeks prior to collections was nearly double in September then June/July. No rainfall > 2cm within 24hrs. was recorded in the three weeks prior to the July collections, while one rainfall event >2cm occurred 11 days prior to the September collections. Daily mean precipitation was the same prior to the July and September collections. Mean high, low and average temperatures over the three weeks prior to collections did not differ between June/July and September. Mean high and average temperatures one week prior to collections was ~10°C cooler in September compared to July (table 2.4).

Table 2.4 – Meteorological data for the 3 weeks preceding sampling of the July and September collections.

	June-July/ 03		September/ 03	
	07/07-13/07	23/06-13/07	21/09-27/09	07/09-27/09
# of days with precipitation [1]	3	9	7	16
Range of precipitation (mm) [1]	0 - 8	0 - 13.4	0 - 4	0 - 37.8
Total precipitation (mm) [1]	10.8	43	12.4	78.9
Last rainfall > 2cm/24hrs. [1]	-	> 21 @ 29 days	-	11 days ago
Mean precipitation (mm) [1]	1.5 ± 3	2.1 ± 3.8	1.8 ± 1.8	3.8 ± 8.1
Max mean temp. (°C) [2]	20.1 ± 3.4	23.5 ± 3.8	10 ± 3.9	17.5 ± 6.6
Min. mean temp. (°C) [2]	8.6 ± 3.3	11.1 ± 3.5	5.8 ± 3.4	7.5 ± 5.8
Mean period temp. (°C) [2]	14.4 ± 3	17.3 ± 3.2	5.9 ± 1.9	12.5 ± 5.7

[1] = derived from the St. Labre, Manitoba, Environment Canada recording station.
[2] = derived from the La Broquerie, Manitoba, Environment Canada recording station.

Discussion

Fungi from the genera *Alternaria*, *Cladosporium*, *Penicillium*, and *Trichoderma*, as well as *Epicoccum purpurascens* and *Mortierella vinaceae* were previously reported from *Cladonia mitis* and *C. rangiferina* (Petrini *et al.*, 1990). Occurrences of *Absidia coerulea*, *Aspergillus* (alutoceus group), *Cunninghamella elegans*, *Mortierella isabellina* Oudem., *Mucor* spp., and *Rhizopus* spp. from *C. mitis* and *C. rangiferina* mats are reported here for the first time. *Alternaria* spp., *Aspergillis* spp., *Cladosporium* spp., *Cunninghamella* spp., *Mortierella* spp., *Mucor* spp., and *Penicillium* spp. are common inhabitants of soil (Keller & Bidochka, 1998) and as such soil may serve as inoculum for lichen thalli as well as on moss gametophytes.

My data indicated that fungal assemblages found in *Pleurozium schreberi* were different from those of *Cladonia mitis* and *C. rangiferina* (figs. 2.2, 2.3, 2.4, & 2.5). In the moss mats, frequencies of *Alternaria*, *Epicoccum*, and *Trichoderma* were lower then the frequencies of these fungal taxa from lichen mats. *Mucor* and *Mortierella* frequencies in the moss mats were higher then in lichen mats. No significant differences in fungal frequencies between the two species of lichen mats were detected. This is consistent with Petrini *et al.* (1990) who surveyed the fungal assemblages of lichen mats (including seven species of *Cladonia*), and found no fungal taxa preferentially colonizing a given species of lichen forming fungi. The frequencies of *Alternaria*, *Epicoccum*, *Trichoderma*, *Mucor* and *Mortierella* were approximately the same on *C. mitis* and *C. rangiferina* thalli. Despite the often close proximity of moss and lichen samples taken from mixed moss-

lichen plots, sometimes not more than several inches apart, there was a different composition of taxa in the fungal assemblages from lichen and moss mats.

Average fungal species richness was higher in samples of *Pleurozium schreberi* than from either of the two lichen thalli (table 2.1). Lower fungal species richness from lichen samples than from the moss collections was probably a function of higher moisture levels assumed to be present in those plots containing *P. schreberi*. Due to its morphology, *P. schreberi* captures atmospheric precipitation more effectively, and holds moisture longer than either species of *Cladonia* (Ipatov & Trofimets, 1988). Canopies of *Cladonia alpestris* (syn. *C. stellaris*) have been shown to dry out within 9 hours of saturation. The mid-canopy zone takes between 12-18 hours to dry out, and the base of the lichen mat remains at 50% saturation or more 24 hours after wetting (Kershaw & Rouse, 1971). Mats of *P. schreberi* and *Cladonia* spp., growing virtually intertwined amongst each other, as observed in the mixed moss-lichen plots, have less of a moisture gradient than in the pure lichen plots. Thus, the lichen bases and canopies of thalli included in a moss mat tend to have a similar humidity level and are wetter than large pure stands of lichens where there is a definite gradient of lower moisture (canopy) to higher moisture (base).

In this study, wetter sites were shown to favour fungi which are adapted to higher moisture levels. Fungi better adapted to dryer conditions survive in the pure lichen plots where the lichen canopies dry out faster (Kershaw & Rouse, 1971), and a moisture gradient is established. Concomitantly, a moisture gradient promotes a greater diversity of fungi, due to moist habitat niches at the lichen bases, and dry habitat niches in the lichen canopies. Trends in my data tended to confirm this, since even though *Pleurozium schreberi* had the highest species richness on average, the overall number of unaveraged fungal taxa found in the pure lichen plots for both samples of *Cladonia rangiferina* and *C. mitis* were higher (table 2.3).

Though there was a significant difference between fungal assemblages found in *Pleurozium* samples and lichen species, September collected thalli of *Cladonia rangiferina* in pure stands showed the highest species richness. The overall increase in species richness and occurrence in the September collections as compared to the July

collections may be explained by increased September rainfall and cooler temperatures (table 2.4), which facilitate fungal growth (Keller & Bidochka, 1998).

Increased number of rainfall events and total precipitation over the three weeks prior to September collections were nearly doubled compared to the three weeks prior to July collections. While total precipitation within one week of collections was similar for July and September, both max. and average mean temperatures were ~10 °C cooler one week prior to collections for September compared to July (table 2.4). Cooler temperatures combined with increased frequencies of rainfall events decrease the amount of evaporation which takes place, resulting in a wetter forest floor in September collections. Moss and lichen mats may have become saturated, resulting in a more homogenous moisture regime in both the mixed moss-lichen and pure lichen plots. Thus the increased moisture holding capacity of *Pleurozium schreberi* (Ipatov & Trofimets, 1988) no longer creates differences in moisture levels between plot types in the September collections, as increased precipitation rates keep pure lichen plot canopies more mesic. This would explain the greater equality in species richness among the collection type groups in the September collections. Finally increased evenness in the September fungal assemblages, contrasted with the July mycota, could be a result of the increased occurrences of fungi due to the cooler temperatures, increased precipitation, and drying events.

The low coefficients of association between the five types of collections (Pure stands of *Cladonia mitis*; pure stands of *C. rangiferina*; *C. mitis* in moss; *C. rangiferina* in moss; *Pleurozium schreberi*) for the July collections indicate a lack of association between the groups. (fig. 2.6). The assemblages were neither similar nor different from each other. However, the coefficients of association for the September fungal assemblages were reasonably strong. These associations indicated that the *P. schreberi* and *Cladonia* assemblages were similar. Again the similarity was most likely a result of the September moisture and temperature regimes and the biotic and physical nature of the mats (Ipatov & Trofimets, 1988).

The fact that the nature of moisture gradients in lichen mats affect fungal taxa richness, evenness, and assemblage associations begs the question of assemblage differences, and similarities, in mat canopies and bases due to potential moisture gradients. Are fungal assemblages in the canopy and base of individual lichens different?

Chapter 3

Spatial comparisons of the fungal assemblages found in mats of *Cladonia mitis* and *C. rangiferina* in southern Manitoba

Introduction

Vertical stratification within fruticose lichen mats has been demonstrated for a number of *Cladonia* species, including *C. arbuscula* and *C. rangiferina* (Kershaw & Field, 1975; Kershaw & Harris, 1971). Three microclimate variables, i.e. temperature, moisture, and light, are important not only to the fungal assemblages present within *Cladonia* mats, but are also metabolically important to the lichens themselves, varying considerably within lichen mats (Kershaw & field, 1975). For example, under conditions of partial cloud cover, a temperature difference of 17°C was observed between the canopy and base within a *C. alpestris* mat (Kershaw & Field, 1975). Canopies of *Cladonia alpestris* have been shown to dry out within 9 hours of saturation, where as the midcanopy zone of *C. alpestris* takes 12-18 hours to dry out. The base of the lichen mat remains at 50% saturation or more 24 hours later. Such differential drying creates a pronounced moisture gradient within the mat (Kershaw & Rouse, 1971). Light intensities are highly stratified within heavily branched *Cladonia* mats. Along the vertical profile of *C. alpestris*, light intensities have been shown to drop from 100% full illumination at the top of the canopy to 10% in the first one-third of the height of the mat thalli (Kershaw & Harris, 1971). Concomitantly, living symbiotic algal cells were not observed in the bottom two-thirds of the thalli (Kershaw & Harris, 1971). *Cladonia arbuscula* has demonstrated a similar light profile, dropping from 100% full illumination to 10% approximately two-fifths the way down the mat thalli, with similar loss of living algal cells (Kershaw & Harris, 1971). *Cladonia rangiferina* has a more open canopy, dropping from 100% full illumination to 10% three-quarters of the height of the lichen mat thalli (Kershaw & Harris, 1971). Despite increased light penetration, living algal cells in *C. rangiferina* mats are not observed at illumination levels of 40%. This suggests that some other parameter, e.g. age of the podetia, rather than light intensity affects the presence and absence of living algal cells (Kershaw & Harris, 1971).

Microhabitat differences, as a function of light intensities, temperature, moisture and the physiology and biochemistry of the living algal symbiont, establishes different living conditions for fungal assemblages in the canopy and base portions of the lichen mats.

Fungal assemblages have been surveyed from soil (Keller & Bidochka, 1998) and the canopies of lichen and moss mats (Petrini *et al.*, 1990; Thormann *et al.*, 2004; Thormann *et al.*, 2001). In the cited studies, fungi from thalli bases have often not been studied with the view that fungal inhabitants of the soil and thalli lichen bases are the same. Petrini *et al.* (1990) proposed surveying epiphytic rather than terrestrial lichens for fungal assemblages, to avoid results confounded by soil fungal communities, but never verified the presence of soil fungal communities in the bases of lichen mats. To date, study of the vertical profile of the fungal assemblages within *Cladonia* mats has yet to be reported in the literature.

Materials and Methods

Collection Sites:

Sites and plots within the 8 x 8 kilometer region of the Sandilands Provincial forest, as described in Chapter 2 (sect. *collection sites*), were utilized in this study.

Lichen Assemblages

Surveying for Fungal Assemblages and Data Analysis:

Collection, isolation and identification methods for fungal materials were employed as described in Chapter 2 (sect. *surveying fungal assemblages*). September data from the previous chapter was left segregated into canopy and base fractions. Frequency characterization, sums of squares cluster analyses, principle component analyses and multiple discriminate analyses were performed. Frequency characterizations were calculated as previously detailed (Chapter 2 – sect. *data analysis*). Sums of squares cluster analyses were conducted mathematically as previously described (Chapter 2 – sect. *data analysis*), separately on *Cladonia mitis* and *C. rangiferina* samples (Kenkel & Booth, 1992). Log transformed fungal assemblage frequency data was subjected to principle component analyses (PCA) separately on both the *C. mitis* and *C. rangiferina* samples, as well as for the combined data set. PCA analyses were conducted mathematically the same as previously described (Chapter 2 – sect. *data analysis*). The first three component axes from the principle component analyses were subjected to multiple discriminate analyses (MDA), examining any separation of the fungal

assemblage species composition found between canopies and bases of each lichen species, and canopies or bases across lichen species (Kenkel & Booth, 1992). MDA analyses were conducted mathematically in the same manner previously described (Chapter 2 – sect. *data analysis*).

Species richness, and evenness, occurrence, and coefficients of association were determined for each of the canopy and base groups. Detailed explanations for their calculation is provided in Chapter 2 (sect. *data analysis*), and by Krebs (1972). Average species richness was derived by summing the total number of fungal taxa in each plot of a specific canopy or base group and dividing it by the total number of plots in the group (five).

Occurrence, expressed as a percentage of the number of observations of a fungal taxon divided by the total number of collections (usually 15), was calculated for the mat vertical profile group, i.e. canopy and base portions of the thalli.

Data organization

Fungal assemblage frequency data was further organized by grouping mat samples together, constructing eight 'canopy and base' groups. Canopy groups included: 1) *Cladonia mitis* canopy samples from pure lichen plots (LMC); 2) *C. mitis* canopy samples from mixed moss-lichen plots (MMC); 3) *C. rangiferina* canopy samples from pure lichen plots (LRC); 4) *C. rangiferina* canopy samples from mixed moss-lichen plots (MRC). Base groups included: 5) *C. mitis* base samples from pure lichen plots (LMB); 6) *C. mitis* base samples from mixed moss-lichen plots (MMB); 7) *C. rangiferina* base samples from pure lichen plots (LRB); and 8) *C. rangiferina* base samples from mixed moss-lichen plots (MRB).

Soil Assemblages

Surveying for Fungal Assemblages:

Collection of substratum

Fifteen soil samples from beneath *Cladonia rangiferina* mats were collected, using a 4cm deep, 5cm diameter bulk density ring, from sampling three of the ten selected 5x5m plots (five samples per plot) previously studied for lichen assemblage fungi. Similarly, 15 soil

samples were taken beneath mats of *Cladonia mitis* from three of the ten selected plots previously studied. The five samples per plot were randomized over 25 1 x 1m quadrats within the selected plots. Samples were placed in labelled plastic bags and returned to the laboratory for further processing.

Isolation of fungi

Soil samples were sifted through a clean 1.7mm sieve. Captured litter was transferred to a sterilized Sartorius filter apparatus and washed in 20 changes of sterile distilled water to remove fungal spores (Muhsin & Booth, 1987). For each soil sample, 20 washed pieces, 4-5 mm in length, of litter (senescent lichen material, rotted twigs etc.) were subsequently removed aseptically from the filter apparatus and equidistantly arranged in lots of five per petri plate on rose Bengal agar (Malloch, 1981). Four petri plates per sample were incubated at ~ 20°C and checked daily for developing fungi. Developing fungal colonies were observed and hyphae transferred to separate petri plates of potato dextrose agar (Malloch, 1981).

Occurrence

Utilizing a frequency table (appendix 11a), occurrence was calculated for the combined soil fungal assemblages of *Cladonia rangiferina* and *C. mitis* by plot type, (i.e. the number of lichen samples containing a specific fungal species collected from a specific plot type, over the total number of lichen samples of that plot type). Frequencies of individual fungal taxa on washed soil litter were taken as the number of finds divided by the number of starts (in this case 20) from each of the soil samples.

Results

Cluster analyses of the frequencies of fungal taxa over all canopy and base groups for *Cladonia mitis* (appendix 9a) and *C. rangiferina* (appendix 10a) mat collections showed separation of collections of the thallus parts with their different fungi. *C. mitis* canopy samples grouped together predominantly in cluster B, while bases grouped in cluster A (fig. 3.1). *C. rangiferina* canopy samples clustered in 'A' and base collections grouped in 'B' (fig. 3.2).

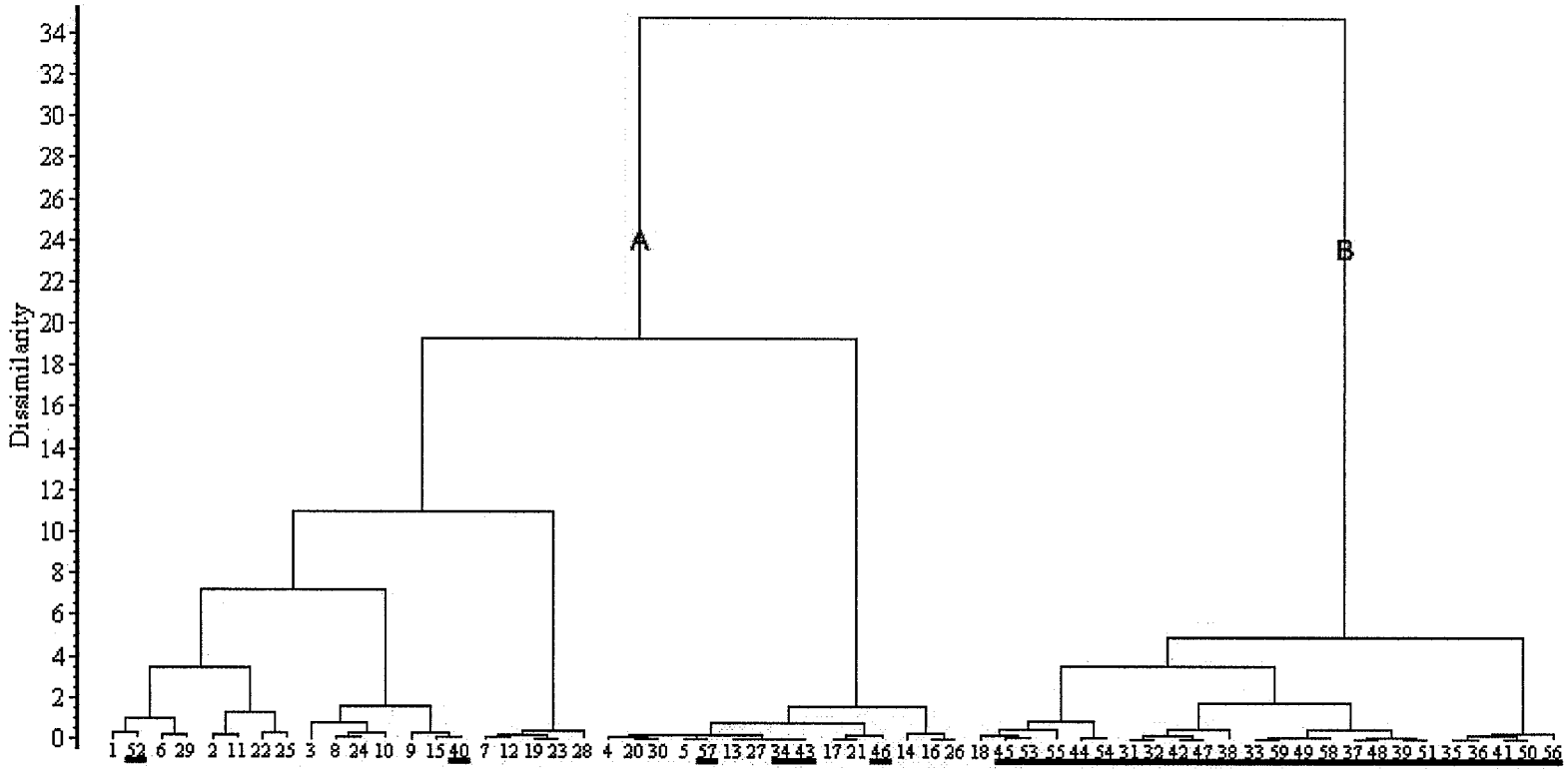


Fig. 3.1 – Sums of squares cluster analysis of *Cladonia mitis* samples using chord distance as the association measure. Black bars represent *C. mitis* canopy samples, while the other samples are *C. mitis* bases. Numbers represent samples in order of appearance in appendix 7a.

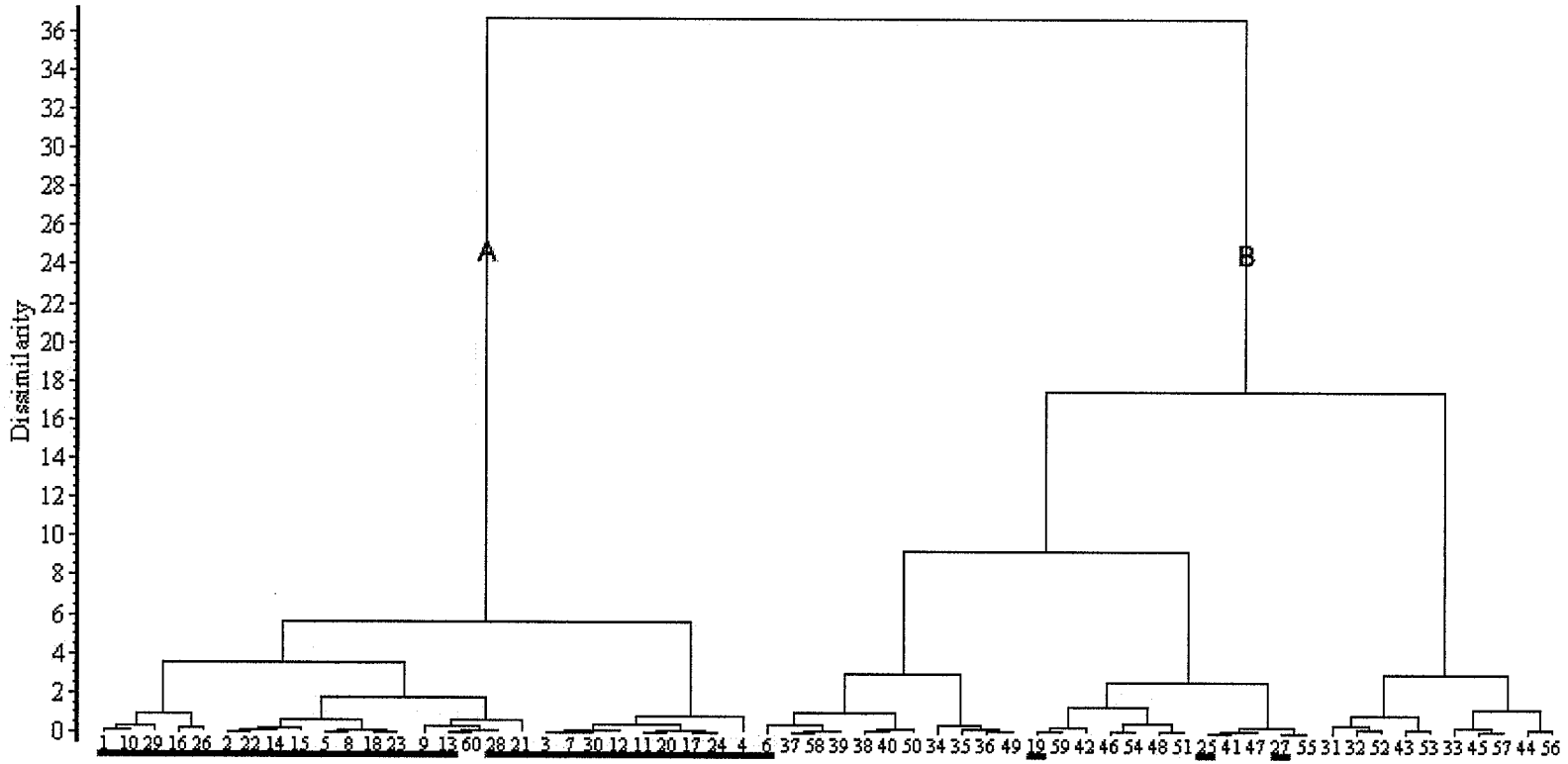


Fig. 3.2 – Sums of squares cluster analysis of *Cladonia rangiferina* samples using chord distance as the association measure. Black bars represent *C. rangiferina* canopy samples, while the other samples are *C. rangiferina* bases. Numbers represent samples in order of appearance in appendix 8a.

Multiple discriminate analyses of the canopy portion of both lichens for the matrix of frequencies of fungal taxa by thallus samples, showed no significant differences between *Cladonia mitis* and *C. rangiferina*. Similarly, comparisons of *C. mitis* and *C. rangiferina* fungal frequencies on thallus base portion samples yielded no differences between the two lichens.

Conversely, comparisons of the fungal assemblages of the canopies of *C. mitis* with its lower base strata yielded significant differences in the fungal assemblages. Additionally, multiple discriminate analysis comparisons of the canopies of *C. rangiferina* with its corresponding lower base strata showed significant differences (appendices 9a-c & 10a-c). Although there were no significant differences in canopy and base samples across the two lichens, comparisons of multiple discriminate analysis results from canopy samples and base collections yielded significant differences for thalli of *C. mitis* and *C. rangiferina* separately.

Diversity indices, determined for the canopy and base fungal assemblages, indicated little difference in average species richness (6.4 – 7.8) or evenness (0.92 - 0.95) (table 3.1). All the assemblages are markedly even in their fungal species distributions and average in species richness.

Table 3.1 - Species Richness & Shannon-Wiener diversity indices for the grouped data set.

Pooled Samples	Species Richness	Evenness
LMC:	6.8	0.95
MMC:	7.2	0.92
LMB:	7.8	0.93
MMB:	7.2	0.94
LRC:	7.2	0.95
MRC:	6.4	0.95
LRB:	6.8	0.93
MRB:	7.6	0.94

LMC = *Cladonia mitis* canopy samples from pure lichen plots
MMC = *Cladonia mitis* canopy samples from mixed moss-lichen plots
LMB = *Cladonia mitis* base samples from pure lichen plots
MMB = *Cladonia mitis* base samples from mixed moss-lichen plots
LRC = *Cladonia rangiferina* canopy samples from pure lichen plots
MRC = *Cladonia rangiferina* canopy samples from mixed moss-lichen plots
LRB = *Cladonia rangiferina* base samples from pure lichen plots
MRB = *Cladonia rangiferina* base samples from mixed moss-lichen plots

The number of taxa over all the canopy and base fungal assemblages ranged from 11-14 ($\bar{x} = 12 \pm 1$) with LRC at 14 taxa and falling more then one standard deviation higher than the mean (table 3.2). Included among taxa which were encountered on $\geq 25\%$ of the samples from all canopy and base groups were: *Alternaria* spp.; *Penicillium* spp. (non-sclerotial); and *Trichoderma* spp..

Table 3.2 - List of fungal assemblages and occurrences for canopy and base groups.

	LMC	LMB	LRC	LRB	MMC	MMB	MRC	MRB
Absidia coerulea	0.13	0.6	0.07	0.47	0.14	0.33	-	0.33
Alternaria spp.	0.8	0.27	1	0.27	1	0.33	0.87	0.4
Aspergillus alutaceus group	-	-	-	-	-	-	0.07	0.07
Cladosporium # 1	0.8	0.4	0.87	0.07	0.86	0.13	0.73	0.13
Cladosporium # 2	0.07	-	0.27	-	0.07	0.07	-	-
Cunninghamella elegans	-	-	-	0.07	-	-	-	-
Epicoccum purpurascens	0.53	0.2	0.8	0.07	0.36	0.13	0.4	0.07
Mortierella isabellina/vinacea complex	0.33	0.53	0.2	0.53	0.43	0.53	0.13	0.53
Mucor # 1	0.13	0.47	0.2	0.6	0.43	0.73	0.27	0.67
Mucor # 4	-	0.07	0.13	-	0.07	0.07	-	-
Mucor # 5	-	-	0.07	-	-	-	-	-
Mucor # 10	0.13	0.67	0.07	0.67	0.14	0.47	0.27	0.67
Penicillium (non-sclerotial)	0.33	0.67	0.27	0.47	0.5	0.47	0.53	0.47
Penicillium (sclerlotial)	0.13	0.07	0.13	0.13	-	-	0.13	0.2
Rhizopus oryzae	0.13	0.07	0.07	0.07	0.14	-	-	0.07
Rhizopus # 2	-	-	-	-	0.14	-	0.07	-
Trichoderma spp.	0.47	0.6	0.27	0.6	0.5	0.73	0.4	0.73

LMC = *Cladonia mitis* canopy samples from pure lichen plots
MMC = *Cladonia mitis* canopy samples from mixed moss-lichen plots
LMB = *Cladonia mitis* base samples from pure lichen plots
MMB = *Cladonia mitis* base samples from mixed moss-lichen plots
LRC = *Cladonia rangiferina* canopy samples from pure lichen plots
MRC = *Cladonia rangiferina* canopy samples from mixed moss-lichen plots
LRB = *Cladonia rangiferina* base samples from pure lichen plots
MRB = *Cladonia rangiferina* base samples from mixed moss-lichen plots

Alternaria spp. and *Cladosporium* # 1 had high (> 70%) occurrence across all canopy collections of both lichens (table 3.2). On average, *Epicoccum purpurascens* had slightly lower occurrence (>50%) in canopy collections. *Absidia coerulea*, *Mortierella isabellina/vinacea complex*, *Mucor* # 1, *Mucor* # 10, *Penicillium* spp. (non-sclerotial), and *Trichoderma* spp. occurred in more than 45% of thallus base samples of the two lichens.

Zygomycetes, including: *Absidia coerulea*; *Mortierella isabellina/vinacea complex*; *Mucor* # 1, 4 ,5, & 10; *Rhizopus oryzae*; and *Rhizopus* # 2; accounted for 36% (MRC) to 54% (MMC) of the taxa in the assemblages across all canopy and base groups (table 3.2). Mean occurrences of the Zygomycetes were 45% for MRB; 43% for MMB; 40% for LMB and LRB; 21% for MMC; 18% for MRC; 17% for LMC; and 11% for LRC. At coefficients of association for the fungal assemblages in canopy and base groups at >70% (fig. 3.3) MRB, LMB, and LRB are strongly associated as a group. Also, MMB and MMC are strongly associated as an independent pairing at >70% occurrence. The pure lichen canopies (LMC and LRC) of *C. mitis* and *C. rangiferina* are strongly associated with one another and have association with MRB, LMB and LRB. The assemblages of MRC show no association with any of the assemblages on the other canopy and base groups.

	LMC	**MMB**	**MMC**	**LRB**	**LRC**	**MRB**	**MRC**
LMB	0.72	0.60	0.56	0.72	0.72	0.72	0.33
LMC		0.60	0.56	0.72	0.72	0.72	0.33
MMB			0.75	0.33	0.63	0.33	0.23
MMC				0.25	0.47	0.25	0.17
LRB					0.38	0.72	0.33
LRC						0.38	-0.02
MRB							0.60

Fig. 3.3 - Coefficients of association for fungal assemblages in canopy and base group pairings.

Absidia coerulea, *Mortierella isabellina/vinacea complex*, and *Rhizopus oryzae* showed increased average occurrences on thalli of *Cladonia mitis* as contrasted with isolations from *C. rangiferina* (table 3.2). *Mortierela isabellina/vinacea complex* occurred at 40% on *C. mitis* and ~ 15% on *C. rangiferina*. *Absidia coerulea* and *R. oryzae* occurred at ~ 15% on *C. mitis* and around 5% on *C. rangiferina*.

Fungi isolated from washed litter pieces removed from soil samples collected underneath *Cladonia mitis* and *C. rangiferina* mats (appendix 11a) totalled 11 taxa (table 3.3). Of these taxa, six were previously isolated from washed thalli of the two lichens. *Cladosporium* # 1; *Mortierella isabellina/vinacea* complex; *Mucor* # 10; *Penicillium* (non-sclerotial); *Penicillium* (sclerotial); and *Trichoderma* spp. fell into this category. High occurrence (>40%) featured *Mucor* # 10 and *Trichoderma* spp. Five taxa (i.e.

Absidia californica; *Aureobasidium pullulans*; *Mucor* # 12; *Trichoderma* # 1 and *Zygorhynchus exponens*) of the 11 soil litter isolates were encountered only in soil collections. Eleven fungal taxa were only encountered in the lichen mats and not in the soil litter layers (i.e. *Absidia coerulea*; *Alternaria* spp.; *Aspergillis alutaceus* group; *Cladosporium* # 2; *Cunninghamella elegans*; *Epicoccum purpurascens*; *Mucor* # 1, 4, & 5; *Rhizopus oryzae*; *Rhizopus* # 2) Among fungi isolated from lichen thalli and not collected from soil litter *Alternaria* spp., *Epicoccum purpurascens*; and *Mucor* # 1 were of high occurrence (table 3.2) (photographs of select fungal taxa can be seen in appendix 15a).

Table 3.3 - List of fungi and their occurrences on washed litter from soil samples taken beneath *Cladonia mitis* and *C. rangiferina* mats.

Fungal Taxa	Pure Lichen Plot	Mixed Moss-Lichen Plot
Absidia californica Ellis & Hesseltine	0.13	0.07
Aureobasidium pullulans	0.07	0.07
Cladosporium # 1	0.13	0.07
Mortierella isabellina/vinacea	0.13	0.2
Mucor # 12	0.13	0.2
Mucor # 10	0.6	0.47
Penicillium spp. (sclerotial)	0.07	-
Penicillium spp. (non-sclerotial)	0.07	0.07
Trichoderma # 1	0.2	0.07
Trichoderma spp.	0.47	0.47
Zygorhynchus exponens Burgeff	0.07	-

Discussion:

Although there were no significant differences in the fungal assemblages on *Cladonia mitis* and *C. rangiferina* (Chapter 2, fig. 2.3), confirming Petrini's (1990) findings demonstrating no fungal taxa preferentially colonizing eight *Cladonia* taxa and *Stereocaulon dactylophyllum* Florke, in my study, assemblages on canopy portions of the lichen were significantly different from those on the base section of the thalli (figs. 3.1-3.2). Species richness (diversity) and evenness of the taxa (table 3.1) were generally the same for each of the canopy and base assemblage groups. However, assemblages associated with the *C. rangiferina* canopies of pure lichen plots were more diverse in fungi than the other canopy and base groups, again suggesting differences among assemblages along the length of the lichen thalli (table 3.2). This was corroborated by the

generally higher occurrence of *Alternaria* spp., *Cladosporium* # 1 and *Epicoccum purpurascens* in canopy collections than in base samples, consistent with the literature, in that *Alternaria* spp., *Epicoccum purpurascens*, and *Cladosporium* spp. all commonly have epiphytic tendencies (adapted to high light intensities, frequent desiccation, and high temperature fluctuations) (Petrini *et al.*, 1990; Pugh, 1980).

That *Absidia coerulea*, *Mortierella isabellina/vinacea complex*, *Mucor* # 1, *Mucor* # 10, *Penicillium* (non-sclerotial), and *Trichoderma* spp. occurred in more than 45% of thallus base samples, is further evidence for an environmental gradient in the lichen mats. *Absidia coerulea*, *Mortierella isabellina/vinacea complex*, *Mucor* # 1, and *Mucor* # 10 are all Zygomycetes, which generally prefer moist environments (Chowdhery *et al.*, 1982). It is known that highly branched *Cladonia* lichen mats maintain humidity, particularly in the base region of the thalli (Kershaw & Field, 1975). Dense canopies of lichen mats also shade the environment, protecting it from ultraviolet radiation (Kershaw & Harris, 1971). The combination of high humidity levels and shading of the bases of highly branched *Cladonia* mats helps to maintain temperature, reducing the temperature fluctuations that occur in ambient air, protecting organisms in the microhabitat from heat stress (Kershaw & Field, 1975; Kershaw & Harris, 1971).

Despite the tendencies of highly branched *Cladonia* lichen mats to retain moisture, it is known that the thalli differentially dehydrate from the highly branched canopy to the base (Kershaw & Rouse, 1971). The complexity of branching within the lichen canopy determines the amelioration of moisture within the mat. In highly branched and enclosed canopies of *C. mitis* sufficient moisture may be trapped to create minor differences in moisture along a gradient between bases and canopies. In contrast, the greater openness of canopies of *C. rangiferina* (Kershaw & Harris, 1971) suggests the possibility of greater differential drying within the canopy versus the base of the thalli, and establishment of a more defined moisture gradient.

Moisture gradients are well known to influence the presence and absence of fungi. Zygomycete occurrences (table 3.2) and coefficients of association (fig. 3.3) indicated differences attributable to moisture availability, moisture retention of humidity, and moisture gradient establishment. All base collections in the mats were higher (>40%) in Zygomycete occurrences, indicating that moisture levels were sufficient to support these

fungi. Canopy collections across the collection type groups were lower (11-21%) in Zygomycetes suggesting less than optimal conditions of moisture for these fungi. It is interesting to note that canopies of pure *Cladonia rangiferina* stands (LRC) had the highest diversity of mycota among all the groups. This may reflect the fact that this mat group had a moisture gradient of wide spatial and temporal range. Also, the diversity of taxa were highest for the pure stands of *C. rangiferina* among the collection type groups (Chapter 2, table 2.2).

Further indication of the influence of moisture along a gradient, as related to lichen architecture (degree of branching of podetia), was seen in the higher occurrences of *Absidia coerulea*, *Mortierella isabellina/vinacea complex*, and *Rhizopus oryzae* from *Cladonia mitis* than *C. rangiferina*. Branching of *C. mitis* is more complex than in *C. rangiferina*, suggesting the maintenance of a moisture gradient for a longer time in the former over the later.

In addition to the establishment of moisture gradients in lichen mats, nitrogen and other potential nutrients occur in gradations along the mat thalli. Nitrogen is mobilized in older regions of the lichen thalli and transported to the growing apices (Dahlman *et al.*, 2002; Hyvarinen & Crittenden, 2000) where it accumulates. This can produce pronounced vertical gradients of nitrogen, as seen in *Cladonia stellaris* and *Stereocaulon paschale* (L.) Hoffm. (Crittenden, 1988). During the fungal isolation phase of my work it was observed that *Cladosporium* #'s 1 & 2 required nitrogen (peptone) to sporulate in culture. The high canopy occurrence of *Cladosporium* #'s 1 & 2 can be partially explained in light of the accumulation of nitrogen the lichen mat canopies. Vertical gradients of nitrogen may not only affect the sporulation of *Cladosporium* but may even determine occurrence of other fungi in the canopy and base regions of lichen mat thalli.

As mentioned earlier, highly branched fruticose lichens are effective particulate filters, due to their highly branched structure which creates abundant traps for particulates. Particulates are trapped in considerable quantity on the external surface, and accumulate in the internal spaces of fruticose lichen thalli by gravity and item flow (Longton, 1988). Such captured particulates include pollen and a variety of litter, i.e. conifer needles and bark chips. Pollen production is episodic, and anthesis in most species lasts approximately two weeks (Lee *et al.*, 1996b). A jack pine stand, typical of

the Sandilands region, can release 24.6 kg of pollen per hectare yearly (Lee *et al.*, 1996a). Once trapped, pollen and litter substrata gravimetrically and hydrologically settle in the lichen mats and establish gradients of various attendant nutrients. Massive inputs of pollen, which initially accumulates in the mat canopy, are known to support such fungi as *Alternaria* spp., *Cladosporium* spp. *Penicillium* spp., and *Rhizopus* spp. (Venugopal Rao & Manoharachary, 1985).

Other nutrient inputs occurring sporadically come from the leakage of metabolites, occurring from the lichen with the onset of rainfall (Farrar & Smith, 1976). Lichen thallus hydration results in the depletion of glucose from the phycobiont, as it is transferred to the mycobiont during hydration periods. Frequent alternating of wetting and drying cycles allows the transfer of glucose to the mycobiont without depleting the resources of the phycobiont (Kershaw, 1985). Fungi, other than the mycobiont of the lichen thallus, likely utilize these leachates when they are released during hydration of the lichen thallus. Zygomycetes, such as *Absidia* spp., *Cunninghamella* spp., *Mortierella* spp., *Mucor* spp., and *Rhizopus* spp. are widely known to be highly active procurers of glucose.

The highly varied environment within lichen mats suggests that organisms living within them are presented with a range of niches. Fungi in niches are strategically adapted to specific conditions. Grime (1979) recognized that groups of organisms demonstrate strategies in response to nutrients (stress) and perturbation (disturbance) regimes which characterize generalized niches.

Pugh (1980), theorizing on fungal ecology, proposes adoption of Grime's (1979) strategies, utilizing the concepts of stress and disturbance. Stress, which is any factor that affects the ability to procure nutrients includes, among other factors: 1) moisture; 2) temperature extremes; 3) ultraviolet light; and 4) allelopathy. Disturbance, a dramatic alteration of the norm, such that fungal biomass is reduced either by its destruction, or the superimposition of a new environment over an existing one, encompasses such factors as: 1) sudden massive additions or deletions of nutrients; 2) water saturation with concomitant anoxic conditions; 3) extirpation; or 4) addition of antifungal substances.

Lichen mats in forest ecosystems are high disturbance environments. With periodic inputs of needle litter, bark etc., high levels of nutrients are common in mats on

the forest floor. With episodic deposition of massive levels of nutrients and substratum during periods of pollination and rainfall, conditions for high disturbance are met. As further evidence of high disturbance of lichen mats, extirpation (removal) commonly occurs by grazing or trampling. Lichens are particularly vulnerable since they lack root systems (Brodo *et al.*, 2001), and can be removed easily. Thirdly, water saturation and anoxic conditions, particularly in mat bases, are common features during and immediately after heavy rainfall (Kershaw and Field, 1975). Finally both *C. rangiferina* and *C. mitis* also produce secondary compounds which have antibacterial and antifungal properties (Brodo *et al.*, 2001; Elix, 1996).

Four major life strategies: competitors, stress tolerant, ruderals, and survivors-escapers are recognized by Pugh (1980). Competitors are fungi living in a regime of low stress and low disturbance. Stress tolerant fungi survive under high stress and low disturbance. Survivors-escapers thrive in high stress/high disturbance environments and ruderals grow in low stress/high disturbance environments. With lichen mats being high disturbance microhabitats, conditions for fungi living within them are restricted to low stress/high disturbance (bases of thalli) and high stress/high disturbance (thallus canopy) microhabitats.

Lichen mat canopies are high stress environments in contrast to lichen bases, which are taken to be low stress environments. The surface temperatures of lichen canopies often reach much higher temperatures than the surrounding ambient air. Surface temperatures of *Cladonia stellaris* mats have been seen to reach 40°C during solar noon, when the ambient air temperature is as low as 20°C (Tegler & Kershaw, 1980). Fungi found in lichen canopies are provided with little protection from ultraviolet radiation, and lichen canopies dry out much more rapidly than the lower portions of the mat (Kershaw & Field, 1975; Kershaw & Harris, 1971).

High occurrences of *Alternaria* spp., *Cladosporium* # 1 & 2 and *Epicoccum purpurascens* in the canopies of *C. mitis* and *C. rangiferina*, indicat that these fungi are survivors-escapers (high stress/high disturbance). Taxa of *Alternaria*, *Cladosporium*, and *Epicoccum* have been previously classified as survivors/escapers, being found in such high stress/high disturbance environments such as living leaves; exposed to widely fluctuating temperatures, high ultraviolet radiation, and general desiccation (Pugh, 1980).

Ruderal propagules germinate readily and mycelial growth is rapid. These factors enable ruderals to act as pioneering species, colonizing easily decomposed, non-refractive substrata (Pugh, 1980). High humidity levels and shading of the bases of highly branched *Cladonia* mats, as well as reduced temperature fluctuations, protect organisms in the microhabitat from desiccation, ultraviolet radiation, and heat stress (Kershaw & Field, 1975; Kershaw & Harris, 1971). *Absidia coerulea*, *Mortierella isabellina/vinacea complex*, and *Mucor* # 1 & # 10, utilizing high levels of glucose in culture, growing rapidly, and of high occurrence in the bases of lichen mats, are most likely ruderals.

Pugh (1980) also identifies many Zygomycetes as ruderals, and suggests that most soil fungi fall into this strategic category. Keller & Bidochka (1998) also record *Cunninghamella* spp., *Mortierella* spp., *Mucor* spp., and *Rhizopus* spp. as common components in soil. That some representatives of these Zygomycetes i.e. *Mortierella isabellina/vinacea* complex and *Mucor* # 10, were of high occurrence on lichen mat bases and washed litter from soil collections beneath the lichen mats in my study (tables 3.2 & 3.3), suggests that they are opportunistic and thriving in environments of low stress.

Comparisons of the soil and lichen mat fungi surveyed shows a total of 22 fungal taxa isolated from *Cladonia mitis* thalli, *C. rangiferina* mats and underlying soil samples (tables 3.2 & 3.3). Eleven taxa occur only in the lichen mats (base and canopy), while five taxa are restricted to the soil. Thus, 65% (11 of 17) of the mycota isolated from lichen mats are unique to the lichen thalli.

Highly branched *Cladonia* mats may create a unique microhabitat in the soils beneath them. Soil litter restricted presence of *Absidia californica*; *Aureobasidium pullulans*; *Mucor* # 12; *Trichoderma* # 1; and *Zygorhynchus exponens* indicated such a phenomenon.

Examination of the soil fungi found from surveying soil litter layers beneath the lichen mats revealed that *Cladosporium* # 1 was both in the soil litter mycota and at high occurrence in the canopy portion of the lichen thalli. Among the fungi of high occurrence on bases of lichen mat thalli, *Mortierella isabellina/vinacea*; *Mucor* # 10; *Penicillium* (non-sclerotial); and *Trichoderma* spp. are routinely isolated from soil borne litter. My results suggest that soil litter fungi may act as an inoculum for not only lichen mat base assemblages but also for canopy assemblages. Furthermore, fungal inhabitants of soils

are known to change over time (Ananda & Sridhar, 2004; Keller & Bidochka, 1998; Zak & Parkinson, 1984) which may serve to partially explain my earlier results demonstrating temporal changes in lichen mats (Chapter 2, tables 2.1-2.3).

Alternaria spp. and *Epicoccum purpurascens*, of high occurrence in lichen canopy assemblages, were not soil litter inhabitants as determined in this study. These taxa are known epiphytes (Petrini *et al.*, 1990; Pugh, 1980) with large dermateaceous spores adopted to survival and life in environments with high light (Durrell & Shields, 1960; Joensson, 1967; Pugh, 1980). Found in the assemblages of lichen mat bases at high occurrence, *Mucor* # 1 was not present in the soil litter mycota. As a Zygomycete, this taxon may be responding to the unique moisture gradients within lichen mats.

Though circumstantial, it is probable to conclude that a portion of the lichen mat canopy and base fungal assemblages was derived from soil borne inoculua. However, what was the inoculum for higher occurrence fungi i.e. *Alternaria* spp.; *Epicoccum purpurascens*; and *Mucor* # 1, which were not found from soil litter? These fungal taxa may sporulate and spread by wind or water dispersal of propagules. Inoculum may also arise by dispersal of existing lichen fragments infested by these fungi. In the ensuing chapters of this thesis the question of lichen dispersal and its possible impact on fungal assemblages associated with lichen mats is examined. Secondly, in the construction of the two *Cladonia* species studied, are there differences in fungal assemblages across a range of different types of fruticose and foliose lichens?

Chapter 4

Intraspecific Variation Within the nuclear SSU rDNA for Populations of *Cladonia arbuscula* in Northern Manitoba

Introduction

Inoculum responsible for the presence of fungal assemblages within lichen mats at least partially arises from the soil litter layers beneath the mats (Keller & Bidochka, 1998; Pugh, 1980; Chapter 3 – table 3.3). Differences seen in fungal taxa between the soil and lichen mat assemblages (Chapter 3 – tables 3.2-3.4) leave unresolved questions as to their inoculum source. Sporulating fungal taxa can spread by wind or water dispersal of propagules. Inoculum may arise by dispersal of existing fungus (spores and hyphae) attached to lichen fragments (thalli and soredia).

Fruticose lichens are effective traps for foreign materials, including lichen fragments, along with needle and twig litter, pollen, rain and insect exoskeleton parts. Lichen fragments, along with their attendant fungi, are trapped on external surfaces, and may accumulate in the internal spaces of fruticose lichen thalli (Longton, 1988). Once in favourable microhabitats, such as within a lichen mat, it is assumed that the transported fungal propagules begin to develop into mycelia. Fragments of lichen thalli transporting fungal inoculum, can be derived from separate lichen patches. Co-dispersal of lichen thallus fragments and mat associated fungal taxa, can potentially introduce new fungal inoculum into lichen mats. Thus, in terms of propagule dispersal and infestation of lichen mats by other fungi, the dispersal strategies of lichens (thallus fragments and soredia vs. sexually produced spores) must be examined for their degree of occurrence.

Sexual reproduction, resulting in the production of apothecia and ascospores, while important in maintaining genetic diversity, is assumed to occur less frequently in the 'reindeer lichens' (*Cladonia* sect. *Cladina* as per Stenroos *et al.*, 2002) than asexual or vegetative reproduction (Yarranton, 1975). Reindeer lichens are assumed to reproduce primarily by fragmentation as a result of disturbance (Yarranton, 1975). Fragments of the lichen thallus can then be dispersed by wind, water or animals (Kiss, 1985). Thomson (1972) noted flow lines of arctic melt water carrying lichen fragments, and Westman (1973) reported lichen fragments in arctic drift ice, suggesting that these fragments could be carried a shore by birds. Richardson ((1975) as cited in Bailey (1976)) listed several species of birds incorporating lichens in nests.

Dispersal studies were once limited to experimental lab conditions such as wind tunnels (Armstrong, 1994; Bailey, 1966), or short distance field experiments using

marked lichen thalli (Heinken, 1999) or spore samplers (Marshall, 1996). While these studies provided direct and valuable information, the data were incomplete. Modern molecular techniques allow lichen dispersal to be indirectly assessed by measuring genetic differentiation among populations (Hageman & Fahselt, 1992; Printzen, 2003). The SSU rDNA of lichen-forming fungi contains a large amount of variation even within species, due to optional group I intron insertions (DePriest, 1993a). Within the genus *Cladonia*, these introns have been mapped for a number of species including *C. subtenuis*, *C. gracilis*, *C. rangiferina* and the *C. chlorophaea* complex, finding various combinations of these introns being present or absent along five positions on the rDNA repeat (Beard & DePriest, 1996; Depriest & Been, 1992; DePriest 1993b; Piercey-Normore, 2004). Since these introns are often spliced out during the transcription of rDNA to rRNA, the introns potentially are not subjected to evolution's natural selection processes, allowing for higher sequence evolution rates and greater length variations. Restriction fragment length polymorphisms (RFLPs), microsatellites, and sequencing techniques have all been used with introns in population studies (Booton *et al.*, 2004; Coates *et al.*, 2002; Järvinen *et al.*, 2003). The presence or absence of the introns within the SSU rDNA, provides additional length and nucleotide sequence variation to conduct population studies, which is particularly useful as rDNA is highly conserved within species.

Cladonia arbuscula (Wallr.) Flot. is a reindeer lichen, with highly branched hollow podetia, no cortex, and yellowish-green in color. *C. arbuscula* has no vegetative propagules such as soredia or isidia, though it does have pycnidia. Pycnidia are hollow enclosures lined with conidiophores which produce conidia. The conidia disperse independently of the lichen photobiont and re-associate with a new algal partner to form a new lichen thallus. *C. arbuscula* reproduces vegetatively by fragmentation and pycnidia, and sexually by apothecia. *C. arbuscula* grows abundantly in northern regions, including northern Manitoba, usually over rocky ground or thin soil (Brodo *et al.*, 2001). *C. arbuscula* is commonly mistaken for *C. mitis*, but tends to have denser crowns, the presence of fumarprotocetraric acid (absent in some populations, particularly in Manitoba), more prominent browning of the tips, and a tendency for those tips to curve in one direction, giving a comb like appearance (Brodo *et al.*, 2001). Some lichenologists do not separate the two lichen forming fungi (Brodo *et al.*, 2001). Indeed, separation of the

two lichen forming fungal taxa, even at the molecular level was not seen (Stenroos *et al.*, 2002). This study examined variation in the SSU rDNA of populations of *C. arbuscula*, to infer dispersal of thallus fragments. *C. mitis* was not used as *C. arbuscula* was much more prevalent in the area, and the collector was under the assumption the *C. arbuscula* samples collected were *C. mitis* and could not identify the subtle differences distinguishing them under field conditions.

Materials & Methods

Lichen samples

Fifty specimens of *Cladonia arbuscula* were collected along ten transects from boreal Manitoba, five samples per transect (fig. 4.1). Samples were collected from separate mats of *C. arbuscula*, with distances between mats varying from 1-8 meters. Five of the transects were placed on separate islands, with water separating these transects. Additional verification of sample identifications was done by detection of secondary compounds using thin-layer chromatography (TLC) (appendix 14c) according to methods described by Culberson *et al.* (1972; 1974) (vouchers of scanned images for the TLC plates are available in molecular laboratory of Dr. Piercey-Normore, U of M).

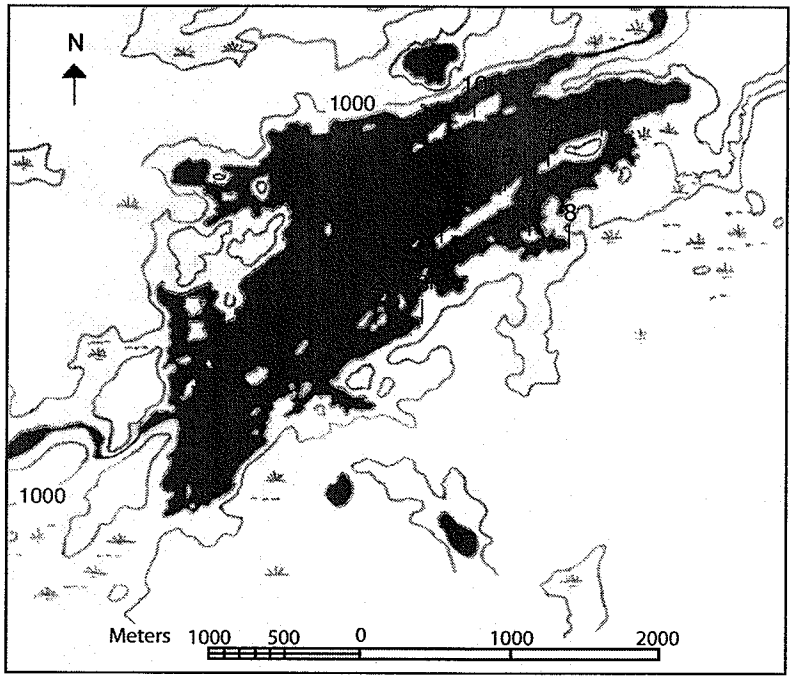


Fig. 4.1 - Topographic map of Payuk lake, northern Manitoba. Transect sites are labelled.

Site Descriptions

Dominant vegetation in the Payuk lake regions sampled consisted mainly of Jack pine growing on Precambrian shield. *Pleurozium, Peltigera, Stereocaulon, Vulpicida, Evernia, Usnea* and *Cladonia* were common undergrowth and epiphytic vegetation present. Two independent estimates of the percentage of each transect shaded by tree cover were taken and averaged. Transects were then categorized into three groups based on the average percentage cover: Wooded (75-100% cover), Partially Exposed (25-75% cover), and Fully Exposed (<25% cover). Compass headings were taken to determine the direction each transect ran, and aspects of any slopes present were noted (table 4.1).

Table 4.1 – Site descriptions of transects from the Payuk lake region.

Transect	Transect Direction and Topography	Slope Aspect	Exposure
1	N-S	N	Fully exposed
2	S-N	E	Fully exposed
3	S-N (10°-190°)	N	Wooded
4	N-S (330°-150°) over a ridge	NNW-SSE	Wooded
5	N-S (350°-170°)	N & S	Partially exposed
6	NE-SW (50°-230°) along bedrock sloping to the water's edge. Open to lake.	SE	Fully exposed
7	S-N	No slope	Fully exposed
8	N-S with large boulders	N	Wooded
9	ENE-WSW (60°-240°) along a rock bluff.	WSW-ENE	Partially exposed
10	NE-SW (45°-225°) along a ridge crest	SE	Partially exposed

DNA extraction

Isolation of genomic DNA was performed following the methods of Grube *et al.* (1995). Genomic DNA was extracted by grinding approximately 0.5cm^2 of lichen thallus per sample in TES buffer (100mM Tris, pH 8, 10mM EDTA, 2% SDS), using plastic blue pestles and 1.5ml eppindorf tubes. After grinding, NaCl and CTAB (cetyltrimethylammonium bromide) were added to 986mM and 0.99% concentrations respectively. Samples were vortexed, followed by incubation for 1hr. at 65 °C. Upon adding an equal volume of chloroform : isoamyl alcohol (24:1) and mixing for 1 min., samples were centrifuged at 5,000 rpm for 5 min. The supernatant was transferred to a clean eppindorf and the chloroform : isoamyl alcohol extraction was repeated with the supernatant again being transferred a clean eppindorf tube. DNA was precipitated by the

addition of 0.2 vol. of NaCl and 2.5 vol. of 100% ethanol to the transferred supernatant. Samples were allowed to stand for 5 min., followed by centrifuging for 5 min. at 13,000 rpm. The supernatant was discarded and the DNA pellet washed with cold 80% ethanol. Eppindorf tubes with the DNA pellet were turned upside down and allowed to air dry for 25 min. at room temperature. The DNA pellet was then resuspended in warm sterile distilled water.

Electrophoresis & Quantification of DNA

A 1% agarose gel containing 0.5mg/ml ethidium bromide was prepared in 1x TBE buffer (Tris 89mM, Boric acid 89mM, and EDTA 20mM). Three µl of each DNA sample was mixed with one drop of sterilized distilled water and one drop of 6x methylene blue loading dye on parafilm, and placed into separate wells. Half a µg of a 1 Kb Plus DNA ladder (BRL) was also mixed with one drop of sterilized distilled water and a drop of 6x methylene blue loading dye on parafilm and placed in the first well of the gel. The gel was run at 120 volts for approximately 35 min., inside an electrophoresis unit (Fisher Biotech electrophoresis system mini horizontal unit FB-SB-710) containing 1x TBE buffer, until the loading dye was approximately 1.5cm from the bottom of the gel. The gel was removed from the electrophoresis unit and DNA bands were visualized under a UV transluminator. DNA concentrations were determined by comparing the band intensities of the DNA with the intensity of the 1650bp band of the 1 Kb Plus DNA ladder. The 1650bp band of the 1 Kb Plus DNA ladder represents approximately 40ng of DNA when 0.5µg of ladder is loaded.

PCR amplification of DNA

PCR was performed on all samples. Primers (nu-SSU-1427-5') 5'-TTTGA GGCAA TAACA GGT-3'; (nu-SSU-1583-5') 5'-CAACG AGGAA TTCCT AGT-3'; (nu-SSU-1580-3') 5'-GATGA CTCGC GCTTA CTA-3'; and (nu-SSU-1750-3') 5'-AAACC TTGTT ACGAC TTTTA-3' (Gargas & DePriest, 1996) were used for amplifying the 3' half of the SSU rDNA (fig. 4.2). Primers nu-SSU-1427-5' and nu-SSU-1583-5' indicated the presence or absence of introns, while DNA fingerprint banding patterns were produced using primers nu-SSU-1580-3' and nu-SSU-1750-3' (18 & 20 bp primers

respectively) as a result of non-specific binding of the primers due to introns. The PCR protocol utilized 1.6 units of Taq polymerase (BRL), the buffer (50mM KCl, 10mM Tris-HCl, pH 8.3), 2 mM $MgCl_2$, 0.5 μM of primer, and 2 mM of dNTP's, in a 50 μl reaction volume. Following an initial denaturation step of 5 min. at 94 °C, the reactions underwent 33 cycles of denaturation at 94 °C for 45 sec., annealing at 56 °C (for primers nu-SSU-1583-5' & nu-SSU-1750-3') or 54 °C (for primers nu-SSU-1427-5' & nu-SSU-1580-3') for 45 sec., and extension at 72 °C for 2 min in a Fisher Scientific Techne Genius thermocycler. PCR product was run on 1% agarose gel stained with ethidium bromide, and visualized under a UV transluminator (see electrophoresis section above). Both 1kb and 50bp ladders were used as size standards.

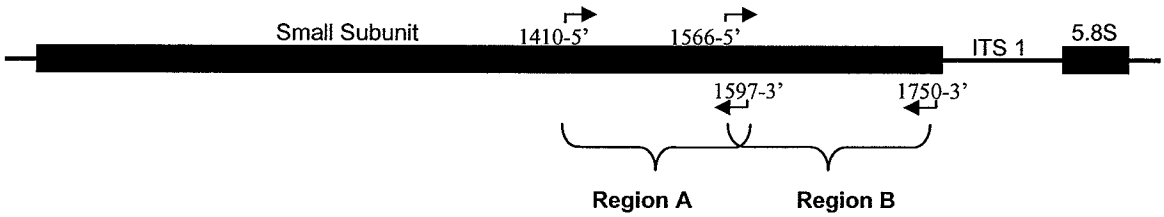


Fig. 4.2 - Diagram of a portion of the nuclear ribosomal DNA repeat unit, showing location and direction of primers.

Data analysis

DNA bands visualized under the UV transluminator were scored as either present or absent for each of the samples for region B. Data collected from region B was then subjected to a sums of squares cluster analysis, conducted mathematically as previously described (Chapter 2 – sect. *data analysis*). DNA bands visualized from amplification of region A were scored as either: intron absent (0), intron present (1), or intron both present and absent (2) for each of the samples. Intron data and transect number were mapped onto the dendrogram produced from the cluster analysis of the region B data set. Multiple analysis of variances (MANOVAs) were also run on the scored data for both regions A and B combined, comparing variances by transects and by exposure, using the program JMP IN 4.04 (SAS institute, 2001). MANOVA tests the equality of mean vectors for multiple populations, assessing group differences across multiple variables based on a categorical grouping of populations. The technique is analogous to ANOVA

for univariate data, except groups are compared on multiple response variables simultaneously.

Results

TLC revealed the presence of usnic acid, and the absence of fumarprotocetraric acid in 48 of the 50 samples, and the presence of both usnic and fumarprotocetraric acid in the remaining two samples. Those two samples containing fumarprotocetraric acid were subsequently removed from the analyses.

DNA amplification of region A yielded one of three banding patterns: the presence of a short band approximately 200bp in length, the presence of a longer band approximately 400bp in length, or the presence of both bands (fig. 4.3). Samples having the 400bp band present may contain one 200bp intron within region A of the SSU.

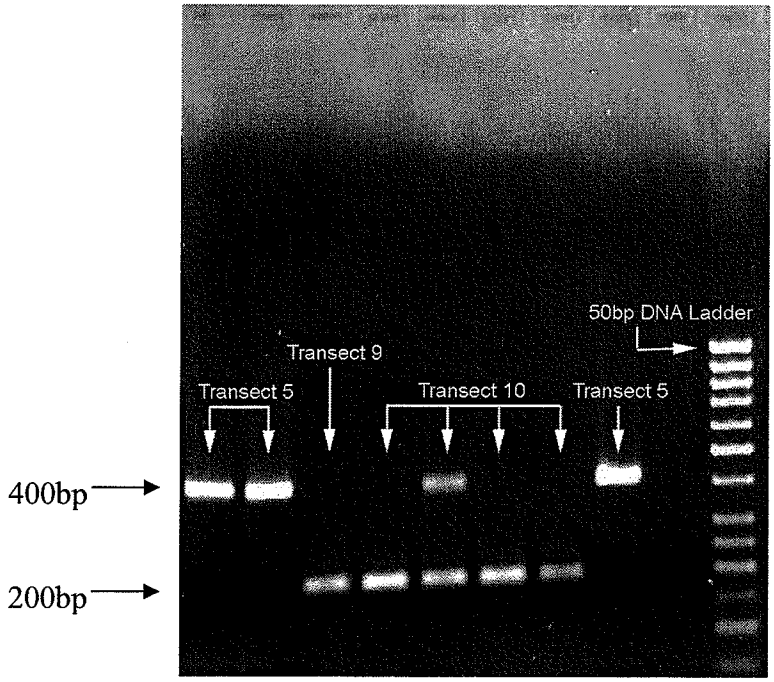


Fig. 4.3 - Electrophoresis of PCR product from region A, showing the presence and absence of introns, on a 1% agarose gel with a 50bp ladder.

DNA amplification of region B yielded 23 different banding patterns across all the samples, each sample having a minimum of two bands present, producing a 'DNA

fingerprint' for the samples (fig. 4.4). One sample failed to amplify using these primers and was subsequently removed from the analyses.

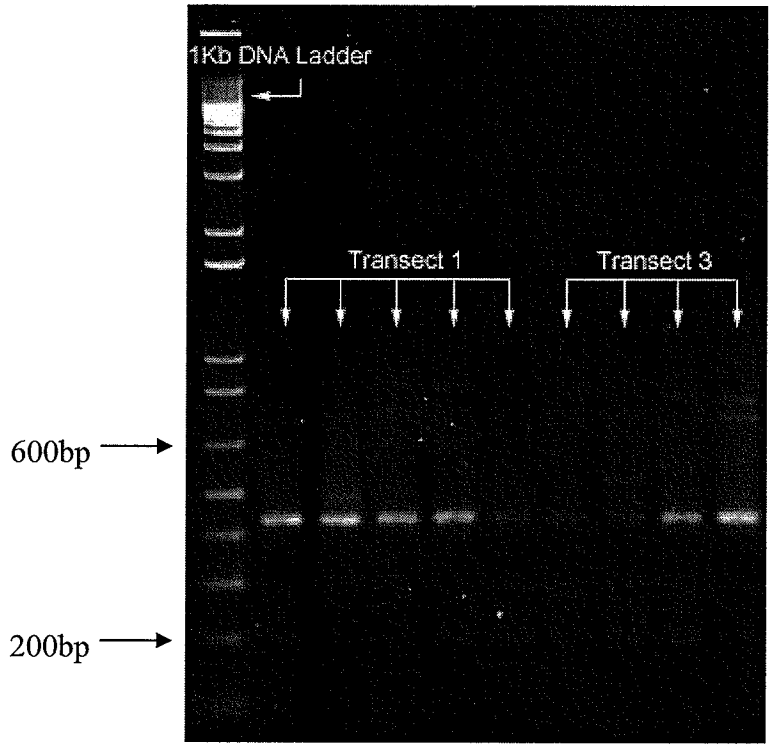


Fig. 4.4 - Electrophoresis of PCR product from region B, showing banding pattern variation on a 1.5% agarose gel with a 1kb ladder.

Geographic distribution did not correspond with the dendrogram branching patterns (figs. 4.5 & 4.6). The number of identical samples was greater between transects than within transects. Only five transects contained samples along the same transect with identical banding patterns produced from region B, one pair from each transect. As well, three of those five pairs of identical samples were non-identical when examining both region A and B data sets (appendix 12a). Results from a MANOVA showed no significant differences between transects ($F = .8388$, $Prob>F = .5857$, $p = 0.05$). Percentage cover of the transects also displayed no pattern with the genetic variation, as revealed by a MANOVA ($F = .5045$, $Prob>F = .6072$, $p = 0.05$).

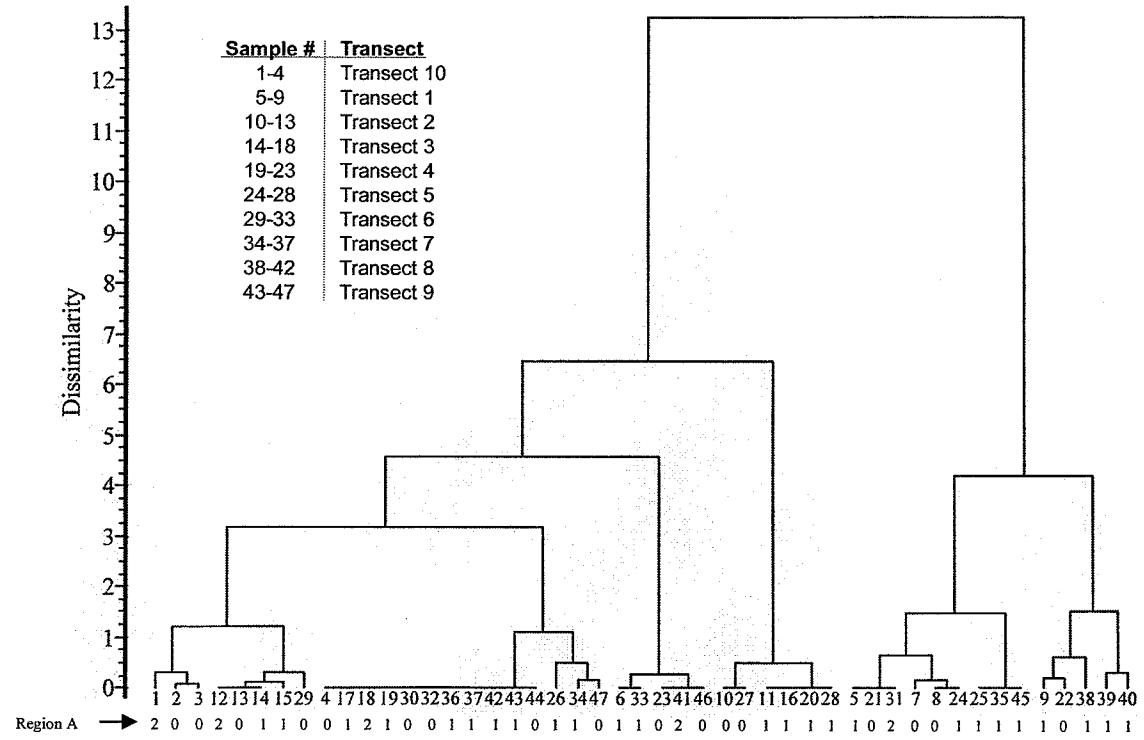


Fig. 4.5 - Sums of squares cluster analysis of data set from region B using chord distance as the association measure (fig. 4.2). Intron data from region A (fig. 4.2) is mapped on below the samples.

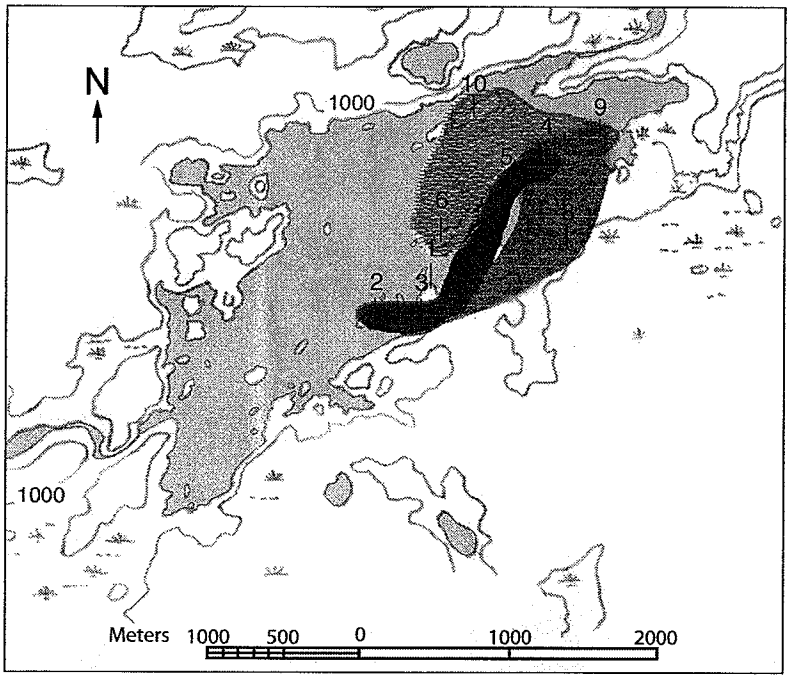


Fig. 4.6 -Topographic map of Payuk lake, northern Manitoba. Transect sites are labeled and textured dispersal patterns for 3 identical clusters displayed.

Mapping of the intron data from region A onto the cluster analysis dendrogram constructed from sequence variation of region B, demonstrated a lack of correspondence between the two regions (fig. 4.5).

Discussion

Chemistry

The two samples containing fumarprotocetraric acid were excluded from the analysis because of variation within *C. arbuscula*. *C. mitis* and *C. arbuscula* are not always recognized as separate species, thus variation with the lichen acids may reflect several subspecies of *C. arbuscula* in these samples (Brodo, 2001; Stenroos, 2002).

DNA

Amplification of region B revealed insertions within that segment of the SSU as bands greater than 200bp were observed. Insertions in the SSU rDNA of both the *Cladonia chlorophaea* complex and *C. subtenuis* have been shown to be group I introns, based on their secondary structure and conserved sequences (Beard & DePriest, 1996; DePriest, 1993a). In *C. subtenuis*, a 'reindeer lichen' related to *C. arbuscula* (Stenroos, 2002), intron insertions were 200bp long (Beard & DePriest, 1996). Thus those samples of *C. arbuscula* having the 400bp band present, likely contained one 200bp intron within the 200bp region of the SSU amplified. As well, variation in DNA fingerprint banding patterns from region A, did not correspond with the presence or absence of introns from region B (fig. 4.5). The DNA fingerprint banding patterns displayed sequence variation of both the SSU coding region and any introns that were present. The heterogeneous presence and absence of introns may explain the lack of agreement between the patterns in region A and B. On the other hand, the 2 regions may be considered 2 different loci with different introns. The lack of agreement may suggest horizontal intron transfer or recombination in rDNA in the distant past. Direct decent of introns have been observed within some fungal species, making presence or absence of group I introns potentially useful as a marker designating population strains (Coates *et al.*, 2002).

Eleven percent of the samples amplified contained both 200bp and 400bp bands from region A (fig. 4.2 & 4.5). This suggests several possibilities: 1) Samples having both bands present may represent heterokaryons. Heterokaryons are fungi with hyphae containing two or more nuclei of different genotypes. Similar results were found by Gobbi et al. (2003) for fungus *Cryphonectria parasitica*. 2) The podetia sampled may have contained two or more strains of the lichen fungus. Although Beard and DePriest (1996) showed that mats of *Cladonia subtenuis* were genetically homogeneous, it is possible that the podetium of *C. arbuscula* sampled may contain multiple strains of the lichen fungus, one with the intron and one without the intron. Heterothallism has been inferred in some lichen species such as *C. floerkeana*, *C. portentosa*, *C. galindezii*, *Xanthoria calcicola*, *X. ectaneoides*, and *X. polycarpa* (Honeggar *et al.*, 2004; Seymour *et al.*, 2005). *C. portentosa* is a 'reindeer lichen', similar in morphology to *C. arbuscula*. If *C. arbuscula* is heterothallic, like *C. portentosa*, it would be advantageous for the podetia to be composed of two strains of the lichen fungus, of different mating types, or if the podetium contained an ascogonium it may also have had the dikaryon. 3) Regions of the tandemly repeated rDNA may differ, some repeats containing the intron insertion and others without it. The nuclear rDNA locus in ascomycetous fungi is a tandem array of 100-200 rDNA repeats (Warner, 1989). The multiple copies of these rDNA repeats evolve together and thus should be identical (Arnheim *et al.*, 1980). If this is not the case, eg. horizontal transfer of the group I intron occurred in only part of the tandomly repeated rDNA, both the presence and absence of the intron would appear.

Scale

Geographic distance did not correspond with genetic patterns. Transects closer to each other geographically did not cluster together (figs. 4.5 & 4.6). Five transects contained identical banding patterns from different lichen mats within the transects for region B (fig. 4.2). However, intron patterns from region A (fig. 4.2) differed among identical banding patterns from region B on three of the five transects. The number of identical samples was greater among transects than within transects, inferring efficient dispersal between transects. Thus, the scale upon which this study was conducted is likely too small, and samples are too close together, resulting in random mixing of genotypes.

Dispersal

As is seen from the results, fragment polymorphism within a transect was no less than the genetic variation between transects (fig. 4.5). Separation of transects by bodies of water do not appear to prevent dispersal, confirming observations and hypotheses mentioned earlier (Thomson, 1972; Westman, 1973). Wind, water, and insects or birds may be good vectors for transport of thallus fragments. Given that the transects were on islands or along lake shores, transects may also be frequently exposed to high winds, which could carry away broken fragments from disturbed lichen mats. As well, no relation between cover of the transect and fragment polymorphism was seen. Variation in fragment polymorphism in wooded transects was the same as in those transects fully or partially exposed. Heinken (1999) noted a large reduction in thallus fragment dispersal in forested areas as opposed to exposed grassland. In this study, high winds created from the open lake, and the small size of the islands may explain why fragment polymorphism was similar between wooded (75%-100% cover) and more open (0-75% cover) transects. The limited forest present on the islands and shore edges may have been an insufficient buffer against the high winds.

Given the efficient gene flow seen in *C. arbuscula* across a 2 km region, co-dispersal of infested lichen thalli and their corresponding fungal assemblage taxa may be an efficient dispersal method for fungi growing in lichen mats. However, gene flow, a strong indicator of lichen dispersal, also occurs by ascospore production and subsequent growth of thalli in a new location. Cursory examination of *C. arbuscula* showed that 40% of the samples had apothecia present. While fragmentation has been thought a major form of reproduction in reindeer lichens, with apothecia infrequently produced (Brodo *et al.*, 2001), sexual reproduction seems to be occurring in *C. arbuscula* around Payuk lake. Study is required to determine the method gene flow, as well as thallus fragment dispersal mechanisms and distances in the region studied. Also, direct evidence of inclusion of lichen mat mycota in dispersal of lichen fragments is required by comparing population structure of the lichen forming fungi to the associated the fungal assemblage population structure.

Conclusions

This study conducted sampling transects over an area spanning approximately 2 km. Identical samples were found between transects as far apart as 1.5 km. Given the large number of clusters of identical samples found among transects (21 of the 48 samples taken were identical with at least one other individual on a separate transect), the size of the geographic area sampled may be too small to adequately assess dispersal. Future researchers may wish to place transects further apart from each other. As well, given the lack of identical samples within a transect, each sampling site may not have been adequately characterized in terms of its' genetic variation. Conducting a number of smaller plots as opposed to one long transect might better assess the site genetic variation. The scale on which transects were sampled potentially requires adjustment, as well as the scale upon which transects were laid.

As well, introns within the DNA region used to assess genetic variation could pose problems. Genetic variation is present when the introns are present. However, when introns are absent, genetic variation is based strictly on the rDNA coding region. Thus the absence of the introns decreases the potential genetic variation that can occur in those samples, which may skew the results. One means of testing this and confirming the results found in this study would be to examine another region such as the ITS region. This region contains sequence variation without the presence of introns as a confounding factor.

Finally, further environmental characterization of the transect sites might help to answer questions involving the dispersal patterns seen, such as a measure of average wind speeds, pH, aspect, slope, and moisture regimes.

Chapter 5

Fungal assemblages associated with selected ground dwelling and epiphytic lichens

Introduction

Fungi developing on and in lichen thalli were studied by Petrini *et al.* (1990), who examined 8 lichen taxa: *Cladonia arbuscula*; *C. arbuscula* ssp. *mitis* Sandst.; *C. arbuscula ssp. squarrosa* (Wallr.) Ruoss; *C. furcata* (Huds.) Schrad.; *C. gracilis* (L.) Willd.; *C. rangiferina*; *C. squamosa* (Scop.) Hoffm.; and *Stereocaulon dactylophyllum*. No fungal taxa preferentially colonizing a given lichen species were encountered. Results in this thesis (Chapters 2, figs. 2.3 & 2.4) seem to confirm the findings of Petrini *et al.* (1990), as multiple discriminate analyses of the fungal assemblages isolated from the fruticose lichens *C. mitis* and *C. rangiferina* revealed no significant differences ($p = 0.05$) between them. With apparent lack of preference among fungi colonizing a range of fruticose lichen forming fungi, and my results indicating no significant differences between fungal assemblages on two fruticose lichens, one might assume the microenvironment within fruticose lichens to be similar.

Girlanda *et al.* (1997) conducted a study of the fungal assemblages on two foliose lichens: *Parmelia taractica* Krempelh. and *Peltigera praetextata* (Florke *ex* Sommerf.) Zopf. Significant differences between the fungal assemblages were found on the two lichens. A far greater number of taxa were resident on *P. taractica* (39) then on *P. praetextata* (26). Also there was significant dissimilarity between the fungal assemblages on the two lichens.

Petrini *et al.* (1990) and Girlanda *et al.* (1997) proposed that fungal colonizers of lichens are insensitive to the secondary compounds found in their hosts. However, production of secondary compounds likely plays a role in influencing microhabitat. A wide range of secondary compounds are produced by lichens (Brodo *et al.*, 2001), and the compounds vary in their effectiveness as antibacterial and antifungal compounds (Konig & Wright, 1999). Usnic acid for example, is well known for its antibacterial and antifungal activities (Elix, 1996; Konig & Wright, 1999). Absence of such antibacterial lichen substances would allow for greater bacterial colonization and correspondingly smaller fungal colonization on lichens (Girlanda *et al.*, 1997). As well, antifungal properties could decrease fungal competition and influence fungal assemblages seen in the lichen mats (Elix, 1996).

Thus questions arise as to possible differences between fungal assemblages across a range of lichens. Does lichen habitat and microenvironment determine the nature of associated non-mycobiont fungi? Do growth form (fruticose vs. foliose vs. crustose) and physiology (including green-algal vs. cyanobacterial lichens) factor into the nature of "free living" fungi found associated with lichens? Do lichens control the presence and absence of fungi, or the frequency of fungi by production of lichen acids, thus determining the configuration of associated fungal assemblages?

Examination of these questions follows. Four species of ground dwelling, mat forming, green-algal, fruticose lichens of the genus *Cladonia*, i.e. *C. amaurocraea*; *C. arbuscula; C. rangiferina*; and *C. stellaris*, were surveyed for their fungal assemblages. *C. arbuscula* is selected over *C. mitis* due to the prevalence of *C. arbuscula* in the sampling region, and the difficulty separating the two lichen forming fungal species in the field. Two additional fruticose lichens are included: 1) *Stereocaulon alpinum* (ground dwelling, mat forming, cyanobacterial and green-algal) and 2) *Evernia mesomorpha* (epiphytic, tufted, and green-algal). Additionally, two leafy-foliose lichens are also included, i.e. *Vulpicida pinastri* (epiphytic/wood colonizing, and green-algal) and *Peltigera* spp. (ground dwelling, cyanobacterial and sometimes green-algal).

Materials and Methods

Substratum Sampling and Fungal Isolation:

Samples from mats of *Cladonia amaurocraea*, *C. arbuscula*, *C. rangiferina*, *C. stellaris*, *Evernia mesomorpha*, *Vulpicida pinastri, Stereocaulon alpinum*, and *Peltigera* spp. were collected (August 5th, 2003) from the Payuk lake transects as previously described (Chapter 4 – sect. *site descriptions*, & fig. 4.1). Samples were transported in labelled plastic bags to University of Manitoba laboratories for further processing. Simultaneously with initiation of fungal isolations, the lichen collections were surveyed for secondary compounds using thin-layer chromatography (TLC) (appendix 14d), according to the methods of Culberson *et al.* (1972; 1974), so as to confirm field identifications. All identifications were confirmed except two of the five *Cladonia amaurocraea* collections (vouchers of scanned images for the TLC plates are available in molecular laboratory of

Dr. Piercey-Normore, U of M). Isolation methods for the fungi were as previously outlined in Chapter 2 (sect. *Surveying Fungal Assemblages*) for *Pleurozium schreberi.*

Data Analysis:

Frequency calculations, principle component analysis (PCA), multiple discriminate analysis (MDA), and occurrence computations were performed. Frequencies for each fungal taxon on 20 washed lichen pieces derived from each sample of the eight lichen taxa were calculated as previously detailed (Chapter 2 – sect. *data analysis*). Log transformed fungal assemblage frequency data was subjected to PCA for fungal data from the eight lichen taxa. The first three component axes from the PCA analysis were subjected to a MDA analysis, examining any separation of the fungal assemblage species composition found between samples of *Cladonia rangiferina*, *C. arbuscula*, *C. amaurocraea*, *C. stellaris*, *Evernia mesomorpha*, *Vulpicida pinastri*, *Stereocaulon alpinum*, and *Peltigera* spp. (Kenkel & Booth, 1992). PCA and MDA analyses were carried out as previously described (Chapter 2 – sect. *data analysis*).

Occurrences of each of the fungal taxa were taken as the number of samples a particular taxon was encountered in the five samples taken for each lichen (Krebs, 1972). In the case of *C. amaurocraea*, there were fungal occurrences over only three samples.

Results

Multiple discriminate analysis comparisons between the various lichen taxa sampled yielded significant differences among fungal assemblages ($p = 0.05$) (appendices 13a-c). The chi squared value was $\chi^2 = 71.09$, d.f. = 24. The scatter plot constructed from the multiple discriminate analysis (fig. 5.1) shows the assemblages on *Cladonia arbuscula* and *C. stellaris* to be the most closely associated lichens in the study. The fungal assemblages of *Peltigera* appear to be distinctly different from the other lichens sampled, as virtually no over lap was seen at 95% confidence levels. The fungal assemblages of *C. rangiferina* and *Vulpicida pinastri* also appear to be closely associated. The assemblage associated with *Stereocaulon alpinum* is similar to those of *C. rangiferina* and *V. pinastri*. *Evernia mesomorpha* and *Peltigera* spp. mycota tended to differ from the assemblages of *C. rangiferina*; *S. alpinum*; and *V. pinastri.*

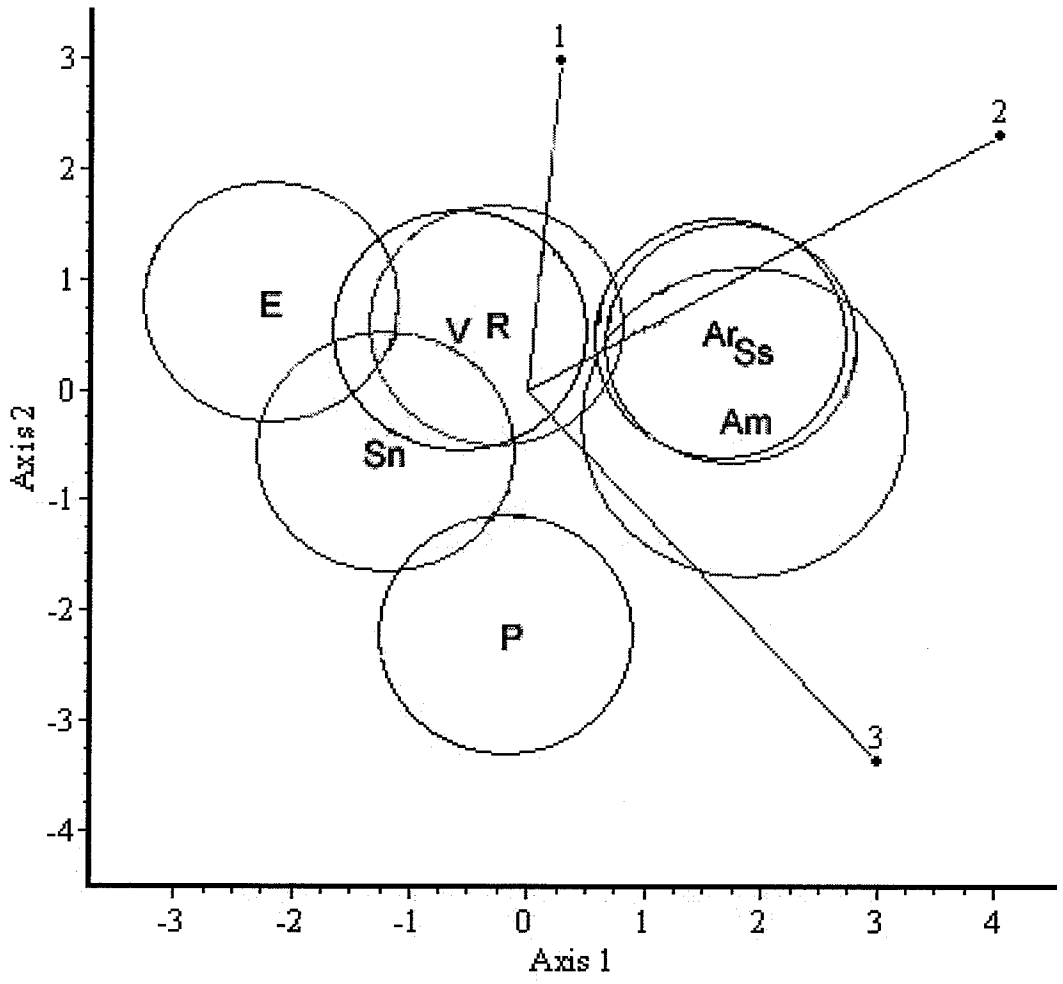


Fig. 5.1 - Graph of ordinations from a multiple discriminate analysis of the object scores from a principle component analysis for the fungal assemblages found in *Cladonia amaurocraea* (Am), *C. arbuscula* (Ar), *C. rangiferina* (R), *C. stellaris* (Ss), *Evernia mesomorpha* (E), *Peltigera* spp. (P), *Stereocaulon alpinum* (Sn), & *Vulpicida pinastri* (V) for data set. Circles represent the 95% confidence intervals (C.I.) for the respective lichen samples of each taxa.

Fungi previously shown (Chapters 2 & 3, tables 2.2 & 3.2) to be of high occurrence in lichen mats, i.e. *Absidia coerulea*; *Alternaria* spp.; *Cladosporium* # 1; *Epicoccum purpurascens*; *Mucor* # 1; *Mucor* #10; *Mortierella isabellina/vinacea* complex; *Penicillium* (non-sclerotial); and *Trichoderma* spp. were generally above a mean occurrence of 50% for *Alternaria* spp. (81%), *Cladosporium* # 1 (49%); and *Penicillium* (non-sclerotial) (91%); across all 8 lichens in this study (table 5.1). *Absidia*

coerulea (12%); *Mortierella/vinacea* complex (46%); and *Mucor* # 1 (5%) were at < 50% occurrences. *Epicoccum purpurascens* occurred at < 5% (on *Evernia mesomorpha* only) and *Trichoderma* was not isolated from any of the lichens in this study (photographs of select fungal taxa can be seen in appendix 15a).

Table 5.1 - Occurrence of fungal taxa on various lichens. Species of lichen forming fungi are labelled across the top, and taxa of non-lichen forming fungi associated with the lichens are labelled on the left.

Fungal Taxa	*Peltigera spp.*	*Cladonia amaurocrea*	*Cladonia arbuscula*	*Cladonia stellaris*	*Cladonia rangiferina*	*Vulpicida pinastri*	*Stereocaulon alpinum*	*Evernia mesomorpha*
Absidia coerulea	0	0.33	0.2	0.2	0	0	0.2	0
Alternaria spp.	1	0.67	0.8	0.8	0.8	0.6	0.6	1
Cladosporium # 1	0.8	0.33	0.4	0.2	0.2	0.6	0.4	1
Cladosporium # 2	0.2	0	0	0	0	0.4	0	0
Cunninghamella elegans	0	0.33	0.6	0.4	0.2	0	0.2	0
Epicoccum purpurascens	0	0	0	0	0	0	0	0.2
Mortierella isabellina/vinacea complex	0.6	0.67	0.8	0.6	0.4	0.4	0.2	0
Mortierella # 7	0.2	0	0	0	0	0	0	0
Mucor # 1	0.2	0	0	0	0	0	0.2	0
Mucor # 4	0.4	0.33	0	0	0	0	0	0
Mucor # 5	0.6	0	0	0	0	0	0	0
Mucor # 10	1	1	0.8	0.8	0.6	0.8	0.8	0.2
Mucor spinosus	0.6	0.33	0.2	0.4	0.2	0	0	0
Penicillium (non-sclerotial)	0.8	1	1	1	1	1	1	0.4
Penicillium (sclerotial)	0	0.33	0.4	0.4	0.4	0.4	0.2	0.2
Penicillium # 5	0	0	0	0.2	0	0.2	0	0
Phycomyces blaksleanus	0.2	0	0	0	0	0	0	0
Sphareopsidales # 23	0.2	0	0	0	0	0	0	0
Mycelia Sterilia # 1	0	0	0.2	0	0.4	0	0.4	0
Mycelia Sterilia # 14	0	0	0.2	0	0.2	0	0	0
Mycelia Sterilia # 24	0	0	0	0.2	0	0	0	0
Mycelia Sterilia # 38	0	0	0	0	0	0.2	0	0

Diversity of fungi across all the lichens studied, ranged from 6-13 taxa (table 5.1). Mean number of taxa was ten, with a standard deviation of two. At 13 taxa, the number of fungal taxa composing the fungal assemblages on *Peltigera* spp. were more than one standard deviation above the mean value of the mycota. *Evernia mesomorpha*, containing six fungal taxa among its fungal assemblages, was two standard deviations below the mean. Mean occurrences of the 22 fungal taxa on the lichens ran from 31% (*Peltigera* spp.) down to 14% (*E. mesomorpha*) with an average of 22.4%. At a standard deviation

of 5.1%, the occurrences of the assemblages on *Peltigera* spp. were somewhat significantly greater than the mean of means.

Zygomycetes were 62% (8 of 13) of the *Peltigera* spp. assemblages; 60% (6 of 10) of the *Cladonia amaurocrea* assemblages; 50% (5 of 10) of the *Stereocaulon alpinum* mycota; 45% (5 of 11) of the *C. arbuscula* and *C. stellaris* assemblages; 40% (4 of 10) of the *C. rangiferina* assemblages; 22% (2 of 9) of the *Vulpicida pinastri* assemblages and 17% (1 of 6) of *Evernia mesomorpha* mycota (table 5.1). The Zygomycete taxa across the lichens ranged from 1-8 ($\bar{x} = 4.5 \pm 2.2$). The assemblage of *Peltigera* spp. was high, more than one standard deviation greater than the mean for Zygomycetes, and assemblages of *Vulpicida pinastri* and *Evernia mesomorpha* were low (more than one standard deviation below the mean) in the number of Zygomycetes.

Among the low occurrence Zygomycetes and other fungi, *Mortierella* # 7; *Mucor* # 5; and *Phycomyces blakesleeanus* occurred solely on the lichen *Peltigera* spp., along with Sphaeropsidales # 23. *Mucor* # 1 occurred only on the lichens *Peltigera* spp. and *Stereocaulon alpinum*. Pattern of occurrences of non-lichen forming fungi *Absidia coerulea*, *Cunninghamella elegans*, *Mortierella isabellina/vinacea* complex and *Mucor spinosus* generally showed elevated values over the assemblages on the lichens *Cladonia amaurocraea*, *C. arbuscula*, and *C. stellaris*.

Multiple discriminate analysis groupings correspond reasonably well with the presence of secondary compounds seen in the lichen taxa (fig. 5.1). Secondary compounds across the lichens group *Cladonia amaurocraea*, *C. arbuscula*, and *C. stellaris* with usnic acid (table 5.2), as well as, *C. rangiferina* and *Stereocaulon alpinum* with atranorin. Among other secondary compounds produced, *Vulpicida pinastri* produces vulpinic acid, *Evernia mesomorpha* produces divaricatic acid, and *Peltigera* spp. can produce gyrophoric acid.

Table 5.2 – Secondary compounds listed for these lichen taxa were determined in this study or were obtained from the literature (Brodo *et al.*, 2001; Thomson, 1984) as indicated by an asterisk after the species name.

Lichen Taxon	Secondary Compounds
*Peltigera spp.**	Tenuiorin, Methyl Gyrophorate, Gyrophoric acid, & Triterpene
Cladonia amaurocraea	Usnic acid & Barbatic acid
Cladonia arbuscula	Usnic acid
Cladonia stellaris	Usnic acid & Perlatolic acid
*Stereocaulon alpinus**	Atranorin, Lobaric acid, & β-sitosterin
Cladonia rangiferina	Atranorin & Fumarprotocetraric acid
*Vulpicida pinastri**	Vulpinic acid, Pinastric acid, Usnic acid, & Zeorin
*Evernia mesomorpha**	Divaricatic acid

Discussion

The non-Zygomyceteous fungi previously reported (Chapters 2 & 3, tables 2.2 & 3.2) as high in occurrence on *Cladonia mitis* and *C. rangiferina*, *Alternaria* spp., *Cladosporium* # 1, and *Penicillium* (non-sclerotial), showed no pattern of difference across the lichens in this study. Generally the occurrence values of these fungi were high across all lichen forming fungal taxa sampled and promote the general observation that, as is the case for *Cladonia* spp. (Petrini *et al.*, 1990), fungal assemblages may not differ on different lichen forming fungi.

However, among Zygomycetes of high occurrence in lichen mats at different times of season (Chapter 2, table 2.2) and in different parts of the mat profile (canopy and base collections) (Chapter 3, table 3.2) *Absidia coerulea* and *Mortierella isabellina/vinacea* complex were of interest. *Absidia coerulea* had recognizable patterns of elevated occurrences on *Stereocaulon alpinum* and the *Cladonia amaurocraea*, *C. arbuscula*, and *C. stellaris* group. *Mortierella isabellina/vinacea* complex had recognizable patterns of elevated occurrences on *Peltigera* spp. and the *Cladonia amaurocraea*, *C. arbuscula*, and *C. stellaris* group. Occurrences of *Cunninghamella elegans* and *Mucor spinosus*, not previously isolated in earlier studies, were similarly higher on the assemblages of *C. amaurocraea*, *C. arbuscula* and *C. stellaris*. Thus, the Zygomycetes demonstrate significant differences among assemblages on different species of lichen forming fungi, consistent with the observations of Girlanda *et al.* (1997) that

indicate strong dissimilarity of fungal assemblages on two foliose lichens, i.e. *Parmelia taractica* and *Peltigera praetextata*. Assemblage groups detected by multiple discriminate analysis in this study (fig. 5.1) also corroborated differences associated fungal assemblages across a range of lichen forming fungi.

Absence of *Trichoderma* spp., which had high occurrences in the previous studies (Chapters 2 & 3, tables 2.2 & 3.2), across the Payuk lake lichens of this study, likely reflect the fact that the collection sites were mainly along bedrock slopes. *Trichoderma* spp. are known competitors with Zygomycetatious fungi of *Mortierella*. Bedrock slopes are well drained locations, with high potential for extreme drying and wetting cycles. This can limit *Trichoderma* spp., as *Mortierella* spp. out competes *Trichoderma* in well drained sites (Christensen, 1969).

Diversity of the lichen associated fungal assemblages, delineated the assemblages on *Peltigera* spp., *Evernia mesomorpha*, and *Vulpicida pinastri* as distinct from the fungal assemblages of the other lichens studied. Additionally, the low occurrence fungi: *Mortierella* # 7; *Mucor* # 1; *Mucor* # 5; and *Phycomyces blakesleeanus* (Zygomycetes) and Sphaeropsidales # 23 (Deuteromycete) were found either solely on *Peltigera* spp. or on *Peltigera* spp. and *Stereocaulon alpinum*. This suggests possibly the Zygomycetes in the assemblages on *Peltigera* spp. and *S. alpinum* were favourably supplied with nutrients.

Among nutrients known to influence fungi, moisture levels in a lichen mat are known to vary according to the complexity of thallus branching in the canopy (Kershaw & Field, 1975; Kershaw & Harris, 1971; Kershaw & Rouse, 1971). Also, lichen epiphytes live in a drier habitat and represent a more xeric microenvironment the lichen associated fungal assemblages would be subjected to (Pugh, 1980). When looking across the lichens in my study both form and habitat of the thalli suggested a gradient of moisture for the grouped assemblages on: 1) *Peltigera* spp. (foliose, ground dwelling); 2) *Cladonia amaurocraea*, *C. arbuscula*, *C. stellaris* (fruticose, closed mats); 3) *Stereocaulon alpinum* (fruticose, open mats), *C. rangiferina* (fruticose, open mats), and *Vulpicida pinastri* (foliose, epiphytic); and 4) *Evernia mesomorpha* (fruticose, epiphytic). Generally ground dwelling lichens have a more diverse mycota in their assemblages than epiphytic lichens, particularly for Zygomycetes which are known inhabitants of moist

environments (Chowdhery *et al.*, 1982). The Zygomycete *Cunninghamella elegans* was found only on fruticose ground dwelling lichens.

Nitrogen as a nutrient is reported to be transported from older portions of lichen thalli to the growing canopy (fruticose lichens) or edge of the thallus (foliose lichens) (Dahlman *et al.*, 2002; Hyvarinen & Crittenden, 2000). This mobilized nitrogen may be used to promote growth and sporulation of non-lichen forming fungi associated with the lichen. As previously mentioned, *Cladosporium* # 1 and *Cladosporium* # 2 seem to require provision of nitrogen into growth media in order to sporulate (Chapter 3 – sect. Discussion). In this study, *Mucor* # 1, occurred solely on the cyanobacteria containing lichens *Peltigera* spp. and *Stereocaulon alpinum*. Further evidence for fungal response to available nitrogen was seen in the previously mentioned occurrences of *Mortierella* # 7, *Mucor* # 1, *Mucor* # 5, *Phycomyces blakesleeanus* and Sphaeropsidales # 23 on both or one of *Peltigera* spp. and *Stereocaulon alpinum*.

Lichen acids, as determined in the laboratory or from the literature for the eight lichens (table 5.2), demonstrate a possible trend related to the multiple discriminate analysis of the assemblage frequencies of the fungi (fig. 5.1). Somewhat striking is the fact that the grouping of assemblages found on *Cladonia amaurocraea*, *C. arbuscula* and *C. stellaris* was an usnic acid exposed group, as all three of these lichens were experimentally determined to produce the acid. Usnic acid, in terms of its antibacterial activity, is effective more so than atranorin and vulpinic acid (Konig & Wright, 1999). Reduced competition for substrates (i.e. carbohydrates, particularly simple sugars), due to decreased bacterial colonization and diminished activity for affected fungi, may explain the higher occurrences of fungi, including Zygomycetes, on lichens (Brodie & Blakeman, 1976; Girlanda *et al.*, 1997). *Stereocaulon alpinum* and *C. rangiferina* both produce atranorin (table 5.2), and the assemblages group along with that of *Vulpicida pinastri* which produces vulpinic acid (table 5.2), a known poison for carnivores, insects and molluscs (Elix, 1996). Zygomycetes may be severely limited by vulpinic acid which might explain the virtual absence of these fungi on *Vulpicida pinastri* (tables 5.1 & 5.2).

Chapter 6

Discussion

Major Conclusions

A wide variety of fungal assemblages can be found both on lichens and within lichen mats. A total of 31 fungal taxa were found associated with nine lichen taxa. The non-lichen forming fungi belong to the genera *Absidia*, *Alternaria*, *Aspergillus*, *Cladosporium*, *Cunninghamella*, *Epicoccum*, *Penicillium*, *Phycomyces*, *Mortierella*, *Mucor*, *Rhizopus*, and *Trichoderma*, as well as fungal taxa from the class Sphaeropsidales (Chapters 2, 3 & 5). All of these fungal taxa are considered lichen-associated taxa rather than true lichenicolous fungi. Lichenicolous fungi are fungi found only on lichens, and tend to be primarily non-lichenized fungi, eg. *Lichenopeltella*, or are secondarily lichenized, eg. *Arthonia* (Lucking & Bernecker-Lucking, 2002). Lichenicolous fungi utilize the host photobiont, acting as commensalists or eventually damaging the lichen while developing reproductive structures (Lucking & Bernecker-Lucking, 2002). The fungal taxa isolated from this paper, on the other hand, are mainly soil and litter inhabiting fungi (Keller & Bidochka, 1998; Petrini *et al.*, 1990), and are not extremely host specific. They are likely present due to the microhabitats offered by the various lichen taxa.

Overall temporal, spatial, morphological and physiological changes were seen to affect the fungal assemblages found within lichen mats. Lichen mats form unique microhabitats, allowing for the colonization of fungi which otherwise might not be present. The influence of microhabitat on the presence of individual fungi in fungal assemblages was interpreted to be related to: 1) temperature and moisture interactions and gradients; 2) nitrogen physiology in and along lichen thalli; 3) lichen acids; and 4) fungal inoculum and dispersal.

Changes in the moisture levels on lichen surfaces, as inferred by their morphology and habitat, appears to be the most microenvironmentally influential factor controlling the fungal assemblages associated with lichen thalli. The most distinguishing feature among the fungal assemblages across the various lichen forming fungal taxa surveyed, was the number and frequency of Zygomycetes (Chapters 2, 3 & 5) which are indicators of predominantly highly moist environments (Chowdhery *et al.*, 1982). Also, overall increases in species richness and occurrence of most of the fungi studied were seen in

mats of *Cladonia mitis* and *C. rangiferina* in the September collections as compared to the July collections (Chapter 2), likely due to the increased rainfall and cooler temperatures associated with the fall season, which facilitate fungal growth (Keller & Bidochka, 1998).

Moisture gradients promote a greater diversity of fungi in mats, due to moist habitat niches in the region of lichen bases, and dry habitat niches in the lichen canopies. Vertical stratification of moisture occurs in highly branched mats of *Cladonia*, creating a stratified microhabitat (Kershaw & Rouse, 1971), and this thesis (Chapter 3) supports the idea that such stratification causes spatial changes in the fungal assemblages found within lichen mats. In general lichen associated fungi respond by either employing a 'survivor-escaper' life strategy (Pugh, 1980), found predominantly in the canopies of highly branched *Cladonia* mats, or by employing a 'ruderal' life strategy (Pugh, 1980), found predominantly in the lower strata of highly branched *Cladonia* mats (Chapter 3).

Pleurozium schreberi forms more highly branched mats which capture and retain moisture better than *Cladonia mitis* and *C. rangiferina* mats (Ipatov & Trofimets, 1988). *Pleurozium schreberi* mats differed from mats of *Cladonia mitis* and *C. rangiferina*, in the increased presence of *Mucor* and *Mortierella*, Zygomycetes and 'ruderals', while *C. rangiferina* and *C. mitis* had an increased presence of *Alternaria* and *Epicoccum*, ascomycetes and 'survivors-escapers' (Pugh, 1980; Chapter 2). The presence of *P. schreberi* may be partially responsible for the increased number of Zygomycetes seen in *Peltigera* spp., as compared with the other lichen forming fungal taxa sampled in northern Manitoba, as *Peltigera* samples were always collected on *P. schreberi* mats (Chapter 5). As well *Cladosporium* # 2 was seen to be associated with foliose lichens in northern Manitoba, being absent from all fruticose lichen samples in northern Manitoba, and *Cunninghamella elegans* was restricted to ground dwelling fruticose lichens, further emphasizing that growth form and morphology affect moisture, and hence fungi, by alteration of microhabitat (Chapter 5).

The photobiont cells of lichens are suspected to play a major role in determination of the microhabitat available to fungi by lichens. Excess glucose is leaked from algal cells onto and within the lichen thallus, establishing a gradient by stem flow or dilution. Additionally, in lichen mats, nitrogen is mobilized in the base region of the thalli and

passively transported along the podetium to the canopy, thereby establishing a nitrogen gradient. Some lichen photobionts are nitrogen fixing cyanobacteria rather than green algal cells. Cyanobacteria increase the availability of nitrogen in lichens due to the nitrogen fixing photobionts (Nash III, 1996). Perhaps fungi respond to this available nitrogen as indicated by the fact that *Mucor* # 1 was only isolated from *Stereocaulon alpinum* and *Peltigera* spp. in northern Manitoba (Chapter 5), both of which have cyanobionts (Brodo *et al.*, 2001; Thomson, 1984).

Lichen secondary compounds can also have antifungal properties, which could decrease the fungal competition and partially explain the high occurrence of 'ruderal' and 'survivor' fungal taxa seen in the lichen mats (Elix, 1996; Pugh, 1980). Though circumstantial, the presence of specific lichen secondary compounds may affect the composition of fungal assemblages (Chapter 5). Petrini *et al.* (1990) and Girlanda *et al.* (1997) concluded that fungal colonizers of lichens are insensitive to the secondary compounds found in their hosts. While the majority of fungi occurring in fungal assemblages appear to be unaffected by the secondary compounds produced (Chapters 2, 3, & 5), confirming these results, the production of secondary compounds likely play a more general role in influencing microhabitat. Usnic acid for example, found in six of the nine lichen taxa sampled, is well known for its antibacterial activities (Elix, 1996). Limitation of bacteria favors colonization of substrata by antibiotic resistant fungi, which are otherwise competitors for lichen sugar exudates (Brodie & Blakeman, 1976; Girlanda *et al.*, 1997).

Inoculum, responsible for establishing the presence of fungal assemblages within lichen mats, at least partially arises from the soil litter layers beneath lichen mats as living hyphae, asexual spores and sexually produced spores (Chapter 3). Inoculum may be dispersed by wind, insects, or water, or incorporated with lichen fragments. Reindeer lichens particularly reproduce frequently by fragmentation as a result of disturbance (Yarranton, 1975). Gene dispersal in *Cladonia arbuscula* was seen to be quite efficient over the 2 km area surveyed in northern Manitoba (Chapter 4). This dispersal could be due to pycnidia, thallus fragmentation, or apothecia. While fragmentation of thallus has been assumed to be the dominant form of reproduction in reindeer lichens, as apothecia are infrequently produced (Brodo *et al.*, 2001), this may not be the case in this situation

as 40% of the *C. arbuscula* samples examined had apothecia present (Chapter 4). Thus little inference about the effectiveness and distance of the dispersal of lichen thallus fragments and the role of fragments in providing lichen associated fungal inoculum is possible (Chapter 4).

Future Studies

Future researchers may wish to examine spatial relations, temporal changes and inoculum sources in lichen mats more closely. Lichens sampled could be dissected by years of growth (Ahti, 1959), subsamples being examined separately for fungal assemblages, as age of the lichen thallus affects its physiology (Kershaw & Harris, 1971). This might shed further light on the niche requirements of the fungal taxa found, as base and canopy subdivisions used in this thesis were crude classifications of mat 'architecture' and demonstrated spatial overlap among canopy and base fungal taxa (Chapter 3). Soil litter layers directly beneath where lichens are sampled could be sampled simultaneously, eliminating potential spatial and temporal effects (Chapters 2 & 3) that might influence the fungal assemblages found in the soil litter layer as compared with the lichen mat. If soil litter samples could be tied to specific lichen samples, direct comparisons, as opposed to the inferences seen in Chapter 3, could be made between the fungal taxa in the mat and beneath it. As well, lichen mats could be marked where they were sampled so that when sampling multiple times to investigate temporal effects, sampling can be restricted to the exact mat previously sampled, as opposed to the same 5x5m plot (Chapter 2), reducing potential variability in microhabitat.

Habitat itself could also be better classified, with data collected on daily precipitation and temperature factors at the actual collection sites, and probes monitoring internal humidity and temperature levels of the mat, since moisture gradients appear to be a key influencing factor in the assemblages of fungal taxa in the lichen mats (Chapters 2, 3, & 5). As well, effects of the type of photobiont present, cyanobacteria or green-algae, could also be more closely examined by expanding the number of cyanobiont lichens sampled for fungi, and ensuring similar habitats and growth form between phycobiont and cyanobiont lichens sampled, as nitrogen also appears to be an influential factor in the determination of fungal assemblages present in lichen mats (Chapters 3 & 5).

Dispersal could also be examined more clearly if both the mycobiont and photobiont were examined. If genotypes of mycobiont and photobiont correlated, it is more probable that the mycobiont and photobiont were dispersed together, which implies vegetative reproduction via fragmentation in *C. arbuscula* since *C. arbuscula* does not produce isidia or soredia.

Literature Cited

Ahti, T. 1997. Lichens of the boreal coniferous zone. *In* Lichen ecology. *Edited by* M. R. D. Seaward. Academic Press, New York.

Ahti, T. 1959. Studies on the caribou lichen stands of Newfoundland. Ann. Bot. Soc. 'Vanamo' 30: 1-44.

Ainsworth, G. C. 1973. Introduction and keys to higher taxa. *In* The fungi: An advanced treatise IVB. *Edited by* G. C. Ainsworth, F. K. Sparrow, & A. S. Sussman. Academic Press, New York.

Ananda, K., & Sridhar, K. R. 2004. Diversity of filamentous fungi on decomposing leaf and woody litter of mangrove forests in the southwest coast of India. Curr. Sci. **87**: 1431-1437.

Armstrong, R. A. 1994. Dispersal of soredia from individual soralia of the lichen Hypogymnia physodes (L.) Nyl. in a simple wind tunnel. Environ. & Exper. Bot. **34**: 39-46.

Arnheim, N., Krystal, M., Schmickel, R., Wilson, G., Ryder, O., & Zimmer, E. 1980. Molecular evidence for genetic exchanges among ribosomal genes on non-homologous chromosomes in man and apes. Proc. Nat. Acad. Sci. USA **77**: 7323-7327.

Azmi, O. R., & Seppelt, R. D. 1998. The broad-scale distribution of microfungi in the Windmill islands region, continental Antarctica. Polar Biol. **19**: 92-100.

Bailey, R. H. 1976. Ecological aspects of dispersal and establishment in lichens. *In* Lichenology: Progress and Problems. *Edited by* D. H. Brown, D. L. Hawksworth, and R. H. Bailey. Academic Press, New York.

Bailey, R. H. 1966. Studies on the dispersal of lichen soredia. J. Linn. Soc. Bot. **59**: 479-490.

Barnett, H. L., & Hunter, B. B. 1972. Illustrated genera of imperfect fungi (3rd ed.). Burgess Publishing Company, Minneapolis.

Beard, K. H., & DePriest, P. T. 1996. Genetic variation within and among mats of the reindeer lichen *Cladina subtenuis*. Lichenologist **28**: 171-182.

Bhat, P. R., & Kaveriappa, K. M. 1998. Aerial surface mycoflora of *Myristica fatua* var. magnifica and *Myristica malabarica* - the two endangered taxa of the western ghats. J. mycol. Plant Pathol. **28**: 288-293.

Booton, G. C., Floyd, G. L., & Fuerst, P. A. 2004. Multiple group I introns detected in the nuclear small subunit rDNA of the autosporic green alga *Selenastrum capricornutum*. Curr. Genet. **46**: 228-234.

Brodie, I. D. S., & Blakeman, J. P.1976. Competition for exogenous substrates in-vitro by leaf surface microorganisms and germination of conidia of *Botrytis cinerea*. Phys. Plant Pathol. **9**: 227-239.

Brodo, I. M., Sharnoff, S. D., & Sharnoff, S. 2001. Lichens of North America. Yale University Press, New Haven & London.

Carleton, T. J., & Dunham K. M. M. 2003. Distillation in a boreal mossy forest floor. Can. J. For. Res. **33**: 663-671.

Carleton, T. J., & Read, D. J. 1991. Ectomycorrhizae and nutrient transfer in conifer – feather moss ecosystems. Can. J. Bot. **69**: 778-785.

Chowdhery, H. J., Garg, K. L., & Jaitly, A. K. 1982. Occurrence of fungi in rhizosphere, rhizoplane, and non-rhizosphere zones of some mangroves. Indian J. Mar. Sci. **11**: 138-142.

Christensen, M. 1969. Soil microfungi of dry to mesic conifer-hardwood forests in northern Wisconsin. Ecology **50**: 9-27.

Coates, B. S., Hellmich, R. L., & Lewis, L. C. 2002. Nuclear small subunit rRNA group I intron variation among *Beauveria* spp. provide tools for strain identification and evidence of horizontal transfer. Curr. Genet. **41**: 414-424.

Cole, M. S., & Hawksworth, D. L. 2001. Lichenicolous fungi, mainly from the USA, including *Patriciomyces* gen. nov. Mycotaxon **77**: 305-338.

Crittenden, P. D. 1988. Nitrogen relations of mat-forming lichens. *In* Nitrogen, phosphorus and sulphur utilization by fungi. *Edited by* L. Boddy, R. Marchant & D. J. Read. Cambridge University Press, New York.

Culberson, C. F. 1972. Improved conditions and new data for the identification of lichen products by a standardized thin layer chromatographic method. J. Chromatogr. **72**: 113-125.

Culberson, C. F. 1974. Conditions for the use of merck silica gel 60-F-254 plates in the standardized thin layer chromatographic technique for lichen products. J. Chromatogr. **97**: 107-108.

Dahlman, L., Nasholm, T., & Palmqvist, K. 2002. Growth, nitrogen uptake, and resource allocation in the two tripartite lichens *Nephroma arcticum* and *Peltigera aphthosa* during nitrogen stress. New Phytol. **153**: 307-315.

Dennis, R. W. G. 1968. British Ascomycetes (Rev. ed.). Verlag Von J. Cramer, Lehre.

DePriest, P. T. 1993a. Molecular innovations in lichen systematics: The use of ribosomal and intron nucleotide sequences in the *Cladonia chlorophaea* complex. Bryologist **96**: 314-325.

DePriest, P. T. 1993b. Small subunit rDNA variation in a population of lichen fungi due to optional group-I introns. Gene (Amsterdam) **134**: 67-74.

Depriest, P. T., & Been, M. D. 1992. Numerous group I introns with variable distributions in the ribosomal DNA of a lichen fungus. J. Mol. Biol. **228**: 315-321.

Dobbeler, P. 2001. *Polytrichadelphus magellanicus* sensu lato (Musci) and its ascomycetes – different fungi on different hosts. Sendtnera **7**: 35-45.

Dobbeler, P. 1997. Biodiversity of bryophilous ascomycetes. Biodivers. Conserv. **6**: 721-738.

Durrell, L. W. & Shields, L. M. 1960. Fungi isolated in culture from soils of the Nevada test site. Mycologia **52**: 636-641.

Elix, J. A. 1996. Biochemistry and secondary metabolites. *In* Lichen biology. *Edited by* T. H. Nash III. Cambridge University Press, UK.

Ellis, M. B. 1976. More dematiaceous hyphomycetes. Commonwealth mycological institute, England.

Ellis, M. B. 1971. Dematiaceous hyphomycetes. Commonwealth mycological institute, England.

Environment Canada 2003. Climate data online [online]. Available from http://www.climate.weatheroffice.ec.gc.ca/advanceSearch/searchHistoricDataStations_e.html [cited 7 July 2005].

Farrar, J. F., & Smith, D. C. 1976. Ecological physiology of the lichen *Hypogymnia physodes*. III. The importance of the rewetting phase. New Phytol. **77**: 115-125.

Felix, H. 1988. Fungi on bryophytes, a review. Bot. Helv. **98**: 239-269.

Gargas, A., & DePriest, P.T. 1996. A nomenclature for fungal PCR primers with examples from intron-containing SSU rDNA. Mycologia **88**: 745-748.

Girlanda, M. Isocrono, S. Bianco, C., & Luppi-Mosca, A. M. 1997. Two foliose lichens as microfungal ecological niches. Mycologia **89**: 531-536.

Gobbi, E., Firrao, G., Carpanelli, A., Locci, R., & Van Alfen, N. K. 2003. Mapping and characterization of polymorphism in mtDNA of *Cryphonectria parasitica*: Evidence of the presence of an optional intron. Fungal Genet. Biol. **40**: 215-224.

Greenfield, L. G. 1999. Weight loss and release of mineral nitrogen from decomposing pollen. Soil Biol. Biochem. **31**: 351-361.

Greenfield, L. G. 1993. Decomposition studies on New Zealand and antarctic lichens. Lichenologist **25**: 73-82.

Grime, J. P. 1979. Plant strategies and vegetation processes. Wiley, New York.

Grube, M., DePriest, P. T., Gargas, A., & Hafellner, Y. 1995. DNA isolation from lichen ascomata. Mycol. Res. **99**: 1321-1324.

Hageman, C., & Fahselt, D. 1992. Geographical distribution of enzyme polymorphisms in the lichen *Umbilicaria mammulata*. Bryologist **95**: 316-323.

Hale, M. E. 1983. The biology of lichens. Edward Arnold, London.

Heinken, T. 1999. Dispersal patterns of terricolous lichens by thallus fragments. Lichenologist **31**: 603-612.

Hesseltine, C. W. 1955. Genera of Mucorales with notes on their synonymy. Mycologia **47**: 344-363.

Hesseltine, C. W., & Ellis, J. J. 1973. Introduction and keys to higher taxa. *In* The fungi: An advanced treatise IVB. *Edited by* G. C. Ainsworth, F. K. Sparrow, & A. S. Sussman. Academic Press, New York.

Honegger, R., Zippler, U., Gansner, H., & Scherrer, S. 2004. Mating systems in the genus *Xanthoria* (lichen-forming ascomycetes). Mycol. Res. **108**: 480-488.

Hyvarinen, M., & Crittenden, P. D. 2000. P-33 translocation in the thallus of the mat-forming lichen *Cladonia portentosa*. New Phytol. **145**: 281-288.

Ing, B. 1994. Tansley Review 62. The phytosociology of myxomycetes. New Phytol. **126**: 175-201.

Ipatov, V. S., & Trofimets, V. I. 1988. Effect of lichen and green moss covers on water regimen in the upper root-inhabited soil layer in dry pine forests. Ekologiya (Moscow) **1**: 19-23.

Järvinen, P., Lemmetyinen, J., Savolainen, O. & Sopanen, T. 2003. DNA sequence variation in *BpMADS2* gene in two populations of *Betula pendula*. Mol. Ecol. **12**: 369-384.

Joensson, A. G. 1967. Pilot-plant production of protease by Alternaria Tenuissima. Appl. Microbiol. **15**: 319-324.

Johnson, E. A. 1981. Vegetation organization and dynamics of lichen woodland communities in the Northwest Territories, Canada. Ecology **62**: 202-215.

Keller, L., & Bidochka, M. J. 1998. Habitat and temporal differences among soil microfungal assemblages in Ontario. Can. J. Bot. **76**: 1798-1805.

Kenkel, N. C., & Booth, T. 1992. Multivariate analysis in fungal ecology. *In* The fungal community – Its organization and role in the ecosystem (2nd ed.). *Edited by* Carrol, George C., & Donald T. Wicklow. Marcel Deeker inc., New York.

Kershaw, K. A. 1985. Physiological ecology of lichens. Cambridge University Press, Cambridge.

Kershaw, K. A., & Field, G. F. 1975. Studies on lichen-dominated systems. XV. The temperature and humidity profiles in a *Cladina alpestris* mat. Can. J. Bot. **53**: 2614-2620.

Kershaw, K. A., & Harris, G. P. 1971. A technique for measuring the light profile in a lichen canopy. Can. J. Bot. **49**: 609-611.

Kershaw, K. A., & Rouse, W. R. 1971. Studies on lichen-dominated systems. I. The water relations of *Cladonia alpestris* in spruce-lichen woodland in northern Ontario. Can. J. Bot. **49**: 1389-1399.

Kiss, T. 1985. The life-strategy system of lichens – a proposal. Abstr. Bot. 9: 56-66.

Knoops, J. M. H., Nash III, T. H., Boucher, V. L., & Schlesinger, W. H. 1991. Mineral cycling and epiphytic lichens: implications at the ecosystem level. Lichenologist **23**: 309-321.

Konig, G. M., & Wright, A. D. 1999. ^{1}H and ^{13}C-NMR and biological activity investigations of four lichen-derived compounds. Phytochem. Anal. **10**: 279-284.

Krebs, C. J. 1972. Ecology. The experimental analysis of distribution and abundance. Harper & Row Publishing, New York.

La Roi, G. H., & Stringer, M. H. L. 1976. Ecological studies in the boreal spruce-fir forests of the North American tiaga. II. Analysis of the bryophyte flora. Can. J. Bot. **54**: 619-643.

Lauhn-Jensen, D. M. 1987. Geological highway map of Manitoba. Province of Manitoba, Manitoba.

Lee, E., Kenkel, N. C., & Booth, T. 1996a. Atmospheric deposition of macronutrients by pollen in the boreal forest. Ecoscience **3**: 304-309.

Lee, E., Kenkel, N. C., & Booth, T. 1996b. Pollen deposition in the boreal forest of west-central Canada. Can. J. Bot. **74**: 1265-1272.

Linskens, H. F., Bargagli, R., Focardi, S., & Cresti, M. 1991. Antarctic moss turf as pollen traps. Proc. Kon. Ned. Akad. V. Wetensch. **94**: 233-241.

Longton, R. E. 1988. The biology of polar bryophytes and lichens. Cambridge University Press, New York.

Lucking, R., & Bernecker-Lucking, A. 2000. Lichen feeders and lichenicolous fungi: Do they affect dispersal and diversity in tropical foliicolous lichen communities?. Ecotropica **6**: 23-41.

Malloch, D. 1981. Molds: Their isolation, cultivation, and identification. University of Toronto Press, Toronto.

Maltzahn, v. K. E., & McQuarrie, J. G. 1958. The effect of gibberellic acid on the growth of protonema in Splachnum ampullaceum (L.) Hedw. Nature 181: 1139-1140.

Marshall, W. A. 1996. Aerial dispersal of lichen soredia in the maritime antrctic. New Phytol. **134**: 523-530.

Martin, G. W., & Alexopoulos, C. J. 1969. The myxomycetes. University of Iowa Press: Iowa.

Miller, O. K. jr. 1984. Mushrooms of north America. E. P. Dutton, New York.

Moller, C., & Dreyfuss, M. M. 1996. Microfungi from Antarctic lichens, mosses, and vascular plants. Mycologia 88: 922-933.

Moore, D. S. 2000. The basic practice of statistics (2nd ed.). W.H. Freeman & Company, New York.

Moore, T. R. 1984. Litter decomposition in a subarctic spruce-lichen woodland, eastern Canada. Ecology **65**: 299-308.

Muhsin, T. M., & Booth, T. 1987. Fungi associated with halophytes of an inland salt marsh, Manitoba, Canada. Can. J. Bot. **65**: 1137-1151.

Nash III, T. H. 1996. Nitrogen, its metabolism and potential contribution to ecosystems. *In* Lichen Biology. *Edited by* Thomas H. Nash III. Cambridge University Press, UK.

Oechel, W. C., & Van Cleve, K. 1986. The role of bryophytes in nutrient cycling in the tiaga. *In* Forest ecosystems in the Alaskan tiaga: a synthesis of structure and function. *Edited by* K. Van Cleve, F. S. Chapin III, P. W. Flanagan, L. A. Viereck, & C. T. Dyrness. Ecol. Stud. Anal. Synth. **57**: 121-137.

Parinkina, O. M., Pereverzev, V. N., & Piyn, T. Kh. 1995. Features of decomposition of soil surface lichens in the mountain tundra and northern and southern tiaga. Eurasian Soil Sci. **27**: 70-81.

Petrini, O., Hake, U., & Dreyfuss. M. M. 1990. An analysis of fungal communities isolated from fruticose lichens. Mycologia **82**: 444-451.

Piercey-Normore, M. D. 2004. Group I intron-like insertions in SSU rDNA of *Cladonia gracilis* and *C. rangiferina*. Lichenologist **36**: 365-380.

Piercey-Normore, M. D. 2003. A field survey of the genus Cladonia (Ascomycotina) in Manitoba, Canada. Mycotaxon **86**: 233-247.

Pocock, K., Duckett, J. G., Grolle, R., Mohamed, M. A. H., & Pang, W. C. 1984. Branched and swollen rhizoids in hepatics from montane rain forest in peninsular Malaya. J. Bryol. **13**: 241-246.

Podani, J. 2001. *SYN-TAX 2000.* Scientia Publishing, Budapest.

Printzen, C., & Ekman, S. 2003. Local population subdivision in the lichen *Cladonia subcervicornis* as revealed by mitochondrial cytochrome oxidase subunit 1 intron sequences. Mycologia **95**: 399-406.

Pugh, G. J. F. 1980. Presidential address. Strategies in fungal ecology. Trans. Br. Mycol. Soc. **75**: 1-14.

Richardson, D. H. S. 1975. The vanishing lichens. David & Charles, Vancouver.

Ritchie, J. C. 1956a. The vegetation of northern Manitoba. I. Studies in the southern spruce forest zone. Can. J. Bot. **34**: 523 – 561.

Ritchie, J. C. 1956b. The native plants of Churchhill, Manitoba, Canada. Can. J. Bot. **34**: 269-320.

Ritchie, J. C. 1960a. The vegetation of northern Manitoba. IV. The Caribou lake region. Can. J. Bot. **38**: 185-197.

Ritchie, J. C. 1960b. The vegetation of northern Manitoba. VI. The lower Hayes river region. Can. J. Bot. **38**: 769-788.

SAS Institute 2001. JMP In 4.04. Duxbury, Canada.

Seymour, F. A., Critten, P. D., Dickinson, M. J., Paoletti, M., Montiel, D., Cho, L., & Dyer, P. S. 2005. Breeding systems in the lichen-forming fungal genus *Cladonia*. Fungal Genet. Biol. **42**: 554-563.

Shay, C. T. 1984. The history of Manitoba's vegetation. *In* Natural Heritage of Manitoba: Legacy of the ice age. Edited by J. T. Teller.

Singer, R. 1986. The Agaricales in modern taxonomy. Koeltz Scientific Books, Germany.

Stenroos, S., Hyvonen, J., Myllys, L., Thell, A., & Ahti, T. 2002. Phylogeny of the genus *Cladonia* s.lat. (Cladoniaceae, Ascomycetes) inferred from molecular, morphological, and chemical data. Cladistics **18**: 237-278.

Tegler, B., & Kershaw, K. A. 1980. Studies on lichen-dominated systems. XXIII. The control of seasonal rates of net photosynthesis by moisture, light, and temperature in *Cladonia rangiferina*. Can. J. Bot. **58**: 1851-1858.

Thomson, J. W. 1984. American Arctic Lichens. 1. The Macrolichens. Columbia University Press, New York.

Thomson, J. W. 1972. Distribution patterns of American Arctic lichens. Can. J. Bot. **50**: 1135-1156.

Thormann, M. N., Randolph, S. C., & Bayley, S. E. 2004. Patterns of distribution of microfungi in decomposing bog and fen plants. Can. J. Bot. **82**: 710-720.

Thormann, M. N., Randolph, S. C., & Bayley, S. E. 2001. Microfungi isolated from *Sphagnum fuscum* from a southern boreal bog in Alberta, Canada. Bryologist **104**: 548-559.

Topham, P. B. 1997. Colonization, growth, succession, and competition. *In* Lichen ecology. *Edited by* M. R. D. Seaward. Academic press, New York.

Vaarama, A., & Taren, N. 1959. The effect of gibberellic acid and fungi on spore germination and protonema growth in mosses. Bot. Not. **112**: 481-488.

Venugopal Rao, K., & Manoharachary, C. 1985. Studies on pollen-fungal association. Indian Bot. Rep. **4**: 20-22.

Von Arx, J. A. 1982. On Mucoraceae s. str. and other families of the Mucorales. Sydowia **35**: 10-26.

Warner, J. R. Synthesis of ribosomes in *Saccharomyces cerevisiae*. Microbiol. Rev. **53**: 256-271.

Westman, L. 1973. Notes on the taxonomy and ecology of an arctic lichen *Lecanora symmicta* var. *sorediosa.* Lichenologist **5**: 457-460.

Yarranton, G. A. 1975. Population growth in *Cladonia stellaris* (Opiz.) Pouz. And Vezda. New Phytol. **75**: 99-110.

Zak, J. C., & Parkinson, D. 1984. Patterns of initial saprophytic fungal colonization of grass roots from two severely disturbed soils. Can. J. Bot. **62**: 596-602.

Appendix 1a

July Fungal Assemblage Frequency Data Pooled By Sample

Appendix 1a- Fungal assemblage frequency data for the July collection, from southern Manitoba, pooled by sample (canopy and base) (Cont. next pg.).

	Fungus (out of 40)																			
	Alternaria spp.	*Aureobasidium pullulans*	*Cladosporium # 1*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mortierella # 3*	*Mortierella # 4*	*Mucor # 1*	*Mucor # 4*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerlotial)*	*Sphaeopsidales #23*	*Sphaeropsidales #36*	*Sphaeropsidales #37*	*sterile # 1*	*sterile # 14*	*Sterile # 24*	*Sterile # 38*	*Trichoderma spp.*
Sample	1	2	3	4	5	6	7	8	10	9	11	12	13	14	15	16	17	18	19	20
1lm	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0
1lm	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0
5lm	0	0	0	0	0	0	0	1	0	1	0	0	0	0	0	3	0	0	0	0
7lm	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	6
8lm	0	0	0	1	0	0	0	0	0	2	0	1	0	0	0	0	0	0	0	4
9lm	0	0	0	0	0	0	19	7	0	0	0	0	0	0	0	0	0	0	0	0
9lm	16	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0
8lm	1	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	1	0	0
3mm	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0
5mm	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	3	0	0	0	1
5mm	0	0	0	0	1	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
6mm	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0
6mm	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
7mm	0	0	0	0	0	0	0	1	0	0	0	0	1	0	0	0	1	0	1	3

Sample Legend:
_ = # (plot number)
_ = plot type (mixed moss-lichen [m] or pure lichen [l]plot)
_ = Genus/species (*Cladonia mitis* [m], *C. rangiferina* [r], & *Pleurozium scherberi* [p])

Appendix 1a (cont.) - Fungal assemblage frequency data for the July collection, from southern Manitoba, pooled by sample (canopy and base).

	Fungus (out of 40)																			
	Alternaria spp.	*Aureobasidium pullulans*	*Cladosporium # 1*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mortierella # 3*	*Mortierella # 4*	*Mucor # 1*	*Mucor # 4*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerlotial)*	*Sphaeopsidales # 23*	*Sphaeropsidales # 36*	*Sphaeropsidales # 37*	*sterile # 1*	*sterile # 14*	*Sterile # 24*	*Sterile # 38*	*Trichoderma spp.*
Sample	1	2	3	4	5	6	7	8	10	9	11	12	13	14	15	16	17	18	19	20
1lr	0	0	0	0	0	0	0	0	0	1	4	0	0	0	0	0	1	0	0	0
5lr	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
7lr	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
8lr	0	0	0	0	0	0	0	6	0	10	0	0	0	0	0	0	8	1	0	0
8lr	4	0	0	0	1	0	0	1	3	0	2	0	0	1	0	1	10	2	0	1
9lr	0	0	0	0	0	2	0	5	0	0	0	0	0	0	0	0	0	0	0	1
9lr	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
9lr	2	0	0	0	5	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0
5mr	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
6mr	0	0	0	0	1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
7mr	0	1	2	0	1	0	0	0	0	0	0	0	0	0	0	0	1	1	0	1
7mr	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
6mp	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
6mp	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
7mp	0	0	0	0	1	0	0	1	0	4	0	0	1	0	0	0	2	0	0	0
7mp	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0
7mp	0	0	1	0	0	0	0	1	0	1	0	0	0	0	0	0	5	0	0	0
8mp	0	0	2	0	0	0	0	0	1	1	1	0	0	0	0	0	4	0	0	0
8mp	0	0	0	0	2	0	0	0	0	7	2	0	0	0	0	0	2	0	0	0
8mp	0	0	0	0	0	1	0	0	1	7	0	0	0	0	0	0	3	0	0	0

Sample Legend:

_ = # (plot number)

_ = plot type (mixed moss-lichen [m] or pure lichen [l]plot)

_ = Genus/species (*Cladonia mitis* [m], *C. rangiferina* [r], & *Pleurozium scherberi* [p])

Appendix 2a

September Fungal Assemblage Frequency Data Pooled By Sample

Appendix 2a- Fungal assemblage frequency data for the September collection, from southern Manitoba, pooled by sample (canopy and base) (cont. on next pg.).

	Fungus (out of 40)																
	Absidia coerulea	*Alternaria spp.*	*Aspergillus alutaceus group*	*Cladosporium # 1*	*Cladosporium # 2*	*Cunninghamella elegans*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mucor # 1*	*Mucor # 4*	*Mucor # 5*	*Mucor # 10*	*Penicillium (Non-sclerotial)*	*Penicillium (Sclerotial)*	*Rhizopus oryzae*	*Rhizopus # 2*	*Trichoderma spp.*
sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
l1mit	0	9	0	3	0	0	3	1	0	0	0	0	6	1	0	0	0
l1mit	2	7	0	4	0	0	6	0	0	0	0	0	2	1	0	0	4
l1mit	2	6	0	5	0	0	0	2	0	0	0	0	3	1	0	0	0
l2mit	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	18
l2mit	1	8	0	1	2	0	2	0	1	0	0	1	0	0	0	0	12
l2mit	2	10	0	4	0	0	3	0	0	1	0	1	4	0	0	0	8
l5mit	0	5	0	2	0	0	0	0	2	0	0	1	0	0	0	0	0
l5mit	3	7	0	3	0	0	2	10	6	0	0	1	1	0	0	0	0
l5mit	0	5	0	5	0	0	0	5	1	0	0	3	2	0	1	0	3
l7mit	2	0	0	0	0	0	1	11	5	0	0	5	10	0	1	0	2
l7mit	2	8	0	2	0	0	3	1	1	0	0	1	0	0	0	0	5
l7mit	0	8	0	2	0	0	4	3	9	0	0	4	1	0	0	0	0
l9mit	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	20
l9mit	3	4	0	1	0	0	1	1	0	0	0	2	2	0	1	0	5
l9mit	0	6	0	1	0	0	1	7	1	0	0	2	7	0	0	0	5
m2mit	0	3	0	2	1	0	0	2	3	1	0	1	0	0	1	0	10
m2mit	0	9	0	0	0	0	5	3	0	0	0	0	0	0	0	0	7
m2mit	0	14	0	2	0	0	0	5	1	1	0	0	3	0	0	0	0
m4mit	0	6	0	5	0	0	0	0	11	0	0	2	3	0	0	0	1
m4mit	0	6	0	3	1	0	0	1	1	0	0	0	0	0	0	0	13
m4mit	1	2	0	0	0	0	0	1	1	0	0	0	0	0	0	0	7
m6mit	6	7	0	4	0	0	2	0	0	0	0	2	2	0	0	0	0
m6mit	0	3	0	2	0	0	2	0	7	0	0	3	6	0	0	0	0
m6mit	0	6	0	4	0	0	3	10	2	0	0	0	4	0	0	0	1
m7mit	7	5	0	2	0	0	0	2	0	0	0	11	3	0	1	0	5
m7mit	1	4	0	2	0	0	0	1	3	0	0	3	2	0	0	1	2
m7mit	0	4	0	0	0	0	0	0	1	0	0	0	0	0	0	1	13
m8mit	0	1	0	1	0	0	0	1	7	0	0	1	0	0	0	0	10
m8mit	3	8	0	5	0	0	2	0	2	0	0	0	4	0	0	0	4
m8mit	1	5	0	5	0	0	0	1	1	0	0	0	2	0	0	0	6

Sample Legend:

_ = plot type (mixed moss-lichen [m] or pure lichen [l]plot)

_ = # (plot number)

_ = Genus/species (*Cladonia mitis* [mit], *C. rangiferina* [rang], & *Pleurozium scherberi* [pleuro])

Appendix 2a (cont.) - Fungal assemblage frequency data for the September collection, from southern Manitoba, pooled by sample (canopy and base) (cont. on next pg.).

	Fungus (out of 40)																
	Absidia coerulea	*Alternaria spp.*	*Aspergillus alutaceus group*	*Cladosporium # 1*	*Cladosporium # 2*	*Cunninghamella elegans*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mucor # 1*	*Mucor # 4*	*Mucor # 5*	*Mucor # 10*	*Penicillium (Non-sclerotial)*	*Penicillium (Sclerotial)*	*Rhizopus oryzae*	*Rhizopus # 2*	*Trichoderma spp.*
sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
l1rang	0	10	0	5	0	0	4	5	0	0	0	4	3	0	0	0	3
l1rang	0	7	0	6	0	0	5	5	0	0	0	0	3	0	0	0	1
l1rang	1	7	0	0	1	2	3	2	1	0	0	0	2	0	0	0	2
l2rang	0	6	0	0	5	0	4	0	2	1	0	9	0	1	0	0	0
l2rang	1	8	0	6	0	0	1	1	0	0	0	4	0	0	1	0	2
l2rang	1	2	0	2	0	0	3	0	7	0	0	5	0	0	1	0	2
l5rang	0	7	0	2	1	0	3	0	9	0	0	3	0	0	0	0	0
l5rang	0	8	0	8	0	0	0	0	2	1	0	1	0	0	0	0	1
l5rang	0	4	0	2	1	0	0	0	8	0	1	5	1	0	0	0	0
l7rang	1	5	0	4	0	0	2	0	5	0	0	1	0	0	0	0	6
l7rang	0	8	0	2	0	0	2	2	0	0	0	0	0	0	0	0	9
l7rang	0	10	0	2	0	0	3	2	6	0	0	2	4	0	0	0	5
l9rang	5	6	0	3	0	0	0	7	0	0	0	0	9	0	0	0	1
l9rang	3	7	0	6	0	0	2	4	0	0	0	1	6	8	0	0	0
l9rang	2	6	0	4	0	0	2	8	2	0	0	0	8	2	0	0	4
m2rang	1	6	0	0	0	0	0	3	7	0	0	1	0	0	0	0	8
m2rang	0	12	0	0	0	0	1	1	0	0	0	0	0	0	0	0	9
m2rang	0	8	0	4	0	0	1	2	2	0	0	2	0	0	0	0	2
m4rang	0	2	0	2	0	0	0	0	3	0	0	5	1	0	0	0	8
m4rang	3	8	0	2	0	0	1	0	6	0	0	3	0	0	0	0	3
m4rang	0	5	0	2	0	0	0	3	2	0	0	4	2	4	1	1	6
m6rang	0	8	0	5	0	0	3	4	0	0	0	2	5	1	0	0	0
m6rang	2	9	0	6	0	0	0	5	0	0	0	1	1	1	0	0	0
m6rang	0	9	0	1	0	0	0	3	3	0	0	0	1	0	0	0	8
m7rang	0	0	1	0	0	0	0	0	0	0	0	0	2	0	0	0	19
m7rang	6	6	1	1	0	0	0	2	1	0	0	2	10	3	0	0	5
m7rang	1	0	0	0	0	0	0	4	0	0	0	0	10	0	0	0	15
m8rang	0	3	0	3	0	0	0	0	6	0	0	1	3	1	0	0	0
m8rang	0	6	0	3	0	0	1	0	1	0	0	3	2	0	0	0	11
m8rang	0	13	0	5	0	0	3	0	1	0	0	1	4	0	0	0	0

Sample Legend:

_ = plot type (mixed moss-lichen [m] or pure lichen [l]plot)

_ = # (plot number)

_ = Genus/species (*Cladonia mitis* [mit], *C. rangiferina* [rang], & *Pleurozium scherberi* [pleuro])

Appendix 2a (cont.)- Fungal assemblage frequency data for the September collection, from southern Manitoba, pooled by sample (canopy and base).

Fungus (Frequency out of 20)

sample	1 *Absidia coerulea*	2 *Alternaria spp.*	3 *Cladosporium # 1*	4 *Cladosporium # 2*	5 *Epicoccium purpurans*	6 *Mortierella isabellina/vinacea*	7 *Mucor # 1*	8 *Mucor # 4*	9 *Mucor # 10*	10 *Penicillium (Non-sclerotial)*	11 *Penicillium (Sclerotial)*	12 *Rhizopus oryzae*	13 *Trichoderma spp.*
m2pleuro	0	0	0	0	0	13	2	0	1	0	0	0	17
m2pleuro	0	0	1	1	0	10	2	0	1	5	0	0	15
m2pleuro	2	2	2	0	3	13	3	0	6	4	0	1	10
m6pleuro	4	2	2	0	0	8	5	0	0	10	5	0	3
m6pleuro	3	0	0	0	1	3	8	1	7	1	0	0	3
m6pleuro	2	0	0	0	0	7	4	0	2	2	0	0	12
m7pleuro	7	1	0	0	0	3	3	0	2	1	2	1	14

Sample Legend:

_ = plot type (mixed moss-lichen [m] or pure lichen [l]plot)

_ = # (plot number)

_ = Genus/species (*Cladonia mitis* [mit], *C. rangiferina* [rang], & *Pleurozium scherberi* [pleuro])

Appendix 3a

July Fungal Assemblage Frequency Data

Appendix 3a - Fungal assemblage frequency data for the July collection, from southern Manitoba (Cont. on next pg.).

	Fungus (out of 20)																			
	Alternaria spp.	*Aureobasidium pullulans*	*Cladosporium # 1*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mortierella # 3*	*Mortierella # 4*	*Mucor # 1*	*Mucor # 4*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (Sclerlotia)*	*Sphaeopsidales # 23*	*Sphaeropsidales # 36*	*Sphaeropsidales # 37*	*sterile # 1*	*sterile # 14*	*Sterile # 24*	*Sterile # 38*	*Trichoderma spp.*
Sample	1	2	3	4	5	6	7	8	10	9	11	12	13	14	15	16	17	18	19	20
1lmb	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0
1lmb	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0
5lmb	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	3	0	0	0	0
7lmb	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6
8lmb	0	0	0	0	0	0	0	0	2	0	0	1	0	0	0	0	0	0	0	4
9lmb	0	0	0	0	0	0	19	7	0	0	0	0	0	0	0	0	0	0	0	0
9lmb	16	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0
1lmt	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
5lmt	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
7lmt	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
8lmt	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
8lmt	1	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	1	0	0
3mb	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0
5mb	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	3	0	0	0	1
5mb	0	0	0	0	1	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
6mb	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
6mb	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
7mb	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3
6mt	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0
7mt	0	0	0	0	0	0	0	1	0	0	0	0	1	0	0	0	1	0	1	0

Sample Legend:

_ = # (plot number)

_ = plot type (mixed moss-lichen [m] or pure lichen [l]plot)

_ = Genus/species (*Cladonia mitis* [m], *C. rangiferina* [r], & *Pleurozium scherberi* [p])

_ = canopy or base of sample (canopy[t], base [b])

Appendix 3a (cont.) - Fungal assemblage frequency data for the July collection, from southern Manitoba.

	Fungus (out of 20)																			
	Alternaria spp.	*Aureobasidium pullulans*	*Cladosporium # 1*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mortierella # 3*	*Mortierella # 4*	*Mucor # 1*	*Mucor # 4*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (Sclerlotia)*	*Sphaeopsidales #23*	*Sphaeropsidales #36*	*Sphaeropsidales #37*	*sterile #1*	*sterile # 14*	*Sterile # 24*	*Sterile # 38*	*Trichoderma spp.*
Sample	1	2	3	4	5	6	7	8	10	9	11	12	13	14	15	16	17	18	19	20
1lrb	0	0	0	0	0	0	0	0	1	0	4	0	0	0	0	0	0	0	0	0
5lrb	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
7lrb	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
8lrb	0	0	0	0	0	0	0	6	10	0	0	0	0	0	0	0	8	0	0	0
8lrb	1	0	0	0	1	0	0	1	0	3	2	0	0	0	0	1	10	1	0	1
9lrb	0	0	0	0	0	2	0	5	0	0	0	0	0	0	0	0	0	0	0	0
9lrb	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
9lrb	2	0	0	0	5	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0
1lrt	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
7lrt	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
8lrt	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0
8lrt	3	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	1	0	0
9lrt	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
5rb	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
6rb	0	0	0	0	1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
7rb	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
7rb	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
7rt	0	1	2	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	0	0
6mp	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
6mp	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
7mp	0	0	0	0	1	0	0	1	4	0	0	0	1	0	0	0	2	0	0	0
7mp	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0
7mp	0	0	1	0	0	0	0	1	1	0	0	0	0	0	0	0	5	0	0	0
8mp	0	0	2	0	0	0	0	0	1	1	1	0	0	0	0	0	4	0	0	0
8mp	0	0	0	0	2	0	0	0	7	0	2	0	0	0	0	0	2	0	0	0
8mp	0	0	0	0	0	1	0	0	7	1	0	0	0	0	0	0	3	0	0	0

Sample Legend:
_ = # (plot number)
_ = plot type (mixed moss-lichen [m] or pure lichen [l]plot)
_ = Genus/species (*Cladonia mitis* [m], *C. rangiferina* [r], & *Pleurozium scherberi* [p])
_ = canopy or base of sample (canopy[t], base [b])

Appendix 3b

Principal Component Analysis Summary Of The July Fungal Assemblage Frequency Data

PRINCIPAL COMPONENTS ANALYSIS

July data set, southern Manitoba

INPUT AND RUN PARAMETERS

NUMBER OF ROWS =	46
NUMBER OF COLS =	20
TYPE OF ANALYSIS =	PCA FROM COVARIANCES
NO. OF COMPONENTS RETAINED =	3
LABELS FOR OBJECTS =	NOT USED
LABELS FOR VARIABLES =	NOT USED
CORRESP. ANALYSIS =	NOT APPLICABLE
MATRIX =	NOT SAVED
PRINTOUT =	SHORT
VAR. SCORE OPTION =	EIGENVECTORS AS COORDINATES OF VAR.
OBJ. SCORE =	NORMALIZED TO LAMBDA

VARIABLES STATISTICS

POOLED VARIANCE = .4725

VARIABLE	MEAN	STANDARD DEVIATION	VARIANCE	VARIANCE AS %
1	.0633	.2161	.0467	9.881
2	.0065	.0444	.0020	.417
3	.0377	.1255	.0157	3.331
4	.0065	.0444	.0020	.417
5	.0835	.1748	.0305	6.465
6	.0169	.0823	.0068	1.435
7	.0283	.1918	.0368	7.787
8	.1138	.2288	.0523	11.075
9	.0262	.1067	.0114	2.409
10	.1136	.2667	.0711	15.050
11	.0659	.1662	.0276	5.847
12	.0065	.0444	.0020	.417
13	.0131	.0621	.0039	.815
14	.0065	.0444	.0020	.417
15	.0065	.0444	.0020	.417
16	.0496	.1506	.0227	4.799
17	.2080	.2979	.0888	18.786
18	.0327	.0947	.0090	1.899
19	.0065	.0444	.0020	.417
20	.0728	.1934	.0374	7.918

NUMBER OF POSITIVE EIGENVALUES = 20

SUM OF POSITIVE EIGENVALUES = 0.47252374E+0

EIGENVALUES

0.1275E+00	0.7361E-01	0.5822E-01	0.4153E-01	0.3751E-01
0.2959E-01	0.2517E-01	0.1959E-01	0.1721E-01	0.1274E-01
0.8747E-02	0.6796E-02	0.4332E-02	0.2635E-02	0.1956E-02
0.1824E-02	0.1347E-02	0.1161E-02	0.6352E-03	0.3864E-03

EIGENVALUES AS PERCENT

26.99	15.58	12.32	8.79	7.94
6.26	5.33	4.15	3.64	2.70
1.85	1.44	.92	.56	.41
.39	.28	.25	.13	.08

CUMULATIVE PERCENTAGE OF EIGENVALUES

26.99	42.57	54.89	63.68	71.62
77.88	83.20	87.35	90.99	93.69
95.54	96.98	97.90	98.45	98.87
99.25	99.54	99.78	99.92	100.00

SQUARE ROOTS OF EIGENVALUES

.357122	.271317	.241290	.203786	.193681
.172005	.158641	.139960	.131187	.112887
.093523	.082436	.065816	.051333	.044224
.042709	.036696	.034069	.025203	.019658

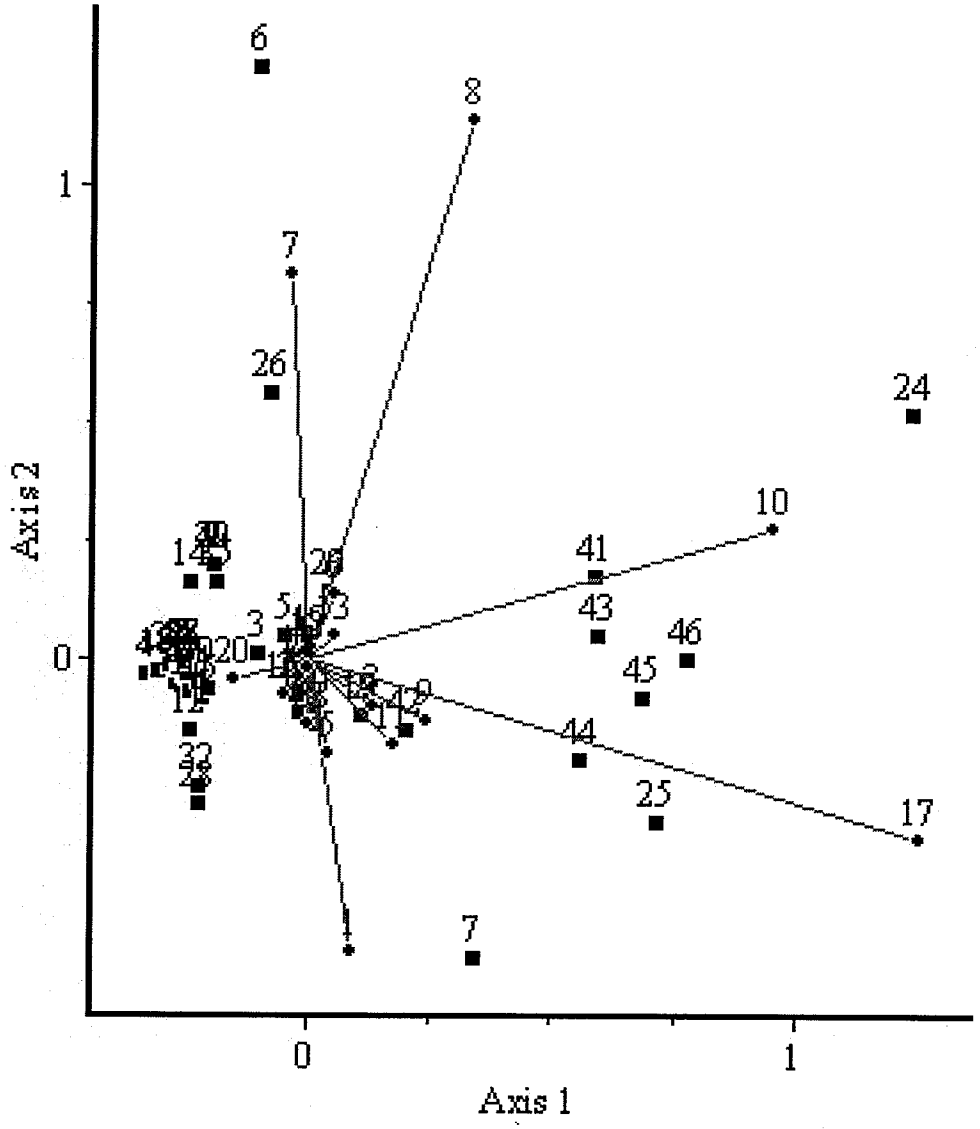
COMPONENT SCORES

1	-.207	-.064	-.124
2	.105	-.122	.080
3	-.103	.010	-.193
4	-.335	-.033	-.261
5	-.048	.050	-.495
6	-.105	1.253	.624
7	.341	-.637	.863
8	-.257	-.011	-.051
9	-.196	.197	.040
10	.009	-.111	.045
11	-.261	-.011	-.053
12	-.244	-.154	.166
13	.105	-.122	.080
14	-.243	.160	-.017
15	-.218	-.082	-.115
16	-.022	-.119	.014
17	-.245	-.070	-.079
18	-.313	-.027	-.200
19	.105	-.122	.080
20	.043	.139	.123
21	-.011	-.039	-.318
22	-.196	.197	.040
23	-.257	-.011	-.051
24	1.236	.519	-.089
25	.716	-.349	.408
26	-.084	.565	.180
27	-.257	-.011	-.051
28	-.227	-.310	.197
29	-.029	-.081	.031
30	-.225	-.044	-.097
31	-.257	-.037	-.017
32	-.227	-.271	.349

33	-.285	-.019	-.126
34	-.029	-.081	.031
35	-.189	.160	.022
36	-.277	-.056	-.143
37	-.250	-.048	-.069
38	.009	-.141	.082
39	-.266	-.025	-.042
40	-.196	.197	.040
41	.585	.171	-.223
42	.199	-.152	.114
43	.590	.045	.101
44	.558	-.214	-.038
45	.685	-.087	-.502
46	.772	-.005	-.354

SCORES FOR VARIABLES

VARIABLE 1		
.055	-.376	.587
VARIABLE 2		
0.000	-.013	.009
VARIABLE 3		
.079	-.063	.028
VARIABLE 4		
-.014	-.001	-.006
VARIABLE 5		
.024	-.125	-.059
VARIABLE 6		
.034	.081	-.008
VARIABLE 7		
-.024	.492	.310
VARIABLE 8		
.202	.690	.301
VARIABLE 9		
.145	-.083	.049
VARIABLE 10		
.575	.163	-.532
VARIABLE 11		
.104	-.111	-.153
VARIABLE 12		
-.003	.005	-.057
VARIABLE 13		
.033	.028	-.011
VARIABLE 14		
-.012	-.025	.040
VARIABLE 15		
-.013	-.014	.019
VARIABLE 16		
-.032	-.048	.030
VARIABLE 17		
.758	-.234	.273
VARIABLE 18		
0.000	-.086	.113
VARIABLE 19		
.002	.013	.014
VARIABLE 20		
-.093	-.027	-.248



Appendix 3b - Principle component analysis biplot of the fungal assemblages for the *Cladonia rangiferina, C. mitis, & Pleurozium schreberi* July data collection. Fungal species variables are represented by the circles along the axes, while samples are represented by squares.

Appendix 3c

Multiple Discriminate Analysis Summary Of The July Fungal Assemblage Frequency Data, Separating Groups By Mat Species

CANONICAL VARIATES ANALYSIS (MULTIGROUP DISCRIMINANT ANALYSIS)

July data set for southern Manitoba by species

NUMBER OF VARIABLES = 3
NUMBER OF GROUPS = 3
NUMBER OF OBSERVATIONS = 46
LABELS FOR VARIABLES = NOT USED
LABELS FOR OBJERCTS = NOT USED
SCORES ARE = SPHERIZED

UNIVARIATE F RATIOS WITH 2 AND 43 D.F.

VARIABLE	AMONG GROUP SSQ	WITHIN GROUP SSQ	F RATIO
1	.66	.10	6.479
2	0.00	.08	.010
3	.06	.06	1.068

EIGENVALUES

0.3863613D+00 0.6607841D-03

CAN. VAR.	EIGENVALUE	E.V. AS %	CAN. CORR.
1	.39	99.83	.528
2	0.00	.17	.026

CHI-SQUARE TESTS WITH SUCCESSIVE VARIATES REMOVED

	CAN. VAR. REMOVED	CHISQ	DEGREES OF FREEDOM	WILKS LAMBDA
UP TO	0	13.75	6	.7208
UP TO	1	.03	2	.9993

DISCRIMINANT WEIGHTS (CANONICAL VARIATES)

C.V. 1 0.2937425E+01-0.1355394E+00-0.1957057E+01
C.V. 2 0.9099272E+00-0.1913291E+01 0.3159333E+01

CORRELATION MATRIX FOR THE TOTAL SAMPLE

1	1.000	0.000	0.000
2	0.000	1.000	0.000
3	0.000	0.000	1.000

CORRELATIONS OF VARIABLES WITH CANONICAL VARIATES

VAR.	1	.911	.332
VAR.	2	-.032	-.531
VAR.	3	-.410	.780

COMMUNALITIES OF VARIABLES FOR 2 CANONICAL VARIATES

```
   1   .941    2   .283    3   .776

PERCENTAGE OF TR{R} ACCOUNTED FOR BY EACH EIGENVALUE

   1 33.332    2 33.333

CENTROID FOR GROUP    1 IN    2 DIMENSIONAL CANONICAL SPACE

   1  -.359    2  -.024

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID =    .547


CENTROID FOR GROUP    2 IN    2 DIMENSIONAL CANONICAL SPACE

   1  -.178    2   .030

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID =    .577


CENTROID FOR GROUP    3 IN    2 DIMENSIONAL CANONICAL SPACE

   1  1.297    2  -.007

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID =    .865


THE 95% ISODENSITY CIRCLE AROUND EACH CENTROID
HAS A RADIUS OF 2.45


SPHERIZED SCORES OF OBJECTS ON CANONICAL VARIATES

GROUP     1
       1      -.357      -.459
       2       .168       .581
       3       .075      -.722

       4      -.470     -1.064
       5       .821     -1.703
       6     -1.701      -.522
       7      -.602      4.256
       8      -.653      -.374
       9      -.680      -.430
      10      -.046       .362
      11      -.662      -.383
      12     -1.021       .597
      13       .168       .581
      14      -.701      -.582
      15      -.405      -.405
      16      -.073       .250
      17      -.556      -.339
      18      -.523      -.866
      19       .168       .581
      20      -.134       .161
```

GROUP	2	
	21	.595 -.940
	22	-.680 -.430
	23	-.653 -.374
	24	3.734 -.151
	25	1.353 2.609
	26	-.673 -.591
	27	-.653 -.374
	28	-1.011 1.010
	29	-.135 .228
	30	-.467 -.427
	31	-.717 -.216
	32	-1.314 1.413
	33	-.588 -.620
	34	-.135 .228
	35	-.619 -.408
	36	-.527 -.598
	37	-.592 -.352
	38	-.114 .536
GROUP	3	
	39	-.697 -.327
	40	-.680 -.430
	41	2.130 -.498
	42	.383 .831
	43	1.531 .769
	44	1.744 .797
	45	3.006 -.795
	46	2.962 -.407

Appendix 4a

September Fungal Assemblage Frequency Data

Appendix 4a - Fungal assemblage frequency data for *Cladonia mitis* from the September collection in southern Manitoba (Cont. on next pg.).

	Fungus (Frequency out of 20)													
sample	1 *Absidia coerulea*	2 *Alternaria spp.*	3 *Cladosporium # 1*	4 *Cladosporium # 2*	5 *Epicoccum purpurascens*	6 *Mortierella isabellina/vinacea*	7 *Mucor # 1*	8 *Mucor # 4*	9 *Mucor # 10*	10 *Penicillium (non-sclerotial)*	11 *Penicillium (sclerotial)*	12 *Rhizopus oryzae*	13 *Rhizopus # 2*	14 *Trichoderma spp.*
l1mitt1	0	7	3	0	3	0	0	0	0	1	1	0	0	0
l1mitt2	0	7	2	0	3	0	0	0	0	0	1	0	0	0
l1mitt3	0	6	4	0	0	0	0	0	0	2	0	0	0	0
l2mitt1	0	0	0	0	0	0	0	0	0	0	0	0	0	10
l2mitt2	0	8	1	2	2	0	0	0	0	0	0	0	0	5
l2mitt3	0	9	2	0	3	0	0	0	0	0	0	0	0	5
l5mitt1	0	5	2	0	0	0	0	0	0	0	0	0	0	0
l5mitt2	2	3	3	0	2	1	0	0	0	0	0	0	0	0
l5mitt3	0	5	3	0	0	0	1	0	0	0	0	1	0	0
l7mitt1	0	0	0	0	0	5	2	0	2	6	0	1	0	2
l7mitt2	0	7	2	0	0	0	0	0	0	0	0	0	0	3
l7mitt3	0	5	2	0	4	1	0	0	0	0	0	0	0	0
l9mitt1	0	0	0	0	0	0	0	0	0	0	0	0	0	10
l9mitt2	2	4	1	0	1	1	0	0	0	2	0	0	0	2
l9mitt3	0	6	1	0	1	2	0	0	1	1	0	0	0	0
m2mitt1	0	3	2	1	0	1	1	1	0	0	0	1	0	5
m2mitt2	0	6	0	0	5	1	0	0	0	0	0	0	0	0
m2mitt3	0	9	2	0	0	1	0	0	0	0	0	0	0	0
m4mitt1	0	6	5	0	0	0	1	0	0	1	0	0	0	1
m4mitt2	0	6	3	0	0	0	0	0	0	0	0	0	0	5
m6mitt1	0	5	3	0	1	0	0	0	0	0	0	0	0	0
m6mitt2	0	3	2	0	2	0	0	0	0	3	0	0	0	0
m6mitt3	0	6	3	0	2	3	1	0	0	2	0	0	0	0
m7mitt1	1	5	2	0	0	2	0	0	2	2	0	1	0	3
m7mitt2	1	4	2	0	0	1	2	0	2	2	0	0	1	0
m7mitt3	0	4	0	0	0	0	1	0	0	0	0	0	1	3
m8mitt1	0	1	1	0	0	0	1	0	0	0	0	0	0	7
m8mitt2	0	6	5	0	2	0	0	0	0	1	0	0	0	2
m8mitt3	0	5	5	0	0	0	0	0	0	2	0	0	0	0

Sample Legend:

_ = l (lichen plot), m (mixed moss-lichen plot)

_ = # (plot number)

_ = rang(*Cladonia rangiferina*), mit (*Cladonia mitis*), pleuro (*Pleurozium schreberi*)

_ = b(base), t(canopy)

_ = #(sample #)

Appendix 4a (Cont.) - Fungal assemblage frequency data for *Cladonia mitis* from the September collection in southern Manitoba (Cont. on next pg.).

	Fungus (Frequency out of 20)													
	Absidia coerulea	*Alternaria spp.*	*Cladosporium # 1*	*Cladosporium # 2*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mucor # 1*	*Mucor # 4*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Rhizopus oryzae*	*Rhizopus # 2*	*Trichoderma spp.*
sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14
l1mitb1	0	2	0	0	0	1	0	0	0	5	0	0	0	0
l1mitb2	2	0	2	0	3	0	0	0	0	2	0	0	0	4
l1mitb3	2	0	1	0	0	2	0	0	0	1	1	0	0	0
l2mitb1	2	0	0	0	0	0	0	0	0	0	0	0	0	8
l2mitb2	1	0	1	0	0	0	1	0	1	0	0	0	0	7
l2mitb3	2	1	2	0	0	0	0	1	1	4	0	0	0	3
l5mitb1	0	0	0	0	0	0	2	0	1	0	0	0	0	0
l5mitb2	1	0	0	0	0	9	6	0	1	1	0	0	0	0
l5mitb3	0	0	2	0	0	5	0	0	3	2	0	0	0	3
l7mitb1	2	0	0	0	1	6	3	0	3	4	0	0	0	0
l7mitb2	2	1	0	0	3	1	1	0	1	0	0	0	0	2
l7mitb3	0	3	0	0	0	2	9	0	4	1	0	0	0	0
l9mitb1	0	0	0	0	0	0	0	0	0	1	0	0	0	10
l9mitb2	1	0	0	0	0	0	0	0	2	0	0	1	0	3
l9mitb3	0	0	1	0	0	5	1	0	1	6	0	0	0	5
m2mitb1	0	0	0	0	0	1	2	0	1	0	0	0	0	5
m2mitb2	0	3	0	0	0	2	0	0	0	0	0	0	0	7
m2mitb3	0	5	0	0	0	4	1	1	0	3	0	0	0	0
m4mitb1	0	0	0	0	0	0	10	0	2	2	0	0	0	0
m4mitb2	0	0	0	1	0	1	1	0	0	0	0	0	0	8
m4mitb3	1	2	0	0	0	1	1	0	0	0	0	0	0	7
m6mitb1	6	2	1	0	1	0	0	0	2	2	0	0	0	0
m6mitb2	0	0	0	0	0	0	7	0	3	3	0	0	0	0
m6mitb3	0	0	1	0	1	7	1	0	0	2	0	0	0	1
m7mitb1	6	0	0	0	0	0	0	0	9	1	0	0	0	2
m7mitb2	0	0	0	0	0	0	1	0	1	0	0	0	0	2
m7mitb3	0	0	0	0	0	0	0	0	0	0	0	0	0	10
m8mitb1	0	0	0	0	0	1	6	0	1	0	0	0	0	3
m8mitb2	3	2	0	0	0	0	2	0	0	3	0	0	0	2
m8mitb3	1	0	0	0	0	1	1	0	0	0	0	0	0	6

Sample Legend:

_ = l (lichen plot), m (mixed moss-lichen plot)

_ = # (plot number)

_ = rang(*Cladonia rangiferina*), mit (*Cladonia mitis*), pleuro (*Pleurozium schreberi*)

_ = b(base), t(canopy)

_ = #(sample #)

Appendix 4a (Cont.) - Fungal assemblage frequency data for *Cladonia rangiferina* from the September collection in southern Manitoba (Cont. on next pg.).

	Fungus (Frequency values out of 20)																
	Absidia coerulea	*Alternaria spp.*	*Aspergillus alutaceus group*	*Cladosporium # 1*	*Cladosporium # 2*	*Cunninghamella elegans*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mucor # 1*	*Mucor # 4*	*Mucor # 5*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Rhizopus oryzae*	*Rhizopus # 2*	*Trichoderma spp.*
sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
l1rangt1	0	10	0	5	0	0	4	0	0	0	0	0	0	0	0	0	3
l1rangt2	0	7	0	6	0	0	4	0	0	0	0	0	1	0	0	0	0
l1rangt3	0	7	0	0	1	0	3	0	0	0	0	0	0	0	0	0	0
l2rangt1	0	6	0	0	5	0	4	0	0	1	0	0	0	0	0	0	0
l2rangt2	1	8	0	6	0	0	1	0	0	0	0	0	0	0	1	0	0
l2rangt3	0	2	0	2	0	0	3	0	5	0	0	2	0	0	0	0	0
l5rangt1	0	7	0	2	1	0	3	0	0	0	0	0	0	0	0	0	0
l5rangt2	0	8	0	8	0	0	0	0	0	1	0	0	0	0	0	0	0
l5rangt3	0	4	0	2	1	0	0	0	2	0	1	0	1	0	0	0	0
l7rangt1	0	4	0	4	0	0	2	0	0	0	0	0	0	0	0	0	3
l7rangt2	0	8	0	2	0	0	2	1	0	0	0	0	0	0	0	0	0
l7rangt3	0	8	0	2	0	0	3	2	2	0	0	0	0	0	0	0	1
l9rangt1	0	6	0	3	0	0	0	0	0	0	0	0	2	0	0	0	1
l9rangt2	0	5	0	5	0	0	2	0	0	0	0	0	0	2	0	0	0
l9rangt3	0	3	0	4	0	0	2	1	0	0	0	0	2	2	0	0	0
m2rangt1	0	6	0	0	0	0	0	2	2	0	0	1	0	0	0	0	3
m2rangt2	0	8	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
m2rangt3	0	8	0	4	0	0	1	0	0	0	0	0	0	0	0	0	0
m4rangt1	0	2	0	2	0	0	0	0	2	0	0	2	1	0	0	0	6
m4rangt2	0	8	0	2	0	0	1	0	1	0	0	0	0	0	0	0	0
m4rang.t3	0	5	0	2	0	0	0	0	0	0	0	1	1	4	0	1	0
m6rangt1	0	5	0	5	0	0	2	0	0	0	0	1	2	0	0	0	0
m6rangt2	0	8	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0
m6rangt3	0	2	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0
m7rangt1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	10
m7rangt2	0	5	1	1	0	0	0	2	0	0	0	0	3	0	0	0	2
m7rangt3	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	8
m8rangt1	0	3	0	3	0	0	0	0	1	0	0	0	3	1	0	0	0
m8rangt2	0	4	0	3	0	0	1	0	0	0	0	0	2	0	0	0	6
m8rangt3	0	6	0	1	0	0	3	0	0	0	0	0	0	0	0	0	0

Sample Legend:

_ = l (lichen plot), m (mixed moss-lichen plot)

_ = # (plot number)

_ = rang(*Cladonia rangiferina*), mit (*Cladonia mitis*), pleuro (*Pleurozium schreberi*)

_ = b(base), t(canopy)

_ = #(sample #)

Appendix 4a (Cont.) - Fungal assemblage frequency data for *Cladonia rangiferina* from the September collection in southern Manitoba (Cont. on next pg.).

	Fungus (Frequency values out of 20)																
	Absidia coerulea	*Alternaria spp.*	*Aspergillus alutaceus group*	*Cladosporium # 1*	*Cladosporium # 2*	*Cunninghamella elegans*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mucor # 1*	*Mucor # 4*	*Mucor # 5*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Rhizopus oryzae*	*Rhizopus # 2*	*Trichoderma spp.*
sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
l1rangb1	0	3	0	0	0	0	0	5	0	0	0	4	3	0	0	0	0
l1rangb2	0	0	0	0	0	0	1	5	0	0	0	0	2	0	0	0	1
l1rangb3	1	1	0	0	0	2	0	2	1	0	0	0	2	0	0	0	2
l2rangb1	0	0	0	0	0	0	0	0	2	0	0	9	0	1	0	0	0
l2rangb2	1	0	0	0	0	0	0	1	0	0	0	4	0	0	0	0	2
l2rangb3	1	0	0	0	0	0	0	0	2	0	0	3	0	0	1	0	2
l5rangb1	0	0	0	0	0	0	0	0	9	0	0	3	0	0	0	0	0
l5rangb2	0	0	0	0	0	0	0	0	2	0	0	1	0	0	0	0	1
l5rangb3	0	0	0	0	0	0	0	0	6	0	0	5	0	0	0	0	0
l7rangb1	1	1	0	0	0	0	0	0	5	0	0	1	0	0	0	0	3
l7rangb2	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	9
l7rangb3	0	2	0	0	0	0	0	0	4	0	0	2	4	0	0	0	4
l9rangb1	5	0	0	0	0	0	0	7	0	0	0	0	7	0	0	0	0
l9rangb2	3	2	0	1	0	0	0	4	0	0	0	1	6	6	0	0	0
l9rangb3	2	3	0	0	0	0	0	7	2	0	0	0	6	0	0	0	4
m2rangb1	1	0	0	0	0	0	0	1	5	0	0	0	0	0	0	0	5
m2rangb2	0	4	0	0	0	0	0	1	0	0	0	0	0	0	0	0	9
m2rangb3	0	0	0	0	0	0	0	2	2	0	0	2	0	0	0	0	2
m4rangb1	0	0	0	0	0	0	0	0	1	0	0	3	0	0	0	0	2
m4rangb2	3	0	0	0	0	0	0	0	5	0	0	3	0	0	0	0	3
m4rangb3	0	0	0	0	0	0	0	3	2	0	0	3	1	0	1	0	6
m6rangb1	0	3	0	0	0	0	1	4	0	0	0	1	3	1	0	0	0
m6rangb2	2	1	0	2	0	0	0	5	0	0	0	1	1	1	0	0	0
m6rangb3	0	0	0	0	0	0	0	3	3	0	0	0	0	0	0	0	8
m7rangb1	0	0	1	0	0	0	0	0	0	0	0	0	2	0	0	0	9
m7rangb2	6	1	0	0	0	0	0	0	1	0	0	2	7	3	0	0	3
m7rangb3	1	0	0	0	0	0	0	4	0	0	0	0	6	0	0	0	7
m8rangb1	0	0	0	0	0	0	0	0	5	0	0	1	0	0	0	0	0
m8rangb2	0	2	0	0	0	0	0	0	1	0	0	3	0	0	0	0	5
m8rangb3	0	7	0	4	0	0	0	0	1	0	0	1	4	0	0	0	0

Sample Legend:

_ = l (lichen plot), m (mixed moss-lichen plot)

_ = # (plot number)

_ = rang(*Cladonia rangiferina*), mit (*Cladonia mitis*), pleuro (*Pleurozium schreberi*)

_ = b(base), t(canopy)

_ = #(sample #)

Appendix 4a (Cont.) - Fungal assemblage frequency data for *Pleurozium schreberi* from the September collection in southern Manitoba.

	Fungus (Frequency out of 20)												
sample	*Absidia coerulea* 1	*Alternaria spp.* 2	*Cladosporium # 1* 3	*Cladosporium # 2* 4	*Epicoccium purpurans* 5	*Mortierella isabellina/vinacea* 6	*Mucor # 1* 7	*Mucor # 4* 8	*Mucor # 10* 9	*Penicillium (non-sclerotial)* 10	*Penicillium (sclerotial)* 11	*Rhizopus oryzae* 12	*Trichoderma spp.* 13
m2pleuro1	0	0	0	0	0	13	2	0	1	0	0	0	17
m2pleuro2	0	0	1	1	0	10	2	0	1	5	0	0	15
m2pleuro3	2	2	2	0	3	13	3	0	6	4	0	1	10
m6pleuro1	4	2	2	0	0	8	5	0	0	10	5	0	3
m6pleuro2	3	0	0	0	1	3	8	1	7	1	0	0	3
m6pleuro3	2	0	0	0	0	7	4	0	2	2	0	0	12
m7pleuro1	7	1	0	0	0	3	3	0	2	1	2	1	14

Sample Legend:
_ = l (lichen plot), m (mixed moss-lichen plot)
_ = # (plot number)
_ = rang(*Cladonia rangiferina*), mit (*Cladonia mitis*), pleuro (*Pleurozium schreberi*)
_ = #(sample #)

Appendix 4b

Principal Component Analysis Summary Of The September Fungal Assemblage Frequency Data

PRINCIPAL COMPONENTS ANALYSIS

September data set, southern Manitoba

INPUT AND RUN PARAMETERS

NUMBER OF ROWS =	126
NUMBER OF COLS =	17
TYPE OF ANALYSIS =	PCA FROM COVARIANCES
NO. OF COMPONENTS RETAINED =	3
LABELS FOR OBJECTS =	NOT USED
LABELS FOR VARIABLES =	NOT USED
CORRESP. ANALYSIS =	NOT APPLICABLE
MATRIX =	NOT SAVED
PRINTOUT =	SHORT
VAR. SCORE OPTION =	EIGENVECTORS AS COORDINATES OF VAR.
OBJ. SCORE =	NORMALIZED TO LAMBDA

VARIABLES STATISTICS

POOLED VARIANCE = .9238

VARIABLE	MEAN	STANDARD DEVIATION	VARIANCE	VARIANCE AS %
1	.1370	.2390	.0571	6.180
2	.4347	.3895	.1517	16.425
3	.0048	.0378	.0014	.154
4	.2587	.2894	.0837	9.065
5	.0243	.1021	.0104	1.128
6	.0038	.0425	.0018	.196
7	.1522	.2395	.0574	6.210
8	.2461	.3281	.1076	11.651
9	.2432	.3023	.0914	9.893
10	.0143	.0644	.0041	.448
11	.0024	.0268	.0007	.078
12	.1974	.2670	.0713	7.714
13	.2549	.2979	.0888	9.608
14	.0513	.1589	.0253	2.733
15	.0239	.0817	.0067	.722
16	.0072	.0461	.0021	.230
17	.4053	.4028	.1623	17.563

NUMBER OF POSITIVE EIGENVALUES = 17

SUM OF POSITIVE EIGENVALUES = 0.92384052E+00

EIGENVALUES

0.3320E+00	0.1526E+00	0.1240E+00	0.6602E-01	0.5746E-01
0.5014E-01	0.3601E-01	0.3213E-01	0.2981E-01	0.1932E-01
0.9996E-02	0.5529E-02	0.3203E-02	0.1916E-02	0.1726E-02
0.1310E-02	0.6064E-03			

EIGENVALUES AS PERCENT

35.94	16.52	13.42	7.15	6.22
5.43	3.90	3.48	3.23	2.09
1.08	.60	.35	.21	.19
.14	.07			

CUMULATIVE PERCENTAGE OF EIGENVALUES

35.94	52.46	65.88	73.03	79.25
84.68	88.58	92.05	95.28	97.37
98.45	99.05	99.40	99.61	99.79
99.93	100.00			

SQUARE ROOTS OF EIGENVALUES

.576226	.390696	.352117	.256940	.239704
.223916	.189767	.179257	.172651	.138991
.099981	.074359	.056593	.043771	.041550
.036196	.024625			

COMPONENT SCORES

1	.136	-.378	.153
2	-.097	.209	.309
3	-.100	-.364	.090
4	.897	-.117	.060
5	.882	.038	-.096
6	.735	-.152	.087
7	-.481	.648	.093
8	-.476	.487	-.117
9	-.107	-.030	.287
10	-.449	.797	.063
11	.480	.601	.210
12	.593	.570	.259
13	-.198	-.047	-.709
14	-.643	-.759	-.211
15	-.474	-.224	.330
16	.661	.097	-.177
17	.745	-.146	.109
18	.618	.019	-.268
19	-.603	-.953	-.069
20	-.186	.053	-.115
21	-.236	-.627	-.579
22	-.751	-.534	.193
23	.472	.475	.125
24	.745	-.047	.004
25	-.481	.650	.194
26	-.419	.336	-.248
27	-.671	-.274	.515
28	-.449	.797	.063
29	.206	-.053	.430
30	.475	-.327	.075
31	-.620	.318	-.207
32	-.150	.498	.338
33	.148	-.535	.200
34	.050	.404	.177

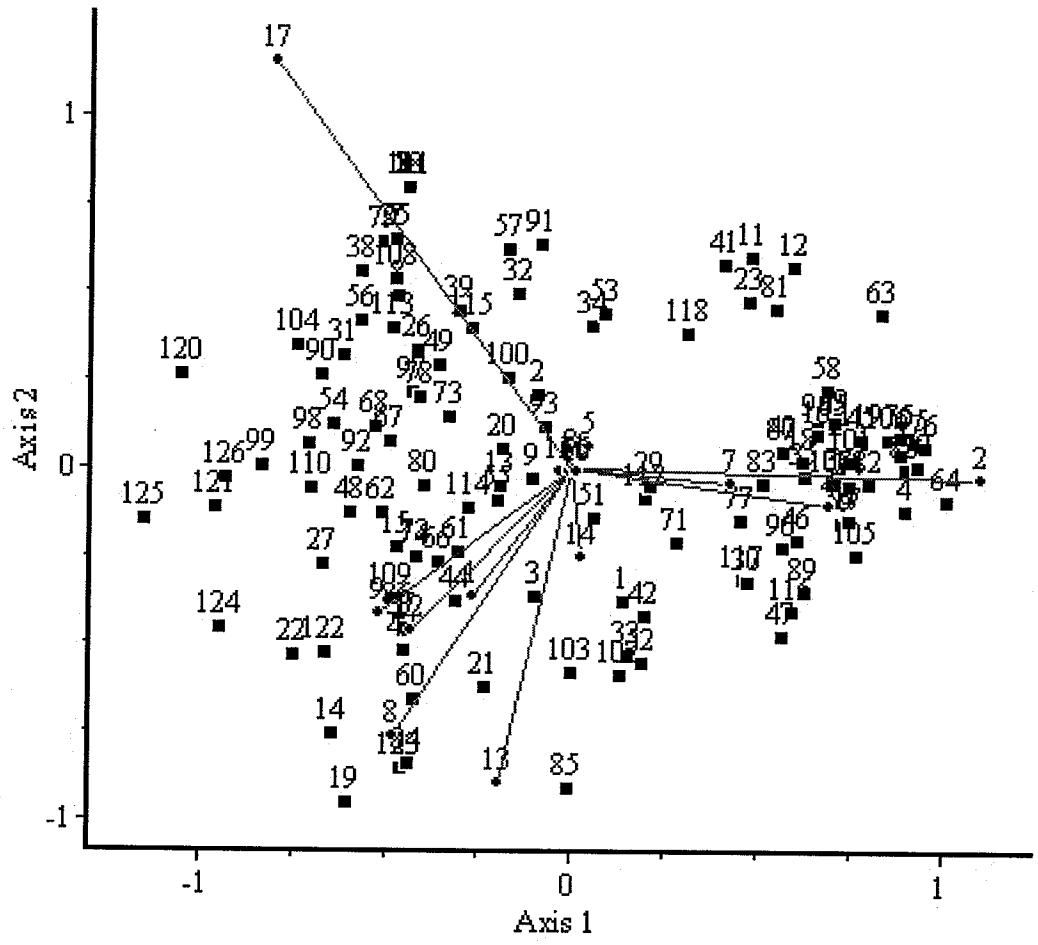
35	.623	-.022	-.073
36	.716	-.034	.007
37	-.451	-.455	-.785
38	-.569	.557	.045
39	-.309	.446	.155
40	.566	.052	.045
41	.406	.580	.220
42	.193	-.419	-.077
43	-.455	-.516	-.716
44	-.313	-.379	.309
45	.779	.082	-.136
46	.602	-.207	.078
47	.565	-.480	.257
48	-.597	-.131	-.271
49	-.358	.295	-.417
50	-.449	.797	.063
51	.055	-.141	.426
52	.191	-.555	-.149
53	.083	.437	-.130
54	-.647	.123	-.436
55	-.201	-.092	.052
56	-.572	.418	.037
57	-.177	.618	-.005
58	.685	.220	.273
59	.725	-.155	.090
60	-.427	-.663	-.019
61	-.309	-.240	.311
62	-.510	-.124	.160
63	.833	.438	.249
64	1.008	-.092	.089
65	.703	.118	-.213
66	-.361	-.268	-.914
67	-.492	.075	-.241
68	-.530	.116	-.538
69	.703	.131	-.216
70	.928	.003	-.006
71	.284	-.212	-.670
72	-.419	-.243	-1.017
73	-.330	.143	-.570
74	-.417	-.253	-1.013
75	.884	.090	-.119
76	.950	.063	-.049
77	.453	-.149	-.253
78	-.411	.200	-.450
79	-.510	.645	.186
80	-.397	-.052	-.137
81	.544	.454	.157
82	.799	-.045	.025
83	.514	-.041	.062
84	-.440	-.839	.514
85	-.011	-.911	.485
86	-.469	-.428	.691
87	.566	.045	.207
88	.894	.001	-.053
89	.627	-.352	.247
90	-.678	.267	-.185
91	-.088	.632	.319

92	-.576	.009	-.321
93	-.079	.118	-.045
94	.657	.114	-.220
95	.922	.073	-.083
96	.569	-.227	-.091
97	-.430	.218	-.516
98	-.710	.069	-.562
99	-.838	.008	.003
100	-.175	.258	-.060
101	.752	.017	-.248
102	.128	-.588	.229
103	-.003	-.584	.225
104	-.741	.352	.065
105	.764	-.244	.018
106	.852	.081	-.100
107	.696	-.044	-.038
108	-.482	.539	.256
109	-.486	-.371	.205
110	-.707	-.059	.740
111	-.449	.797	.063
112	.194	-.085	.501
113	-.484	.400	.327
114	-.284	-.116	-.830
115	-.274	.400	-.286
116	.589	-.407	-.025
117	.457	-.313	-.021
118	.308	.384	.448
119	.775	.091	-.165
120	-1.053	.264	.411
121	-.964	-.113	.737
122	-.665	-.529	.680
123	-.462	-.854	.799
124	-.951	-.459	-.300
125	-1.154	-.147	.362
126	-.935	-.031	.374

SCORES FOR VARIABLES

VARIABLE 1		
-.148	-.199	.146
VARIABLE 2		
.604	-.018	.190
VARIABLE 3		
-.002	.007	.015
VARIABLE 4		
.380	-.059	.196
VARIABLE 5		
.023	.035	.003
VARIABLE 6		
-.006	-.003	.005
VARIABLE 7		
.234	-.025	.055
VARIABLE 8		
-.264	-.421	.471
VARIABLE 9		
-.285	-.228	-.401

```
VARIABLE  10
      .006        -.007         .002
VARIABLE  11
      .003        -.002        -.005
VARIABLE  12
     -.239        -.256        -.328
VARIABLE  13
     -.106        -.490         .435
VARIABLE  14
      .014        -.139         .080
VARIABLE  15
     -.018        -.005         .015
VARIABLE  16
      .006        -.005        -.007
VARIABLE  17
     -.440         .633         .463
```



Appendix 4b - Principle component analysis biplot of the fungal assemblages for the *Cladonia rangiferina, C. mitis, & Pleurozium schreberi* September data collection. Fungal species variables are represented by the circles along the axes, while samples are represented by squares.

Appendix 4c

Multiple Discriminate Analysis Summary Of The September Fungal Assemblage Frequency Data, Groups Separated By Mat Species

CANONICAL VARIATES ANALYSIS (MULTIGROUP DISCRIMINANT ANALYSIS)

September data set for southern Manitoba by species

NUMBER OF VARIABLES = 3
NUMBER OF GROUPS = 3
NUMBER OF OBSERVATIONS = 126
LABELS FOR VARIABLES = NOT USED
LABELS FOR OBJERCTS = NOT USED
SCORES ARE = SPHERIZED

UNIVARIATE F RATIOS WITH 2 AND 123 D.F.

VARIABLE	AMONG GROUP SSQ	WITHIN GROUP SSQ	F RATIO
1	2.94	.29	10.162
2	.30	.15	2.024
3	.77	.11	6.823

EIGENVALUES

0.3580044D+00 0.1000378D-01

CAN. VAR.	EIGENVALUE	E.V. AS %	CAN. CORR.
1	.36	97.28	.513
2	.01	2.72	.100

CHI-SQUARE TESTS WITH SUCCESSIVE VARIATES REMOVED

	CAN. VAR. REMOVED	CHISQ	DEGREES OF FREEDOM	WILKS LAMBDA
UP TO	0	38.55	6	.7291
UP TO	1	1.21	2	.9901

DISCRIMINANT WEIGHTS (CANONICAL VARIATES)

C.V. 1 -0.1471273E+01-0.8971839E+00 0.1997422E+01
C.V. 2 0.6005623E-01 0.2238129E+01 0.1356111E+01

CORRELATION MATRIX FOR THE TOTAL SAMPLE

1	1.000	0.000	0.000
2	0.000	1.000	0.000
3	0.000	0.000	1.000

CORRELATIONS OF VARIABLES WITH CANONICAL VARIATES

VAR.	1	-.733	.035
VAR.	2	-.303	.877
VAR.	3	.608	.479

COMMUNALITIES OF VARIABLES FOR 2 CANONICAL VARIATES

1 .539 2 .861 3 .600

PERCENTAGE OF TR{R} ACCOUNTED FOR BY EACH EIGENVALUE

1 33.333 2 33.333

CENTROID FOR GROUP 1 IN 2 DIMENSIONAL CANONICAL SPACE

1 -.056 2 .105

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = .319

CENTROID FOR GROUP 2 IN 2 DIMENSIONAL CANONICAL SPACE

1 -.226 2 -.097

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = .316

CENTROID FOR GROUP 3 IN 2 DIMENSIONAL CANONICAL SPACE

1 2.414 2 -.057

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = .925

THE 95% ISODENSITY CIRCLE AROUND EACH CENTROID
HAS A RADIUS OF 2.45

SPHERIZED SCORES OF OBJECTS ON CANONICAL VARIATES

GROUP	1	
1	.444	-.631
2	.573	.881
3	.654	-.700
4	-1.095	-.128
5	-1.522	.007
6	-.772	-.177
7	.312	1.546
8	.029	.902
9	.758	.314
10	.071	1.844
11	-.827	1.658
12	-.868	1.662
13	-1.082	-1.079
14	1.207	-2.024
15	1.558	-.082
16	-1.413	.016
17	-.747	-.134
18	-1.462	-.284
19	1.605	-2.264

20	-.005	-.049
21	-.246	-2.203
22	1.969	-.978
23	-.870	1.263
24	-1.046	-.057
25	.512	1.689
26	-.180	.391
27	2.263	.044
28	.071	1.844
29	.603	.478
30	-.256	-.602
31	.213	.395
32	.450	1.566
33	.664	-.917
34	-.083	1.146
35	-1.042	-.112
36	-1.008	-.023
37	-.497	-2.110
38	.426	1.274
39	.364	1.191
40	-.790	.211
41	-.679	1.623
42	-.062	-1.031
43	-.298	-2.155
44	1.417	-.449
45	-1.490	.046
46	-.544	-.322
47	.113	-.691
48	.453	-.696
49	-.572	.072
50	.071	1.844
51	.896	.267
52	-.079	-1.432
53	-.774	.807
54	-.030	-.355
55	.481	-.147
56	.541	.952
57	-.304	1.366
58	-.660	.904
59	-.748	-.181
GROUP 2		
60	1.184	-1.535
61	1.291	-.133
62	1.182	-.092
63	-1.120	1.368
64	-1.222	-.025
65	-1.565	.018
66	-1.055	-1.861
67	.174	-.188
68	-.400	-.501
69	-1.584	.043
70	-1.379	.054
71	-1.566	-1.366
72	-1.198	-1.949
73	-.780	-.471
74	-1.183	-1.965

75	-1.619	.093
76	-1.553	.133
77	-1.039	-.648
78	-.473	-.187
79	.543	1.664
80	.356	-.326
81	-.895	1.261
82	-1.086	-.019
83	-.595	.024
84	2.426	-1.208
85	1.802	-1.381
86	2.453	-.049
87	-.458	.415
88	-1.422	-.018
89	-.113	-.416
90	.388	.306
91	.199	1.842
92	.198	-.450
93	-.080	.198
94	-1.509	-.004
95	-1.588	.106
96	-.815	-.597
97	-.594	-.238
98	-.139	-.651
99	1.231	-.030
100	-.095	.486
101	-1.617	-.251
102	.797	-.997
103	.977	-1.002
104	.903	.830
105	-.871	-.476
106	-1.525	.096
107	-1.061	-.109
108	.737	1.526
109	1.456	-.582
110	2.571	.831
111	.071	1.844
112	.791	.500
113	1.008	1.309
114	-1.136	-1.401
115	-.527	.491
116	-.551	-.909
117	-.432	-.702
118	.097	1.485
119	-1.553	.026

GROUP	3	
120	2.134	1.087
121	2.993	.690
122	2.812	-.302
123	3.042	-.856
124	1.213	-1.491
125	2.553	.093
126	2.149	.382

Appendix 5a

Pooled July Fungal Assemblage Frequency Data By Collection Type Group

Appendix 5a - Pooled fungal assemblage frequencies by collection type group data for the July collection, from southern Manitoba.

	Fungus (out of 120)																			
	Alternaria spp.	*Aureobasidium pullulans*	*Cladosporium # 1*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mortierella # 3*	*Mortierella # 4*	*Mucor # 1*	*Mucor # 4*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Sphaeopsidales # 23*	*Sphaeropsidales #36*	*Sphaeropsidales # 37*	*sterile # 1*	*sterile # 14*	*Sterile # 24*	*Sterile # 38*	*Trichoderma spp.*
Sample	1	2	3	4	5	6	7	8	10	9	11	12	13	14	15	16	17	18	19	20
1lm	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	2	0	0	0
5lm	0	0	0	0	0	0	0	1	1	0	0	0	0	0	0	3	0	0	0	0
7lm	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	6
8lm	1	0	0	1	0	0	0	0	2	0	0	1	0	0	1	0	0	1	0	4
9lm	16	0	0	0	0	0	19	7	0	0	0	0	0	0	0	0	4	0	0	0
3mm	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0
5mm	0	0	0	0	1	0	0	1	0	0	1	0	0	0	0	3	0	0	0	1
6mm	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0
7mm	0	0	0	0	0	0	0	1	0	0	0	0	1	0	0	0	1	0	1	3
8mm	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
1lr	0	0	0	0	0	0	0	0	1	0	4	0	0	0	0	0	1	0	0	0
5lr	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
7lr	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
8lr	4	0	0	0	1	0	0	7	10	3	2	0	0	1	0	1	18	3	0	1
9lr	2	0	0	0	5	2	0	5	0	0	0	0	0	0	0	2	0	0	0	1
3mr	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
5mr	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
6mr	0	0	0	0	1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
7mr	0	1	2	0	2	0	0	0	0	0	0	0	0	0	0	0	1	1	0	1
8mr	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
6mp	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
6mp	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
6mp	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
7mp	0	0	0	0	1	0	0	1	4	0	0	0	1	0	0	0	2	0	0	0
7mp	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0
7mp	0	0	1	0	0	0	0	1	1	0	0	0	0	0	0	0	5	0	0	0
8mp	0	0	2	0	0	0	0	0	1	1	1	0	0	0	0	0	4	0	0	0
8mp	0	0	0	0	2	0	0	0	7	0	2	0	0	0	0	0	2	0	0	0
8mp	0	0	0	0	0	1	0	0	7	1	0	0	0	0	0	0	3	0	0	0

Sample Legend:

_ = # (plot number)

_ = plot type (mixed moss-lichen [m] or pure lichen [l]plot)

_ = Genus/species (*Cladonia mitis* [m], *C. rangiferina* [r], & *Pleurozium scherberi* [p])

Appendix 6a

Pooled September Fungal Assemblage Frequency Data By Collection Type Group

Appendix 6a - Pooled fungal assemblage frequencies by collection type group data for the September collection, from southern Manitoba.

	Fungus (out of 120)																
	Absidia coerulea	*Alternaria spp.*	*Aspergillus alutaceus group*	*Cladosporium # 1*	*Cladosporium # 2*	*Cunninghamella elegans*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mucor # 1*	*Mucor # 4*	*Mucor # 5*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Rhizopus oryzae*	*Rhizopus # 2*	*Trichoderma spp.*
sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
l1mit	4	22	0	12	0	0	9	3	0	0	0	0	11	3	0	0	4
l2mitis	5	18	0	6	2	0	5	0	1	1	0	2	4	0	0	0	38
l5mit	3	17	0	10	0	0	2	15	9	0	0	5	3	0	1	0	3
l7mit	4	16	0	4	0	0	8	15	15	0	0	10	11	0	1	0	7
l9mit	3	10	0	3	0	0	2	8	1	0	0	4	10	0	1	0	30
m2mit	0	26	0	4	1	0	5	10	4	2	0	1	3	0	1	0	17
m4mit	1	14	0	8	1	0	0	2	13	0	0	2	3	0	0	0	21
m6mit	6	16	0	10	0	0	7	10	9	0	0	5	12	0	0	0	1
m7mit	8	13	0	4	0	0	0	3	4	0	0	14	5	0	1	2	20
m8mit	4	14	0	11	0	0	2	2	10	0	0	1	6	0	0	0	20
l1rang	1	24	0	11	1	2	12	12	1	0	0	4	8	0	0	0	6
l2rang	3	16	0	8	5	0	8	1	9	1	0	18	0	1	2	0	4
l5rang	0	19	0	12	2	0	3	0	19	1	1	9	1	0	0	0	1
l7rang	1	23	0	8	0	0	7	4	11	0	0	3	4	0	0	0	20
l9rang	10	19	0	13	0	0	4	19	2	0	0	1	23	10	0	0	5
m2rang	1	26	0	4	0	0	2	6	9	0	0	3	0	0	0	0	19
m4rang	3	15	0	6	0	0	1	3	11	0	0	12	3	4	1	1	17
m6rang	2	26	0	12	0	0	3	12	3	0	0	3	7	2	0	0	8
m7rang	7	6	2	1	0	0	0	6	1	0	0	2	22	3	0	0	39
m8rang	0	22	0	11	0	0	4	0	8	0	0	5	9	1	0	0	11
m2pleuro	2	2	0	3	1	0	3	36	7	0	0	8	9	0	1	0	42
m6pleuro	9	2	0	2	0	0	1	18	17	1	0	9	13	5	0	0	18
m7pleuro	7	1	0	0	0	0	0	3	3	0	0	2	1	2	1	0	14

Sample Legend:

_ = l (lichen plot), m (mixed moss-lichen plot)

_ = # (plot number)

_ = rang(*Cladonia rangiferina*), mit (*Cladonia mitis*), pleuro (*Pleurozium schreberi*)

Appendix 7a

Temperature Data For La Broquerie, Prior To July & September Collections

Appendix 7a – Meterological data collected from Environment Canada, La Broquerie collecting station, prior to July and September collections in southern Manitoba.

Date	Max Temp	Min Temp	Mean Temp	Date	Max Temp	Min Temp	Mean Temp
22/06/03	24.1	14.4	19.3	06/09/03	27.5	7.9	17.7
23/06/03	25.2	8.6	16.9	07/09/03	30.3	10	20.2
24/06/03	21.4	11.9	16.7	08/09/03	29.7	15.8	22.8
25/06/03	15.2	9.5	12.4	09/09/03	28.1	18.3	23.3
26/06/03	17.5	8.9	13.2	10/09/03	21.2	16.3	18.8
27/06/03	21.9	3.1	12.5	11/09/03	20	12.9	16.5
28/06/03	21.6	12.4	17	12/09/03	22.3	12.2	17.3
29/06/03	23.4	9.2	16.3	13/09/03	19.4	11	15.2
30/06/03	27.7	9.2	18.5	14/09/03	13	6.3	9.7
01/07/03	29.7	14.3	22	15/09/03	20.4	4	12.2
02/07/03	28.9	19.3	24.1	16/09/03	20.8	9.1	15
03/07/03	26	14.9	20.5	17/09/03	19.1	11	15.1
04/07/03	23.9	13.3	18.6	18/09/03	10.9	7.1	9
05/07/03	25.9	13.4	19.7	19/09/03	13.6	4.2	8.9
06/07/03	24.7	12.3	18.5	20/09/03	15.1	0.6	7.9
07/07/03	19.7	11.7	15.7	21/09/03	12.5	7.3	9.9
08/07/03	19.3	7.3	13.3	22/09/03	12.3	6.6	9.5
09/07/03	21.9	6.9	14.4	23/09/03	19	-1.7	8.7
10/07/03	22	13.1	17.6	24/09/03	8.6	5	6.8
11/07/03	23.4	9.6	16.5	25/09/03	11.8	-1.6	5.1
12/07/03	26.5	8.5	17.5	26/09/03	10.3	1	5.7
13/07/03	28.7	14.9	21.8	27/09/03	8.2	2.3	5.3
14/07/03	24.7	14.1	19.4	28/09/03	10.7	-2.3	4.2

Appendix 8a

Precipitation Data From St. Labre, Prior To July & September Collections

Appendix 8a – Meterological data collected from Environment Canada, St. Labre collecting station, prior to July and September collections in southern Manitoba.

Date	Daily Rainfall	Total Rainfall	Date	Daily Rainfall	Total Rainfall
23/06/03	0.4	0.4	07/09/03	0	0
24/06/03	1.2	1.6	08/09/03	0	0
25/06/03	7.2	8.8	09/09/03	1.6	1.6
26/06/03	0	8.8	10/09/03	6.8	8.4
27/06/03	0	8.8	11/09/03	0	8.4
28/06/03	8.6	17.4	12/09/03	8	16.4
29/06/03	0	17.4	13/09/03	2	18.4
30/06/03	0	17.4	14/09/03	1.3	19.7
01/07/03	0	17.4	15/09/03	0	19.7
02/07/03	13.4	30.8	16/09/03	2.6	22.3
03/07/03	0	30.8	17/09/03	37.8	60.1
04/07/03	1.4	32.2	18/09/03	2	62.1
05/07/03	0	32.2	19/09/03	0	62.1
06/07/03	0	32.2	20/09/03	4.4	66.5
07/07/03	0	32.2	21/09/03	3.6	70.1
08/07/03	0	32.2	22/09/03	3.2	73.3
09/07/03	0	32.2	23/09/03	0.4	73.7
10/07/03	T	32.2	24/09/03	4	77.7
11/07/03	8	40.2	25/09/03	1.2	78.9
12/07/03	0	40.2	26/09/03	T	78.9
13/07/03	2.8	43	27/09/03	T	78.9

T = Trace rainfall

Appendix 9a

Fungal Assemblage Frequency Data For *Cladonia mitis* From The September Data Set

Appendix 9a - Fungal assemblage frequency data for *Cladonia mitis* from the September collection in southern Manitoba (Cont. on next pg.).

	Fungus (Frequency out of 20)													
	Absidia coerulea	*Alternaria spp.*	*Cladosporium # 1*	*Cladosporium # 2*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mucor # 1*	*Mucor # 4*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Rhizopus oryzae*	*Rhizopus # 2*	*Trichoderma spp.*
sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14
l1mitt1	0	7	3	0	3	0	0	0	0	1	1	0	0	0
l1mitt2	0	7	2	0	3	0	0	0	0	0	1	0	0	0
l1mitt3	0	6	4	0	0	0	0	0	0	2	0	0	0	0
l2mitt1	0	0	0	0	0	0	0	0	0	0	0	0	0	10
l2mitt2	0	8	1	2	2	0	0	0	0	0	0	0	0	5
l2mitt3	0	9	2	0	3	0	0	0	0	0	0	0	0	5
l5mitt1	0	5	2	0	0	0	0	0	0	0	0	0	0	0
l5mitt2	2	3	3	0	2	1	0	0	0	0	0	0	0	0
l5mitt3	0	5	3	0	0	0	1	0	0	0	0	1	0	0
l7mitt1	0	0	0	0	0	5	2	0	2	6	0	1	0	2
l7mitt2	0	7	2	0	0	0	0	0	0	0	0	0	0	3
l7mitt3	0	5	2	0	4	1	0	0	0	0	0	0	0	0
l9mitt1	0	0	0	0	0	0	0	0	0	0	0	0	0	10
l9mitt2	2	4	1	0	1	1	0	0	0	2	0	0	0	2
l9mitt3	0	6	1	0	1	2	0	0	1	1	0	0	0	0
m2mitt1	0	3	2	1	0	1	1	1	0	0	0	1	0	5
m2mitt2	0	6	0	0	5	1	0	0	0	0	0	0	0	0
m2mitt3	0	9	2	0	0	1	0	0	0	0	0	0	0	0
m4mitt1	0	6	5	0	0	0	1	0	0	1	0	0	0	1
m4mitt2	0	6	3	0	0	0	0	0	0	0	0	0	0	5
m6mitt1	0	5	3	0	1	0	0	0	0	0	0	0	0	0
m6mitt2	0	3	2	0	2	0	0	0	0	3	0	0	0	0
m6mitt3	0	6	3	0	2	3	1	0	0	2	0	0	0	0
m7mitt1	1	5	2	0	0	2	0	0	2	2	0	1	0	3
m7mitt2	1	4	2	0	0	1	2	0	2	2	0	0	1	0
m7mitt3	0	4	0	0	0	0	1	0	0	0	0	0	1	3
m8mitt1	0	1	1	0	0	0	1	0	0	0	0	0	0	7
m8mitt2	0	6	5	0	2	0	0	0	0	1	0	0	0	2
m8mitt3	0	5	5	0	0	0	0	0	0	2	0	0	0	0

Sample Legend:

_ = l (lichen plot), m (mixed moss-lichen plot)
_ = # (plot number)
_ = mit (*Cladonia mitis*)
_ = b(base), t(canopy)
_ = #(sample #)

Appendix 9a (Cont.) - Fungal assemblage frequency data for *Cladonia mitis* from the September collection in southern Manitoba.

	Fungus (Frequency out of 20)													
	Absidia coerulea	*Alternaria spp.*	*Cladosporium # 1*	*Cladosporium # 2*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mucor # 1*	*Mucor # 4*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Rhizopus oryzae*	*Rhizopus # 2*	*Trichoderma spp.*
sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14
l1mitb1	0	2	0	0	0	1	0	0	0	5	0	0	0	0
l1mitb2	2	0	2	0	3	0	0	0	0	2	0	0	0	4
l1mitb3	2	0	1	0	0	2	0	0	0	1	1	0	0	0
l2mitb1	2	0	0	0	0	0	0	0	0	0	0	0	0	8
l2mitb2	1	0	1	0	0	0	1	0	1	0	0	0	0	7
l2mitb3	2	1	2	0	0	0	0	1	1	4	0	0	0	3
l5mitb1	0	0	0	0	0	0	2	0	1	0	0	0	0	0
l5mitb2	1	0	0	0	0	9	6	0	1	1	0	0	0	0
l5mitb3	0	0	2	0	0	5	0	0	3	2	0	0	0	3
l7mitb1	2	0	0	0	1	6	3	0	3	4	0	0	0	0
l7mitb2	2	1	0	0	3	1	1	0	1	0	0	0	0	2
l7mitb3	0	3	0	0	0	2	9	0	4	1	0	0	0	0
l9mitb1	0	0	0	0	0	0	0	0	0	1	0	0	0	10
l9mitb2	1	0	0	0	0	0	0	0	2	0	0	1	0	3
l9mitb3	0	0	1	0	0	5	1	0	1	6	0	0	0	5
m2mitb1	0	0	0	0	0	1	2	0	1	0	0	0	0	5
m2mitb2	0	3	0	0	0	2	0	0	0	0	0	0	0	7
m2mitb3	0	5	0	0	0	4	1	1	0	3	0	0	0	0
m4mitb1	0	0	0	0	0	0	10	0	2	2	0	0	0	0
m4mitb2	0	0	0	1	0	1	1	0	0	0	0	0	0	8
m4mitb3	1	2	0	0	0	1	1	0	0	0	0	0	0	7
m6mitb1	6	2	1	0	1	0	0	0	2	2	0	0	0	0
m6mitb2	0	0	0	0	0	0	7	0	3	3	0	0	0	0
m6mitb3	0	0	1	0	1	7	1	0	0	2	0	0	0	1
m7mitb1	6	0	0	0	0	0	0	0	9	1	0	0	0	2
m7mitb2	0	0	0	0	0	0	1	0	1	0	0	0	0	2
m7mitb3	0	0	0	0	0	0	0	0	0	0	0	0	0	10
m8mitb1	0	0	0	0	0	1	6	0	1	0	0	0	0	3
m8mitb2	3	2	0	0	0	0	2	0	0	3	0	0	0	2
m8mitb3	1	0	0	0	0	1	1	0	0	0	0	0	0	6

Sample Legend:

_ = l (lichen plot), m (mixed moss-lichen plot)
_ = # (plot number)
_ = mit (*Cladonia mitis*)
_ = b(base), t(canopy)
_ = #(sample #)

Appendix 9b

Principal Component Analysis Summary Of Fungal Assemblage Frequency Data For *Cladonia mitis* From the September Data Set

PRINCIPAL COMPONENTS ANALYSIS

September collection of *Cladonia mitis*, southern Manitoba

INPUT AND RUN PARAMETERS

NUMBER OF ROWS =	59
NUMBER OF COLS =	14
TYPE OF ANALYSIS =	PCA FROM COVARIANCES
NO. OF COMPONENTS RETAINED =	3
LABELS FOR OBJECTS =	NOT USED
LABELS FOR VARIABLES =	NOT USED
CORRESP. ANALYSIS =	NOT APPLICABLE
MATRIX =	NOT SAVED
PRINTOUT =	SHORT
VAR. SCORE OPTION =	EIGENVECTORS AS COORDINATES OF VAR.
OBJ. SCORE =	NORMALIZED TO LAMBDA

VARIABLES STATISTICS

POOLED VARIANCE = .8013

VARIABLE	MEAN	STANDARD DEVIATION	VARIANCE	VARIANCE AS %
1	.1393	.2313	.0535	6.675
2	.4228	.3830	.1467	18.309
3	.2637	.2676	.0716	8.935
4	.0183	.0819	.0067	.837
5	.1471	.2390	.0571	7.126
6	.2201	.2861	.0818	10.214
7	.2057	.2844	.0809	10.094
8	.0153	.0667	.0044	.555
9	.1590	.2406	.0579	7.224
10	.2412	.2738	.0750	9.354
11	.0153	.0667	.0044	.555
12	.0255	.0846	.0071	.892
13	.0102	.0549	.0030	.377
14	.4077	.3887	.1511	18.852

NUMBER OF POSITIVE EIGENVALUES = 14

SUM OF POSITIVE EIGENVALUES = 0.80129160E+00

EIGENVALUES

0.2649E+00	0.1931E+00	0.7704E-01	0.6995E-01	0.5405E-01
0.3845E-01	0.3062E-01	0.2824E-01	0.2283E-01	0.7621E-02
0.4968E-02	0.3676E-02	0.3268E-02	0.2597E-02	

EIGENVALUES AS PERCENT

33.06	24.09	9.61	8.73	6.75
4.80	3.82	3.52	2.85	.95
.62	.46	.41	.32	

CUMULATIVE PERCENTAGE OF EIGENVALUES

33.06	57.15	66.77	75.50	82.24
87.04	90.86	94.39	97.24	98.19
98.81	99.27	99.68	100.00	

SQUARE ROOTS OF EIGENVALUES

.514706	.439376	.277560	.264486	.232484
.196085	.175000	.168060	.151092	.087300
.070483	.060631	.057165	.050961	

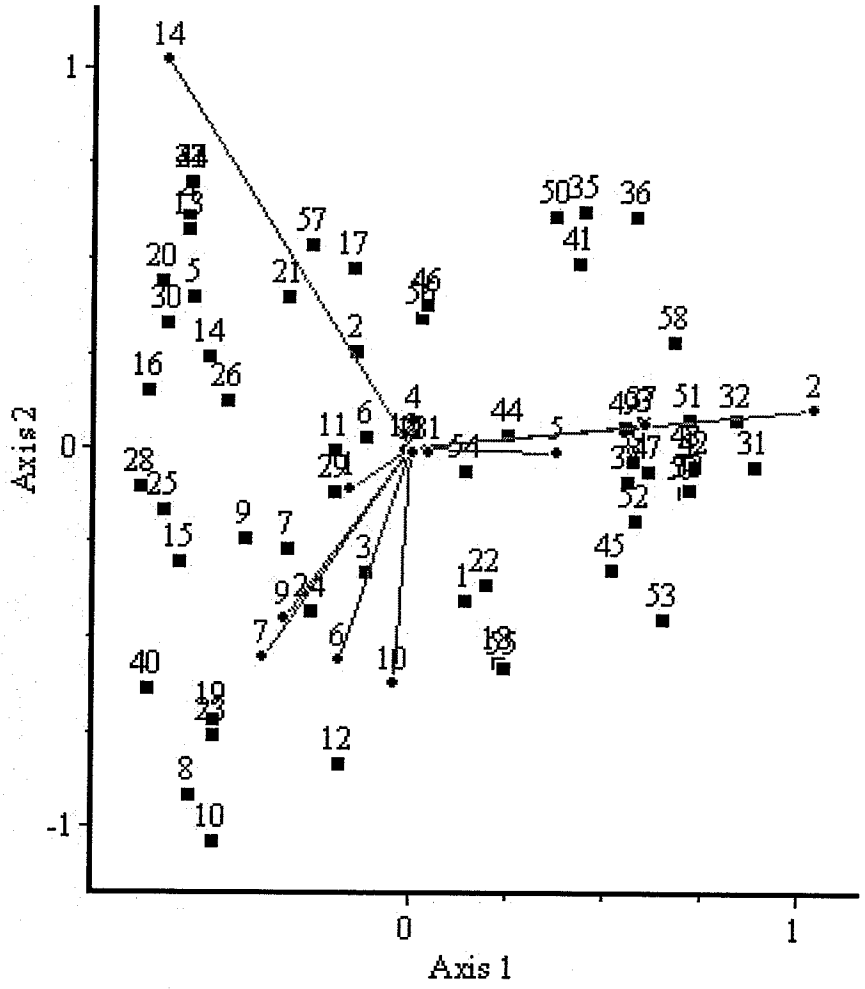
COMPONENT SCORES

1	.139	-.401	.117
2	-.142	.255	.634
3	-.116	-.327	.340
4	-.575	.609	.236
5	-.555	.399	.060
6	-.114	.031	.594
7	-.315	-.265	-.373
8	-.567	-.916	-.259
9	-.425	-.240	.321
10	-.505	-1.039	.255
11	-.197	-.008	.122
12	-.183	-.834	-.584
13	-.571	.578	.101
14	-.515	.240	.160
15	-.590	-.299	.256
16	-.674	.149	-.285
17	-.149	.473	-.151
18	.228	-.566	-.177
19	-.507	-.720	-.456
20	-.638	.437	-.221
21	-.320	.398	-.125
22	.192	-.359	.656
23	-.502	-.759	-.306
24	-.258	-.428	.061
25	-.635	-.164	.668
26	-.468	.121	-.223
27	-.563	.702	-.033
28	-.693	-.104	-.512
29	-.198	-.112	.168
30	-.624	.326	-.045
31	.881	-.045	.070
32	.839	.074	-.089
33	.716	-.111	.054
34	-.563	.702	-.033
35	.442	.622	-.101
36	.574	.609	-.029
37	.594	.072	-.190
38	.557	-.085	.242

39	.566	-.034	-.335
40	-.669	-.633	.087
41	.430	.487	-.148
42	.731	-.044	-.050
43	-.563	.702	-.033
44	.249	.036	.392
45	.515	-.324	.002
46	.038	.377	-.249
47	.607	-.060	-.143
48	.710	-.024	-.227
49	.552	.058	-.145
50	.368	.607	-.093
51	.719	.075	-.106
52	.575	-.192	.202
53	.647	-.451	-.034
54	.140	-.061	.333
55	.240	-.582	.003
56	.026	.342	-.385
57	-.254	.532	-.211
58	.673	.284	.137
59	.701	-.113	.083

SCORES FOR VARIABLES

VARIABLE 1		
-.101	-.073	.582
VARIABLE 2		
.687	.068	-.182
VARIABLE 3		
.399	.041	.203
VARIABLE 4		
.002	.048	-.042
VARIABLE 5		
.248	-.007	.195
VARIABLE 6		
-.120	-.370	.013
VARIABLE 7		
-.251	-.365	-.564
VARIABLE 8		
.003	-.004	.011
VARIABLE 9		
-.212	-.299	.137
VARIABLE 10		
-.027	-.412	.444
VARIABLE 11		
.031	-.008	.022
VARIABLE 12		
-.009	-.003	0.000
VARIABLE 13		
.005	-.006	-.026
VARIABLE 14		
-.415	.675	.108



Appendix 9b - Principle component analysis biplot of the fungal assemblages for the *Cladonia mitis* September data collection from southern Manitoba. Fungal species variables are represented by the circles along the axes, while samples are represented by squares.

Appendix 9c

Multiple Discriminate Analysis Of Fungal Assemblage Frequency Data For The *Cladonia mitis* September Data Set, Groups Separated By Canopy & Base

CANONICAL VARIATES ANALYSIS (MULTIGROUP DISCRIMINANT ANALYSIS)

September collection of *Cladonia mitis* – Canopy vs. base, southern Manitoba

NUMBER OF VARIABLES = 3
NUMBER OF GROUPS = 2
NUMBER OF OBSERVATIONS = 59
LABELS FOR VARIABLES = NOT USED
LABELS FOR OBJERCTS = NOT USED
SCORES ARE = SPHERIZED

UNIVARIATE F RATIOS WITH 1 AND 57 D.F.

VARIABLE	AMONG GROUP SSQ	WITHIN GROUP SSQ	F RATIO
1	8.26	.12	66.334
2	.54	.19	2.887
3	.07	.08	.874

EIGENVALUES

0.1507158D+01

CAN. VAR.	EIGENVALUE	E.V. AS %	CAN. CORR.
1	1.51	100.00	.775

CHI-SQUARE TESTS WITH SUCCESSIVE VARIATES REMOVED

	CAN. VAR. REMOVED	CHISQ	DEGREES OF FREEDOM	WILKS LAMBDA
UP TO	0	51.01	3	.3989

DISCRIMINANT WEIGHTS (CANONICAL VARIATES)

C.V. 1 0.2884645E+01 0.1011628E+01-0.8963438E+00

CORRELATION MATRIX FOR THE TOTAL SAMPLE

1	1.000	0.000	0.000
2	0.000	1.000	0.000
3	0.000	0.000	1.000

CORRELATIONS OF VARIABLES WITH CANONICAL VARIATES

VAR. 1 .946
VAR. 2 .283
VAR. 3 -.159

COMMUNALITIES OF VARIABLES FOR 1 CANONICAL VARIATES
1 .895 2 .080 3 .025

PERCENTAGE OF TR{R} ACCOUNTED FOR BY EACH EIGENVALUE
1 33.334

CENTROID FOR GROUP 1 IN 1 DIMENSIONAL CANONICAL SPACE
1 -1.186

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = .447

CENTROID FOR GROUP 2 IN 1 DIMENSIONAL CANONICAL SPACE
1 1.227

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = .454

THE 95% ISODENSITY CIRCLE AROUND EACH CENTROID
HAS A RADIUS OF 2.45

SPHERIZED SCORES OF OBJECTS ON CANONICAL VARIATES

GROUP	1
1	-.109
2	-.721
3	-.969
4	-1.255
5	-1.251
6	-.829
7	-.842
8	-2.328
9	-1.755
10	-2.735
11	-.685
12	-.847
13	-1.153
14	-1.387
15	-2.234
16	-1.537
17	.184
18	.245
19	-1.782
20	-1.201
21	-.408
22	-.399
23	-1.942
24	-1.233
25	-2.597
26	-1.029
27	-.884
28	-1.646
29	-.836
30	-1.429

GROUP 2

31	2.432
32	2.575
33	1.903
34	-.884
35	1.994
36	2.296
37	1.957
38	1.304
39	1.898
40	-2.649
41	1.867
42	2.110
43	-.884
44	.403
45	1.157
46	.713
47	1.818
48	2.227
49	1.780
50	1.758
51	2.245
52	1.284
53	1.442
54	.043
55	.101
56	.765
57	-.005
58	2.105
59	1.835

Appendix 10a

Fungal Assemblage Frequency Data For *Cladonia rangiferina* From The September Data Set

Appendix 10a - Fungal assemblage frequency data for *Cladonia rangiferina* from the September collection in southern Manitoba (Cont. on next pg.).

	Fungus (Frequency values out of 20)																
	Absidia coerulea	*Alternaria spp.*	*Aspergillus alutaceus group*	*Cladosporium # 1*	*Cladosporium # 2*	*Cunninghamella elegans*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mucor # 1*	*Mucor # 4*	*Mucor # 5*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Rhizopus oryzae*	*Rhizopus # 2*	*Trichoderma spp.*
sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
l1rangt1	0	10	0	5	0	0	4	0	0	0	0	0	0	0	0	0	3
l1rangt2	0	7	0	6	0	0	4	0	0	0	0	0	1	0	0	0	0
l1rangt3	0	7	0	0	1	0	3	0	0	0	0	0	0	0	0	0	0
l2rangt1	0	6	0	0	5	0	4	0	0	1	0	0	0	0	0	0	0
l2rangt2	1	8	0	6	0	0	1	0	0	0	0	0	0	0	1	0	0
l2rangt3	0	2	0	2	0	0	3	0	5	0	0	2	0	0	0	0	0
l5rangt1	0	7	0	2	1	0	3	0	0	0	0	0	0	0	0	0	0
l5rangt2	0	8	0	8	0	0	0	0	0	1	0	0	0	0	0	0	0
l5rangt3	0	4	0	2	1	0	0	0	2	0	1	0	1	0	0	0	0
l7rangt1	0	4	0	4	0	0	2	0	0	0	0	0	0	0	0	0	3
l7rangt2	0	8	0	2	0	0	2	1	0	0	0	0	0	0	0	0	0
l7rangt3	0	8	0	2	0	0	3	2	2	0	0	0	0	0	0	0	1
l9rangt1	0	6	0	3	0	0	0	0	0	0	0	0	2	0	0	0	1
l9rangt2	0	5	0	5	0	0	2	0	0	0	0	0	0	2	0	0	0
l9rangt3	0	3	0	4	0	0	2	1	0	0	0	0	2	2	0	0	0
m2rangt1	0	6	0	0	0	0	0	2	2	0	0	1	0	0	0	0	3
m2rangt2	0	8	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
m2rangt3	0	8	0	4	0	0	1	0	0	0	0	0	0	0	0	0	0
m4rangt1	0	2	0	2	0	0	0	0	2	0	0	2	1	0	0	0	6
m4rangt2	0	8	0	2	0	0	1	0	1	0	0	0	0	0	0	0	0
m4rang.t3	0	5	0	2	0	0	0	0	0	0	0	1	1	4	0	1	0
m6rangt1	0	5	0	5	0	0	2	0	0	0	0	1	2	0	0	0	0
m6rangt2	0	8	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0
m6rangt3	0	2	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0
m7rangt1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	10
m7rangt2	0	5	1	1	0	0	0	2	0	0	0	0	3	0	0	0	2
m7rangt3	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	8
m8rangt1	0	3	0	3	0	0	0	0	1	0	0	0	3	1	0	0	0
m8rangt2	0	4	0	3	0	0	1	0	0	0	0	0	2	0	0	0	6
m8rangt3	0	6	0	1	0	0	3	0	0	0	0	0	0	0	0	0	0

Sample Legend:

_ = l (lichen plot), m (mixed moss-lichen plot)
_ = # (plot number)
_ = rang(*Cladonia rangiferina*)
_ = b(base), t(canopy)
_ = #(sample #)

Appendix 10a (Cont.) - Fungal assemblage frequency data for *Cladonia rangiferina* from the September collection in southern Manitoba.

	Fungus (Frequency values out of 20)																
	Absidia coerulea	*Alternaria spp.*	*Aspergillus alutaceus group*	*Cladosporium # 1*	*Cladosporium # 2*	*Cunninghamella elegans*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mucor # 1*	*Mucor # 4*	*Mucor # 5*	*Mucor # 10*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Rhizopus oryzae*	*Rhizopus # 2*	*Trichoderma spp.*
sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
l1rangb1	0	3	0	0	0	0	0	5	0	0	0	4	3	0	0	0	0
l1rangb2	0	0	0	0	0	0	1	5	0	0	0	0	2	0	0	0	1
l1rangb3	1	1	0	0	0	2	0	2	1	0	0	0	2	0	0	0	2
l2rangb1	0	0	0	0	0	0	0	0	2	0	0	9	0	1	0	0	0
l2rangb2	1	0	0	0	0	0	0	1	0	0	0	4	0	0	0	0	2
l2rangb3	1	0	0	0	0	0	0	0	2	0	0	3	0	0	1	0	2
l5rangb1	0	0	0	0	0	0	0	0	9	0	0	3	0	0	0	0	0
l5rangb2	0	0	0	0	0	0	0	0	2	0	0	1	0	0	0	0	1
l5rangb3	0	0	0	0	0	0	0	0	6	0	0	5	0	0	0	0	0
l7rangb1	1	1	0	0	0	0	0	0	5	0	0	1	0	0	0	0	3
l7rangb2	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	9
l7rangb3	0	2	0	0	0	0	0	0	4	0	0	2	4	0	0	0	4
l9rangb1	5	0	0	0	0	0	0	7	0	0	0	0	7	0	0	0	0
l9rangb2	3	2	0	1	0	0	0	4	0	0	0	1	6	6	0	0	0
l9rangb3	2	3	0	0	0	0	0	7	2	0	0	0	6	0	0	0	4
m2rangb1	1	0	0	0	0	0	0	1	5	0	0	0	0	0	0	0	5
m2rangb2	0	4	0	0	0	0	0	1	0	0	0	0	0	0	0	0	9
m2rangb3	0	0	0	0	0	0	0	2	2	0	0	2	0	0	0	0	2
m4rangb1	0	0	0	0	0	0	0	0	1	0	0	3	0	0	0	0	2
m4rangb2	3	0	0	0	0	0	0	0	5	0	0	3	0	0	0	0	3
m4rangb3	0	0	0	0	0	0	0	3	2	0	0	3	1	0	1	0	6
m6rangb1	0	3	0	0	0	0	1	4	0	0	0	1	3	1	0	0	0
m6rangb2	2	1	0	2	0	0	0	5	0	0	0	1	1	1	0	0	0
m6rangb3	0	0	0	0	0	0	0	3	3	0	0	0	0	0	0	0	8
m7rangb1	0	0	1	0	0	0	0	0	0	0	0	0	2	0	0	0	9
m7rangb2	6	1	0	0	0	0	0	0	1	0	0	2	7	3	0	0	3
m7rangb3	1	0	0	0	0	0	0	4	0	0	0	0	6	0	0	0	7
m8rangb1	0	0	0	0	0	0	0	0	5	0	0	1	0	0	0	0	0
m8rangb2	0	2	0	0	0	0	0	0	1	0	0	3	0	0	0	0	5
m8rangb3	0	7	0	4	0	0	0	0	1	0	0	1	4	0	0	0	0

Sample Legend:

_ = l (lichen plot), m (mixed moss-lichen plot)
_ = # (plot number)
_ = rang(*Cladonia rangiferina*)
_ = b(base), t(canopy)
_ = #(sample #)

Appendix 10b

Principal Component Analysis Summary Of Fungal Assemblage Frequency Data For *Cladonia rangiferina* From the September Data Set

PRINCIPAL COMPONENTS ANALYSIS

September collection of *Cladonia rangiferina*, southern Manitoba

INPUT AND RUN PARAMETERS

NUMBER OF ROWS =	60
NUMBER OF COLS =	17
TYPE OF ANALYSIS =	PCA FROM COVARIANCES
NO. OF COMPONENTS RETAINED =	3
LABELS FOR OBJECTS =	NOT USED
LABELS FOR VARIABLES =	NOT USED
CORRESP. ANALYSIS =	NOT APPLICABLE
MATRIX =	NOT SAVED
PRINTOUT =	SHORT
VAR. SCORE OPTION =	EIGENVECTORS AS COORDINATES OF VAR.
OBJ. SCORE =	NORMALIZED TO LAMBDA

VARIABLES STATISTICS

POOLED VARIANCE = .9002

VARIABLE	MEAN	STANDARD DEVIATION	VARIANCE	VARIANCE AS %
1	.0981	.2082	.0433	4.813
2	.4776	.3845	.1478	16.421
3	.0100	.0545	.0030	.330
4	.2630	.3175	.1008	11.199
5	.0280	.1186	.0141	1.563
6	.0080	.0616	.0038	.421
7	.1600	.2441	.0596	6.618
8	.1939	.2930	.0858	9.536
9	.2319	.2990	.0894	9.935
10	.0100	.0545	.0030	.330
11	.0050	.0389	.0015	.168
12	.2031	.2708	.0733	8.148
13	.2382	.3057	.0934	10.378
14	.0717	.1875	.0352	3.905
15	.0151	.0662	.0044	.486
16	.0050	.0389	.0015	.168
17	.3337	.3745	.1402	15.579

NUMBER OF POSITIVE EIGENVALUES = 17

SUM OF POSITIVE EIGENVALUES = 0.90016690E+00

EIGENVALUES

0.3352E+00	0.1588E+00	0.1218E+00	0.6430E-01	0.4741E-01
0.3614E-01	0.3497E-01	0.3199E-01	0.2714E-01	0.1954E-01
0.9341E-02	0.3813E-02	0.3558E-02	0.2505E-02	0.1943E-02
0.1068E-02	0.7055E-03			

EIGENVALUES AS PERCENT

37.24	17.64	13.53	7.14	5.27
4.01	3.89	3.55	3.01	2.17
1.04	.42	.40	.28	.22
.12	.08			

CUMULATIVE PERCENTAGE OF EIGENVALUES

37.24	54.88	68.40	75.55	80.81
84.83	88.71	92.27	95.28	97.45
98.49	98.91	99.31	99.59	99.80
99.92	100.00			

SQUARE ROOTS OF EIGENVALUES

.578966	.398457	.348956	.253581	.217731
.190103	.187012	.178867	.164737	.139781
.096651	.061751	.059649	.050046	.044081
.032674	.026562			

COMPONENT SCORES

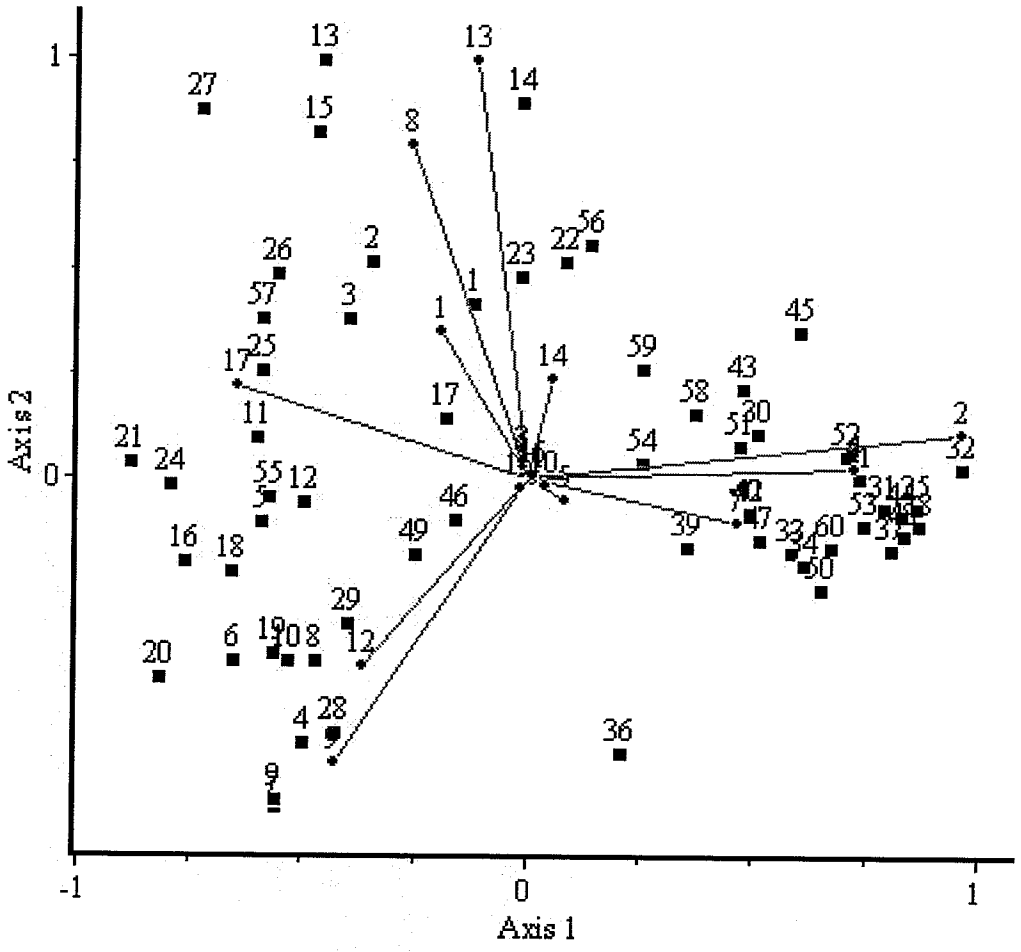
1	-.122	.416	-.556
2	-.350	.515	-.057
3	-.400	.376	.011
4	-.501	-.632	-.649
5	-.593	-.107	-.100
6	-.653	-.439	-.109
7	-.560	-.788	-.523
8	-.472	-.439	-.065
9	-.559	-.772	-.561
10	-.531	-.441	.077
11	-.607	.095	.641
12	-.499	-.061	.022
13	-.463	.993	-.593
14	-.021	.893	-.783
15	-.472	.822	-.013
16	-.764	-.202	.248
17	-.186	.142	.717
18	-.662	-.227	-.079
19	-.565	-.424	-.012
20	-.819	-.480	-.136
21	-.888	.033	.066
22	.080	.513	-.434
23	-.021	.476	-.525
24	-.801	-.019	.430
25	-.596	.251	.594
26	-.559	.485	-.361
27	-.734	.873	.248
28	-.430	-.608	-.359
29	-.400	-.351	.272
30	.508	.104	-.301
31	.790	-.074	.585
32	.959	.024	.040
33	.585	-.181	.079
34	.609	-.211	.089

35	.857	-.072	.014
36	.209	-.660	-.307
37	.802	-.173	.092
38	.866	-.114	.051
39	.353	-.164	-.156
40	.482	-.083	.524
41	.732	-.001	.032
42	.488	-.086	.147
43	.474	.215	.153
44	.827	-.088	-.050
45	.599	.346	-.229
46	-.163	-.104	.179
47	.513	-.147	.053
48	.831	-.136	.071
49	-.252	-.187	.282
50	.649	-.268	.004
51	.468	.076	-.351
52	.706	.055	-.155
53	.743	-.115	.043
54	.251	.035	-.087
55	-.575	-.049	.723
56	.135	.552	.175
57	-.592	.376	.499
58	.369	.151	-.272
59	.252	.258	.585
60	.672	-.170	.078

SCORES FOR VARIABLES

VARIABLE 1		
-.122	.218	-.203
VARIABLE 2		
.603	.067	.109
VARIABLE 3		
-.007	.026	.032
VARIABLE 4		
.454	.016	.026
VARIABLE 5		
.050	-.034	.010
VARIABLE 6		
-.010	.019	.001
VARIABLE 7		
.291	-.068	.095
VARIABLE 8		
-.164	.498	-.169
VARIABLE 9		
-.270	-.428	-.206
VARIABLE 10		
.022	-.010	.006
VARIABLE 11		
.005	-.005	-.007
VARIABLE 12		
-.232	-.282	-.395
VARIABLE 13		
-.076	.624	-.225
VARIABLE 14		
.032	.147	-.254

```
VARIABLE  15
     -.010        -.015        -.001
VARIABLE  16
      .007         .002        -.015
VARIABLE  17
     -.408         .136         .771
```



Appendix 10b - Principle component analysis biplot of the fungal assemblages for the *Cladonia rangiferina* September data collection from southern Manitoba. Fungal species variables are represented by the circles along the axes, while samples are represented by squares.

Appendix 10c

Multiple Discriminate Analysis Of Fungal Assemblage Frequency Data For The *Cladonia rangiferina* September Data Set, Groups Separated By Canopy & Base

CANONICAL VARIATES ANALYSIS (MULTIGROUP DISCRIMINANT ANALYSIS)

September collection of *Cladonia rangiferina* - Canopy vs. Base, southern Manitoba

NUMBER OF VARIABLES = 3
NUMBER OF GROUPS = 2
NUMBER OF OBSERVATIONS = 60
LABELS FOR VARIABLES = NOT USED
LABELS FOR OBJERCTS = NOT USED
SCORES ARE = SPHERIZED

UNIVARIATE F RATIOS WITH 1 AND 58 D.F.

VARIABLE	AMONG GROUP SSQ	WITHIN GROUP SSQ	F RATIO
1	12.40	.13	97.522
2	.07	.16	.413
3	.56	.11	4.873

EIGENVALUES

0.2467787D+01

CAN. VAR.	EIGENVALUE	E.V. AS %	CAN. CORR.
1	2.47	100.00	.844

CHI-SQUARE TESTS WITH SUCCESSIVE VARIATES REMOVED

	CAN. VAR. REMOVED	CHISQ	DEGREES OF FREEDOM	WILKS LAMBDA
UP TO	0	70.26	3	.2884

DISCRIMINANT WEIGHTS (CANONICAL VARIATES)

C.V. 1 0.2993528E+01-0.4618467E+00 0.1746114E+01

CORRELATION MATRIX FOR THE TOTAL SAMPLE

1	1.000	0.000	0.000
2	0.000	1.000	0.000
3	0.000	0.000	1.000

CORRELATIONS OF VARIABLES WITH CANONICAL VARIATES

VAR. 1 .939
VAR. 2 -.100

VAR. 3 .330

COMMUNALITIES OF VARIABLES FOR 1 CANONICAL VARIATES
1 .881 2 .010 3 .109

PERCENTAGE OF TR{R} ACCOUNTED FOR BY EACH EIGENVALUE
1 33.334

CENTROID FOR GROUP 1 IN 1 DIMENSIONAL CANONICAL SPACE
1 -1.545

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = .447

CENTROID FOR GROUP 2 IN 1 DIMENSIONAL CANONICAL SPACE
1 1.545

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = .447

THE 95% ISODENSITY CIRCLE AROUND EACH CENTROID
HAS A RADIUS OF 2.45

SPHERIZED SCORES OF OBJECTS ON CANONICAL VARIATES

GROUP	1
1	-1.530
2	-1.385
3	-1.354
4	-2.341
5	-1.900
6	-1.943
7	-2.225
8	-1.322
9	-2.295
10	-1.250
11	-.744
12	-1.429
13	-2.878
14	-1.843
15	-1.814
16	-1.760
17	.630
18	-2.016
19	-1.517
20	-2.467
21	-2.558
22	-.755
23	-1.200
24	-1.637
25	-.864
26	-2.526
27	-2.169
28	-1.632
29	-.560

	30	.948
GROUP	2	
	31	3.419
	32	2.929
	33	1.973
	34	2.076
	35	2.623
	36	.395
	37	2.640
	38	2.734
	39	.862
	40	2.398
	41	2.246
	42	1.757
	43	1.588
	44	2.429
	45	1.232
	46	-.126
	47	1.697
	48	2.674
	49	-.176
	50	2.072
	51	.754
	52	1.816
	53	2.353
	54	.585
	55	-.435
	56	.455
	57	-1.076
	58	.561
	59	1.656
	60	2.225

Appendix 11a

Soil Fungal Assemblage Frequency Data For Samples Collected Beneath Mats Of *Cladonia mitis* & *C. rangiferina*

Appendix 11a - Fungal assemblage frequency data for soil samples collected beneath mats of *Cladonia rangiferina* and *C. mitis* in southern Manitoba.

	Fungus (out of 20)										
	Absidia californica	*Aureobasidium pullulans*	*Cladosporium # 1*	*Mortierella isabellina/vinacea*	*Mucor # 10*	*Mucor # 12*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Trichoderma # 1*	*Trichoderma spp.*	*Zygorhynchus exponens*
Sample	1	2	3	4	5	6	7	8	9	10	11
m4mit1	0	0	0	0	2	0	0	0	0	1	0
m4mit2	0	0	0	0	3	0	0	0	0	0	0
m4mit3	0	0	0	0	0	3	0	0	4	0	0
m4mit4	0	0	0	0	3	0	0	0	0	0	0
m4mit5	0	0	0	1	0	0	0	0	0	0	0
m6mit1	0	0	0	0	0	0	0	0	0	2	0
m6mit2	0	2	0	1	4	0	3	0	0	0	0
m6mit3	0	0	0	0	0	3	0	0	0	3	0
m6mit4	0	0	1	3	0	0	0	0	0	3	0
m6mit5	0	0	0	0	3	0	0	0	0	4	0
m7rang1	0	0	0	0	0	0	0	0	0	5	0
m7rang2	0	0	0	0	0	5	0	0	0	0	0
m7rang4	4	0	0	0	0	0	0	0	0	4	0
l2mit1	0	0	1	0	0	0	0	0	0	0	0
l2mit2	0	0	0	0	0	0	0	0	0	3	0
l2mit3	0	0	0	0	3	0	1	0	0	1	0
l2mit4	0	0	0	0	3	0	0	0	0	5	0
l2mit5	0	0	0	2	0	5	0	0	4	2	0
l5rang1	0	0	0	0	0	1	0	0	5	3	0
l5rang2	0	0	0	0	5	0	0	0	0	0	0
l5rang3	0	0	0	0	4	0	0	0	0	1	0
l5rang4	0	2	0	0	3	0	0	0	0	0	0
l5rang5	0	0	0	0	0	5	0	0	1	0	0
l9rang1	2	0	0	0	0	0	0	0	0	0	0
l9rang2	0	0	0	0	0	0	0	0	0	2	0
l9rang3	0	0	1	1	0	0	0	2	0	0	0
l9rang4	0	0	0	0	1	0	0	0	0	0	3
l9rang5	2	0	0	0	5	1	0	0	0	0	0

Sample Legend:

_ = l (lichen plot), m (mixed moss-lichen plot)
_ = # (plot number)
_ = rang(*Cladonia rangiferina*), mit (*Cladonia mitis*)
_ = #(sample #)

Appendix 12a

Molecular Data For *Cladonia arbuscula*

Appendix 12a – Molecular data for samples of *C. arbuscula* from northern Manitoba (cont. on next pg.).

Collection No.	Presence/absence of intron Primers 1410 & 1597	Primers 1566 & 1750												
		1	2	3	4	5	6	7	8	9	10	11	12	13
N10lm1so3	2	1	1	1	0	0	0	1	1	0	0	0	1	1
N10lm2so3	0	0	1	0	0	0	1	1	1	0	0	0	1	1
N10lm4so3	0	0	1	0	0	0	0	1	1	0	0	0	1	1
N10lm5so3	0	0	0	0	0	0	0	1	1	0	0	0	1	1
N1lm1so3	1	0	0	0	0	0	0	0	1	0	0	0	1	1
N1lm2so3	1	0	0	0	0	0	0	0	1	0	0	1	1	1
N1lm3so3	0	0	0	0	0	0	1	0	1	0	0	0	1	1
N1lm4so3	0	0	0	0	0	0	1	0	1	0	1	0	1	1
N1lm5so3	1	0	0	0	0	0	0	0	0	0	0	0	1	1
N2lm1so3	0	0	0	0	0	0	0	1	0	0	0	0	0	1
N2lm2so3	1	0	0	0	0	0	0	1	0	0	0	0	1	1
N2lm3so3	2	0	0	0	0	0	1	1	1	0	0	0	1	1
N2lm4so3	0	0	0	0	0	0	1	1	1	0	0	0	1	1
N3lm1so3	1	0	0	0	0	0	1	1	1	0	0	0	1	1
N3lm2so3	1	0	0	0	0	0	1	1	1	1	0	0	1	1
N3lm3so3	1	0	0	0	0	0	0	1	0	0	0	0	1	1
N3Lm4so3	1	0	0	0	0	0	0	1	1	0	0	0	1	1
N3lm5so3	2	0	0	0	0	0	0	1	1	0	0	0	1	1
N4lm1so3	1	0	0	0	0	0	0	1	1	0	0	0	1	1
N4lm2so3	1	0	0	0	0	0	0	1	0	0	0	0	1	1
N4lm3so3	0	0	0	0	0	0	0	0	1	0	0	0	1	1
N4lm4so3	0	0	0	0	0	0	0	0	0	0	1	0	1	1
N4lm5so3	0	0	0	0	0	0	0	1	1	0	0	1	1	1

N1 = Hook point
N2 = Middle 3 Islands
N3 = Three Island Bay
N4 = My Island South
N5 = Long Island North
N6 = Long Island South
N7 = Red Rock Big Zen Island
N8 = East Red Rock Creek
N9 = My Island North
N10 = Mistik Channel West Island

Appendix 12a (cont.) – Molecular data for samples of *C. arbuscula* from northern Manitoba.

Collection No.	Presence/absence of intron Primers 1410 & 1597	Primers 1566 & 1750												
		1	2	3	4	5	6	7	8	9	10	11	12	13
N5lm1so3	1	0	0	0	0	0	1	0	1	0	1	0	1	1
N5lm2so3	1	0	0	0	0	0	0	0	1	0	0	0	0	1
N5lm3so3	1	0	0	0	1	1	0	1	1	0	1	0	1	1
N5lm4so3	0	0	0	0	0	0	0	1	0	0	0	0	0	1
N5lm5so3	1	0	0	0	0	0	0	1	0	0	0	0	1	1
N6lm1so3	0	0	0	0	0	0	1	1	0	0	0	0	1	1
N6lm2so3	0	0	0	0	0	0	0	1	1	0	0	0	1	1
N6lm3so3	2	0	0	0	0	0	0	0	1	0	0	0	1	1
N6lm4so3	0	0	0	0	0	0	0	1	1	0	0	0	1	1
N6lm5so3	1	0	0	0	0	0	0	0	1	0	0	1	1	1
N7lm1so3	1	0	0	0	0	0	0	1	1	0	0	0	0	1
N7lm2so3	1	0	0	0	0	0	0	0	1	0	0	0	0	1
N7lm3so3	1	?	?	?	?	?	?	?	?	?	?	?	?	?
N7lm4so3	1	0	0	0	0	0	0	1	1	0	0	0	1	1
N7lm5so3	1	0	0	0	0	0	0	1	1	0	0	0	1	1
N8lm1so3	1	0	0	0	0	0	1	0	0	0	0	1	1	1
N8lm2so3	1	0	0	0	0	0	0	0	0	0	0	0	0	1
N8lm3so3	1	0	0	0	0	0	1	0	0	0	0	0	0	1
N8lm4so3	2	0	0	0	0	0	0	1	1	0	0	1	1	1
N8lm5so3	1	0	0	0	0	0	0	1	1	0	0	0	1	1
N9lm1so3	1	0	0	0	0	0	0	1	1	0	0	0	1	1
N9lm2so3	0	0	0	0	0	0	0	1	1	0	0	0	1	1
N9lm3so3	1	0	0	0	0	0	0	0	1	0	0	0	0	1
N9lm4so3	0	0	0	0	0	0	0	1	1	0	0	1	1	1
N9lm5so3	0	0	0	0	0	0	0	1	1	0	1	0	0	1

N1 = Hook point
N2 = Middle 3 Islands
N3 = Three Island Bay
N4 = My Island South
N5 = Long Island North
N6 = Long Island South
N7 = Red Rock Big Zen Island
N8 = East Red Rock Creek
N9 = My Island North
N10 = Mistik Channel West Island

Appendix 13a

Fungal Assemblage Frequency Data For The August Data Set

Appendix 13a - Fungal assemblage frequency data for lichen taxa from the August collections in northern Manitoba (cont. on next pg.).

	Fungus (Frequency out of 20)																				
	Absidia coerulea	*Alternaria spp.*	*Cladosporium # 1*	*Cladosporium # 2*	*Cunninghamella elegans*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mortierella # 7*	*Mucor # 1*	*Mucor # 10*	*Mucor # 4*	*Mucor # 5*	*Mucor spinosus*	*Penicillium # 5*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Phycomyces blakeesleanus*	*Sphaeropsidales # 23*	*Sterile # 1*	*Sterile # 14*	*Sterile # 24*	*Sterile # 38*
Sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22
2stellaris5	0	15	0	0	3	0	0	0	0	0	0	0	0	0	18	2	0	0	0	0	0	0
5stellaris5	0	14	0	0	0	0	0	0	0	4	0	0	0	0	12	1	0	0	0	0	0	0
7stellaris5	0	0	0	0	0	0	15	0	0	1	0	0	1	0	13	0	0	0	0	0	1	0
9stellaris5	0	9	0	0	4	0	2	0	0	5	0	0	1	0	15	0	0	0	0	0	0	0
10stellaris5	1	17	1	0	0	0	1	0	0	2	0	0	0	1	12	0	0	0	0	0	0	0
2stereocaulon1	0	2	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	18	0	0	0
5stereocaulon1	0	10	1	0	1	0	0	0	0	3	0	0	0	0	6	0	0	0	7	0	0	0
7stereocaulon1	1	0	0	0	0	0	7	0	1	4	0	0	0	0	3	0	0	0	0	0	0	0
9stereocaulon1	0	0	11	0	0	0	0	0	0	12	0	0	0	0	7	0	0	0	0	0	0	0
10stereocaulon1	0	9	0	0	0	0	0	0	0	9	0	0	0	0	5	1	0	0	0	0	0	0
5amaurocraea1	0	5	0	0	0	0	0	0	0	10	0	0	1	0	10	7	0	0	0	0	0	0
7amaurocraea1	1	0	4	0	0	0	16	0	0	3	1	0	0	0	17	0	0	0	0	0	0	0
9amaurocraea1	0	13	0	0	7	0	2	0	0	1	0	0	0	0	14	0	0	0	0	0	0	0
10vulpicida1	0	18	2	1	0	0	0	0	0	2	0	0	0	0	5	1	0	0	0	0	0	0
2vulpicida1	0	8	0	1	0	0	0	0	0	0	0	0	0	0	1	1	0	0	0	0	0	0
5vulpicida1	0	13	0	0	0	0	2	0	0	1	0	0	0	0	8	0	0	0	0	0	0	2
7vulpicida1	0	0	7	0	0	0	7	0	0	1	0	0	0	0	14	0	0	0	0	0	0	0
9vulpicida1	0	0	4	0	0	0	0	0	0	1	0	0	0	19	8	0	0	0	0	0	0	0

Sample Legend:

_ = # (transect number)

_ = stellaris (*Cladonia stellaris*), stereocaulon (*Stereocaulon alpinum*), amaurocraea (*Cladonia amaurocraea*), vulpicida (*Vulpicida pinastri*)

_ = #(sample #)

Appendix 13a (cont.) - Fungal assemblage frequency data for lichen taxa from the August collections in northern Manitoba.

	Fungus (Frequency out of 20)																				
	Absidia coerulea	*Alternaria spp.*	*Cladosporium # 1*	*Cladosporium # 2*	*Cunninghamella elegans*	*Epicoccum purpurascens*	*Mortierella isabellina/vinacea*	*Mortierella # 7*	*Mucor # 1*	*Mucor # 10*	*Mucor # 4*	*Mucor # 5*	*Mucor spinosus*	*Penicillium # 5*	*Penicillium (non-sclerotial)*	*Penicillium (sclerotial)*	*Phycomyces blakeesleanus*	*Sphaeropsidales # 23*	*Sterile # 1*	*Sterile # 14*	*Sterile # 24*	*Sterile # 38*
Sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22
10evernia1	0	12	7	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0
1evernia1	0	19	4	0	0	0	0	0	0	0	0	0	0	0	5	0	0	0	0	0	0	0
2evernia1	0	6	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
6evernia1	0	13	6	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0
9evernia1	0	20	9	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0
10arbuscula5	2	16	0	0	1	0	6	0	0	7	0	0	0	0	11	0	0	0	0	0	0	0
2arbuscula5	0	12	0	0	7	0	1	0	0	0	0	0	0	0	20	5	0	0	0	0	0	0
5arbuscula5	0	19	1	0	0	0	0	0	0	1	0	0	0	0	19	1	0	0	0	0	0	0
7arbuscula5	0	0	5	0	0	0	7	0	0	1	0	0	0	0	13	0	0	0	0	1	0	0
9arbuscula5	0	16	0	0	12	0	2	0	0	4	0	0	1	0	4	0	0	0	1	0	0	0
10peltigera1	0	3	0	0	0	0	2	0	0	20	2	1	1	0	6	0	0	0	0	0	0	0
11peltigera1	0	6	12	0	0	0	0	1	3	8	0	1	1	0	9	0	0	2	0	0	0	0
1peltigera1	0	6	1	0	0	0	0	0	0	14	0	0	1	0	0	0	1	0	0	0	0	0
3peltigera1	0	13	2	0	0	0	2	0	0	1	0	0	0	0	9	0	0	0	0	0	0	0
9peltigera1	0	5	3	2	0	0	3	0	0	20	1	1	0	0	1	0	0	0	0	0	0	0
2rangiferina5	0	6	0	0	0	0	0	0	0	2	0	0	0	0	6	1	0	0	0	0	0	0
5rangiferina5	0	14	0	0	0	0	0	0	0	0	0	0	0	0	9	4	0	0	0	0	0	0
7rangiferina5	0	0	7	0	0	0	12	0	0	0	0	0	0	0	8	0	0	0	0	1	0	0
9rangiferina7	0	12	0	0	7	0	2	0	0	1	0	0	0	0	4	0	0	0	5	0	0	0
10rangiferina5	0	6	0	0	0	0	0	0	0	2	0	0	2	0	13	0	0	0	7	0	0	0

Sample Legend:

_ = # (transect number)

_ = evernia (*Evernia mesomorpha*), arbuscula (*Cladonia arbuscula*), peltigera (*Peltigera* spp.), rangiferina (*Cladonia rangiferina*)

_ = #(sample #)

Appendix 13b

Principal Component Analysis Summary Of The Fungal Assemblage Frequency Data For The August Data Set

PRINCIPAL COMPONENTS ANALYSIS

August collection, northern Manitoba

INPUT AND RUN PARAMETERS

NUMBER OF ROWS =	40
NUMBER OF COLS =	22
TYPE OF ANALYSIS =	PCA FROM COVARIANCES
NO. OF COMPONENTS RETAINED =	3
LABELS FOR OBJECTS =	NOT USED
LABELS FOR VARIABLES =	NOT USED
CORRESP. ANALYSIS =	NOT APPLICABLE
MATRIX =	NOT SAVED
PRINTOUT =	SHORT
VAR. SCORE OPTION =	EIGENVECTORS AS COORDINATES OF VAR.
OBJ. SCORE =	NORMALIZED TO LAMBDA

VARIABLES STATISTICS

POOLED VARIANCE = 157.8007

VARIABLE	MEAN	STANDARD DEVIATION	VARIANCE	VARIANCE AS %
1	.1250	.4043	.1635	.104
2	9.0750	6.3906	40.8404	25.881
3	.1000	.3789	.1436	.091
4	2.3250	3.3312	11.0968	7.032
5	1.1750	2.7164	7.3788	4.676
6	.0250	.1581	.0250	.016
7	2.2750	4.0635	16.5122	10.464
8	.1000	.4961	.2462	.156
9	.1000	.3789	.1436	.091
10	3.6250	5.1575	26.5994	16.856
11	.0750	.2667	.0712	.045
12	.2500	.4935	.2436	.154
13	.5250	3.0042	9.0250	5.719
14	8.0250	5.6726	32.1788	20.392
15	.6750	1.4916	2.2250	1.410
16	.0250	.1581	.0250	.016
17	.0500	.3162	.1000	.063
18	.0750	.2667	.0712	.045
19	.0250	.1581	.0250	.016
20	.0500	.3162	.1000	.063
21	.9500	3.2499	10.5615	6.693
22	.0250	.1581	.0250	.016

NUMBER OF POSITIVE EIGENVALUES = 21

SUM OF POSITIVE EIGENVALUES = 0.15780062E+03

EIGENVALUES

0.5068E+02	0.3898E+02	0.2244E+02	0.1372E+02	0.1142E+02
0.8034E+01	0.5847E+01	0.4316E+01	0.1355E+01	0.3229E+00
0.1948E+00	0.1483E+00	0.9947E-01	0.7876E-01	0.6573E-01
0.4906E-01	0.2087E-01	0.1754E-01	0.8936E-02	0.4137E-02
0.2886E-02				

EIGENVALUES AS PERCENT

32.12	24.70	14.22	8.69	7.23
5.09	3.71	2.73	.86	.20
.12	.09	.06	.05	.04
.03	.01	.01	.01	0.00
0.00				

CUMULATIVE PERCENTAGE OF EIGENVALUES

32.12	56.82	71.04	79.73	86.97
92.06	95.76	98.50	99.36	99.56
99.69	99.78	99.84	99.89	99.93
99.97	99.98	99.99	100.00	100.00
100.00				

SQUARE ROOTS OF EIGENVALUES

7.119194	6.243209	4.736922	3.704099	3.378713
2.834511	2.417995	2.077383	1.164253	.568259
.441374	.385054	.315386	.280649	.256379
.221504	.144470	.132429	.094529	.064320
.053725				

COMPONENT SCORES

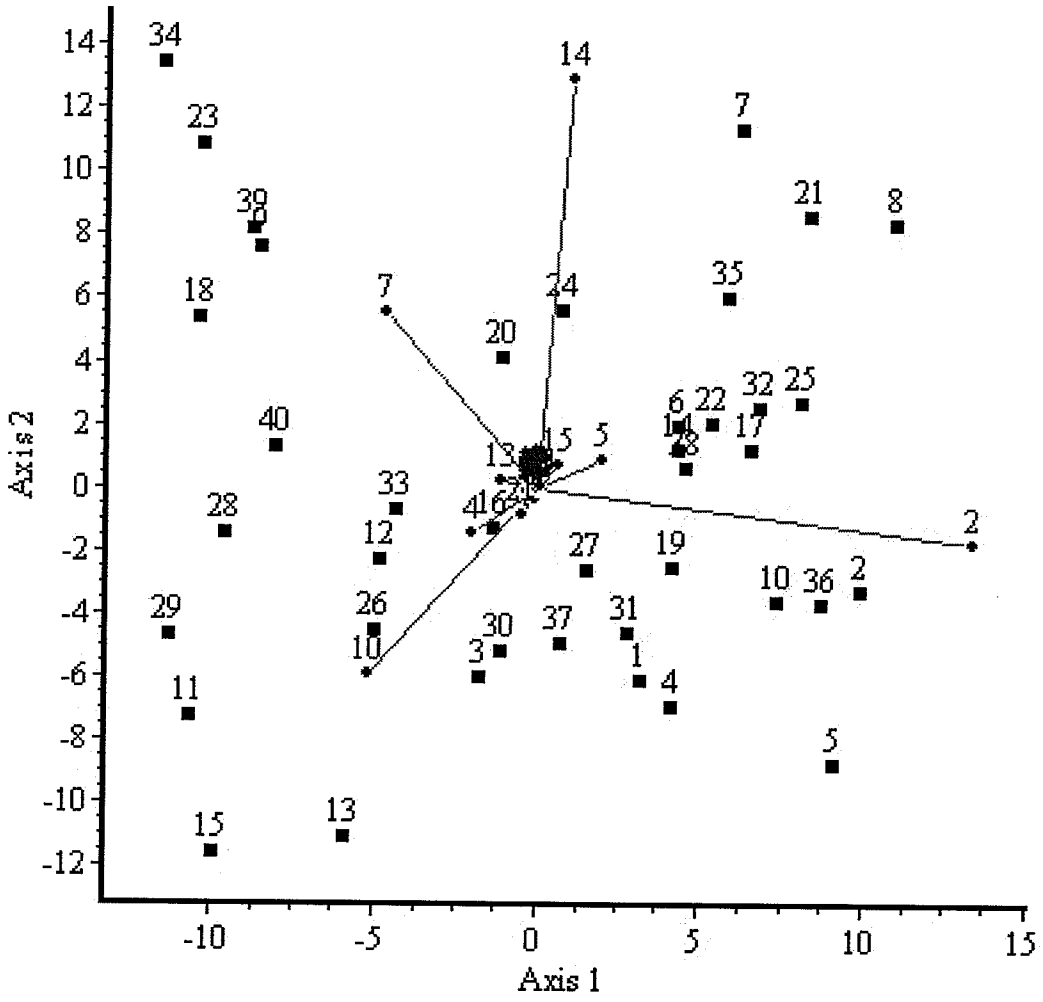
1	3.221	-6.093	-5.518
2	9.965	-3.278	-2.075
3	-1.792	-6.050	-6.798
4	4.194	-6.900	-5.475
5	9.169	-8.791	-2.978
6	4.305	1.964	5.681
7	6.246	11.371	3.807
8	11.031	8.332	4.798
9	-8.553	7.585	-2.571
10	7.376	-3.604	1.187
11	-10.605	-7.236	12.076
12	-4.819	-2.299	2.300
13	-5.907	-11.059	5.297
14	4.363	1.256	-.718
15	-9.885	-11.591	9.936
16	-1.339	-1.309	-1.955
17	6.600	1.177	-.576
18	-10.331	5.391	-6.026
19	4.155	-2.506	-3.498
20	-1.137	4.087	-1.112
21	8.377	8.646	3.206
22	5.353	2.010	3.645
23	-10.306	10.892	-2.340
24	.739	5.593	4.708
25	8.069	2.679	2.178
26	-5.005	-4.490	-10.933

27	1.544	-2.631	-2.406
28	-9.574	-1.441	-3.266
29	-11.280	-4.686	3.651
30	-1.118	-5.173	3.973
31	2.843	-4.602	-1.388
32	6.837	2.519	-.249
33	-4.331	-.661	6.455
34	-11.529	13.441	.209
35	5.857	6.003	2.002
36	8.738	-3.723	-.240
37	.710	-4.960	-5.144
38	4.568	.608	-.809
39	-8.740	8.228	-2.472
40	-8.009	1.301	-6.565

SCORES FOR VARIABLES

VARIABLE 1		
-.002	.012	.012
VARIABLE 2		
.868	-.115	.200
VARIABLE 3		
-.005	-.021	.017
VARIABLE 4		
-.132	-.092	-.153
VARIABLE 5		
.127	.056	.063
VARIABLE 6		
.002	-.004	-.006
VARIABLE 7		
-.301	.355	-.072
VARIABLE 8		
-.012	-.005	.004
VARIABLE 9		
-.022	-.008	.039
VARIABLE 10		
-.331	-.384	.817
VARIABLE 11		
-.013	-.014	.028
VARIABLE 12		
-.012	.002	.031
VARIABLE 13		
-.071	.015	-.142
VARIABLE 14		
.068	.834	.394
VARIABLE 15		
.043	.044	.077
VARIABLE 16		
-.003	-.007	.006
VARIABLE 17		
-.005	-.003	.005
VARIABLE 18		
-.008	.006	-.011
VARIABLE 19		
-.005	.007	-.003

```
VARIABLE  20
      .005          .001         -.002
VARIABLE  21
     -.030         -.057         -.272
VARIABLE  22
     -.002         -.002          .003
```



Appendix 13b - Principle component analysis biplot of the fungal assemblages for the Payuk lake august data collection. Fungal species variables are represented by the circles along the axes, while samples are represented by squares.

Appendix 13c

Multiple Discriminate Analysis Summary Of The Fungal Assemblage Frequency Data For The August Data Set, Groups Separated By Lichen Taxa

CANONICAL VARIATES ANALYSIS (MULTIGROUP DISCRIMINANT ANALYSIS)

August collections - by lichen taxa, northern Manitoba

NUMBER OF VARIABLES = 3
NUMBER OF GROUPS = 9
NUMBER OF OBSERVATIONS = 40
LABELS FOR VARIABLES = NOT USED
LABELS FOR OBJERCTS = NOT USED
SCORES ARE = SPHERIZED

UNIVARIATE F RATIOS WITH 8 AND 31 D.F.

VARIABLE	AMONG GROUP SSQ	WITHIN GROUP SSQ	F RATIO
1	73.99	44.67	1.656
2	111.47	20.27	5.499
3	56.77	13.58	4.180

EIGENVALUES

0.2332839D+01 0.1141838D+01 0.2076750D+00

CAN. VAR.	EIGENVALUE	E.V. AS %	CAN. CORR.
1	2.33	63.35	.837
2	1.14	31.01	.730
3	.21	5.64	.415

CHI-SQUARE TESTS WITH SUCCESSIVE VARIATES REMOVED

	CAN. VAR. REMOVED	CHISQ	DEGREES OF FREEDOM	WILKS LAMBDA
UP TO	0	71.09	24	.1160
UP TO	1	31.36	14	.3866
UP TO	2	6.23	6	.8280

DISCRIMINANT WEIGHTS (CANONICAL VARIATES)

C.V. 1 0.1210321E-01 0.2091944E+00 0.2042394E+00
C.V. 2 0.1082596E+00 0.9504315E-01-0.1835987E+00
C.V. 3 0.1108089E+00-0.6063998E-01 0.9305308E-01

CORRELATION MATRIX FOR THE TOTAL SAMPLE

1	1.000	0.000	0.000
2	0.000	1.000	0.000
3	0.000	0.000	1.000

CORRELATIONS OF VARIABLES WITH CANONICAL VARIATES

VAR.	1	.053	.591	.805
VAR.	2	.802	.455	-.386
VAR.	3	.594	-.667	.450

COMMUNALITIES OF VARIABLES FOR 3 CANONICAL VARIATES

1 1.000 2 1.000 3 1.000

PERCENTAGE OF TR{R} ACCOUNTED FOR BY EACH EIGENVALUE

1 33.333 2 33.333 3 33.333

CENTROID FOR GROUP 1 IN 3 DIMENSIONAL CANONICAL SPACE

1 -2.175 2 .783 3 .501

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = 1.095

CENTROID FOR GROUP 2 IN 3 DIMENSIONAL CANONICAL SPACE

1 1.650 2 .456 3 .381

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = 1.095

CENTROID FOR GROUP 3 IN 3 DIMENSIONAL CANONICAL SPACE

1 -.179 2 -2.230 3 .318

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = 1.095

CENTROID FOR GROUP 4 IN 3 DIMENSIONAL CANONICAL SPACE

1 -.257 2 .569 3 -.373

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = 1.095

CENTROID FOR GROUP 5 IN 3 DIMENSIONAL CANONICAL SPACE

1 1.743 2 .413 3 .122

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = 1.095

CENTROID FOR GROUP 6 IN 3 DIMENSIONAL CANONICAL SPACE

1 -1.199 2 -.571 3 -.507

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = 1.095

CENTROID FOR GROUP 7 IN 3 DIMENSIONAL CANONICAL SPACE

1 -.326 2 .575 3 .523

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = 1.731

CENTROID FOR GROUP 8 IN 3 DIMENSIONAL CANONICAL SPACE

1 1.859 2 -.296 3 -.480

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = 1.413

CENTROID FOR GROUP 9 IN 3 DIMENSIONAL CANONICAL SPACE

1 -.568 2 .528 3 -.362

RADIUS OF 95% CONFIDENCE CIRCLE AROUND CENTROID = 1.095

THE 95% ISODENSITY CIRCLE AROUND EACH CENTROID
HAS A RADIUS OF 2.45

SPHERIZED SCORES OF OBJECTS ON CANONICAL VARIATES

GROUP 1			
1	-2.363	.783	.213
2	-.989	1.148	1.110
3	-2.676	.479	-.464
4	-2.511	.803	.374
5	-2.336	.704	1.272
GROUP 2			
6	1.623	-.390	.887
7	3.232	1.058	.357
8	2.856	1.105	1.164
9	.958	.267	-1.647
10	-.422	.238	1.146
GROUP 3			
11	.824	-4.053	.387
12	-.069	-1.162	-.181
13	-1.303	-2.663	.509
14	.169	.724	.340
15	-.515	-3.996	.532
GROUP 4			
16	-.689	.090	-.251
17	.208	.932	.606
18	-.228	.500	-2.032
19	-1.188	.854	.287
20	.614	.469	-.477

GROUP	5		
21	2.565	1.140	.702
22	1.230	.101	.810
23	1.676	.349	-2.020
24	2.141	-.253	.181
25	1.103	.728	.934
GROUP	6		
26	-3.233	1.039	-1.300
27	-1.023	.359	.107
28	-1.084	-.574	-1.277
29	-.371	-2.337	-.626
30	-.284	-1.342	.560
GROUP	7		
31	-1.212	.125	.465
32	.559	1.025	.582
GROUP	8		
33	1.128	-1.717	.161
34	2.715	-.009	-2.073
35	1.736	.837	.471
GROUP	9		
36	-.722	.636	1.172
37	-2.080	.550	-.099
38	.017	.701	.394
39	1.111	.290	-1.697
40	-1.166	.462	-1.577

Appendix 14a

TLC Data From The July Collections

Appendix 14a – TLC data for the lichen samples from the July collections (cont. on next pg.).

Collection #	TLC #	Lichen Compounds Present	Identification
S1LM1S03	JR2-4	Usnic acid	*Cladonia mitis*
S1LM2S03	JR2-5	Usnic acid	*Cladonia mitis*
S1LM3S03	JR2-6	Usnic acid	*Cladonia mitis*
S1LR1S03	JR21	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S1LR2S03	JR22	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S1LR3S03	JR2-3	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S3MM1S03	JR4-6	Usnic acid	*Cladonia mitis*
S3MM2S03	JR4-7	Usnic acid	*Cladonia mitis*
S3MM3S03	JR4-8	Usnic acid	*Cladonia mitis*
S3MR1S03	JR4-3	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S3MR2S03	JR4-4	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S3MR3S03	JR4-5	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S4MM1S03	JR3-13	Usnic acid	*Cladonia mitis*
S4MM2S03	JR3-14	Usnic acid	*Cladonia mitis*
S4MM3S03	JR3-15	Usnic acid	*Cladonia mitis*
S4MR1S03	JR3-16	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S4MR2S03	JR3-17	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S4MR3S03	JR3-18	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S5LM1S03	JR2-16	Usnic acid	*Cladonia mitis*
S5LM2S03	JR2-17	Usnic acid	*Cladonia mitis*
S5LM3S03	JR2-18	Usnic acid	*Cladonia mitis*
S5LR1S03	JR2-13	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S5LR2S03	JR2-14	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S5LR3S03	JR2-15	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S5MM1S03	JR3-1	Usnic acid	*Cladonia mitis*
S5MM2S03	JR3-2	Usnic acid	*Cladonia mitis*
S5MM3S03	JR3-3	Usnic acid	*Cladonia mitis*
S5MR1S03	JR3-4	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S5MR2SO3	JR3-5	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S5MR3S03	JR3-6	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S6MM1S03	JR3-7	Usnic acid	*Cladonia mitis*
S6MM2S03	JR3-8	Usnic acid, Fumarprotocetraric acid	*Cladonia arbuscula*
S6MM3S03	JR3-9	Usnic acid	*Cladonia mitis*
S6MR1S03	JR3-10	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S6MR2S03	JR3-11	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S6MR3S03	JR3-12	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S7LM1S03	JR2-19	Usnic acid	*Cladonia mitis*
S7LM2S03	JR2-20	Usnic acid	*Cladonia mitis*
S7LM3S03	JR2-21	Usnic acid	*Cladonia mitis*
S7LR1S03	JR2-22	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S7LR2S03	JR2-2	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S7LR3S03	JR2-1	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S7MM1S03	JR3-19	Usnic acid	*Cladonia mitis*
S7MM2S03	JR3-20	Usnic acid	*Cladonia mitis*
S7MM3S03	JR3-21	Usnic acid	*Cladonia mitis*

Appendix 14a (Cont.) – TLC data for the lichen samples from the July collections.

Collection #	TLC #	Lichen Compounds Present	Identification
S7MR1S03	JR3-2	Usnic acid	*Cladonia mitis*
S7MR2S03	JR4-1	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S7MR3S03	JR4-2	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S8LM1S03	JR4-9	Usnic acid	*Cladonia mitis*
S8LM2S03	JR4-10	Usnic acid	*Cladonia mitis*
S8LM3S03	JR4-11	Usnic acid	*Cladonia mitis*
S8LR1S03	JR4-12	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S8LR2S03	JR4-13	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S8LR3S03	JR4-14	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S9LM1S03	JR2-7	Usnic acid	*Cladonia mitis*
S9LM2S03	JR2-8	Usnic acid	*Cladonia mitis*
S9LM3S03	JR2-9	Usnic acid	*Cladonia mitis*
S9LR1S03	JR2-10	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S9LR2S03	JR2-11	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S9LR3S03	JR2-12	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*

Appendix 14b

TLC Data From The September Collections

Appendix 14b – TLC data for the lichen samples from the September collections (Cont. on next pg.).

Collection #	TLC #	Lichen Compounds Present	Identification
S1LM1F03	JR5-7	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S1LM2F03	JR5-8	Usnic acid	*Cladonia mitis*
S1LM3F03	JR5-9	Usnic acid	*Cladonia mitis*
S1LR1F03	JR5-4	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S1LR2F03	JR5-5	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S1LR3F03	JR5-6	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S2LM1F03	JR6-12	Usnic acid	*Cladonia mitis*
S2LM2F03	JR6-13	Usnic acid	*Cladonia mitis*
S2LM3F03	JR6-14	Usnic acid	*Cladonia mitis*
S2LR1F03	JR6-15	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S2LR2F03	JR6-16	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S2LR3F03	JR6-17	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S2MM1F03	JR6-21	Usnic acid	*Cladonia mitis*
S2MM2F03	JR6-22	Usnic acid, Fumarprotocetraric acid	*Cladonia arbuscula*
S2MM3F03	JR7-1	Usnic acid	*Cladonia mitis*
S2MR1F03	JR6-18	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S2MR2F03	JR6-19	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S2MR3F03	JR6-20	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S4MM1F03	JR4-20	Usnic acid	*Cladonia mitis*
S4MM2F03	JR4-21	Usnic acid	*Cladonia mitis*
S4MM3F03	JR4-22	Usnic acid	*Cladonia mitis*
S4MR1F03	JR5-1	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S4MR2F03	JR5-2	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S4MR3F03	JR5-3	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S5LM1F03	JR6-6	Usnic acid	*Cladonia mitis*
S5LM2F03	JR6-7	Usnic acid	*Cladonia mitis*
S5LM3F03	JR6-8	Usnic acid	*Cladonia mitis*
S5LR1F03	JR6-9	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S5LR2F03	JR6-10	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S5LR3F03	JR6-11	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S6MM1F03	JR5-10	Usnic acid	*Cladonia mitis*
S6MM2F03	JR5-11	Usnic acid	*Cladonia mitis*
S6MM3F03	JR5-12	Usnic acid	*Cladonia mitis*
S6MR1F03	JR5-13	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S6MR2F03	JR5-14	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S6MR3F03	JR5-15	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S7LM1F03	JR5-16	Usnic acid	*Cladonia mitis*
S7LM2F03	JR5-17	Usnic acid	*Cladonia mitis*
S7LM3F03	JR5-18	Usnic acid	*Cladonia mitis*
S7LR1F03	JR5-19	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S7LR2F03	JR5-20	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S7LR3F03	JR5-21	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S7MM1F03	JR7-5	Usnic acid	*Cladonia mitis*
S7MM2F03	JR7-6	Usnic acid	*Cladonia mitis*
S7MM3F03	JR7-7	Usnic acid	*Cladonia mitis*

Appendix 14b (cont.) – TLC data for the lichen samples from the September collections.

Collection #	TLC #	Lichen Compounds Present	Identification
S7MR1F03	JR7-2	Usnic acid	*Cladonia mitis*
S7MR2F03	JR7-3	Usnic acid	*Cladonia mitis*
S7MR3F03	JR7-4	Usnic acid	*Cladonia mitis*
S8MM1F03	JR6-2	Usnic acid	*Cladonia mitis*
S8MM2F03	JR6-1	Usnic acid	*Cladonia mitis*
S8MM3F03	JR6-3	Usnic acid	*Cladonia mitis*
S8MR1F03	JR5-22	Usnic acid	*Cladonia mitis*
S8MR2F03	JR6-4	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S8MR3F03	JR6-5	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S9LM1F03	JR7-8	Usnic acid	*Cladonia mitis*
S9LM2F03	JR7-9	Usnic acid	*Cladonia mitis*
S9LM3F03	JR7-10	Usnic acid	*Cladonia mitis*
S9LR1F03	JR7-11	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S9LR2F03	JR7-12	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
S9LR3F03	JR7-13	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*

Appendix 14c

TLC Data From The August Collections For The Molecular Data Sets

Appendix 14c – TLC data for the lichen samples from the August collections used for the molecular data sets (Cont. on next pg.).

Collection #	TLC #	Lichen Compounds Present	Identification
JR8-7	N10LM1S03	Usnic acid	*Cladonia arbuscula*
JR8-8	N10LM2S03	Usnic acid	*Cladonia arbuscula*
JR8-10	N10LM4S03	Usnic acid	*Cladonia arbuscula*
JR7-22	N10LM5S03	Usnic acid	*Cladonia arbuscula*
JR10-1	N1LM1S03	Usnic acid	*Cladonia arbuscula*
JR10-2	N1LM2S03	Usnic acid	*Cladonia arbuscula*
JR10-3	N1LM3S03	Usnic acid	*Cladonia arbuscula*
JR10-4	N1LM4S03	Usnic acid	*Cladonia arbuscula*
JR10-5	N1LM5S03	Usnic acid	*Cladonia arbuscula*
JR9-9	N2LM1S03	Usnic acid	*Cladonia arbuscula*
JR9-10	N2LM2S03	Usnic acid	*Cladonia arbuscula*
JR9-11	N2LM3S03	Usnic acid	*Cladonia arbuscula*
JR9-12	N2LM4S03	Usnic acid	*Cladonia arbuscula*
JR10-22	N2LM5S03	Usnic acid, Fumarprotocetraric acid	*Cladonia arbuscula*
JR9-18	N3LM1S03	Usnic acid	*Cladonia arbuscula*
JR9-19	N3LM2S03	Usnic acid	*Cladonia arbuscula*
JR9-20	N3LM3S03	Usnic acid	*Cladonia arbuscula*
JR9-21	N3LM4S03	Usnic acid	*Cladonia arbuscula*
JR9-22	N3LM5S03	Usnic acid	*Cladonia arbuscula*
JR9-13	N4LM1S03	Usnic acid	*Cladonia arbuscula*
JR9-14	N4LM2S03	Usnic acid	*Cladonia arbuscula*
JR9-15	N4LM3S03	Usnic acid	*Cladonia arbuscula*
JR9-16	N4LM4S03	Usnic acid	*Cladonia arbuscula*
JR9-17	N4LM5S03	Usnic acid	*Cladonia arbuscula*
JR8-11	N5LM1S03	Usnic acid	*Cladonia arbuscula*
JR8-12	N5LM2S03	Usnic acid	*Cladonia arbuscula*
JR10-20	N5LM3S03	Usnic acid	*Cladonia arbuscula*
JR8-14	N5LM4S03	Usnic acid	*Cladonia arbuscula*
JR7-20	N5LM5S03	Usnic acid	*Cladonia arbuscula*
JR10-6	N6LM1S03	Usnic acid	*Cladonia arbuscula*
JR10-7	N6LM2S03	Usnic acid	*Cladonia arbuscula*
JR10-8	N6LM3S03	Usnic acid	*Cladonia arbuscula*
JR10-9	N6LM4S03	Usnic acid	*Cladonia arbuscula*
JR10-10	N6LM5S03	Usnic acid	*Cladonia arbuscula*
JR10-21	N7LM1S03	Usnic acid, unknown	*Cladonia arbuscula*
JR9-6	N7LM2S03	Usnic acid	*Cladonia arbuscula*
JR9-7	N7LM3S03	Usnic acid	*Cladonia arbuscula*
JR9-8	N7LM4S03	Usnic acid	*Cladonia arbuscula*
JR7-19	N7LM5S03	Usnic acid	*Cladonia arbuscula*
JR10-11	N8LM1S03	Usnic acid	*Cladonia arbuscula*
JR10-12	N8LM2S03	Usnic acid	*Cladonia arbuscula*
JR10-13	N8LM3S03	Usnic acid	*Cladonia arbuscula*
JR10-14	N8LM4S03	Usnic acid	*Cladonia arbuscula*
JR10-15	N8LM5S03	Usnic acid	*Cladonia arbuscula*
JR9-1	N9LM1S03	Usnic acid	*Cladonia arbuscula*

Appendix 14c (cont.) – TLC data for the lichen samples from the August collections used for the molecular data sets.

Collection #	TLC #	Lichen Compounds Present	Identification
JR9-2	N9LM2S03	Usnic acid	*Cladonia arbuscula*
JR9-3	N9LM3S03	Usnic acid	*Cladonia arbuscula*
JR9-4	N9LM4S03	Usnic acid	*Cladonia arbuscula*
JR7-21	N9LM5S03	Usnic acid	*Cladonia arbuscula*

Appendix 14d

TLC Data From The August Collections For Lichen Samples Surveyed For Fungi

Appendix 14d – TLC data from the August collections for lichen samples surveyed for fungi.

Collection #	TLC #	Lichen Compounds Present	Identification
JR4-18	N5LU1S03	Usnic acid, Barbatic acid	*Cladonia amaurocraea*
JR4-16	N7LU1S03	Usnic acid, Barbatic acid	*Cladonia amaurocraea*
JR4-17	N9LU1S03	Usnic acid, Barbatic acid	*Cladonia amaurocraea*
JR7-22	N10LM5S03	Usnic acid	*Cladonia arbuscula*
JR7-20	N5LM5S03	Usnic acid	*Cladonia arbuscula*
JR7-19	N7LM5S03	Usnic acid	*Cladonia arbuscula*
JR7-21	N9LM5S03	Usnic acid	*Cladonia arbuscula*
JR10-22	N2LM5S03	Usnic acid, Fumarprotocetraric acid	*Cladonia arbuscula*
JR7-14	N10LR5S03	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
JR7-18	N2LR5S03	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
JR7-15	N5LR5S03	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
JR7-16	N7LR5S03	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
JR7-17	N9LR5S03	Atranorin, Fumarprotocetraric acid	*Cladonia rangiferina*
JR8-1	N10LS5S03	Usnic acid, Perlatolic acid	*Cladonia stellaris*
JR8-5	N2LS5S03	Usnic acid, Perlatolic acid	*Cladonia stellaris*
JR8-3	N5LS5S03	Usnic acid, Perlatolic acid	*Cladonia stellaris*
JR8-4	N7LS5S03	Usnic acid, Perlatolic acid	*Cladonia stellaris*
JR8-2	N9LS5S03	Usnic acid, Perlatolic acid	*Cladonia stellaris*
JR4-15	N10LU1S03	Usnic acid	*Cladonia uncialis*
JR4-19	N2LU1S03	Usnic acid	*Cladonia uncialis*

Appendix 15a

Photographic Plates Of Fungi

Figs. 1-2. *Cladosporium* # 1, 1. A conidiophore with conidium attached. 2. Various Shapes of conidia.

Fig. 3. *Epicoccum purpurascens*, Cushion shaped sporodochium with short conidiophores and conidia.

Figs. 4-5. *Cladosporium* # 2, 4. A conidiophore with conidia attached. 5. Various shapes of conidia.

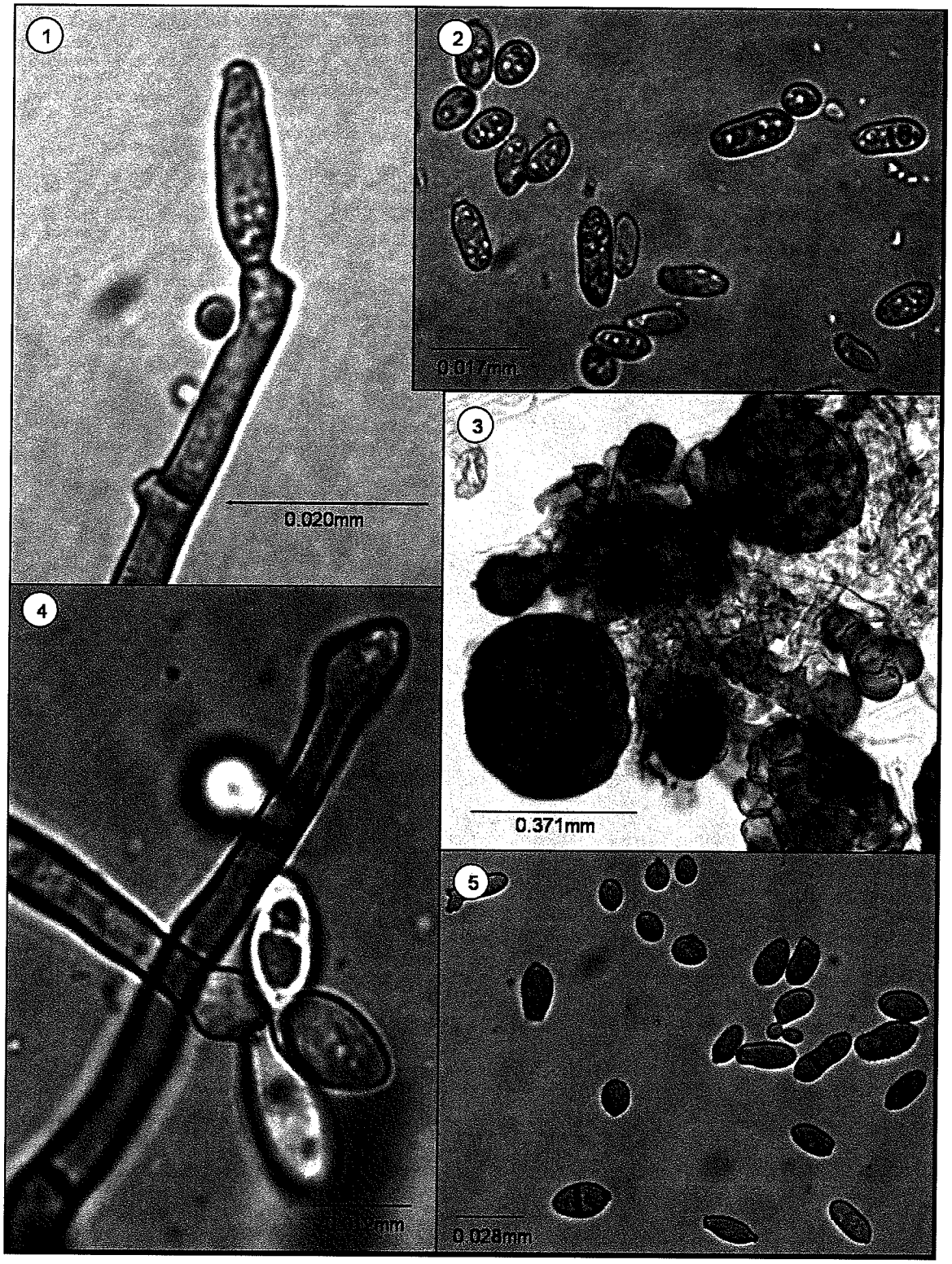

1
0.020mm
2
0.017mm
3
0.371mm
4
5
0.028mm

Fig. 6. *Aspergillus alutaceus* group, Conidiophore with phialides, and a single mature conidium.

Fig. 7. *Penicillium* sp., Conidiophore with phialides, and conidia in basipetal chains.

Fig. 8. *Trichoderma* sp., Conidiophore with phialides and conidia.

Fig. 9. *Cunninghamella elegans*, Sporangiophore with sporangioles attached.

Fig. 10. *Alternaria* sp., Conidiophore with attached conidium.

Fig. 11. *Absidia coerulea*, Sporangiophore with sporangium and sporangiophore with exposed collumella and sporangiospores.

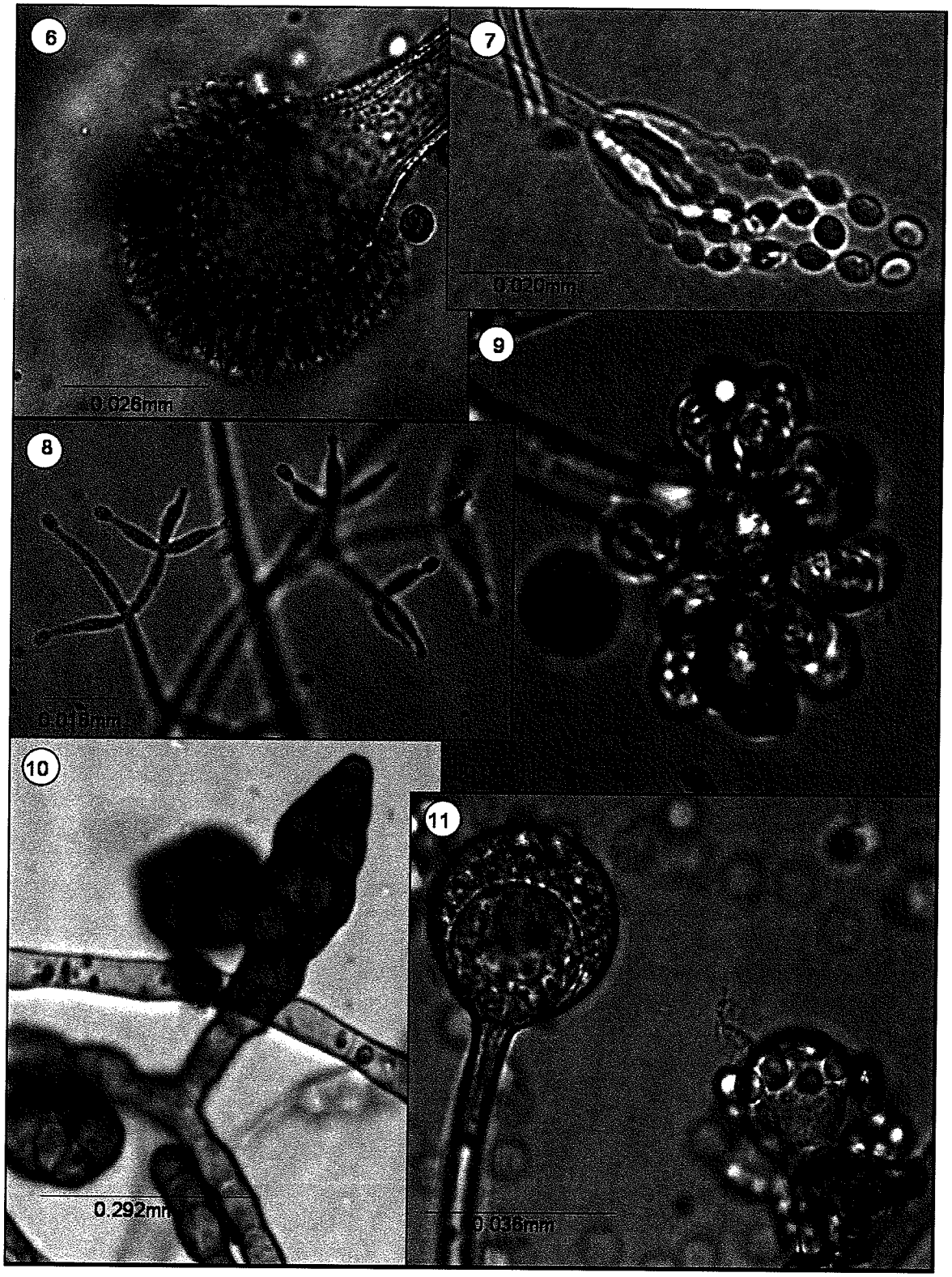

6
0.026mm
7
0.020mm
9
8
0.018mm
10
0.292mm
11
0.036mm

Fig. 12. *Mucor spinosus*, Sporangiophore with ruptured sporangium and sporangiospores.

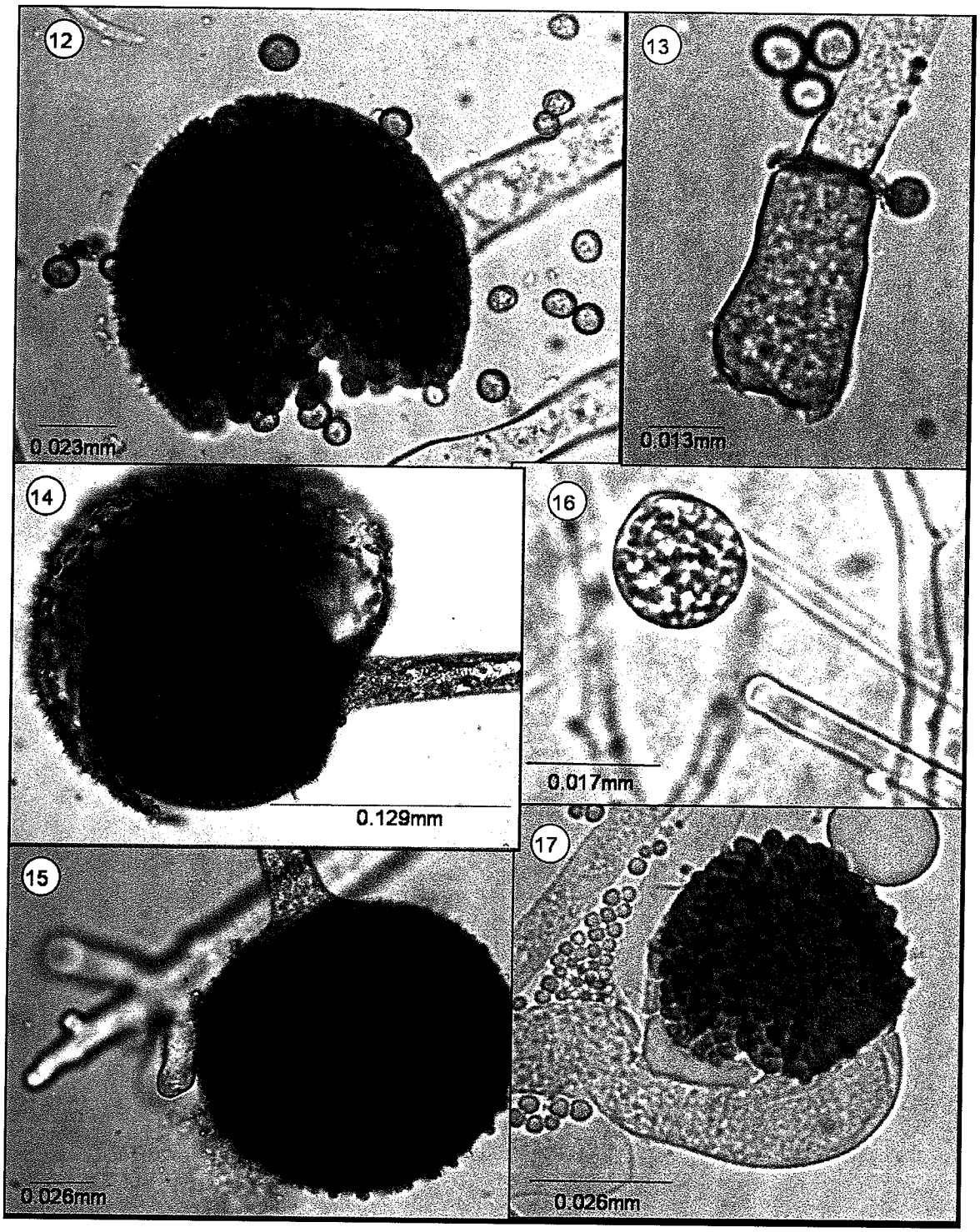
Fig. 13. *Mucor spinosus*, Sporangiospore with exposed spiny collumella, and sporangiospores.

Fig. 14. *Rhizopus oryzae*, Sporangiophore with torn sporangium wall, exposing the collumella.

Fig. 15. *Rhizopus oryzae*, Sporangiophore with sporangium, and sporangiophore rhizoids.

Fig. 16. *Mortierella isabellina/vinacea* complex, Sporangiophore with attached sporangium and a second sporangiophore revealing the absence of a collumella.

Fig. 17. *Zygohynchus exponens*, Zygosporangium with inflated suspensor appendage and sporangiospores.


12
0.023mm
13
0.013mm
14
0.129mm
16
0.017mm
15
0.026mm
17
0.026mm