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A Comparison of Environmental Substrate Gradients and Calcium Selectivity in Plant Species of Calcareous Fens in Massachusetts

Jamie M. Morgan
University of Massachusetts Amherst

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A COMPARISON OF ENVIRONMENTAL SUBSTRATE GRADIENTS AND CALCIUM SELECTIVITY IN PLANT SPECIES OF CALCAREOUS FENS IN MASSACHUSETTS

A Thesis Presented

by

JAMIE MARIE MORGAN

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Department of Plant, Soil, and Insect Sciences

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Approved as to style and content by:

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Peter L. M. Veneman, Chair

____
Lynn Adler, Member

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Allen V. Barker, Member

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Deborah J. Picking, Member

____
Peter L. M. Veneman, Department Head
Department of Plant, Soil, and Insect Sciences

DEDICATION

To my parents, Karen-Marie and Arthur Morgan, and to my grandpa, Trygve Stange, for their endless encouragement and support, and for teaching me the value of hard work.

“Wherever the reign of nature is not disturbed by human interference, the different plant-species join together in communities, each of which has a characteristic form, and constitutes a feature in the landscape of which it is a part. These communities are distributed and grouped together in a great variety of ways, and, like the lines on a man’s face, they give a particular impress to the land where they grow. The species of which a community is composed may belong to the most widely different natural groups of plants. The reason for their living together does not lie in their being of common origin, but in the nature of the habitat. They are forced into companionship not by any affinity to one another but by the fact that their vital necessities are the same… A knowledge of the communities which exist within the realm of plants is of great importance in many ways. It throws a strong light, not only on the mutual relations of the different species which are associated by common or similar needs, but also on the connection of plant-life with local and climatic conditions and with the nature of the soil. It may fairly be said that in the various zones and regions of our earth no kind of phenomenon so thoroughly gives expression to the climate and the constitution of the soil as the presence of particular plant-communities which prevail, and, accordingly, the determination and description of such communities constitutes an important part of geography.”

- Anton Kerner Von Marilaun, from *The Natural History of Plants*, 1895

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Finally, I would like to thank my professors at Rutgers: Dr. Roger Locandro for introducing me to “limestone species” and Dr. Colleen Hatfield for her continuous support and encouragement.

Thank You!

ABSTRACT

A COMPARISON OF ENVIRONMENTAL SUBSTRATE GRADIENTS AND CALCIUM SELECTIVITY IN PLANT SPECIES OF CALCAREOUS FENS OF MASSACHUSETTS

May 2008

JAMIE MARIE MORGAN, B.A., RUTGERS UNIVERSITY
M.S., UNIVERSITY OF MASSACHUSETTS AMHERST

Directed by: Dr. Deborah J. Picking

The distribution and occurrence of plant species within a given region provides insight into the many environmental properties of that region. Although much research has been conducted on plant communities and associated environmental properties, few studies have been conducted on the characteristics of individual plants within those communities. Calcareous fens are wetlands formed by the upwelling of mineral-rich groundwater and often are associated with many unique plant communities and rare species of flora and fauna. Although many studies have documented the vegetation patterns and associated environmental gradients of these fens, none have isolated the specific hydrogeochemical conditions associated with individual species, nor have any studies attempted to document and compare the individual physiological response of species to elevated environmental calcium levels. This research was conducted to estimate environmental calcium requirements for rare as well as common indicator species of calcareous fens of Massachusetts and to examine the relationship between the accumulation of calcium in the tissues of these species to calcium availability in their

environment. These factors will be important when determining required conditions for fen restoration and will further the understanding of why these species often only occur in calcareous fens.

Eight calcareous fen study sites at three different locations were established where calciphiles occur in western Massachusetts. In each site, data were collected on the vegetation patterns and associated soil chemistry, water chemistry, and hydrology. In addition, plant tissues were collected and analyzed for calcium. Species distributions were evaluated as to whether they increased in abundance as environmental calcium did or whether they appeared to occur only once a specific calcium threshold was met. In addition, the concentrations of calcium in the tissues were used to determine the extent to which those plants accumulated calcium and how those levels related to levels of calcium in the substrate environment and to their overall distributions.

It was found that certain calciphiles are calcium specialists, i.e. they are more abundant when environmental calcium levels are elevated, absorb greater quantities of calcium and those quantities correlate to the available environmental supply. These species include *Parnassia glauca, Packera aurea, Geum rivale* and *Carex granularis*. Of these, *Geum rivale* and *Carex granularis,* as well as *Carex sterilis,* did not occur below calcium concentrations of 48 $mg \cdot L^{-1}$. However, other calciphiles are calcium generalists, i.e. they are tolerant of elevated calcium levels but show no other relationship with respect to growth or accumulation. These species include *Carex flava*, *Carex hystericina*, *Juncus nodosus, Solidago patula, Solidago uliginosa,* and *Symphyotrichum puniceum.* In addition, some wetland generalists maintain elevated calcium levels *(Symplocarpus foetidus* and *Mentha arvensis*) whereas most others do not (*Thelypteris palustris* and

Fragaria vesca). Of the calciphile and wetland generalist species, some appear to increase in abundance in calcareous fens in relation to increases in accessory benefits (*Dasiphora fruticosa* and *Juncus brachycephalus* with pH; *Thelypteris palustris* and *Carex flava* with magnesium and possibly *Equisetum fluviatile* with iron). Combined, these findings characterize the growth habits and calcium accumulation of species that grow in calcareous fens and indicate that calciphiles have varying degrees of dependence on calcium.

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CHAPTER I

INTRODUCTION

The distribution and occurrence of plant species within a given community provides insight into the many environmental properties of the climate to which that community is exposed, (i.e., precipitation, temperature, humidity, wind, etc.) as well as to the soil environment where it occurs (i.e. pH, available nutrients, water, disturbance, etc.). Plant communities and individual species occurrences have been documented around the world for hundreds of years, and especially in the last sixty years, numerous efforts have been made to associate those occurrences with specific environmental properties. However, the characteristics of individual plant species that either reflect or influence their environmental habitat selections within those communities are less studied.

Calcareous fens are wetlands that harbor many rare plants and uncommon plant communities, in addition to being core habitat for the federally threatened Bog Turtle (*Glyptemys muhlenbergii*). These associations are due to the unique hydrogeochemical environment caused by the upwelling of mineral-rich groundwater that has passed through bedrock or parent material rich in calcium carbonate, relative to wetlands underlain by other strata. The resulting root-zone contains elevated calcium levels and elevated pH. Since these conditions, along with the limited occurrence of carbonate bedrock in New England, are rare, so too are the associated plant communities. In Massachusetts, calcareous fens harbor many uncommon plant species, six of which are threatened or of special concern (MNHESP, 2004).

For many years, calcareous fens have been a topic of study for their unique chemical properties, plant assemblages, rare fauna and overall ecological importance. These studies have concluded that the unique vegetative assemblages and high occurrence of rare and endangered species are attributable to the minerotrophic properties of the fen wetlands, notably elevated pH and calcium concentrations (Sjörs, 1950; Vitt et al., 1975; Slack et al., 1980; Vitt and Chee, 1990; Motzkin, 1994; Picking and Veneman, 2004). However, no research has yet attempted to isolate the specific geochemical conditions associated with individual species in the fens (Godwin et al., 2002). Certain calciphiles are generalists in terms of calcium requirements, whereas others appear to occur only at distinctly elevated calcium levels (Motzkin, 1994; Picking and Veneman, 2004). It is not yet clear what threshold values are necessary for a given calciphile to occur. In addition, no studies have yet attempted to document and compare the physiological response of individual species in calcareous fens to elevated environmental calcium levels and how this response may be associated with their distributions.

Justification

This research is an effort to determine environmental calcium requirements for rare as well as common indicator species of calcareous fens in Massachusetts. In addition, it seeks to examine the physiological relationship between these plant species and environmental calcium availability by measuring the concentrations of calcium in their tissues. The extent to which these plants selectively absorb calcium and how those levels relate to levels of calcium in the substrate environment and to their overall distribution patterns is also analyzed. Early on, several European researchers compared the relationships between plant tissue calcium in species that preferred to grow on calcareous soils to those that did not (Molisch, 1918; Iljin, 1936; Kinzel, 1963) and others have measured nutrient accumulation in the tissues of wetland plants (Boyd and Hess, 1970;

Auclair, 1979). However, no studies have yet examined calcium accumulation in plant species in calcareous fens.

Knowledge of the suspected calcium requirements of these species is important for conservation efforts as it will allow for a means of identifying specific regions with suitable substrates for these rare communities that then can be prioritized for preservation or targeted for restoration. Understanding the specific limitations for these rare plant species and communities may help to elucidate causes of population changes or local disappearances, whether natural or anthropogenic. Although no definitive conclusions can be made as to locations where plants are notably absent, trends in environmental properties in these regions may provide clues as to the selectivity of the absent species.

There is also a need for a more comprehensive analysis of the geochemical properties of some previously studied calcareous fens. Picking (2002) studied vegetation patterns in a single calcareous sloping fen in Massachusetts by analyzing pore-water and soil chemistry in the plant root-zone. Other researchers studying calcareous fens in North America (Slack et al., 1980; Vitt and Chee, 1990; Motzkin, 1994) have measured surface water chemistry that may not have been reflective of actual pore-water conditions due to the unavoidable "degassing" of the CO_2. This phenomena would have resulted in lower calcium values and higher pH compared to measurements at below-surface depths (Schot and Wassen, 1993). In addition, several researchers have measured the water chemistry but neglected the chemistry of the soil (Vitt et al., 1975; Slack et al., 1980; Vitt and Chee, 1990; Motzkin, 1994; Godwin et al., 2002). Conversely, some have measured the soil chemistry but neglected the water chemistry (Nekola, 2004; Bowles et al., 2005). To analyze the soil environment from the perspective of a plant, it will be important to monitor, as Picking did, the soil chemistry as an indicator of the reserve nutrient pool and the pore-water chemistry (not surface water) to determine the readily available nutrients.

This study is the first of its kind to compare individual plant-tissue calcium levels to substrate calcium levels in calcareous fens and to analyze this relationship in terms of the distribution of plants in relation to substrate calcium. In this context, it is the first to examine the water *and* soil chemical environments for multiple calcareous fens in Massachusetts, including some fen systems that have not been studied previously.

Hypothesis

The hypothesis of this study is that calciphiles select habitats in calcareous fens that have significantly elevated calcium levels and that this trend can be evidenced in how those plants selectively absorb calcium into their tissues. Thus:

- Specific environmental substrate calcium levels can be measured, and for the most selective species, a minimum *threshold* and maximum *range* can be established.
- The most selective calciphile species (calcium specialists) that grow in calcareous fens will have the highest calcium concentrations in their tissues, and these levels will correspond to environmental substrate calcium levels.
- The least selective calciphiles (calcium generalists) will maintain lower tissue calcium levels (than the specialists), and these levels are independent of environmental substrate levels.

Objectives

1) A list of target species was compiled from previous literature reports of vegetation in calcareous fens of Massachusetts. These species were grouped by their

suspected selectiveness to environmental calcium levels (generalists vs. specialists).

2) Field locations were chosen where species from the compiled list naturally occurred. Vegetation was surveyed in each study location, and plant tissues were collected and analyzed for calcium accumulation for selected species.
3) Soil and soil-water samples were analyzed for calcium and other key element concentrations and pH throughout the growing season to determine the nutritional conditions to which the plants are exposed.
4) Hydrologic characteristics of the wetland systems were monitored throughout the growing season and physical properties of the soils were described to be able to identify when environmental properties other than substrate calcium levels may be limiting or controlling plant distributions.
5) Suspected ranges and thresholds of substrate calcium were established for selected species, and these data were compared to the levels of calcium in the plant tissues.

Study Site Layout

Previous studies (Motzkin, 1994; Kearsley, 1999; Picking, 2002) in the Berkshire-Taconic region of New York and Massachusetts have established that many of the wetland communities occurring there are calcareous. The principal bedrock underlying the surficial deposits are various forms of calcitic limestone ($CaCO_3$) and dolomitic limestone ($CaMg(CO_3)_2$), known as the Stockbridge Formation (Appendix A; Zen and Hartshorn, 1966; Zen and Ratcliffe, 1971). The Nature Conservancy oversees many of these properties and was contacted to help identify geographic locations known to contain calcareous wetlands with the desired suite of plant species. Three geographic locations were identified within the Housatonic Watershed (Figure 1.1). In March 2006, eight research sites were established across the three geographic areas where calcareous wetland conditions existed and vegetation was dominantly herbaceous, graminoid, or low shrub cover. Different study sites within one geographic region were chosen by selecting areas within the region where distinct differences were observed in the vegetative

Figure 1.1. Field site geographic locations with watershed and sub-drainage basin boundaries.

community and the hydrologic input, with sufficient area for sampling to occur in triplicate. By mid-April 2006, three replicate plots were chosen in each of the selected sites where data collection took place. The plots were set up approximately 10 m from each other. Plots were arranged so that, when possible, they existed in a triangular arrangement. In some instances the small size of the open fen area did not allow for this arrangement, and thus plots were arranged so that they were linear and followed the contour of the wetland hydrologic gradient.

Two study sites were established at location Jug End; four were established at location Schenob Brook (including one at nearby Bartholomew); and two sites were established at location Shmulsky (Figure 1.1). Photographs of sites are available in Appendix B.

Hydrogeochemical Setting

The Jug End geographic location (Figure 1.2) lies much farther north than the other regions (which cluster around the Schenob Brook). It is surrounded by residential structures to the north and west and agriculture to the south. A long, protected wetland region lies to the east. This region occurs at the base of a hill where water seeps into the lowland. The hill area contains 15-35% calcitic limestone outcrops and has low permeability. The substratum of the fen complex is thin till and has some limestone outcrops (Scanu, 1988). The hill area is comprised of calcitic marble with interbeds of dolomitic limestone, and the bedrock underlying the fen complex is a mix of various forms of calcitic and dolomitic limestone (Zen and Ratcliffe, 1971) (Figure 1.3). The Jug End North site (JN), which lies in the northwestern portion of the fen, is situated at the location where water seeps out from the hill. The other site, Jug End South (JS), lies in the southwestern region of the fen.

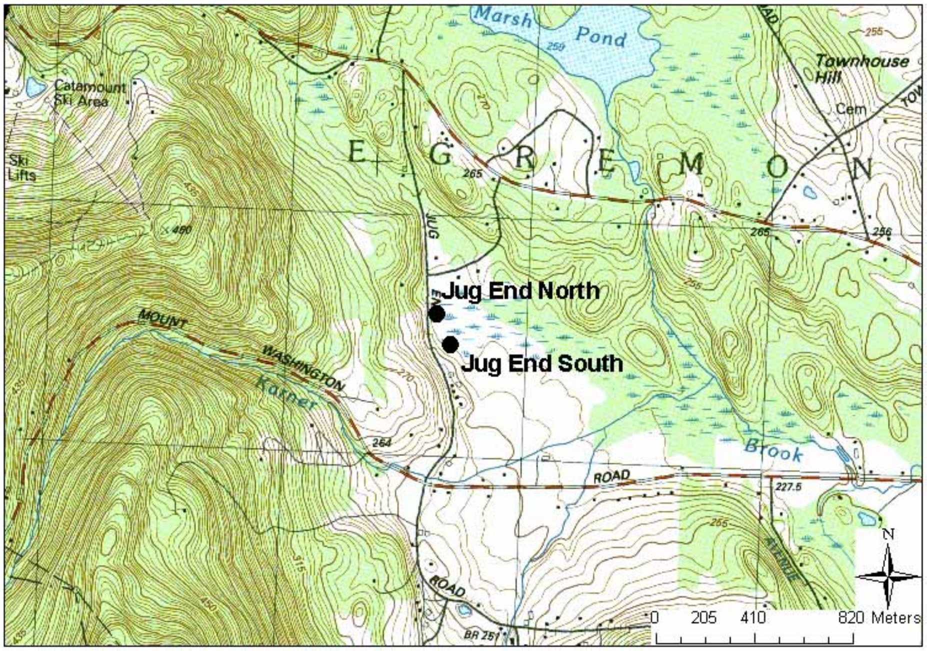


Figure 1.2: Detail of Jug End site locations and topography.

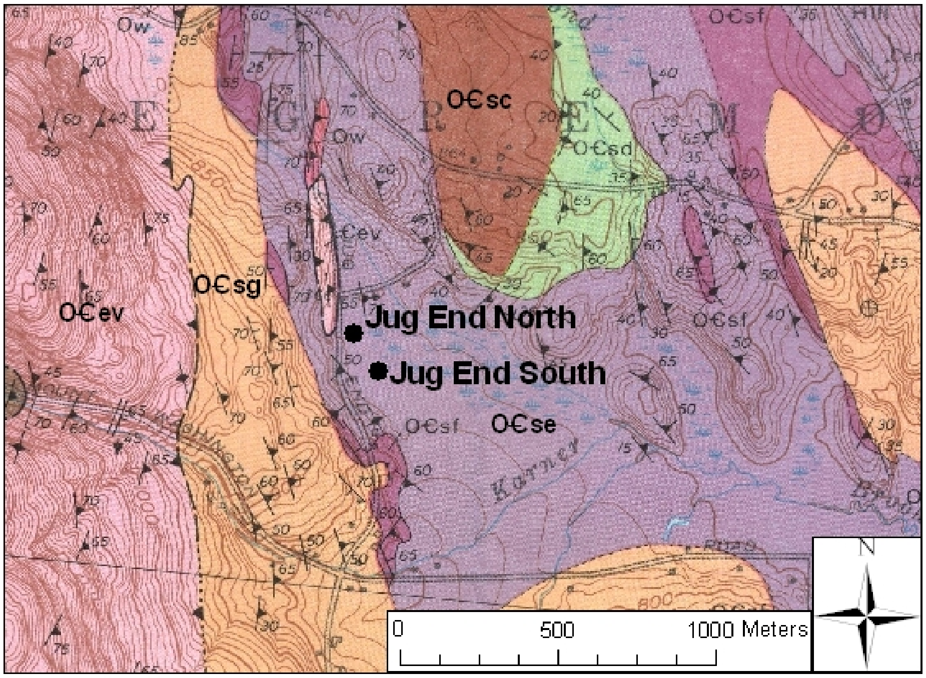


Figure 1.3: Bedrock geology surrounding the Jug End sites. Dominant bedrock types are as follows: OЄev – Quartzose argillite; OЄsg – massive, mottled marble with interbeds of dolostone; OЄse – calcite marble with dolomite masses; OЄsc – massive to sheared, white dolostone; OЄsd – impure limestone, fine-grained dolostone, calcareous siltstone and sandstone; OЄsf – calcareous sandstone, fine-grain limestone, calcareous quartzite, massive dolostone.

The Schenob Brook geographic location (Figure 1.4) is just north of the confluence of the smaller Dry Brook and Schenob Brook. Two of the sites, Schenob Brook Central (SBC) and Schenob Brook North (SBN), occur where a long kame terrace slopes steeply to meet with water-sorted outwash deposits (Zen and Hartshorn, 1966) in a lowland area. At this location, water flows directly from these deposits and into the lowland, creating the wetland fen areas and forming rivulets that separate the vegetation-bearing hummocks. This feature is particularly pronounced at Schenob Brook Central, were the rivulets are comprised of flowing water from the seep areas. The substrates of these two sites are gravelly to sandy glacio-fluvial materials derived from slate, shale, sandstone, limestone, and small amounts of granitic gneiss (Scanu, 1988). These two sites are bordered by the protected fen complex to the south and east and are bordered by open fields and few residential structures to the north and west. The third site, Schenob Brook South (SBS), occurs in a similar landscape position as the other Schenob sites but somewhat closer toward the Dry Brook-Schenob Brook floodplain region. Thus, the substratum is comprised of silty alluvial deposits. This site is surrounded immediately by forest and wetland, but like the other Schenob Brook sites, it has the same agricultural and residential land use 50 m to the west. The principal bedrock surrounding these three Schenob Brook sites is chiefly calcareous in nature, mostly that of dolomitic limestone (Figure 1.5; Zen and Hartshorn, 1966). However, on the ridge areas, the bedrock is comprised of schist or phyllite with small calcareous pockets.

The Bartholomew site (SBB) lies in the Schenob Brook geographic location (Figure 1.4) and is near the Dry Brook, farther downstream from the other Schenob Brook sites. The geomorphology is similar to the Schenob sites (situated at the base of a kame terrace

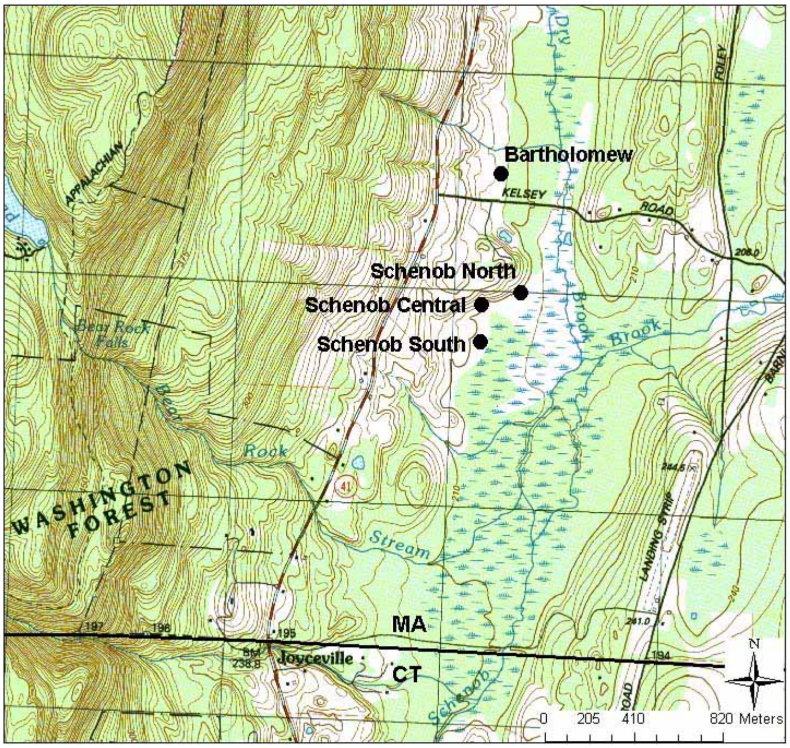


Figure 1.4: Detail of Schenob Brook site locations and topography.

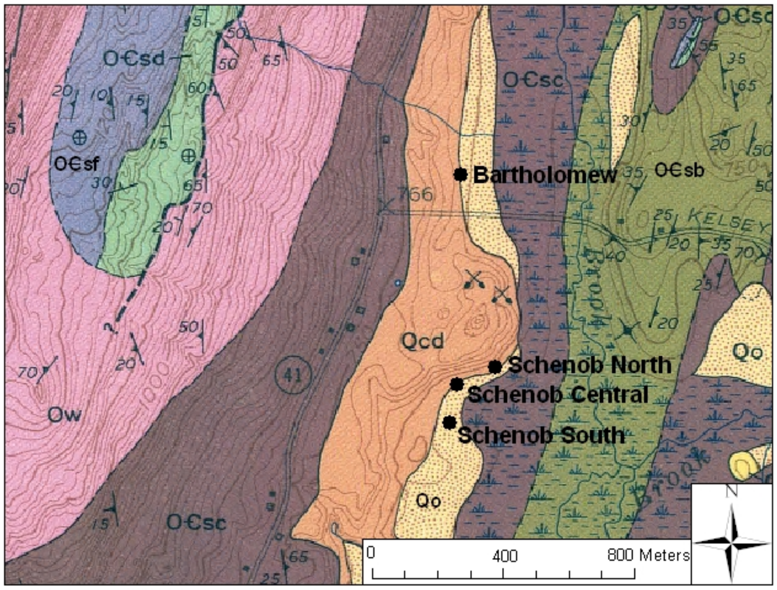


Figure 1.5: Bedrock geology surrounding the Schenob Brook sites. Dominant bedrock types are as follows: Ow – Schist or phyllite, locally calcareous, micaceous or quartzose; OꞒsf – calcareous sandstone, fine-grain limestone, calcareous quartzite, massive dolostone; OꞒsd – impure limestone, fine-grained dolostone, calcareous siltstone and sandstone; OꞒsc – massive to sheared, white dolostone; OꞒsb – uniform, massive dolostone; Qcd – water-laid ice-contact deposits; Qo – outwash.

where water seeps into outwash deposits; Figure 1.5), but the substratum is formed from calcareous, loamy till and contains 10 to 35 percent coarse fragments (Scanu, 1988). As were the Schenob sites, Bartholomew is surrounded by dolomitic limestone (Figure 1.5; Zen and Hartshorn, 1966); however, it is situated much more closely to a pocket of calcitic marble and limestone. The site is bordered to the east by forest and floodplain wetlands and to the west by fields used for vegetable production.

The Shmulsky geographic region (Figure 1.6) occurs several miles downstream and northeast of the Schenob Brook geographic location, draining into a lower reach of the Schenob Brook watershed. Sandy glacio-fluvial deposits form the substratum (Scanu, 1988) in this lowland region that occurs slightly up gradient from the brook and down gradient from a bedrock of schistose marble from the Walloomsac formation and calcareous sandstone, and calcitic and dolomitic limestone from the Stockbridge formation (Figure 1.7; Zen and Hartshorn, 1966). The region is surrounded mainly by wet meadows, forest, and a few residential structures. The site Shmulsky Open (SO) is an open, graminoid-herbaceous fen meadow. The other site, Shmulsky Shrub (SS), is about 100 m north of SO and is characterized by low shrub cover in combination with graminoid and herbaceous vegetation.

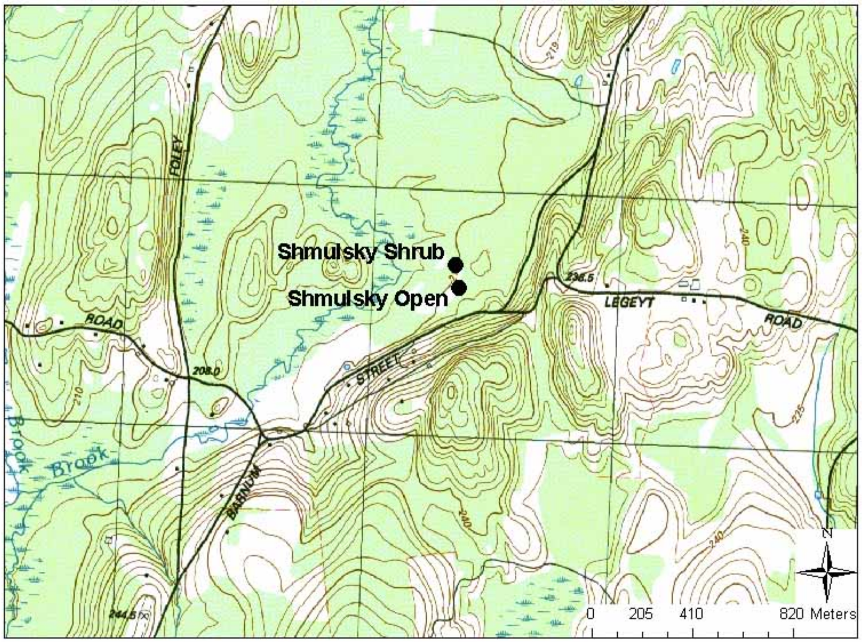


Figure 1.6: Detail of Shmulsky site locations and topography.

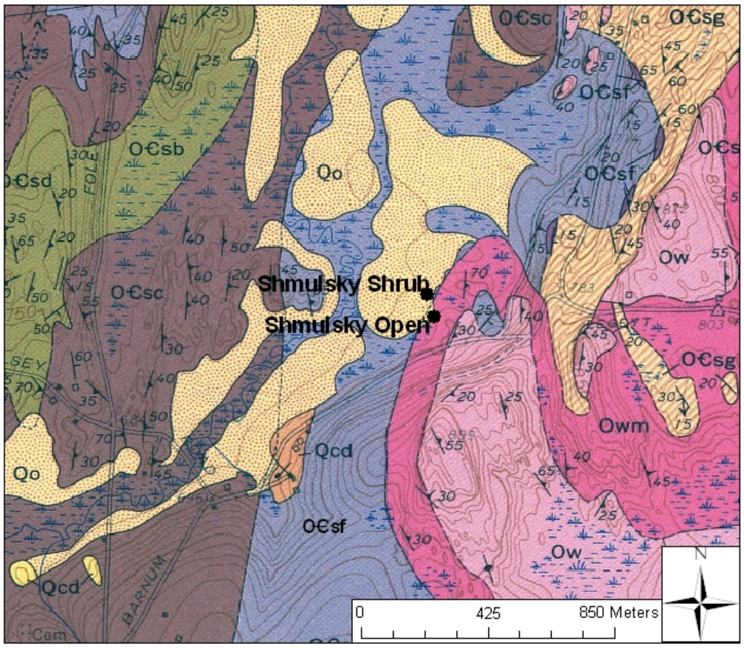


Figure 1.7: Bedrock geology surrounding the Shmulsky sites. Dominant bedrock types are as follows: Owm – Schistose marble mottled by phyllitic masses; Ow – Schist or phyllite, locally calcareous, micaceous or quartzose; OЄsg – massive, mottled marble with interbeds of dolostone; OЄsf – calcareous sandstone, fine-grain limestone, calcareous quartzite, massive dolostone; OЄsc – massive to sheared, white dolostone; OЄsb – uniform, massive dolostone; Qcd – water-laid ice-contact deposits; Qo – outwash.

CHAPTER II

LITERATURE REVIEW

Wetland Hydrology

A wetland is defined as a natural landscape characterized by saturated surface soil and/or shallow standing water, in which unique soil morphological features develop (gleization and organic matter accumulation) and where the vegetation is specifically adapted to such conditions (Mitsch and Gosselink, 1993). Although all wetlands share these characteristics, great variability exists in the mechanisms that cause water to accumulate. At least sixteen different terms have been used to describe the many differences among wetlands; most commonly used are swamp, marsh, bog, and fen. Marshes are often continually inundated areas with emergent vegetation; bogs have no significant inflow and are primarily rain fed; and swamps often are forested regions produced by overflowing streams in low-lying areas or perched water tables (Mitsch and Gosselink, 1993). All of these wetlands are characterized by receiving water from the surface. By contrast, a fen is a wetland that receives water from underground sources that originate in the underlying parent materials or bedrock (Bedford and Godwin, 2003). Although these wetland titles at times are used interchangeably, a *fen* is distinctly different in that it is a groundwater fed system.

Fens are “discharge” wetlands, indicating that the hydraulic head in the wetland is lower than the hydraulic head underlying the surrounding landscape. Fen wetlands often result where groundwater inflow forms a spring or seep at the base of a steep slope or where pressurized groundwater has an outlet to the surface (artesian conditions; Brooks et al., 1997). By contrast “recharge” wetlands like bogs, swamps, and marshes have a downward hydrologic gradient that recharges the groundwater (Mitsch and Gosselink, 1993).

Fen Geochemistry

On average, the pH of wetland soils and waters tends to range from acidic to neutral as they lack a constant supply of base cations from groundwater inputs. This condition, combined with the nature of the vegetation in the wetland (and thus the level of production of organic acids) maintains the pH of most wetlands between 3.5 and 6.5. Bogs are often on the lower end of this range due to organic acid production from sphagnum mosses (*Sphagnum* spp.) (Mitsch and Gosselink, 1993).

Non-fen wetland systems are called ombrotrophic (ombro meaning "rain" and trophic meaning "nourishment") as they rely on precipitation or surface water for their sparse supply of nutrients. The resulting water chemistry reflects in situ chemical processes and rain water characteristics, not the geochemistry of the watershed. However, fens are called minerotrophic (Vitt and Chee, 1990) in that their water chemistry is related directly to the local bedrock and parent material. Fen pH may range from 3.5 (similar to bog environments) to 8.4 (Bedford and Godwin, 2003) depending on the mineralogy of the substratum surrounding it.

In an attempt to qualify the differences in fen pH, terminology has been developed to classify these characteristics. An acidic fen (pH 3.5-5.5) is called a "poor fen"; a circumneutral fen (pH 5.5-7.4) is called a "rich fen"; and when a fen is strongly alkaline (pH 7.5-8.4) it is called an "extreme rich fen" or a "marl fen" (Bedford and Godwin, 2003). One type of rich fen called a "calcareous fen" occurs when the hydrologic input for the fen is supplied by water that has traveled through calcareous bedrock such as calcitic and dolomitic limestone, or metamorphosed forms of either of the two. Many studies have established the water pH range in calcareous fens to be between 6.0 and 8.1 (Slack et al., 1980; Komor, 1994; Motzkin, 1994; Picking and Veneman, 2004), and that fens are characterized as having unusually high levels of dissolved calcium in the water and adsorbed to the soil.

Vitt et al. (1975) concluded that dissolved calcium values greater than 5 $\text{mg}\cdot\text{L}^{-1}$ were characteristic of rich fens; however, other studies have reported higher threshold values. Slack et al. (1980) reported dissolved calcium to range from 18 to 37 $\text{mg}\cdot\text{L}^{-1}$, whereas Motzkin's (1994) rich fen values approached 65 $\text{mg}\cdot\text{L}^{-1}$. The highest values were detected by Komor (1994) who measured values up to 128 $\text{mg}\cdot\text{L}^{-1}$ and by Picking (2002) who reported comparable water calcium values approaching 180 $\text{mg}\cdot\text{L}^{-1}$ and soil calcium values ranging from 2126 $\text{g}\cdot\text{m}^{-3}$ to 4907 $\text{g}\cdot\text{m}^{-3}$.

Calcareous bedrock has a limited distribution in the United States (National Atlas, 2006). It has scattered occurrences in the western United States with most notable areas in northwestern Arizona, south-central New Mexico, and central Texas. In the Upper Midwest it occurs significantly in Iowa, Missouri, and Wisconsin and moderately in Michigan, Indiana, Illinois, and Ohio. Although abundant in Florida and southern Georgia, it has scattered occurrences in the southeastern states of Kentucky and Tennessee. In the Mid-Atlantic and New England, it only occurs in thin bands. For this reason, calcareous fens occur infrequently and exist only as small communities, especially in Massachusetts.

Fen Plant Communities

Eggers and Reed (1987) referred to plant communities associated with calcareous fens as the rarest in North America. As may be expected, many of the plant species that occur in these habitats are adapted specifically to the unique geochemical conditions present, and thus the plant assemblages are unique and often harbor many rare and endangered species. Bowles et al. (2005) reported that the vegetative communities in a 23-hectare prairie fen in Illinois were correlated significantly with soil pH, sodium, magnesium, and calcium concentrations. Motzkin (1994), in a study of western New England fens, established species groupings that have been since used and validated by other researchers (Kearsley, 1999; Picking and Veneman, 2004) to describe the vegetation. These species groupings were related to differences in environmental properties, with depth to the mineral soil being the most important, but also important were pH, magnesium, and calcium in the fen waters. Vitt and Chee (1990) showed a strong correlation between vegetative communities and fen pH, calcium, magnesium, and electrical conductivity in their study of water and peat chemistry of fens in Alberta, Canada. They identified distinctly different species associations between the extreme-rich fens, moderate-rich fens, and poor fens. In addition, vegetation assemblages were further separated by other hydrologic and geomorphologic aspects of the fens. For example, a “calcareous seep” community occurs where calcareous groundwater seeps off of a slope to the soil surface forming rivulets. A “lake-basin” fen community occurs where calcareous groundwater travels through peat that has accumulated in a former lake basin (most likely of glacial origin; Weatherbee, 1996).

The previous studies documented that calcareous fens have high species richness, as do many places with rare and uncommon species. Species richness has often been explained in terms of nutrient availability in wetlands. Bowden (1987) reported that most freshwater wetlands are limited by nitrogen. However, several other researchers (Richardson and Marshall, 1986; Boyer and Wheeler, 1989) proposed that phosphorus is limited under conditions with elevated levels of calcium or aluminum (as these elements bind phosphorus in insoluble forms). In agreement, Bedford et al. (1999) reported that wetlands in North America often are limited by low levels of phosphorus or by a combination of low levels of nitrogen and phosphorus (two of the primary macronutrients required by plants). Bedford et al. (1999) reported that bogs and fens had high total N:P ratios (>16) that were indicative of phosphorus limitation and that swamps and marshes had lower total N:P ratios (<14) indicative of nitrogen as the limiting nutrient. They reported that higher N:P ratios were associated with higher species richness and percent organic matter in the surface soil. Plants that thrive under nutrient enriched conditions are often larger in size and more vigorous in growth than those that require fewer nutrients (Reader, 1990). Thus when conditions are more eutrophic, the nutrient-loving species tend to dominate and species richness is lower.

Calcium in Plants

Calcium is considered a secondary macronutrient for plant survival. The fundamental role of calcium in plants is in the strengthening of the cell walls as calcium pectates, although calcium is also vital in cell nuclear division by providing elasticity to microfibers, stabilizing plant membranes by bridging phosphate and carboxylate groups in phospholipids and proteins (Marschner, 1995), and providing a counter-cation for inorganic and organic anions (White, 1998). Calcium enters plant cells through calcium permeable ion channels in their membranes (White, 2001; White and Broadley, 2003) and is restricted from entry by calcium efflux pumps (Olbe and Sommarin, 1991). Soil solution concentrations of calcium are usually so great that the potential energy required to carry the ions into the root is exceeded. Thus, the electromotive force across the cell membranes facilitates the absorption instead of metabolic energy from ATP. For these reasons, calcium absorption is referred to as passive (Marschner, 1995) but this does not mean that calcium ions flow into the plant with the flow of water (Palta and Stadelmann, 1980). Nevertheless, excess calcium does accumulate in plant tissue and is often precipitated in the vacuoles and apoplasm as calcium oxalate and calcium carbonate (Marschner, 1995).

Although all plants require calcium, some are able to tolerate higher levels more so than others. From an *ecological* standpoint, these plants are called calciphiles, calcicoles, or calciphytes (literally meaning that they are “calcium loving”) (Salisbury, 1920; Eggers and Reed, 1987; White and Broadley, 2003) as they often prefer to grow in environments with elevated calcium levels (Kinzel, 1983). By contrast, plants called calcifuges (or acidophiles) avoid calcareous environments (Kinzel, 1983) and may be poisoned by excess calcium (Kinzel, 1983; White and Broadley, 2003). Thus these ecological definitions of calciphiles and calcifuges are based on the external concentration of calcium. These same terms often have different meanings when referring to the internal concentrations of calcium. Thus a *physiological* calciphile (or also a “calciotrophic” species) is a species that has appreciable amounts of water soluble calcium in its cytoplasm, usually paired with malate. By contrast, a physiological calcifuge (or also a “calciophobic” species) is a species that contains high amounts of oxalate, which facilitates the precipitation of calcium that enters (Kinzel, 1983).

Both physiotypes occur on calcareous soils. True calciotrophic species (physiological calciphiles) are less common as wild plants in North America (compared to the calciophobic species) since they mostly occur in the Crassulaceae, Brassicaeae, and Fabaceae families. Many of the calciphiles that are also calciophobic have a greater capacity to store calcium due to elevated levels of calcium-binding proteins in their cytoplasm (Le Gales et al., 1980). This ability may make them better competitors under calcareous conditions and thus receive the “calciphile” designation. By contrast, calcium toxicity is often a result of a plants’ inability to compartmentalize the calcium or inactivate it physiologically (i.e. producing calcium oxalate, calcium carbonate, etc.) (Marschner, 1995) and this may result in the habitat selections of calcifuges.

Early research on calcium in plants reported that excess calcium had no toxic effect and that toxicity symptoms were due to increased levels of associated anions e.g., Cl^- and NO_3^- (Gauch, 1972). However, more recent studies have concluded that excess calcium in the cytoplasm does prevent stomatal opening (Atkinson, 1991; Ruiz et al., 1993), photosynthetic ability (Portis et al., 1977), and water-use efficiency (Da Silva et al., 1994). Detrimental levels of calcium may exist within a plant if its adaptations to calcium absorption are more efficient in a non-calcareous habitat. For example, if a plants’ absorption of calcium is adapted to calcium-poor conditions (such as is a calcifuge) it may have an overexpression of calcium transporters that act to maximize calcium absorption. When attempting to grow under extremely elevated calcium conditions, it consequently accumulates more calcium than it can effectively tolerate. By contrast, calciphiles are believed to have adaptations that allow them to minimize calcium influx and maximize calcium efflux. When attempting to grow under lower calcium levels however, those calcium restrictive mechanisms are too efficient and do not allow enough calcium for normal growth (Lee, 1999; White and Broadley, 2003).

Fen Plant Nutrition

Although calcium and magnesium are certainly not limited in calcareous fens, the elevated pH of these systems affects the availability of other compounds and plant nutrients. Specifically, when pH is above 7, bicarbonate is elevated, and the availabilities of iron, phosphate, cobalt, and boron are limited. In addition, elevated amounts of divalent cations (i.e. calcium and magnesium ions) may inhibit the ability for plants to absorb potassium (Ehrenberg, 1919; Lee, 1999). Thus, researchers suspect that it may be the ability to tolerate these nutrient limitations, rather than the potential benefits or toxic effects of excess calcium that ultimately result in the calciphile/calcifuge habitat distinction (Kinzel, 1983). For example, studies have shown that calciphiles have special adaptations that allow them to increase absorption of iron by 1) having the ability to promote the reduction of ferric iron to ferrous iron, 2) lower the pH around their roots, or 3) produce chelating agents (Römheld, 1987). Calciphiles also exude low-molecular weight organic acids that act as extractors of iron and phosphate (Tyler and Ström, 1995). By contrast, calcifuge species have been shown to be poisoned by bicarbonate as it inhibits the uptake of iron into the plants (Woolhouse, 1966).

Calcium Uptake

A typical angiosperm contains between 0.1% and 5% calcium on a dry weight basis (Marschner, 1995) but certain species of algae have as high as 11% (Boyd, 1966). Dicots generally have higher tissue calcium than monocots (Loneragan et al., 1968). This difference has been attributed to higher cation exchange capacity (CEC) of the roots of dicots (Broadley et al., 2003) and to the fact that dicots have a lower ability to regulate the passive influx of calcium ions (Kinzel, 1983). The accumulation of calcium in plant leaves is based on the amount of absorption of calcium by the roots and its upward translocation through the xylem. The rates of these processes are affected by the availability of calcium, transpiration rates (Kinzel, 1983), and by the CEC of the roots (Broadley et al., 2003), in which higher root CEC provides increased concentrations of calcium at the root surface (Thompson et al., 1997).

Auclair (1979) measured the macro- and micronutrient tissue concentrations of six species occurring in an emergent marsh and compared them to the soil nutrient concentrations. Auclair reported that calcium concentrations in the six emergent wetland plants varied between 0.27% and 1.8%. This study imparted important knowledge of nutrient absorption in wetland plants, but since only one marsh habitat was studied, inference of how these plants would accumulate nutrients in different geochemical environments is unknown. Boyd and Hess (1970) showed that the calcium percentage in cattail (*Typha latifolia* L.*)* varied from 0.35% to 1.62% across nineteen study sites. These results indicate that there is variability in how much calcium a single species accumulates, yet no comparisons of plant accumulation were made to environmental calcium levels to understand if cattail regulates the amount of calcium it absorbs when environmental levels differ.

In a laboratory experiment, Clarkson (1965) found that certain calciphile and calcifuge species of bent-grass (*Agrostis* spp. L.*)* had equal accumulations of calcium in their shoots and roots at low calcium levels. However, at higher environmental calcium levels, the calcifuge species did not show any change in calcium accumulation whereas the calciphiles had higher levels in their shoots. This trend suggests that some calcifuges exclude calcium from their tissues and some calciphiles exhibit continued accumulation as background levels increase. Perhaps this calcifuge lacked an efficient mechanism with which to properly compartmentalize calcium, and thus could only exist under calcium-rich conditions by excluding calcium altogether. By contrast, the calciphile perhaps had a more efficient mechanism with which to inactivate the calcium physiologically and could therefore absorb greater amounts. Thus a combination of the adaptive properties of a plant, combined with its physiological characteristics, selectively regulate the amount of calcium it absorbs.

Calcium and Plant Ecology

As early as 1865, researchers became aware of differences in competition among plants depending on the substrate where they occurred (Nägeli, 1865). Nägeli observed two species of the genus *Achillea* L. (*A. atrata* L. is a calciphile, and *A. moschata* Wulfen is widely abundant on siliceous soils). When occurring together, the species in its preferred habitat out-competed the other. However in the absence of the more well-suited species, they both did equally well on their own. To further understand the mechanisms behind this, Tansley (1917) conducted laboratory experiments in which he grew bedstraw *Galium hercynicum* Weig. (a calcifuge) and *Galium asperum* Schreb. (a calciphile) alone and together on calcareous or siliceous soils. Both species occur naturally in the British Isles, however *G. asperum* occurs mostly on calcareous soils, and *G. hercynicum* is common on siliceous soils and absent from calcareous soils. Tansley noted that when grown on calcareous soils, many of the calcifuge seedlings that germinated soon became chlorotic and died whereas the calciphiles exhibited normal growth. The shoots of the calciphiles soon suppressed the growth of the few surviving calcifuges. The reverse was true of the calciphiles on an acid substrate. Although it was not determined why these plants responded as they did, Tansley concluded that calcifuges are largely absent from calcium-enriched soils, not entirely because they are poisoned by the calcium, but because the calciphiles are much better competitors under those circumstances.

Subsequent researchers (Hope-Simpson, 1938; Rorison, 1960) later modified Tansley’s findings, suggesting that soil chemistry was the primary factor governing the abundance and growth of calciphiles and calcifuges, but that competition was of some importance. Thus, calciphiles occupy a certain niche that is a combination of their tolerance to potential geochemical toxicities and limitations and their competitive ability under those conditions.

Habitat Conservation

The unique properties of calcareous fens have resulted in their receiving much attention from scientists and conservationists. As a result, many of the properties on which they occur are owned by various government and conservation organizations such as The Nature Conservancy. The mission of The Nature Conservancy is “to preserve the plants, animals and natural communities that represent the diversity of life on Earth by protecting the lands and waters they need to survive” (www.nature.org). Active projects associated with fen conservation from the Berkshire-Taconic chapter of The Nature Conservancy have involved the management of non-native and invasive plants as well as the application of prescribed burns. Purple loosestrife (*Lythrum salicaria* L.), and common reed (*Phragmites australis* Trin. ex Steud.) are two common species considered invasive in calcareous wetlands. They are given this title as they are often able to outcompete the native species since they are not indigenous to the area and thus have fewer adaptive pressures that limit their growth. Some varieties of common reed are indigenous but inbreeding with more vigorous European varieties has resulted in the current invasive nature of this species (Saltonstall, 2002). Without proper removal and treatment, opportunities for the rare native plants will be limited.

Although not fully understood, it has been suspected that recurring fire has been a major component in keeping calcareous wetlands open instead of eventually growing into forested wetlands. Although some fen communities have had similar vegetation for several thousand years, pollen records indicate that the open graminoid fen vegetation of the Schenob Brook location has only been quite similar to the present since European settlers arrived in the area 300 years ago (Shaw, 2003). The suppression of natural forest fires by humans may pose a threat to some of these plant communities and thus The Nature Conservancy is making every effort to recreate post-European settlement conditions.

Although the aforementioned efforts are being made to protect these rare communities, still more difficult challenges face conservationists. Studies have documented significant changes in vegetation of naturally acidic Atlantic white cedar (*Chamaecyparis thyoides* BSP.) swamps when they were subjected to nutrient loading from agriculture and development (Ehrenfeld and Schneider, 1993; Zampella and Laidig, 1997). A similar, opposite trend may be true for calcareous wetlands due to acidic inputs such as acid rain.

Studies of calcareous fens in the Netherlands have shown human usage of groundwater to be a factor in reducing calcium levels in the wetlands (Grootjans et al., 1988). These studies demonstrate the fragile geochemical makeup of a wetland community and show that wetlands may be influenced by many outside sources. Determining suitable calcium ranges for individual calciphiles will be essential to understanding precise changes in plant communities due to associated changes in wetland hydrogeochemistry and may aid in restoration efforts (e.g. returning peat mined fens to their original state) where a knowledge of the precise geochemical conditions that native fen species require is essential (Cobbaert et al, 2004).

CHAPTER III

HYDROLOGY, GEOCHEMISTRY, AND SOILS

Introduction

Calcareous fens are mineral-rich wetlands created by the upwelling of groundwater through carbonate bedrock. Their occurrences are limited through a combination of spatial factors. They must lie in an area where landscape features cause groundwater to move to the surface and where that groundwater moves first through carbonate bedrock to become enriched with dissolved carbonate minerals. Due to these unique conditions, the geochemical properties of these wetlands are also unique, resulting in elevated calcium and pH in the pore waters and soils. Due to the rare occurrence of these conditions, especially in the northeastern United States were carbonate bedrock is quite limited, these wetlands provide interesting study points for understanding bedrock geochemistry as a determinant in the ion composition of surface soils.

This portion of the thesis study evaluated the hydrologic factors that exist in calcareous fens and documented the level of calcium enrichment and pH that occurred in eight study sites across the three geographic locations. The hydrology and geochemistry were compared to examine the relationship between the two. Dissolved primary macronutrients (nitrogen, phosphorus, potassium), iron, and magnesium also were measured to provide a comprehensive understanding of the broad chemical environment and to evaluate whether calcium was the primary defining element in site geochemistry differences. Together, these factors help to further the understanding of the unique geochemical properties in these wetlands and the plant communities that exist there.

Materials and Methods

Hydrologic Measurements

Instrument stations were installed in triplicate plots at each of the eight sites (spread across the three geographic locations) and were installed directly up-gradient from where vegetation was sampled later in the season. Each site was provided with a set of nested piezometers (at depths of 100 cm and 150 cm or as far down as was possible given substrate conditions) installed in a central location to measure the overall vertical groundwater gradients at each site. Each sample plot included a 5-cm diameter PVC well (screened from 0-60 cm) to measure groundwater level fluctuation and a suction lysimeter (Soil Moisture Corporation, Goleta, Ca.; 0.1 MPa air entry value) at a depth of 30 cm to collect pore water.

According to the United States Natural Resources Conservation Service, the maximum frost-free growing season for Great Barrington, Mass., is from April 10 to October 23 (NRCS, 2003). Due to accessibility restrictions posed by The Nature Conservancy (related to critical breeding periods for bog turtles that live there), monitoring began in late-April 2006 for half of the sites and did not begin until the beginning of June for the others where conditions were more critical. Instruments were monitored biweekly until October 18, 2006. A qualitative assessment of soil redoxomorphic conditions was conducted at each visit using α, α-dipyridyl to identify the presence of reduced iron in 2.5-cm soil cores extracted from the top 0 to 15 cm and 15 to 30 cm, respectively.

Water Chemical Analysis

Twice monthly, after removing any passively accumulated water from the device, fresh pore water samples were collected from the lysimeters. A vacuum was applied using a hand pump, and lysimeters were allowed to fill for 2 to 3 hours before collection. Water samples were siphoned from the lysimeters and were placed in acid-washed polyethylene bottles, which were allowed to overflow before capping to minimize any degassing of the samples. Samples were refrigerated for transport back to the laboratory where the pH was measured immediately with a Hach EC-10 portable pH meter followed by nutrient measurements. The general methodology of all water analyses is based on standard methods detailed by Clesceri et al. (1989). One sample each month was analyzed for orthophosphate (as a measure of phosphorus) by the Ascorbic Acid method (4500-P.E); ammonium by Nessler's method (4500-NH_3.C); and total iron by the Ferrover method (3500-Fe.D). These three analyses were carried out with prepackaged reagents produced by The Hach Company (Loveland, Co., 1992) and analyzed using a Hach DR/2000 Model Spectrophotometer (Hach, 1998). Studies have shown nitrate to be almost non-detectable due to the anoxic nature of these systems (Picking, 2002) and for this reason ammonium was the nitrogen species analyzed.

After these first analyses were finished, samples were acidified (with 1µL 1.0 M HNO_3 to prevent microbial growth) for refrigerated storage and later analyzed for calcium (3500-Ca.B), magnesium (3500-Mg.B) and potassium (3500-K.B) by atomic absorption/emission spectrophotometry. All samples were analyzed for calcium and magnesium, however potassium was only analyzed once a month. Lanthanum was added as an ionic suppressant for the calcium and magnesium samples and likewise cesium was added to samples analyzed for potassium.

Soil Analysis

Soil samples were collected at each plot during the vegetation survey period in July 2006. Soil profile descriptions were recorded with standard terminology (Soil Survey Staff, 1993). Samples were collected from each major horizon to a depth of 60 cm. After taken to the laboratory, samples were air-dried and passed through a 2-mm sieve. Samples were characterized physically and chemically, including particle size analysis (pipette method; Gee and Bauder, 1986), bulk density (core method; Blake and Hartge, 1986), percent organic matter (loss on ignition; Dean, 1974), pH (water method; McLean, 1986), and exchangeable calcium, magnesium, and potassium (ammonium acetate method; Thomas, 1986). Extractable phosphorus and iron (ammonium bicarbonate diethylene triamine pentaacetic acid, AB-DTPA method; Soltanpour and Schwab, 1977) and total nitrogen (Dumas method; Bremner and Mulvaney, 1986) also were measured for the uppermost horizons. Early research has determined that the Dumas method overestimates the total nitrogen in organic soils as compared to the Kjeldhal method (Dyck and McKibbin, 1935; Bremmer and Shaw, 1958; Stewart, et al., 1963). However, this has now been attributed to incomplete digestion of the Kjeldhal method (Miller et al., 2007). This fact, along with increased accuracy of elemental analyzers that carry out the Dumas method (early studies were from the mid-twentieth century) have resulted in Dumas being the preferred method of analyzing total nitrogen (Watson and Galliher, 2001), including samples with large amounts of organic nitrogen (Miller et al., 2007).

Data Analysis

Statistical analyses were not applied to hydrologic measurements as these were monitored to be able to view trends in saturation and vertical groundwater gradients. A repeated measures analysis was conducted with statistical software (SAS Version 9.1.3, SAS Institute, 2004) to determine if significant seasonal differences existed among the nutrients in the water depending on the month, month by location and month by site nested within the location. The multivariate test statistic, Wilks' Lambda, was reported to indicate when differences existed. This analysis also included an analysis of variance (ANOVA) to detect significant differences in the nutrients among the different locations and sites. A multivariate analysis of variance (MANOVA) was used to see if there were overall differences in soil properties among the sites and locations and to see if significant differences existed among individual soil properties (exchangeable calcium, organic matter, etc.) among sites and locations. Tukey's Test was applied (to be conservative) to separate differences where they existed, as shown by the ANOVA.

Results and Discussion

Hydrologic Measurements

Well Data

Water table depth measurements indicated that the water tables in each wetland site varied little within each geographic location but varied greatly among geographic locations. The water table at the sites observed in the Shmulsky (Figure 3.1c) location and the Jug End (Figure 3.1a) location appeared to have seasonal differences in that the water table dropped below at least 10 cm from the surface during the warmer, drier, summer months. Of these two locations, the water table dropped less at Jug End than at Shmulsky. The water table at Jug End began to decline in mid-July and then resurfaced in early September and never went below 40 cm in depth. By contrast, at Shmulsky the water table began to drop in mid-June and reached a low 50 to 60 cm below the surface

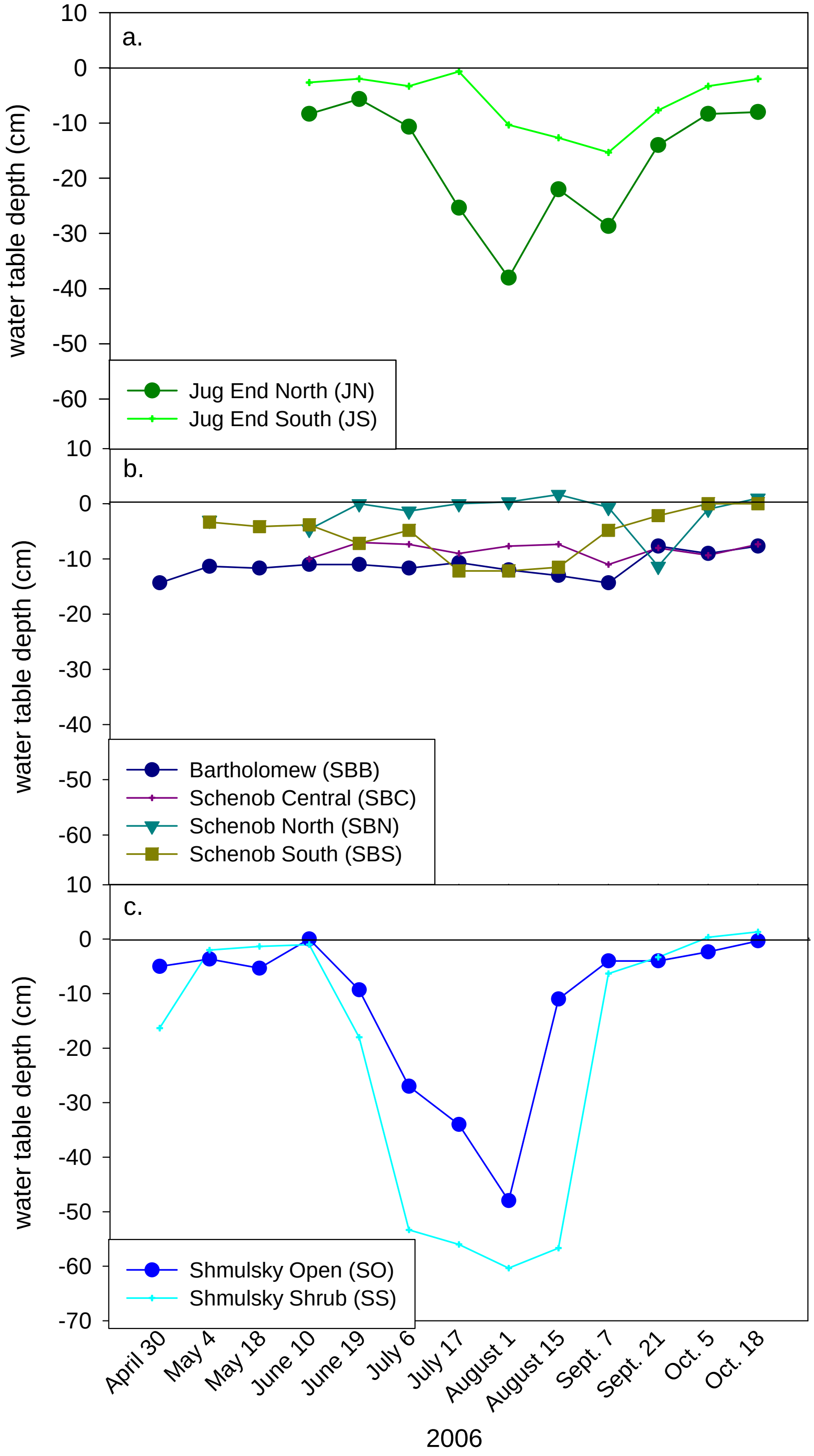


Figure 3.1. Water table depth (cm) at geographic location, a) Jug End, b) Schenob Brook, c) Shmulsky from April 30, 2006 to October 18, 2006. Each point represents a mean of three replicate measurements.

during the driest month of August before resurfacing in early September. The sites observed at the Schenob Brook location all exhibited a fairly stable water table that remained within the upper 15 cm of the surface zone for the entire growing season (Figure 3.1b).

The hydrologic patterns at Shmulsky correspond with those found by Picking (2002) in the last year of a three-year study at this same location. Picking observed a strong drop in the water table beginning in June 1998 after which the water table did not rise again until late September or early October of that year. However in 1996, a fairly steady water table was observed near the surface. In 1997, trends were similar to the following years except for a strong precipitation event that brought the water table almost to the surface in August. According to the National Oceanic and Atmospheric Administration (NOAA), precipitation amounts during the growing season of 2006 were similar to the conditions during Picking's research in 1997 and 1998 (NOAA, 2006), so it makes sense that hydrologic data would be most similar in those years.

It is interesting that in times of low precipitation, the water table at Shmulsky changed quite a bit whereas at Schenob it remained steady and at the surface all summer long. Perhaps the time that Schenob has remained in its current vegetative state (pollen records indicate similar vegetation for the last 2000 years and almost identical vegetation for the last 300 years; Shaw, 2003) is related to its resilience to hydrologic perturbations. Thus the vegetation at Shmulsky and Jug End (to a lesser degree) may be more susceptible to climatic and anthropogenic alterations as its water source is affected easily by overall hydrologic perturbations.

Piezometer Data

Wide variability was observed in the vertical groundwater gradients at the study sites. Contrary to the water table data, trends in groundwater gradients did not appear to be related to the geographic location in which they occurred. At the Jug End location (Figure 3.2a), Jug End South exhibited marginal recharging conditions throughout the entire growing season, whereas Jug End North exhibited a fairly irregular combination of extreme recharge, discharge, and “flow-through”. Flow-through indicates that there is neither discharge nor recharge but that water is moving laterally.

At the Schenob location (Figure 3.2b), Schenob Brook North had fairly constant discharging conditions throughout the growing season. Schenob Brook Bartholomew and Schenob Brook South followed similar trends in that they both exhibited a combination of marginal recharge and discharge (except in early September when they both underwent an extreme drawdown event). Schenob Brook Central had recharging conditions throughout the growing season.

At the Shmulsky location (Figure 3.2c), the site Shmulsky Open exhibited discharging water conditions for all of the growing season except for the dry hot, month of July. At the other Shmulsky site (Shmulsky Shrub), there appeared to be weak recharging (Figure 3.2c) conditions or flow-through (except for August, where the site was strongly recharging).

The results of the piezometric gradients indicate that even though the general hydrologic regimes dictate the water table depth and saturation of the soils, there is variability among sites in the same fen locations as to the direct source of the water. Thus within the same fen area, one site may be in direct contact with groundwater where it upwells but then perhaps a nearby site may receive that same water only after it has traveled on a slope through the soil (flow-through). These distinctions are very important in relation to how the chemical properties of the soil may differ.

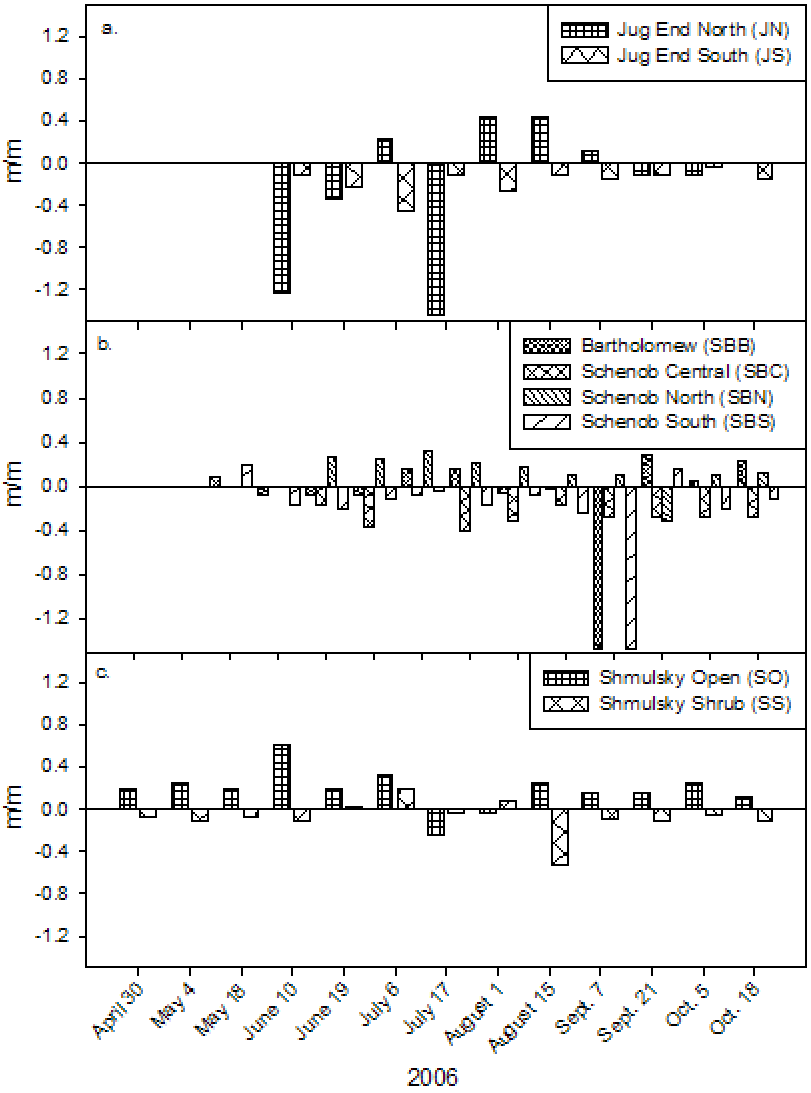


Figure 3.2. Vertical groundwater gradients at locations a) Jug End, b) Schenob Brook, c) Shmulsky. Values greater than zero represent discharge conditions; values less than zero represent recharge conditions. Each bar represents one pair of piezometers at a central location in each site.

Redoximorphic Conditions

Qualitative reduced iron measurements were assessed by allotting degrees of reduced iron to degrees of color change when α, α-dipyridyl was added to a soil sample. A scale of zero to three was applied to increasing degrees of color change, which provided a quantitative "score" for the degree of reduced iron in the soil. These data are presented in Appendix C and will be referenced where they help assist in the understanding of geochemical conditions.

Water Chemical Analyses

pH

Water pH changed significantly across the growing season (Appendix D) (Wilks' Lambda = 0.0002, $F_{5,11}$=11620.3, p=<0.0001); thus the variable "month" significantly explained variation in water pH. There were no significant seasonal differences within locations (month*location interaction: Wilks' Lambda = 0.276, $F_{10,22}$=1.99, p=0.0859) or within sites (month*site interaction: Wilks' Lambda =0.127, $F_{25,42.4}$=1.26, p= 0.2508). This indicates that there were overall differences in pH across the growing season, but these seasonal differences were not a function of the locations or sites. The seasonal mean water pH was significantly different among locations ($F_{2,15}$=7.22, p =0.0064) and among sites within locations ($F_{5,15}$=11.39, p=0.0001) (Table 3.1). The pH at Shmulsky was significantly lower than at Jug End but not significantly different from Schenob. Schenob and Jug End were not different from each other. Overall water pH ranged from a mean of 6.6 at Schenob Brook South to a mean of 7.3 at Schenob Brook Central. Across the entire sampling period, these sites were consistently the lowest and highest, respectively, and the other sites fluctuated around an average pH of 7.0. Interestingly, the sites that represented extremes in pH both occurred in the same geographic location.

Table 3.1. Seasonal range of water pH and mean seasonal water pH for the eight study sites and associated locations. Means with different letters are significantly different by Tukey's Test (p=0.05). Uppercase letters represent differences among locations and lowercase letters represent differences among the eight sites.

Location	Site Code	Seasonal pH Range	Seasonal Mean pH
Jug End	JN	6.7 – 7.3	7.0 bc
	JS	6.8 – 7.7	7.1 ab
	Location mean:		7.0 A
Schenob Brook	SBB	6.7 – 7.3	7.0 abc
	SBC	6.8 - 7.8	7.3 a
	SBN	6.4 – 7.7	7.0 abc
	SBS	6.3 – 6.9	6.6 d
	Location mean:		6.9 AB
Shmulsky	SO	6.6 – 7.2	6.9 cd
	SS	6.5 – 7.3	6.8 cd
	Location mean:		6.8 B

Calcium and Magnesium

Water calcium changed significantly across the growing season (Figures 3.3-3.5; Wilks' Lambda =0.007, F=$_{5,12}$ = 340. 17, p <0.0001); thus the variable "month" significantly explained variation in water calcium. Water calcium was different throughout the season within locations (month*location interaction: Wilks' Lambda = 0.020, $F_{10,24}$=14.55, p=<0.0001) and within sites (month*site interaction: Wilks' Lambda = 0.009, $F_{25,46.1}$=4.68, p=<0.0001). This means that the location and site contributed to the seasonal variation in water calcium. A decline in water calcium was observed at Jug End North (Figure 3.3a) when it dropped 38 $mg^{.}L^{-1}$ to a low of 54 $mg^{.}L^{-1}$ from July to August. Schenob Brook Bartholomew (Figure 3.4a) and Shmulsky Shrub (Figure 3.5a)

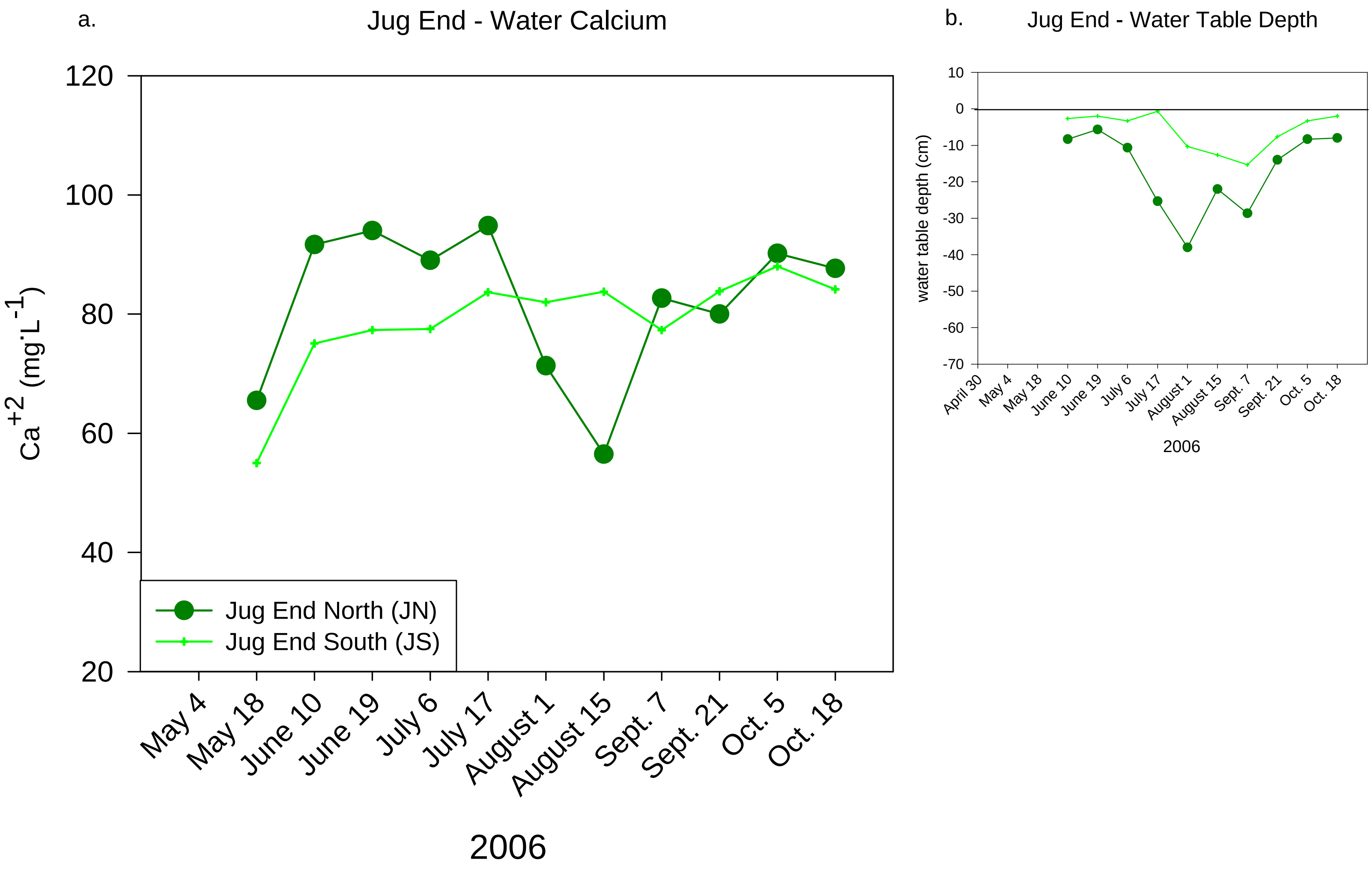
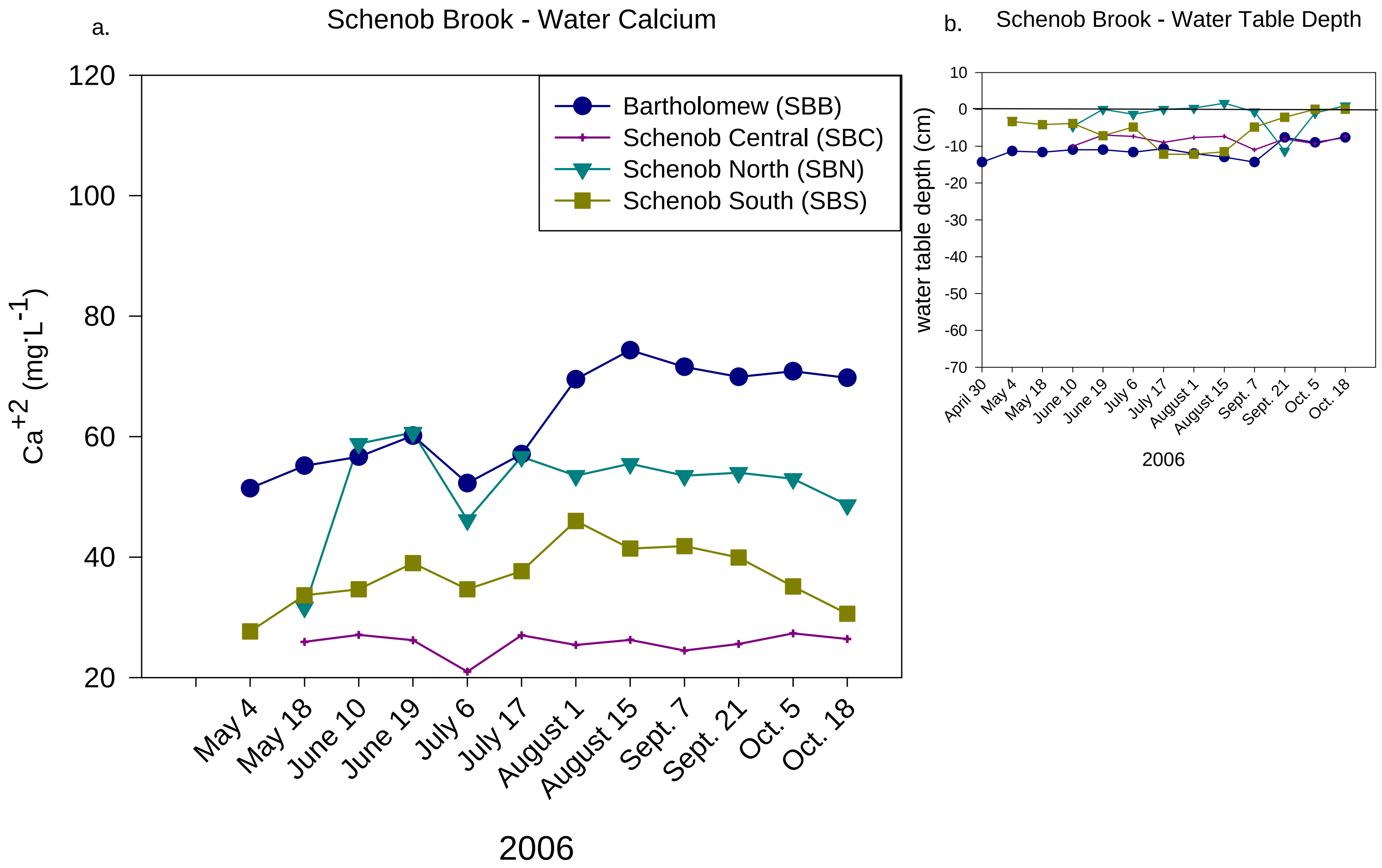


Figure 3.3. a. Seasonal variation in water calcium (mg·L^{-1}) at the Jug End location. Each point represents a mean of three replicate samples. b. A comparison with seasonal water table depth.


a. Schenob Brook - Water Calcium

b. Schenob Brook - Water Table Depth

Figure 3.4. a. Seasonal variation in water calcium ($mg \cdot L^{-1}$) at the Schenob location. Each point represents a mean of three replicate samples. b. A comparison with seasonal water table depth.

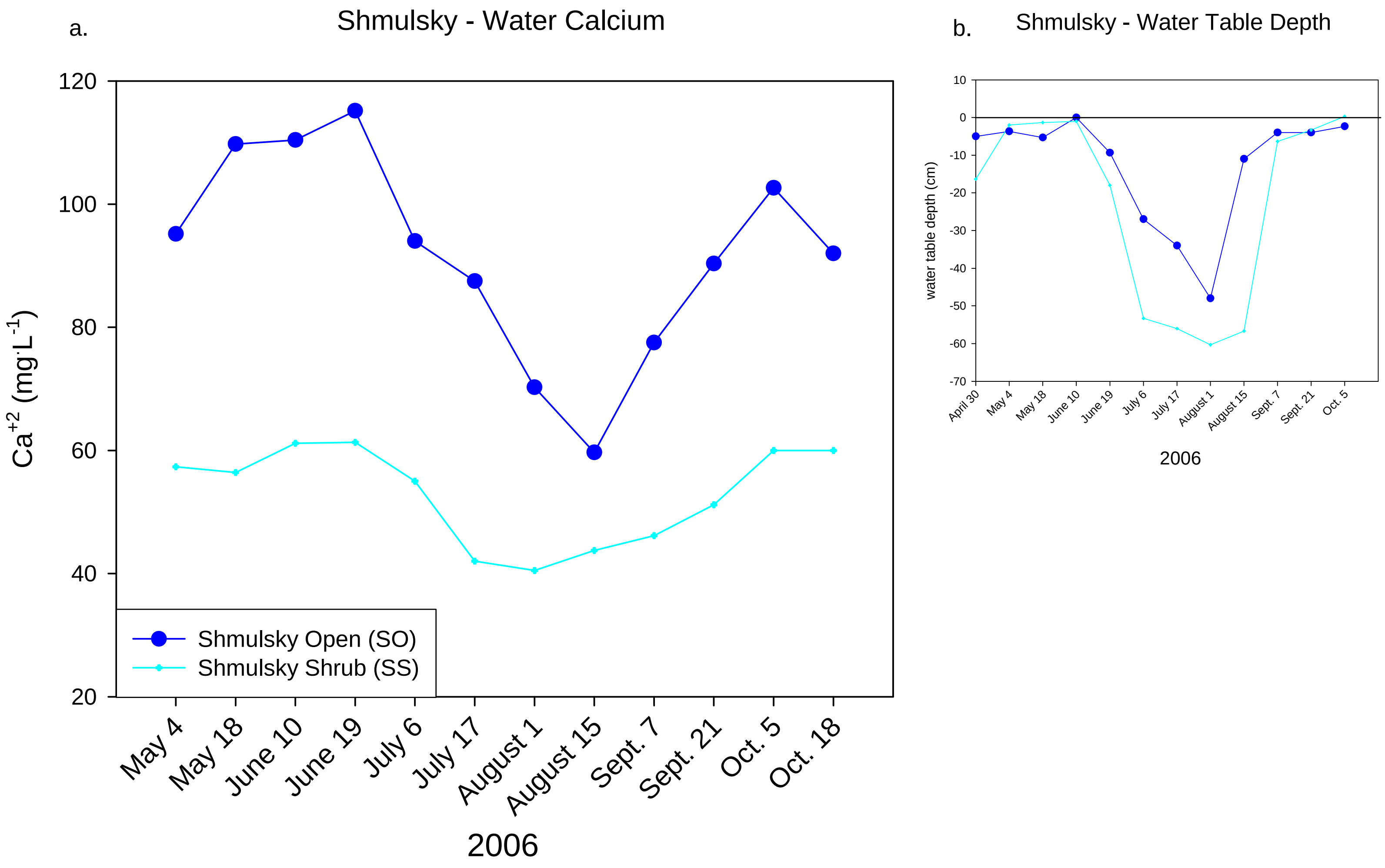


Figure 3.5. a. Seasonal variation in water calcium (mg·L^{-1}) at the Shmulsky location. Each point represents a mean of three replicate samples. b. A comparison with seasonal water table depth.

were the only sites whose calcium values actually increased mid-season. The remaining sites, Schenob Brook North and Central (Figure 3.4a) and Jug End North (Figure 3.3a) maintained steady calcium levels throughout the growing season. The largest seasonal change in water calcium was observed at Shmulsky Open (Figure 3.5a) where values dropped steadily by 55 $mg{\cdot}L^{-1}$ to a low of 38 $mg{\cdot}L^{-1}$ from June to August.

The changes in calcium over the growing season corresponded to characteristics in the water table during the same time periods (Figures 3.3b, 3.4b, 3.5b). At the locations where the water table dropped below the depth at which water was collected (30 cm), groundwater was no longer being sampled directly and thus calcium values were lower as water chemistry was more reflective of unsaturated pore water equilibria instead of groundwater conditions.

The seasonal mean water calcium was significantly different among locations ($F_{2,16}$=62.21, p =<0.0001) and among sites ($F_{5,16}$=21.70, p=<0.0001) (Table 3.2). Schenob Brook had calcium values significantly lower than at Jug End and Shmulsky, which were not different from each other. In general, Shmulsky Open maintained the highest water calcium with a mean of 92 $mg{\cdot}L^{-1}$, and Schenob Brook Central was consistently the lowest with a seasonal mean of 26 $mg{\cdot}L^{-1}$.

Table 3.2. Seasonal range of water calcium and mean seasonal water calcium for the eight study sites and associated locations. Means with different letters are significantly different by Tukey's Test (p=0.05). Uppercase letters represent differences among locations and lowercase letters represent differences among the eight sites.

Location	Site Code	Seasonal Calcium ($mg \cdot L^{-1}$) Range	Seasonal Mean Calcium ($mg \cdot L^{-1}$)
Jug End	JN	37.0 – 112.5	82.1 ab
	JS	47.3 – 95.0	78.9 ab
	Location mean:		80.5 A
Schenob Brook	SBB	42.5 – 78.5	63.2 bc
	SBC	16.3 – 37.8	25.7 e
	SBN	22.0 – 73.5	52.0 cd
	SBS	22.5 – 52.5	36.9 de
	Location mean:		44.4 B
Shmulsky	SO	38.0 – 133.5	92.0 a
	SS	33.5 – 74.5	52.9 cd
	Location mean:		72.4 A

Water magnesium changed significantly across the growing season (Figure 3.6) (Wilks' Lambda =0.164, F=$_{5,12}$ 12.27, p= 0.0002); thus the variable "month" significantly explained the variation in water magnesium. Water magnesium was also significantly different seasonally within locations (month*location interaction: Wilks' Lambda = 0.090, $F_{10,24}$ = 5.57, p=0.0003) and within sites (month*site interaction: Wilks' Lambda = 0.067, $F_{25,46.1}$ = 1.98, p=0.0222). This means that the location and site contributed to the seasonal variation in water magnesium. Shmulsky Open exhibited the largest seasonal change in water magnesium (Figure 3.6c) where values dropped steadily by 25 $mg \cdot L^{-1}$ from June to August. Schenob Brook sites Bartholomew and South (Figure 3.6b) and both Jug End sites (Figure 3.6a) had water magnesium values that increased mid-season. Of these, Bartholomew remained at the elevated level; South returned to the levels it had in the spring, and the two Jug End sites initially returned to spring levels but then increased back to their peak height in October. The other sites, Shmulsky Shrub,

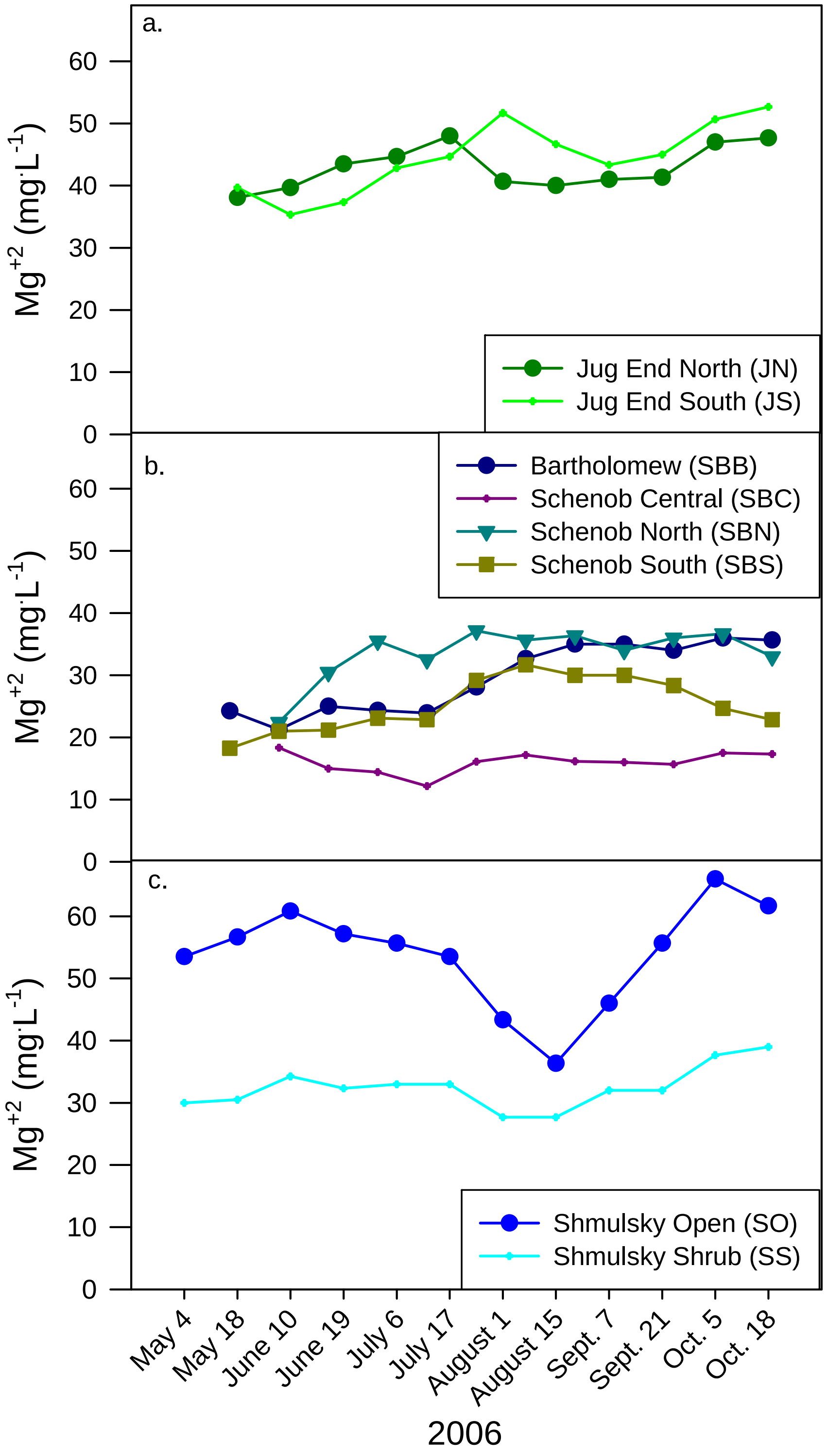


Figure 3.6. Seasonal variation in water magnesium (mg$\cdot$L^{-1}) at a) Jug End, b) Schenob Brook and c) Shmulsky. Each point represents a mean of three replicate samples.

Schenob Brook Central, and Schenob Brook North remained at fairly constant levels throughout the entire growing season. Seasonal changes in magnesium were similar to those of calcium, as would be expected since the bedrock underlying many of these sites contains dolomite. Later, this relationship will be discussed further.

The seasonal mean water magnesium was significantly different among locations ($F_{2,16}$=45.02, p =<0.0001) and among sites ($F_{5,16}$= 11.51, p=<0.0001) (Table 3.3). Schenob Brook had significantly lower magnesium than Jug End and Shmulsky, which were not different from each other. In general, Shmulsky Open maintained the highest water magnesium with a seasonal mean of 54 $mg{\cdot}L^{-1}$, and Schenob Central was consistently the lowest with a seasonal mean of 16 $mg{\cdot}L^{-1}$.

Table 3.3. Seasonal range of water magnesium and mean seasonal water magnesium for the eight study sites and associated locations. Means with different letters are significantly different by Tukey's Test (p=0.05). Uppercase letters represent differences among locations and lowercase letters represent differences among the eight sites.

Location	Site Code	Seasonal Magnesium ($mg{\cdot}L^{-1}$) Range	Seasonal Mean Magnesium ($mg{\cdot}L^{-1}$)
Jug End	JN	30.0 – 60.5	42.5 ab
	JS	32.0 – 59.0	44.1 ab
	Location mean:		43.3 A
Schenob Brook	SBB	17.5 – 42.5	29.1 c
	SBC	9.3 – 30.0	16.2 d
	SBN	14.75 – 42.0	32.7 bc
	SBS	15.3 – 37.5	24.8 cd
	Location mean:		25.7 B
Shmulsky	SO	23.0 – 70.0	54.0 a
	SS	25.0 – 46.5	32.4 bc
	Location mean:		43.2 A

Due to the nature of the bedrock surrounding the study sites (predominantly dolomitic and lesser amounts of calcitic limestone), it is expected that the levels and fluctuations in calcium and magnesium will correspond to each other if they are all delivered with water that has traveled through bedrock with the same mineral composition. In fact, trends in water magnesium across the season do closely follow those of the calcium for each site (Figure 3.7) but with some variability in the spring months. If the groundwater traveled mostly through dolomitic limestone, one would expect Ca:Mg to be about 1.74, the equivalent ratio of the atomic weight of 1 mole of calcium per the atomic weight of 1 mole of magnesium. If the water traveled through calcitic limestone as well as dolomitic limestone, then greater amounts of calcium would be present and the ratio would be greater than 1.74.

Overall, seasonal mean significant differences were observed in the Ca:Mg ratios among the three study locations ($F_{2,16}$=3.68, p=0.0483) and among the eight study sites ($F_{5,16}$=20.92, p=<0.0001) (Table 3.4). Ratios at Jug End were significantly higher than at Schenob and Shmulsky, which were not different from each other. In addition, “month” was a significant causative factor affecting seasonal Ca:Mg variation (Wilks' Lambda =0.031, $F_{5,12}$=74.73, p=<0.0001) (Figure 3.7). However, these differences were only marginally significant within locations (month*location interaction: Wilks' Lambda =0.276, $F_{10,24}$=2.16, p=0.0586) and not in any way within sites (month*site interaction: Wilks' Lambda =0.190, $F_{25,46.1}$=1.04, p= 0.4409). Schenob Brook Bartholomew had the significantly highest Ca:Mg ratio of all of the sites at 2.12. This elevated ratio would be expected as it is the only site that is directly down gradient from a purely calcitic marble/limestone section of bedrock. The sites with the next highest Ca:Mg ratio occurred at Jug End where the surrounding bedrock contained both dolomite and calcite. The Shmulsky sites fell in the middle as their surrounding bedrock also contained both dolomite and calcite. The remaining Schenob sites (Central, North and South) had the lowest range with an average of 1.5, which would also be expected, as the bedrock surrounding them is chiefly comprised of the mineral dolomite.

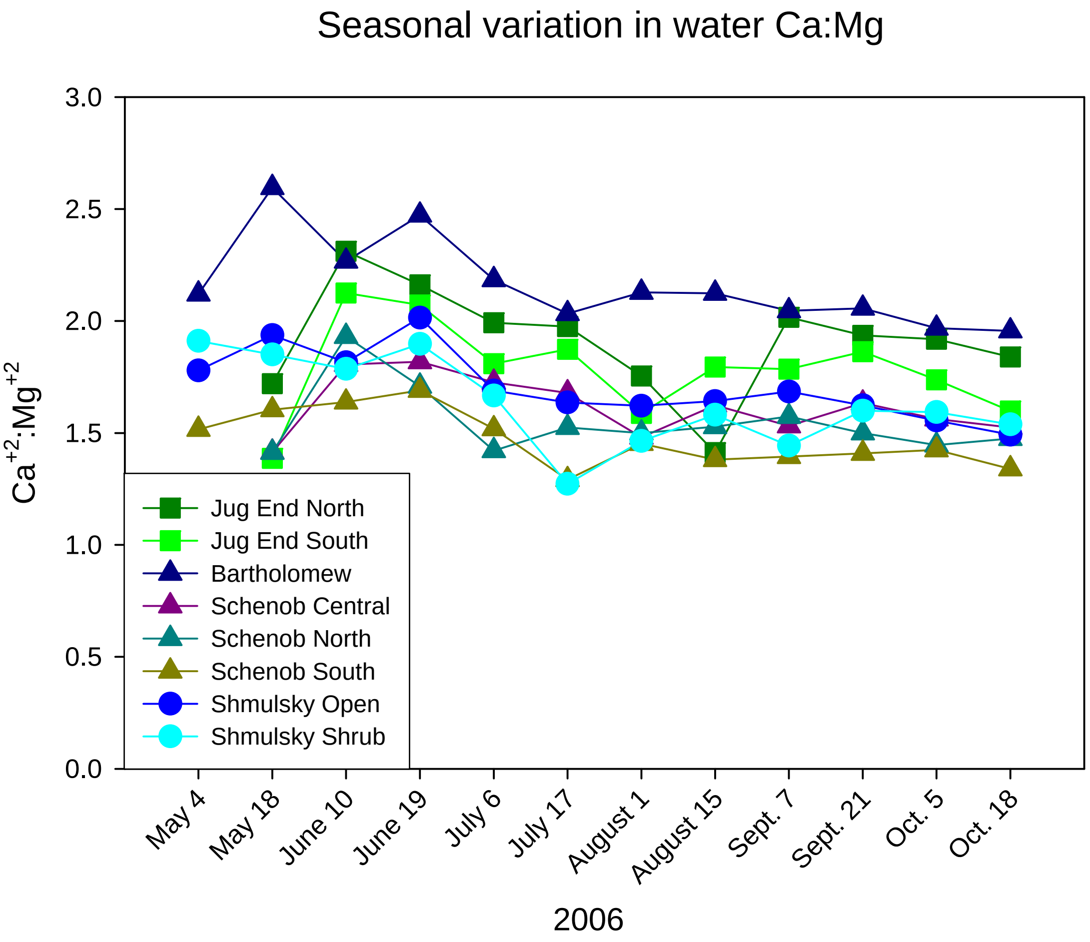


Figure 3.7. Seasonal variation in calcium to magnesium ratios for the eight study sites. Each point represents a mean of three replicate samples.

Table 3.4. Seasonal range of water calcium to magnesium ratios and mean seasonal water calcium to magnesium ratios for the eight study sites and associated locations. Means with different letters are significantly different by Tukey's Test (p=0.05). Uppercase letters represent differences among locations and lowercase letters represent differences among the eight sites.

Location	Site Code	Seasonal Ca:Mg Range	Seasonal Mean Ca:Mg
Jug End	JN	1.41 – 2.31	1.91 ab
	JS	1.39 – 2.13	1.78 bc
	Location mean:		1.85 A
Schenob Brook	SBB	1.96 – 2.60	2.12 a
	SBC	1.41 – 1.82	1.62 cd
	SBN	1.41 – 1.93	1.55 cd
	SBS	1.29 – 1.69	1.47 d
	Location mean:		1.69 B
Shmulsky	SO	1.49 – 2.01	1.71 bcd
	SS	1.27 – 1.91	1.63 bcd
	Location mean:		1.67 C

Iron

No distinct trends were observed in water iron (total iron) across the growing season (Wilks' Lambda =0.365 $F_{5,9}$=3.13 p=0.0656). However, there were significant seasonal differences within locations (month*location interaction: Wilks' Lambda=0.176, $F_{10,18}$ =2,49, p=0.0440) and within sites (month*site interaction: Wilks' Lambda=0.045, $F_{25, 34.9}$=1.83, p =0.0496) (Figure 3.8). Significant differences existed among the sites in June ($F_{5,13}$= 3.11, p=0.0460), and in August among the locations ($F_{2,13}$ =7.12, p=0.0082) and among the sites ($F_{5,13}$=4.23, p=0.0167). Three of the sites at Schenob (Bartholomew, North and South), and Jug End North, exhibited similar seasonal trends in that the iron increased in July and August and then declined in the autumn months. One site at each location, Shmulsky Shrub, Schenob Brook Central, and Jug End South, had very low iron

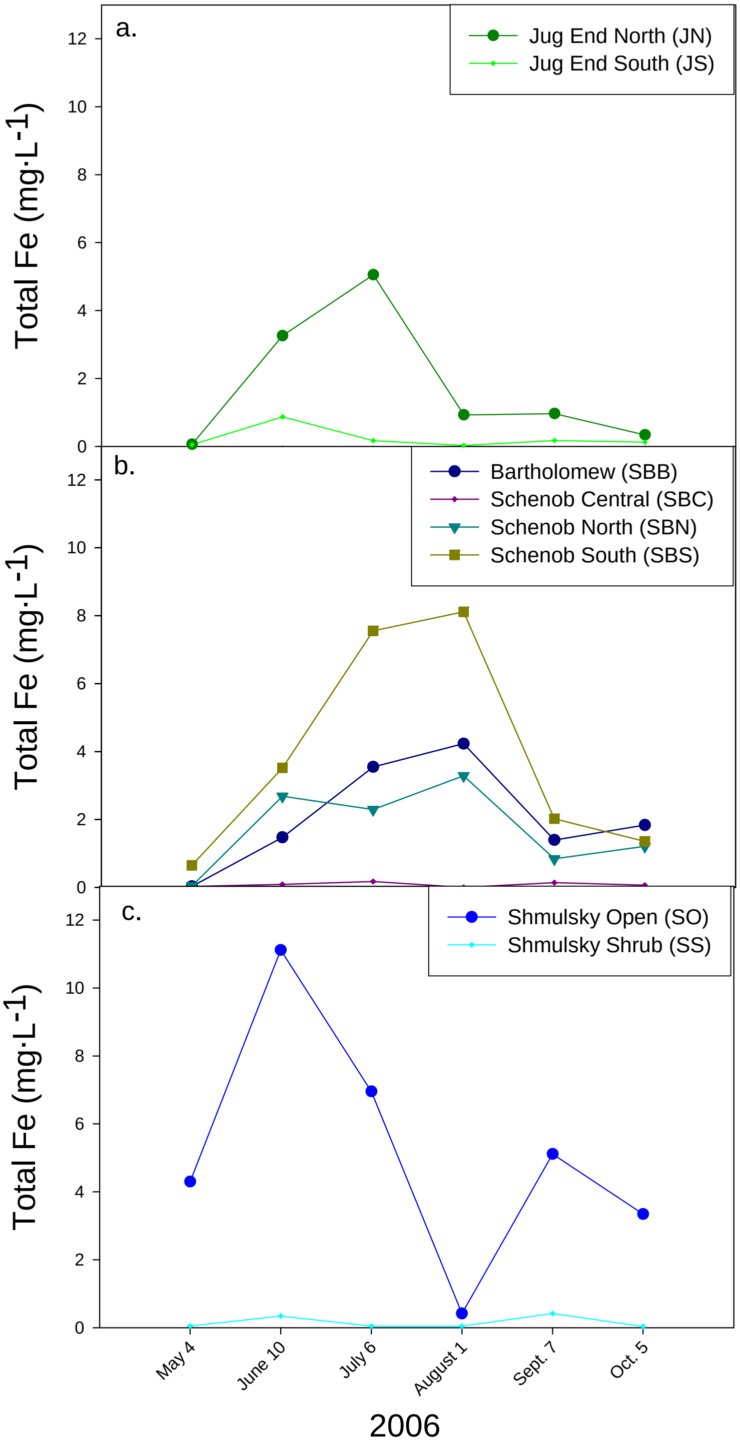


Figure 3.8. Seasonal variation in water iron (mg·L^{-1}) at the a) Jug End, b) Schenob Brook and c) Shmulsky locations. Each point represents a mean of three replicate samples.

levels with no seasonal change. Shmulsky Open exhibited the most unique seasonal trend in comparison to the others, in that it peaked in June, declined greatly in August, and increased again in September (but to only half of the peak it exhibited in June).

Differences in the seasonal mean water iron were not explained by location ($F_{2,13}$=0.34, p=0.7207), nor were they explained by site ($F_{5,13}$=2.43, p=0.0919) (Table 3.5). At many observations, iron was undetectable. The greatest amount of iron was measured during the irregular peak that occurred at Shmulsky Open in which it reached 22 $mg^{\cdot}L^{-1}$ on June 10th. On average, this site also had the highest mean seasonal iron at 5.3 $mg^{\cdot}L^{-1}$. Schenob Brook Central was consistently the lowest with a mean seasonal value of 0.1 $mg^{\cdot}L^{-1}$.

The variation in iron at Shmulsky Open may be explained by the disconnect from groundwater (as indicated by hydrologic measurements) that occurred at that site after June, as well as the loss of strong reducing conditions that occurred in the summer months (Appendix C). Groundwater may provide a source of iron as the bedrock surrounding the sites (Figures 1.4, 1.6 and 1.8) contains micaceous schist, a metamorphic rock with iron-containing silicate minerals (Aldridge and Churchman, 1991). The three sites that showed no seasonal change had lower iron values in general (Jug End South, Schenob Brook Central, Shmulsky Shrub) and also had the most oxidizing conditions throughout the growing season (Appendix C). It is difficult to explain the iron levels at the remaining sites (Jug End North, Schenob Brook North, Schenob Brook Bartholomew, Schenob Brook South), where iron was highest during the summer months and was lower in the spring and fall. For the Schenob sites, the water table remained constant throughout the growing season and reducing conditions were variable in strength, yet persistent.

Table 3.5. Seasonal range of water iron and mean seasonal water iron for the eight study sites and associated locations. nd = iron was < detection limit of 0.02 $mg \cdot L^{-1}$. Means with different letters are significantly different by Tukey's Test (p=0.05). Uppercase letters represent differences among locations and lowercase letters represent differences among the eight sites.

Location	Site Code	Seasonal Iron ($mg \cdot L^{-1}$) Range	Seasonal Mean Iron ($mg \cdot L^{-1}$)
Jug End	JN	nd – 7.4	1.8 a
	JS	nd – 2.3	0.3 a
	Location mean:		2.0 A
Schenob Brook	SBB	nd – 9.5	2.1 a
	SBC	nd – 0.3	0.1 a
	SBN	nd – 6.3	1.6 a
	SBS	0.1 – 15.3	3.8 a
	Location mean:		1.9 A
Shmulsky	SO	nd – 22.0	5.3 a
	SS	nd – 0.9	0.2 a
	Location mean:		2.8 A

Potassium

No significant seasonal differences were detected in water potassium values. Variability among them was not explained by month (Wilks' Lambda =0.508, $F_{5,11}$=2.13, p= 0.1371), month by location (month*location interaction: Wilks' Lambda =0.335, $F_{10,22}$=1.60, p=0.1710) nor month by site (month*site interaction: Wilks' Lambda =0.179, $F_{25,42.4}$=1.60, p= 0.4904) interactions. Overall seasonal mean water potassium was not significantly different among locations ($F_{2,15}$= 3.44, p= 0.0586) but was significantly different among sites within Schenob only ($F_{5,15}$=5.85, p=0.0034) (Table 3.6). Major sources of potassium would be from groundwater that has traveled through mica or feldspar minerals (Brady, 1999) or from fertilizer input. Thus potassium may be supplied to the wetlands by groundwater that has traveled through the micaceous schists surrounding the sites (Figures 1.5 and 1.7) or from adjacent agriculture. These results may indicate that farming played a large role as Shmulsky Open and Schenob Brook Bartholomew had the highest potassium values and were also in closest proximity to various forms of agriculture (e.g. vegetable production, and cattle farms). Schenob Brook Bartholomew was adjacent to vegetable fields and Shmulsky Open was adjacent to cattle grazing areas.

Table 3.6. Seasonal range of water potassium and mean seasonal water potassium for the eight study sites and associated locations. Means with different letters are significantly different by Tukey's Test (p=0.05). Uppercase letters represent differences among locations and lowercase letters represent differences among the eight sites.

Location	Site Code	Seasonal Potassium ($mg \cdot L^{-1}$) Range	Seasonal Mean Potassium ($mg \cdot L^{-1}$)
Jug End	JN	0.4 – 2.7	0.9 bc
	JS	0.6 – 2.3	1.4 abc
	Location mean:		0.6 A
Schenob Brook	SBB	1.5 – 3.1	2.0 a
	SBC	0.7 – 2.1	1.3 abc
	SBN	0.4 – 3.1	1.7 ab
	SBS	0.5 – 2.8	0.9 bc
	Location mean:		1.2 A
Shmulsky	SO	0.5 – 3.9	1.5 abc
	SS	0.3 – 1.5	0.7 c
	Location mean:		1.1 A

Ammonium and Orthophosphate

Seasonal variability in ammonium (Wilks' Lambda =0.322, $F_{3,9}$=2.25, p= 0.0971) and orthophosphate (Wilks' Lambda =0.450, $F_{5,9}$=2.20, p= 0.144) were not observed in this study, nor were seasonal differences detected among sites (ammonium: Wilks' Lambda =0.251, $F_{15,25.3}$=1.09, p= 0.4079; orthophosphate: Wilks' Lambda =0.118, $F_{25,34.9}$=1.08, p= 0.406) or locations (ammonium: Wilks' Lambda =0.336, $F_{6,18}$=2.17, p= 0.095; orthophosphate: Wilks' Lambda =0.384, $F_{10,18}$=1.10, p= 0.410). The average amount of ammonium in the pore water was 0.4 $mg \cdot L^{-1}$ (expressed as ammonium, NH_4^+) but ranged from undetectable levels to 3.0 $mg \cdot L^{-1}$ at Shmulsky Open (Table 3.7). Orthophosphate was present in the pore water at an average concentration of 0.11 $mg \cdot L^{-1}$ (expressed as phosphate, PO_4^{3-}) but ranged from undetectable levels to 1.63 $mg \cdot L^{-1}$ at Schenob Brook Bartholomew (Table 3.8). Due to the proximity of most of these sites to various forms of agriculture it was expected that ammonium and orthophosphate levels would be elevated from fertilizer or manure inputs. Corresponding with findings for elevated potassium, both Shmulsky Open and Schenob Brook Bartholomew (the sites in closest proximity to agricultural land uses) had the highest ammonium and orthophosphate of all of the sites studied. At Shmulsky Open, the peaks in these nutrients occurred in May. This may have coincided with spring fertilizer applications at other locations in the watershed. At Bartholomew these peaks occurred in August (ammonium) and October (orthophosphate) and could be related to increased nitrogen mineralization when temperatures were warmer and/or fall fertilizer applications.

Table 3.7. Seasonal range of water ammonium and mean seasonal water ammonium for the eight study sites and associated locations. nd = ammonium was < detection limit of 0.02 $mg \cdot L^{-1}$. Means with different letters are significantly different by Tukey's Test (p=0.05). Uppercase letters represent differences among locations and lowercase letters represent differences among the eight sites.

Location	Site Code	Seasonal ammonium ($mg \cdot L^{-1}$) Range	Seasonal Mean ammonium ($mg \cdot L^{-1}$)
Jug End	JN	nd – 1.81	0.36 a
	JS	nd – 0.52	0.13 a
	Location mean:		0.24 A
Schenob Brook	SBB	nd – 2.08	0.57 a
	SBC	nd - 0.86	0.14 a
	SBN	nd – 1.42	0.31 a
	SBS	nd – 1.83	0.56 a
	Location mean:		0.40 A
Shmulsky	SO	nd – 3.02	0.86 a
	SS	nd – 0.50	0.17 a
	Location mean:		0.52 A

Table 3.8. Seasonal range of water orthophosphate and mean seasonal water orthophosphate for the eight study sites and associated locations. nd = orthophosphate was < detection limit of 0.02 $mg \cdot L^{-1}$. Means with different letters are significantly different by Tukey's Test (p=0.05). Uppercase letters represent differences among locations and lowercase letters represent differences among the eight sites.

Location	Site Code	Seasonal orthophosphate ($mg \cdot L^{-1}$) Range	Seasonal Mean orthophosphate ($mg \cdot L^{-1}$)
Jug End	JN	nd – 1.19	0.18 a
	JS	nd – 0.50	0.12 a
	Location mean:		0.15A
Schenob Brook	SBB	nd – 1.63	0.17 a
	SBC	nd – 0.96	0.09 a
	SBN	nd – 0.20	0.04 a
	SBS	nd – 0.30	0.08 a
	Location mean:		0.10A
Shmulsky	SO	nd – 1.36	0.13 a
	SS	nd – 0.13	0.05 a
	Location mean:		0.09A

Soil Analysis

Detailed accounts of the physical and chemical properties of the soil at each study plot and for each horizon to a depth of 60cm are available in Appendix E. The following results and discussion refer to the surface soil horizons at each of the study sites unless otherwise stated. Overall, the soil physical and chemical properties differed among sites (Wilks' Lambda = 0.0006, $F_{45,38.9}$= 3.68, p=<0.0001) and among locations (Wilks' Lambda = 0.009, $F_{18,16}$= 8.25, p=<0.0001).

Soil Physical Properties

In general, soils had mucky, organic rich surface horizons and mineral subsurface horizons. The texture of the subsoil varied among the study sites from sand, loamy sand, sandy loam, loam, to silt loam although the majority of the subsurface soils were reflective of their outwash nature as being either loamy sands or sandy loams. Only at Schenob Brook South were all soil samples silt loams. This observation was not surprising as this was the only site comprised of alluvial deposits from the nearby Schenob Brook.

Percent organic matter varied from 8.7% at Schenob Brook Central to 75% at Schenob Brook North. It differed significantly among locations ($F_{2,16}$=6.63, p=0.0080) and among sites within locations ($F_{5,16}$=7.75, p=0.0007) (Table 3.9). The Schenob location was significantly higher than Jug End, but neither of them were significantly different from Shmulsky. Considering that the water table at Schenob never dropped below 30cm, it makes sense that organic matter accumulation would be high. Thus some portion of the surface soils at all of the Schenob sites was saturated during the entire growing season, allowing less decomposition of organic matter as compared to better aerated sites.

Table 3.9. Soil physical and chemical properties for the eight study sites within the three locations. Measurements are represented on a per weight basis. Each number represents a mean of three soil samples collected from three soil pits (one at each plot) at each site. Means with different letters are significantly different by Tukey's Test (p=0.05). Uppercase letters represent differences among locations and lowercase letters represent differences among the eight sites.

Location	Site	pH	Organic Matter (% by wt.)	Total N ($mg \cdot kg^{-1}$)	AB-DTPA Extractable Elements (pH 7.6) ($mg \cdot kg^{-1}$)		Exchangeable cations (NH_4OAc, pH 7) ($mg \cdot kg^{-1}$)			
					P	Fe	Ca	Mg	K	Fe
Jug End	JN	7.3 ab	14.0 bc	5646 bc	1.1 a	99.0 cd	3132 b	335.7 cd	39.6 a	0.7 a
	JS	7.5 a	11.1 c	4653 c	1.3 a	63.9 bd	2649 b	289.7 d	31.5 a	0.8 a
	Mean	7.4 A	12.5 A	5150 A	1.5 A	81.5 A	2891 A	312.7 A	35.6 A	0.7 A
Schenob	SBB	7.0 abc	17.1 bc	7630 abc	2.0 a	137.5 bcd	3914 b	459.8 bcd	59.1 a	1.4 a
	SBC	6.8 abc	10.6 c	3620 c	2.1a	89.4 cd	2483 b	335.2 cd	41.9 a	2.8 a
	SBN	6.4 bc	51.1 a	14843 a	2.5 a	267.0 ab	7968 a	1204.1 a	90.8a	3.7 a
	SBS	6.1 d	35.0 ab	14323 ab	4.2 a	401.3 a	5232 a	780.9 b	216.6 a	6.8 a
	Mean	6.5 B	26.6 B	10104 B	2.7 B	222.3 B	4900 B	695.0 B	104.7 B	3.7 B
Shmulsky	SO	6.4 bc	26.1 abc	10513 abc	1.8 a	162.7 abc	4907 b	681.8 bc	36.4 a	1.3 a
	SS	6.8 abc	13.9 bc	6246 abc	1.3 a	127.1 bcd	2926 b	401.4 cd	34.8 a	1.1 a
	Mean	6.6 B	20.0 AB	8380 AB	1.2 A	144.9 B	3916 AB	541.6 B	35.6 A	1.2 B

The differences in organic matter affected the method of reporting of the soil nutrients. When represented on a per weight basis the concentration of an element in $mg \cdot kg^{-1}$ was more representative of the amount of organic matter in the sample than the actual concentrations per volume of soil. For this reason, the bulk density was measured for each of the soils so that the nutrient measurements could be standardized on a volume basis. Nutrient measurements as a function of weight are presented in Table 3.9. The following discussion of the soil chemical properties will only refer to the nutrient measurements that are on a volume basis which are reported in Table 3.10.

In general, bulk densities ranged from 0.34 $g \cdot cm^{-3}$ for the soils that contained large amounts of organic matter to 0.96 $g \cdot cm^{-3}$ for those with less. Significant differences in bulk density were detected among locations ($F_{2,16}$= 10.34, p=<0.0001) and among sites ($F_{5,16}$=7.98, p=0.0009) (Table 3.10). The bulk density at Schenob was significantly lower than at Jug End and Shmulsky, which were not different from each other.

Soil Chemical Properties

Similar trends were observed for pH, calcium, and magnesium in the soil, as were observed in the water chemistry. Soil pH ranged from 6.1 at Schenob Brook South to 7.5 at Jug End South. Significant differences in pH were observed among locations ($F_{2,16}$=11.14, p=0.0009) and among sites within locations ($F_{5,16}$=2.91, p=0.0467) (Table 3.9). The soil pH at Jug End was significantly higher than at Schenob and Shmulsky, which were not different from each other. Significant differences in soil calcium were observed among locations ($F_{2,16}$=7.97, p=0.0256) and among sites within locations ($F_{5,16}$=10.62 p=0.0137) (Table 3.10). When grouped by location, the soil calcium at

Table 3.10. Physical and chemical properties for surface soils at the eight study sites within the three locations, expressed on a volume basis. Each number represents a mean of three soil samples collected from three soil pits (one at each plot) at each site. Means with different letters are significantly different by Tukey's Test (p=0.05). Uppercase letters represent differences among locations and lowercase letters represent differences among the eight sites.

Location	Site	Bulk Density (g·cm^{-3})	Organic Matter (kg·m^{-3})	Total N (g·m^{-3})	AB-DTPA Extractable Elements (pH 7.6) (g·m^{-3})		Exchangeable cations (NH_4OAc, pH 7) (g·m^{-3})			
					P	Fe	Ca	Mg	K	Fe
Jug End	JN	0.84 a	113.2 bc	4557 ab	0.9 a	82.7 b	2528 ab	272.7 b	32.0 a	0.6 a
	JS	0.84 a	95.2 c	3977 ab	1.1 a	55.8 b	2254 ab	247.6 b	27.2 a	0.7 a
	Mean	0.84 A	104.2 A	4266 A	1.0 A	69.2 A	2390 A	260.1 A	29.6 A	0.6 A
Schenob	SBB	0.76 ab	125.0 bc	5559 ab	1.4 a	97.1 ab	2885 ab	336.8 ab	43.5 a	1.1 a
	SBS	0.48 bc	161.0 a	6559 a	1.9 a	179.9 a	2423 ab	361.9 ab	99.1 a	3.1 a
	SBC	0.71 ab	74.2 bc	2507 b	1.4 a	61.2 b	1724 b	235.8 b	28.6 a	1.9 a
	SBN	0.34 c	143.0 a	4922 ab	0.8 a	89.6 b	3112 a	470.3 a	38.3 a	1.4 a
	Mean	0.57 B	125.8 AB	4886 A	1.4 A	106.9 AB	2536 A	351.2 B	52.4 A	1.9 B
Shmulsky	SO	0.73 ab	188.5 ab	7576 a	1.3 a	119.0 ab	3541 a	489.3 a	26.0 a	0.9 a
	SS	0.96 a	130.9 bc	5874 ab	1.2 a	120.9 ab	2744 ab	376.5 ab	32.2 a	1.1 a
	Mean	0.84 A	160.0 B	7088 B	1.2 A	120.0 B	3142 B	432.9 C	29.1 A	1.0 A

Shmulsky was significantly higher than Schenob and Jug End, which were not different from each other. Schenob Brook Central had the lowest soil calcium at 1554 $g \cdot m^{-3}$ and Shmulsky Open had the highest at 3680.3 $g \cdot m^{-3}$. Significant differences existed in soil magnesium among locations ($F_{2,16}$=16.41, p=0.0003) and among sites ($F_{5,16}$=16.47, p=0.0015) (Table 3.10). All three locations were significantly different from each other.

Although water analyses did not show significant differences in nitrogen (as ammonium) and iron (as total iron), significant differences were observed in total nitrogen and in DTPA-iron in the soil. Total nitrogen was significantly different among locations ($F_{2,16}$=4.80, p=0.0132) and among sites within locations ($F_{5,16}$=5.75, p=0.0234) (Table 3.10). This same trend was also observed in % organic matter (by wt.) suggesting that the nitrogen pool is controlled directly by the contribution from organic matter. Total nitrogen at Shmulsky was significantly higher than at Schenob and Jug End, which were not different from each other. Differences in exchangeable iron were only observed among locations ($F_{2,16}$= 7.14, p=0.0370) as Jug End was significantly lower than Schenob and Shmulsky, which were not different from each other. Significant differences in DTPA iron were observed among locations ($F_{2,16}$=12.68, p=0.0334) and among sites within locations ($F_{5,16}$= 7.43, p=0.0071) (Table 3.10). Schenob was not different from Jug End or Shmulsky, both of which were different from each other. Again, Jug End had the lowest iron levels. This trend makes sense considering that both Schenob and Shmulsky had significant amounts of micaceous schist (a rock with iron-containing silicates) in the surrounding bedrock and Jug End had very little. Thus groundwater would be more likely to carry iron to those sites where it could be stored in exchangeable forms on soil colloids and in amorphous precipitates.

Soil phosphorus and potassium were not significantly different among sites but were significantly different among locations when compared on a weight basis. These differences then were most likely due to differences in the amount of organic matter at those sites. Large differences were not expected for soil phosphorus as it did not differ in the water analysis, however it was somewhat surprising to see that soil potassium values were not different as they were in the water analysis. Although there were no significant differences, soil potassium ranged from 22 to 99 $g \cdot m^{-3}$ and this may reflect the observed differences in its water chemical values.

General Chemical Properties

Water chemistry at Shmulsky corresponded with those found by Picking (2002) at the same location. Levels of water ammonium, orthophosphate, iron, and soil phosphorus potassium, magnesium, and percent organic matter fell within the expected range. Even similar maximum iron levels were observed in the water at Shmulsky (22 $mg \cdot L^{-1}$). It was surprising to observe orthophosphate levels above 1 $mg \cdot L^{-1}$, (especially in soils with a pH above 7) as this is often only associated with heavily fertilized soils (Brady, 1999). Thus the proximity of these sites to agricultural land must result in a large input from fertilizers.

Seasonal trends in calcium also corresponded with Picking (2002) who observed high water calcium in April and May which then dropped to almost half of its original concentration in the mid-summer months when the water table dropped. Maximum calcium values corresponded to Picking's study in 1997 where water calcium never exceeded 120 $mg \cdot L^{-1}$ and also to research by Komor (1994) who measured values up to 128 $mg \cdot L^{-1}$. Shmulsky is an interesting site, because most research on calcareous fens has determined the average range of dissolved calcium (even in what is defined as a "rich fen") of being much lower (only 5 to 65 mg·L^{-1}). Jug End had a range of 37 to 112.5 mg·L^{1} calcium and sites at Schenob ranged from 16.3 to 73.5 mg·L^{-1}. The mid-range of calcium values at these sites were similar to those as studied by Motzkin (1994), and Slack et al. (1980). Picking (2002) observed soil calcium values to range from 2126 g·m^{-3} to 4907 g·m^{-3} at Shmulsky, which corresponded to Shmulsky Open where calcium values were the highest measured in the current study (3541 g·m^{-3}; although maximum values in this study were lower).

Overall, the strongest and most significant difference in the chemistry of the waters and soils of the calcareous fens was due to the differences that could be associated with calcareous groundwater inputs (i.e., calcium, magnesium, and pH). Minor differences were observed in the primary macronutrients. The strongest significant difference in primary macronutrients was observed in total soil nitrogen, although this difference may be a reflection of the differences in organic matter content among the sites. Phosphorus was never significantly different and potassium showed significant yet small differences in the water only. Although water iron was not significantly different among sites or locations, it showed significant seasonal differences. In the soil, differences were only observed from the AB-DTPA iron analysis in which much more iron was chelated. This analysis may have been able to detect those seasonal differences that alone were not significantly different, by giving an indication to the available pool of iron.

Hydrology – Geochemistry Relationships

Calcium and magnesium did not always show similar trends, and sites with the highest magnesium values did not necessarily also have the highest calcium values. This would indicate that there are different sources of water entering the wetland locations. Observing the Ca:Mg ratio seems to support this, as sites whose surrounding bedrock was comprised of more calcite in relation to dolomite also had less magnesium in relation to calcium and thus Ca:Mg ratios above 1.74. It was expected that the different locations would have different sources of groundwater as they were geographically separated from one another. However, without modeling the groundwater flow-paths these statements are purely conjecture.

The site with the highest recorded calcium value and overall average highest calcium levels (Shmulsky Open) also had the most continuous upwelling of groundwater. By contrast, the two sites with the lowest average water calcium values (Schenob Brook sites Central and South) were the only two sites that showed repeated recharge conditions throughout the growing season. These findings support the notion that the major source of calcium entering these wetlands is groundwater and that when water moving from the substratum to the surface is substantial (i.e., when the wetland is in direct contact with the groundwater) the surface waters will have elevated calcium levels. When a site only receives water that flows laterally from other areas of strong upwelling or when those flow-through sources are diluted by surface water, the calcium levels in the wetlands appear to be much lower.

This trend was particularly evident at Shmulsky, where the water table dropped sharply in mid-summer at both sites, but only Shmulsky Open had a significant drop in water calcium. Shmulsky Open generally had very high calcium levels and continuous upwelling whereas the other site, Shmulsky Shrub, had neither upwelling nor recharge and maintained overall lower calcium levels. Thus, Shmulsky Shrub was receiving a supply of calcium only after it had flowed through the soil and attained an equilibrium with it. Thus when Shmulsky Open stopped receiving groundwater inputs, the water calcium values dropped to the levels present at Shmulsky Shrub and most likely soil equilibrium levels.

In general, the sites that exhibited the largest change in water calcium during the growing season, Shmulsky Open and Jug End South, also exhibited the largest drop in water table depth. These two changes occurred simultaneously during the warm dry summer months from June to August when groundwater was no longer in contact with the 30cm sampling depth. By contrast, the sites that maintained fairly constant water tables throughout the growing season (Schenob Brook sites) also had stable water calcium values.

Conclusions

The hydrologic properties of eight calcareous fens sites in Massachusetts were not alike; however, the differences were attributable to calcium and magnesium enrichment in the wetland waters and the level at which those elements varied across the growing season. Significant differences in water and soil calcium, magnesium, and pH were observed among study sites and among locations, as were differing trends in water table depth and vertical groundwater gradients. Interestingly, water table depth and vertical groundwater gradients could not be correlated to each other as it was determined that sites were receiving water through different mechanisms (directly upwelling or lateral flow from upwelling zones). The level of upwelling seemed to control the average water calcium level, but the fluctuation in the water table seemed to govern how that calcium varied across the season.

Although the main focus of this study, as reported in the next chapter, was to document plant patterns and associations with soil calcium, it is important to remember that the observed plant distribution patterns may be in response to many environmental variables other than calcium. By establishing that the strongest environmental gradients in these calcareous fens are those related to the calcareous bedrock geochemistry, i.e., calcium, magnesium, and pH, and that in fact other minor gradients do exist, i.e., nitrogen and iron, a better understanding of the suspected gradients underlying individual species can be achieved.

CHAPTER IV

PLANT COMMUNITY ANALYSIS

Introduction

Naturalists have long been interested in the unique plant communities that develop in soils enriched with calcium (Sendtner, 1860; Contejean, 1881; Bonnier, 1894; Kerner von Marilaun, 1895; Schrimper, 1898; and many others). Recent studies have focused on the plant communities in calcareous fens (Vitt et al., 1975, Slack et al., 1980; Komor, 1994; Motzkin, 1994; Bedford et al, 1999; Picking and Veneman, 2004) where elevated calcium levels exist due to upwelling groundwater that has traveled through calcareous bedrock. These studies have identified unique plant communities that are associated with the geochemical properties of calcareous fens, i.e., elevated calcium and magnesium levels, elevated pH, and low phosphorus availability relative to noncalcareous fens. No two fens are identical with respect to these conditions and thus overall plant communities differ among fens. Little research has yet focused on these differences at the species level; however, literature reports indicate that some fen species occur only at significantly elevated levels of calcium whereas others appear to occur across a wide range. Within these occurrences, plants can be quite abundant or occur infrequently depending on their life cycles. In addition, not all species occurring in the fens are specific to those calcium-loving communities; common wetland plants also occur there as well.

This chapter will focus on organizing species of northeastern calcareous fens into groups based on literature reports of their rarity and affinity for calcium. These groupings are used in subsequent chapters to test hypotheses regarding the distribution and physiology of calciphiles in relation to environmental calcium levels (see Chapter 1).

In addition, this chapter presents the findings of the vegetation survey conducted across eight calcareous fen research sites in three locations in the Berkshire-Taconic region of Massachusetts, including a characterization of the plant communities observed as well as analysis of the species diversity among sites. Species richness was analyzed in terms of the soil N:P ratio to test the hypothesis by Bedford et al. (1999) that fen ecosystems generally have high species richness as they are limited by phosphorus.

Materials and Methods

Vegetation Survey

The vegetation survey was conducted from July 10 to 21, 2006. This timeframe was selected to maximize potential for identifying graminoid species, many of which were flowering during this period. Three vegetation sampling plots were established at each of the eight field sites, and each plot was associated with a water-sampling station. Vegetation distribution was sampled in a 2.5-m radius plot positioned at the center point. This specific size provided a sampling area of 19.6 m^2, which exceeds the minimum sampling area that was developed using a species area curve for similar habitats (Picking, 2002). Plant species occurring within the plots were identified, and their percent cover was estimated visually.

A Priori Species Selection

A list of species was compiled from the three most recent studies of Massachusetts calcareous fen vegetation (Motzkin, 1994; Kearsley, 1999; Picking and Veneman, 2004) to determine species with which to test hypotheses regarding the distribution and physiology of calciphiles in relation to environmental calcium levels. These studies all conducted vegetation surveys and measured a general substrate calcium level associated with the fens where the plants occurred. Species were grouped based on their abundance in relation to substrate calcium levels into one of three categories. Species placed in Category 1 were species determined to be rare or indicator species in high-calcium fens; Category 2 species were dominant or characteristic in high-calcium fens and Category 3 represented species common to a wide range of fen calcium levels, including common wetland plants. Species in *a priori* category groupings were compared with species actually observed at the research sites.

Species Richness and Turnover

Presence-absence data from the vegetation survey were used to calculate species richness at each 19.6 m^2 plot and collectively at each site (58.8m^2). A regression analysis was conducted (SAS Version 9.1.3, SAS Institute, 2004) to assess if the species richness correlated with the ratio of total soil nitrogen to AB-DTPA-extractable phosphorus (see Chapter 3 for method details). Non-normal data were log-transformed prior to analysis. Presence-absence data from the vegetation survey also were used to calculate species turnover by estimating the similarity between each plot and between each site. Sørensen's distance matrix (Sørenson/Bray-Curtis distance measure calculated in PC-ORD Ver. 5) was subtracted from 1 to calculate a similarity index (McCune and Mefford, 1999). This index provided an estimate of the number of species shared between plots and between sites.

Results and Discussion

Community Characterization

One hundred thirty-three vascular plant species were observed in total of which 28 were woody, 58 were herbaceous, 7 were ferns and allies, and 40 were graminoids. A complete species list is included in Appendix F along with species nomenclature authorities (as they are not included in the text of this manuscript). Species observations correspond with previous studies of calcareous fen vegetation in Western New England (Reschke, 1990; Weatherbee and Crow, 1992; Motzkin, 1994; Kearsley, 1999; Picking, 2002). The vegetation at Shmulsky Open, Jug End North and Jug End South appear to be similar to the "Sloping Graminoid Fen Communities" (after Weatherbee and Crow, 1992), "Calcareous Sloping Fens" (after Kearsley, 1999) or "Rich Sloping/graminoid Fens" (after Reschke, 1990) where sedges dominate the site with a mixture of smaller forbs and shrubs. This community type also corresponds with Motzkin's (1994) Group III *Carex interior-C. leptalea-C. flava* type. Sites Shmulsky Shrub and Schenob Brook Bartholomew appear to be "Shrub Fen" (after Weatherbee and Crow, 1992) or "Rich Shrub Fen" (after Reschke, 1990) communities as they are in a somewhat transitional state where shrubs are beginning to dominate. Schenob Brook Central would be described as a "Calcareous Seep Community" where cold calcareous water seeps to the surface and forms small rivulets (after Weatherbee and Crow, 1992). The vegetation at Schenob Brook North appears to be more closely related to a "Lake Basin Graminoid Fen Community" even though this area may not have formed from a former lake. It is characteristic of a community that has peat accumulation and elements of graminoid fens, shrub fens, and bog vegetation (after Weatherbee and Crow, 1992). Schenob Brook

South is the only site that would be considered a non-rich fen. According to Reschke (1990), this site would be called a “Medium Fen” containing many common wetland species but still having a minerotrophic groundwater influence.

Species Composition: *A Priori* Groupings

Three categories of plants were established to identify species with different suspected needs for or tolerances to calcium. All of the selected species occurred in fens with elevated calcium levels but some species were notably absent or occurred at only low frequencies when lower levels of calcium existed. Species placed in Category 1 included species that were considered to be either “rare” or “indicator species” in the high-calcium wetlands (Table 4.1). It is assumed that these species may be the most important to study since they occur in limited amounts and usually only in highly calcareous environments. Six species in this category are listed as endangered, threatened, or of special concern in Massachusetts (MNHESP, 2004). Category 2 includes species that are considered “dominant” or “characteristic” species in the high-calcium fens. Although they are not endangered, threatened, or of special concern, these species are important because they are abundant and provide the dominant vegetative makeup of the calcareous fens and appear to occur only at more elevated calcium levels. Category 3 contains both calciphile and non-calciphile species that appear to occur independent of environmental calcium levels in calcareous fens. Two of these species (*Carex granularis* and *Dasiphora fruticosa*) are known calciphiles (Gleason and Cronquist, 1991), and because they are generalists as to their calcium selectivity, will provide a useful comparison to calcium specialists (from Categories 1 and 2) with respect to calcium accumulation in tissues. The remaining three species in this category (*Symphyotrichum puniceum* var. *puniceum*,

Scientific Name [†]	Common Name	Status MA[+]
Category 1		
Carex sterilis [1, 2, 3]	Dioecious sedge	T
Carex tetanica [2, 3]	Rigid sedge	SC
Cypripedium parviflorum var. *parviflorum* [2]	Lesser yellow ladies slipper	E
Equisetum fluviatile [1]	Water horsetail	
Equisetum scirpoides [2, 3]	Dwarf scouringrush	SC
Eriophorum gracile [2]	Slender cottongrass	
Geum rivale [1, 3]	Water avens	
Juncus nodosus [1, 2]	Knotted rush	
Lobelia kalmii [1, 2, 3, 4]	Kalm's lobelia	
Petasites frigidus var. palmatus [2, 3]	Arctic sweet coltsfoot	E
Solidago uliginosa [1, 4]	Northern bog goldenrod	
Spiranthes romanzoffiana [1, 2]	Hooded ladies tresses	E
Category 2		
Carex flava [1, 2, 3]	Yellow sedge	
Carex hystericina [1, 2, 3]	Bottlebrush sedge	
Carex interior [2, 3]	Inland sedge	
Carex leptalea [1, 2, 3]	Bristly stalked sedge	
Juncus brachycephalus [1]	Smallhead rush	
Packera aurea [1]	Golden ragwort	
Parnassia glauca [1, 2, 3, 4]	Fen grass of Parnassus	
Solidago patula [1, 2, 3]	Roundleaf goldenrod	
Category 3		
Carex granularis [1, 3, 4]	Limestone meadow sedge	
Dasiphora fruticosa ssp. *floribunda* [1, 2, 3, 4]	Shrubby cinquefoil	
Symphyotrichum puniceum var. *puniceum* [1]	Purplestem aster	
Symplocarpus foetidus [1]	Skunk cabbage	
Thelypteris palustris [1, 2, 3]	Eastern marsh fern	

Table 4.1. *A priori* species category groupings. Category 1: Rare or indicator species in fens with high calcium levels. Category 2: Dominant or characteristic species in fens with high calcium levels. Category 3: Species that are common fen species across a range of calcium levels.

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[1] Picking and Veneman (2004)
[2] Kearsley (1999)
[3] Motzkin (1994)
[4] Species known for growth in calcareous regions by Gleason and Cronquist (1991)
[†] All nomenclature follows USDA (2007)
[+] Species status derived from MNHESP (2004). E = Endangered, T= Threatened, SC=Special Concern

Symplocarpus foetidus, and *Thelypteris palustris)* are common wetland species (non-calciphiles) that show no selectivity as to environmental calcium levels. Studying this group will be useful in determining whether calcium tissue storage patterns are unique to calciphiles or if non-calciphiles also show similar responses to environmental calcium levels.

Of the 25 species in the *a priori* categories, 20 were observed in the study plots. Species not observed were *Cypripedium parviflorum* Salisb. var *parviflorum, Equisetum scirpoides* Michx., *Eriophorum gracile* L., *Petasites frigidus* (L.) Fries var. *palmatus* (Ait) Cronq., and *Spiranthes romanzoffiana* Cham. The absence of these species is not surprising as they represent the rarest of the species from the category as three are endangered and one is of special concern in Massachusetts (Table 4.1). A cottongrass similar to *E. gracile, Eriophorum viridicarinatum* was observed. Similarly, the orchid relative of *S. romanzoffiana*, *Spiranthes cernua* was observed.

Species Richness and Turnover

Among each plot, species richness varied from 16 species at Jug End South (JS2) to 52 species at Shmulsky Open (SO3) (Table 4.2). Thirty species occurred at each plot on average. Among all of the sites, Schenob Brook South had the lowest total species richness with 37 species and Jug End North had the highest with 79 species (Table 4.2). Picking (2002) studied some of the same wetland locations (including the Shmulsky fen) and recorded species richness values from 55 to 80 species per 15-m^2 sampling plot. Picking's "Station 2" corresponded to a location very close to the Shmulsky Open site (D. Picking, Pers. Comm.) and at this station, Picking observed a maximum of 68 species per 15-m^2 plot. Thus, it was not surprising to see that Shmulsky Open had the highest species richness of any other study plot, and when all three plots at Shmulsky Open were combined, species, richness was almost identical to Picking's maximum value at 67 (Table 4.2). The general species richness values for the other sites compared with fen richness values reported by Bedford et al. (1999) who observed between 26 to 78 species per 25-m^2 plot in fens of New York State.

Table 4.2. Species richness at each study plot and cumulative for each study site.

Site	Plot	Richness	Site Total
Jug End North	JN1	32	79
	JN2	34	
	JN3	20	
Jug End South	JS1	23	39
	JS2	16	
	JS3	27	
Bartholomew	SBB1	32	50
	SBB2	31	
	SBB3	34	
Schenob Central	SBC1	22	43
	SBC2	28	
	SBC3	26	
Schenob North	SBN1	29	52
	SBN2	38	
	SBN3	33	
Schenob South	SBS1	17	37
	SBS2	18	
	SBS3	25	
Shmulsky Open	SO1	34	67
	SO2	42	
	SO3	52	
Shmulsky Shrub	SS1	37	57
	SS2	33	
	SS3	40	

For each site, combined species richness was always greater than when viewed as individual plots. This trend was the most extreme at Jug End North where total site richness was more than double the maximum value observed at an individual plot (Table 4.2). This finding implies that few species are shared among those plots, which is indicative high β-diversity and high species turnover. Similarity indices were developed to explore this trend further. Species similarity was compared among plots (Table 4.3) and among sites (Table 4.4). Values closer to 0 represent extreme dissimilarity and values closer to 1 indicate identical species composition.

On average, plots shared 38.5% of the same species. This value is quite low but makes sense considering that some plots occurred in sites that were geographically separated and that were also determined to be different fen types. Replicate plots shared more species with each other than with plots in other sites (Table 4.3). On average, replicate plots within the same site shared 59% of the same species. The lowest similarity observed within replicate plots was between Jug End North-3 and Jug End North-2 where they shared only 44% of their species. The strongest overall similarity was observed between two replicate plots, Schenob Brook North-3 and Schenob Brook North-2, in which they shared 79% of their species. Much lower similarity existed when non-replicate plots from different sites were compared. Most notably, the plots at Schenob Brook South did not share one similar species with Schenob Brook Central-1. This finding is interesting as these two sites occurred in the same geographic location; however, Schenob Brook Central was characterized as a "Calcareous Seep Community" and Schenob Brook South was characterized as a "Medium Fen". These two types of fens have very different geochemical properties, and so it is not surprising that these two

Table 4.3. Species similarity matrix as measured by 1 minus the Sorensen/Bray-Curtis distance matrix. Bold numbers indicate within-site comparisons. Values multiplied by 100 indicate the percent species shared between associated plots.

SO1	SO2	SO3	SS1	SS2	SS3	SBB1	SBB2	SBB3	SBS1	SBS2	SBS3	SBC1	SBC2	SBC3	SBN1	SBN2	SBN3	JN1	JN2	JN3	JS1	JS2	JS3	Plot
1	**0.55**	**0.6**	0.59	0.48	0.51	0.3	0.28	0.32	0.24	0.23	0.27	0.25	0.26	0.37	0.38	0.33	0.27	0.45	0.47	0.37	0.39	0.4	0.39	SO1
	1	**0.7**	0.61	0.53	0.49	0.43	0.41	0.53	0.24	0.27	0.27	0.19	0.4	0.38	0.39	0.43	0.37	0.45	0.47	0.39	0.43	0.34	0.46	SO2
		1	0.61	0.54	0.54	0.45	0.43	0.47	0.2	0.37	0.34	0.22	0.3	0.41	0.4	0.38	0.35	0.45	0.51	0.36	0.45	0.35	0.48	SO3
			1	**0.69**	**0.65**	0.32	0.32	0.28	0.26	0.33	0.32	0.17	0.28	0.35	0.36	0.32	0.26	0.4	0.42	0.35	0.37	0.3	0.31	SS1
				1	**0.6**	0.25	0.31	0.27	0.28	0.31	0.34	0.15	0.26	0.34	0.32	0.28	0.24	0.39	0.39	0.3	0.32	0.2	0.3	SS2
					1	0.22	0.37	0.41	0.21	0.21	0.31	0.29	0.32	0.33	0.35	0.33	0.27	0.47	0.43	0.3	0.29	0.36	0.45	SS3
						1	**0.7**	**0.55**	0.33	0.24	0.32	0.15	0.4	0.38	0.39	0.37	0.4	0.34	0.42	0.38	0.47	0.29	0.34	SBB1
							1	**0.68**	0.25	0.16	0.21	0.19	0.41	0.35	0.37	0.38	0.38	0.47	0.52	0.35	0.56	0.38	0.48	SBB2
								1	0.24	0.15	0.2	0.21	0.45	0.37	0.38	0.33	0.39	0.51	0.56	0.41	0.53	0.36	0.56	SBB3
									1	**0.51**	**0.57**	0	0.13	0.19	0.48	0.29	0.28	0.28	0.2	0.16	0.25	0.18	0.23	SBS1
										1	**0.51**	0	0.09	0.18	0.3	0.21	0.2	0.2	0.19	0.16	0.24	0.18	0.09	SBS2
											1	0	0.15	0.2	0.41	0.29	0.24	0.21	0.17	0.13	0.21	0.1	0.12	SBS3
												1	**0.48**	**0.46**	0.27	0.4	0.4	0.18	0.18	0.1	0.22	0.37	0.29	SBC1
													1	**0.74**	0.39	0.58	0.62	0.36	0.35	0.25	0.47	0.41	0.33	SBC2
														1	0.47	0.56	0.61	0.37	0.4	0.22	0.53	0.48	0.34	SBC3
															1	**0.63**	**0.58**	0.32	0.38	0.24	0.5	0.31	0.32	SBN1
																1	**0.79**	0.31	0.31	0.17	0.39	0.37	0.31	SBN2
																	1	0.3	0.33	0.15	0.43	0.33	0.33	SBN3
																		1	**0.6**	**0.53**	0.5	0.45	0.6	JN1
																			1	**0.44**	0.56	0.44	0.59	JN2
																				1	0.37	0.22	0.3	JN3
																					1	**0.56**	**0.48**	JS1
																						1	**0.51**	JS2
																							1	JS3

communities had very different species compositions. On average, plots had only 34% species in common with plots that occurred at different sites.

When plots are combined and species similarity is reported on the site level, similar trends are observed as with plots (Table 4.4) although species similarity is higher overall. Highest similarity within geographic locations was observed at Shmulsky and at Jug End where these sites had 69% and 73% species in common within each location, respectively. The Schenob sites had fewer species in common. Schenob Brook Central had only 18% of its species in common with Schenob Brook South, and this distribution corresponded to the comparison of similarity among plots. Of the Schenob sites, the greatest number of shared species were between Schenob Brook North and Schenob Brook Central (60%). Even though Schenob Brook North had vegetation similar to a "Lake Basin Fen" and Schenob Brook Central was a "Calcareous Seep", they both occurred adjacent to each other and in similar landscape positions.

Table 4.4. Species similarity matrix as measured by 1 minus the Sorensen/Bray-Curtis distance matrix comparing overall sites ($58.8m^2$). Bold numbers indicate within-location comparisons. Values multiplied by 100 indicate the percent species shared between associated sites.

SO	SS	SBB	SBS	SBC	SBN	JN	JS	Site
1	**0.69**	0.58	0.45	0.4	0.47	0.64	0.6	SO
	1	0.49	0.37	0.36	0.39	0.59	0.5	SS
		1	**0.33**	**0.37**	**0.42**	0.65	0.61	SBB
			1	**0.18**	**0.37**	0.25	0.27	SBS
				1	**0.6**	0.4	0.49	SBC
					1	0.41	0.51	SBN
						1	**0.73**	JN
							1	JS

The lack of similarity within sites indicates high species turnover as is associated with multiple environmental gradients (Picking, 2002). Although rare plants are limited by factors other than nutrient availability, (i.e. competition, unique pollination strategies, etc.) significant differences in environmental gradients over small areas allow for more species with different niches to establish. In Chapter 3, it was determined that there were strong differences within and among sites. Not only were the primary fen-defining elements (e.g. calcium, magnesium and pH) significantly different among the sites, but also differences were observed in hydrology, groundwater upwelling, percent organic matter, dissolved potassium, as well as DTPA soil iron and total soil nitrogen. These differences among sites may contribute to differences in plant species composition however, these additional findings suggest that there may also be significant environmental differences within sites to account for the high species dissimilarity there. High species turnover in calcareous fens also was observed by Picking (2002); however, Picking's plot and site similarity were much greater than observed in this study. This difference may have been because Picking's (2002) study involved comparisons within one fen region (the Shmulsky location) whereas the current study viewed three different geographic locations.

Bedford et al. (1999) reported species richness to be correlated to the ratio of total soil nitrogen to total soil phosphorus. When conditions become more nutrient rich (increasing phosphorus levels), species that thrive under nutrient-rich conditions out-compete the smaller, less robust species (Drexler and Bedford, 2002). The opposite occurs in phosphorus-limited environments, allowing many more specialized plants to thrive. Data from the current study is in agreement with this theory and indicates that these fen wetlands are limited in phosphorus. As phosphorus levels decreased, the ratio of nitrogen to phosphorus became larger, and the number of species observed increased (r^2=0.385, p=0.0012; Figure 4.1).

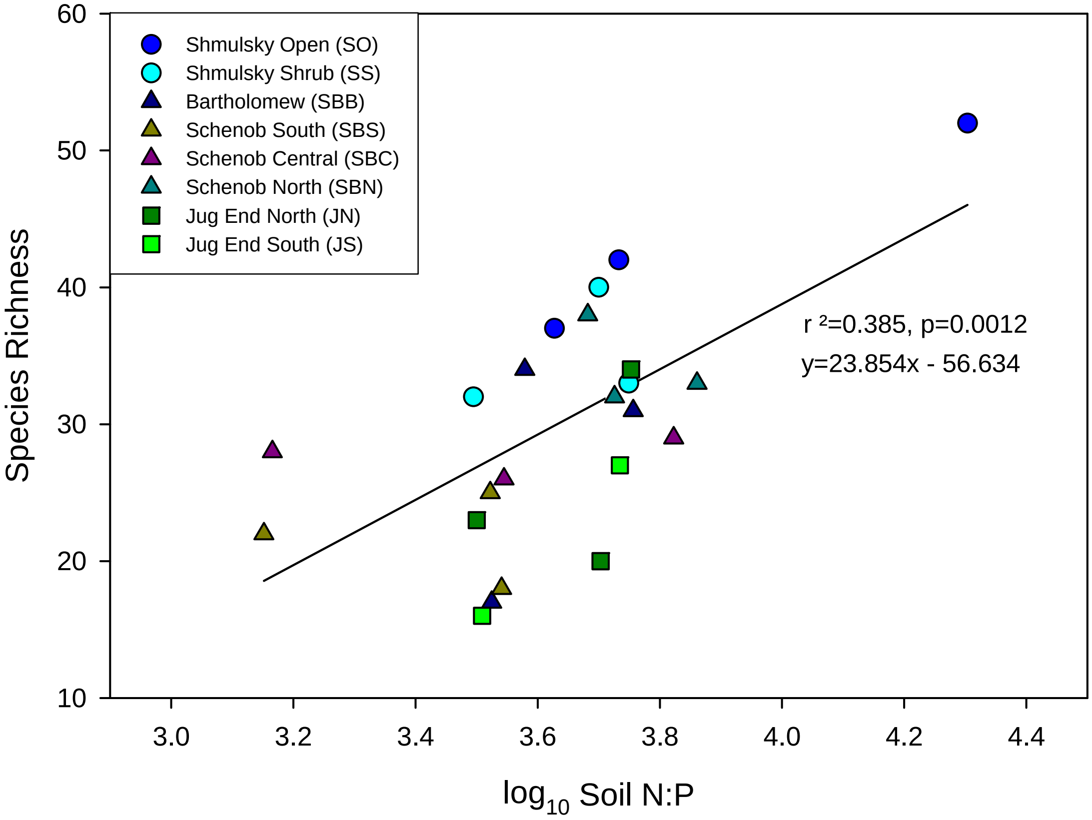

Figure 4.1. The relationship between soil N:P (log of ratio of total nitrogen to AB-DTPA extractable phosphorus) and species richness at the 24 study plots.

These results concur with what Bedford et al. (1999) observed in a study of New York fens. It was interesting to see that this trend was the same because the current study used a different technique for measuring soil phosphorus as compared to Bedford et al. (1999) and other previous related studies. It did not seem ecologically meaningful to measure total phosphorus in the soil (as Bedford did) as much of that is not readily available to plants. Instead, the AB-DTPA extraction method was selected in this study because it is buffered at a pH of 7.6 so that it is a realistic representation of potentially available phosphorus in calcareous fen environments (Jones, 2001). Total nitrogen was measured as all common forms of nitrogen (organic, nitrate, and ammonium) could be readily available depending on mineralization and denitrification rates. Thus, absolute N:P ratios are higher than Bedford's because measurements of extractable phosphorus are naturally lower than total phosphorus. Even though the ratios differ, the relationships to species richness are similar.

Conclusions

Even though study plots were established before the majority of vegetation emerged in April, the species list generated from this study indicates that many common species of calcareous fens were observed. Out of the total 133 species observed, 20 were of the 25 species selected in the *a priori* category groupings. The eight sites represented several different types of calcareous fens as described from previous literature, with only one being considered a non-rich fen. Species richness varied among the 19.6m^2 plots from a low of 16 species to a maximum of 52 species and corresponds to expected richness values for vascular species of calcareous fens. Species richness was significantly higher when plot richness was combined on the site level. Similarity matrices confirmed that on average only 59% of species are in common between adjacent plots. This high species turnover may be indicative of the multiple environmental gradients present in the sites as determined in Chapter 3. Species richness increased log-based linearly as did the N:P in the soil, suggesting that these wetlands are limited by phosphorus and that large, strong nutrient-loving competitors are absent and greater numbers of smaller, less nutrient demanding species are able to coexist.

CHAPTER V

VEGETATION AND ENVIRONMENTAL GRADIENTS

Introduction

The plant communities associated with calcareous fens have been described as some of the rarest in North America (Eggers and Reed, 1987). The species that occur in them are specifically adapted to the unique geochemical conditions, namely elevated soil and water pH, calcium, and magnesium (Slack et al., 1980; Vitt and Chee, 1990; Komor, 1994; Motzkin, 1994; Kearsley, 1999; Picking and Veneman, 2004; Bowles et al., 2005). Motzkin (1994), in a study of western New England fens, established species groupings that have been since used and validated by other researchers (Kearsley, 1999; Picking and Veneman, 2004) to describe the vegetation. These species groupings were correlated significantly to differences in environmental properties, with depth to the mineral soil being the most important, but also with pH, calcium, and magnesium in the fen surface waters. Vitt and Chee (1990) reported a strong correlation between vegetation composition and fen pH, calcium, magnesium, and electrical conductivity in their study of water and peat chemistry in fens of Alberta, Canada. Generalizations have been made as to the types of vegetation communities that are associated with certain levels of water calcium and pH, although few studies have used these findings to focus on the requirements and distributions of individual species in relation to these geochemical factors.

In this chapter, ordination analyses are used to discern where differences in species abundance patterns may exist in relation to site geochemical properties. By identifying trends in environmental calcium levels and identifying where species-abundance patterns correlate to these underlying calcium gradients it may be possible to determine if certain species increase in abundance as calcium levels increase or whether species occur only above a specific calcium threshold. For these species, calcium ranges and thresholds were identified as to the suspected environmental calcium levels required by these plants.

Materials and Methods

Data Analysis

The Bray-Curtis (Polar) Ordination with three axes (following variance-regression endpoints (Beals, 1984), Sørenson's distance measure and Euclidean projection geometry) was used to view patterns in species distributions and environmental variables in relation to the study plots. The ordinations were carried out using multivariate statistical software (PC-ORD Ver. 5, McCune and Mefford, 1999). A general relativization was performed prior to these analyses to normalize the data and then a Mantel Test (Douglas and Endler, 1982) was conducted to compare the original data matrix to the transformed data matrix. A p=<0.05 assured that the transformed matrix did not deviate in trends with the original data structure.

The overlay function in PC-ORD was used to assess the correlations that existed between one set of raw variables and the underlying ordination. For the environmental data ordination, this procedure was accomplished by setting the normalized environmental variables as the main matrix and then using the "overlay" function to view the raw vegetation abundance data (from a secondary matrix) as they correlated to the underlying ordination scores. Similarly, to view the strong underlying environmental gradients, the raw environmental data were overlaid on the main transformed environmental matrix, and resulting axes of correlation and coefficients were recorded.

The same procedure was used for the vegetation ordination. Results of the correlation were assessed visually in the form of joint plots and scatter plots with a Pearson's linear correlation coefficient (r) calculated for each variable. The direction of the vector in the joint plot indicates the direction of the correlation with the underlying axis to which it is parallel. The length of the vector indicates the strength (Pearson's r) of the correlation relationship.

The locations of the 24 study plots were chosen based on the expected vegetation that they would contain, and thus inferential statistics cannot be applied to analyses including them as individual points, as the analysis would be violating the assumptions that the variables are independent of each other. Nevertheless, the 24 plots were ordinated simply to view trends in the similarity among them in relation to their vegetative community structure. More reliable inferences were derived from the ordination of the environmental data, in which the plots were selected without bias with respect to their substrate characteristics.

The environmental data set used in the ordination of the environmental properties included 40 environmental variables organized by the 24 plots. For water chemistry data, the mean, median, minimum, and maximum concentration for each element were included if those elements where shown to change across the growing season (Chapter 3). For elements with no seasonal differences, only the means were used. Soil properties were measured only once, and thus one single representation of each soil measure was used in the data set. For the ordination of the vegetation, species that occurred in fewer than three plots were removed. This reporting resulted in a matrix of the abundance measurements of 85 species organized by the 24 plots. These 85 species are noted in Appendix F.

For species where the ordination indicated they were correlated to an environmental gradient, a regression analysis (SAS Ver. 8.1.3 - SAS Institute, 2004) compared the abundance of that species to that environmental property. Data were transformed by log or square root for normality prior to analysis. Graphs present results for untransformed data; however, the statistics are based on the normalized values.

Species Distribution Patterns

Species that appeared to change significantly along the axes where calcium gradients were significant were considered gradient species, and the numerical range of water calcium in which it occurred was noted. Species whose abundance correlated significantly with calcium gradients (even if it was a low correlation) but occurred only at an extreme of the ordination (where calcium was highest), were considered threshold species. This distribution implied that these species occurred only when calcium values reached a certain elevated level. Once a species of this threshold nature was identified, the lowest calcium values at which the species occurred was reported.

Results and Discussion

Vegetation Community Analysis

The distribution patterns of the eighty-five species generated a matrix in which plots were arranged in "species space" (Figure 5.1). Plots are displayed only as they were arranged on Axis 1 and Axis 3 due to the very low significance of Axis 2 (r^2=0.020). Collectively, Axis 1 and Axis 3 explain 29.3% of the variation in species distribution

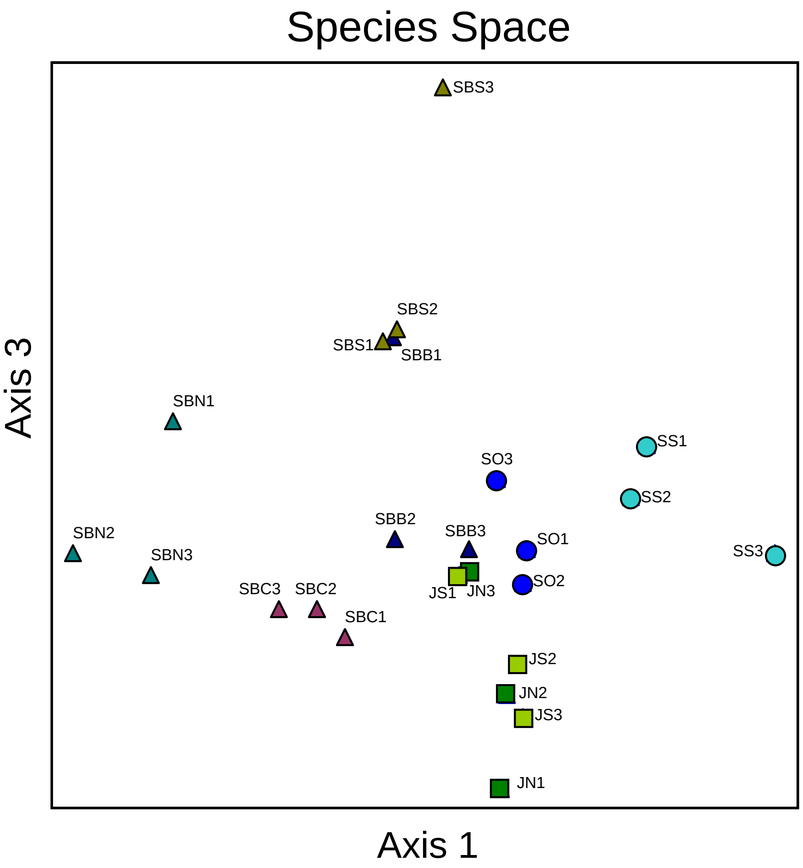

Figure 5.1. Two-dimensional (Axis 1 and Axis 3) ordination of study plots represented in species space. r^2= 0.293 for all variables shown (Axis1: r^2=0.123, Axis3: r^2=0.170). Larger distances between plots represent greater dissimilarity in species composition at those plots. Symbols are coded by location (shapes), and site (colors). Refer to legend in Figure 4.1 for full clarification.

patterns. The axes represent the range of ordination scores assigned to each plot as computed from the vegetation abundance at each of those plots. Thus plots that occurred farthest from each other had the most dissimilar species community structure, and those that occurred closer to one another were more similar in plant species populations. Not surprisingly, the three plots from each study site were grouped closer to each other than they were to plots in other sites. Likewise, the sites that occurred within the same location were grouped closer to each other than to those from different locations.

Schenob Brook Bartholomew, Shmulsky Open and Schenob Brook Central appeared to be quite similar in species community structure as they are clustered in the center of the diagram (Figure 5.1). The Jug End sites (Jug End North and Jug End South) are separated on Axis 3, and are on the opposite end of Axis 3 to Schenob Brook South. Schenob Brook North and Shmulsky Shrub are separated on opposite ends of Axis 1.

These findings correspond with the Sørenson's distance matrices calculated from presence-absence data for the plots (Chapter 4). Plots that shared very few species by Sørenson's distance matrix were also more distant from each other in species space. In addition, the sites that were very dissimilar by Sørenson's distance matrix, Jug End North and Schenob Brook South, are on opposite ends of Figure 5.1. Sites that were very similar, Shmulsky Open and Shmulsky Shrub, and Jug End North and Jug End South are clustered nearby each other in Figure 5.1. The Bray-Curtis ordination uses Sørenson's distance matrix to calculate its ordination scores, and so by definition, these findings are consistent.

Individual species with linear correlations ($r^2 > 0.4$) to the underlying axes are presented in the joint plot in Figure 5.2 and are listed with significant axes of correlation and Pearson’s r values in Table 5.1. Because this ordination is in "Species Space" these strongly correlated species are largely responsible for the underlying plot distribution in the resulting ordination. Two major, and three minor groups of species can be identified as being responsible for the separation among the plots. Six species (*Alnus serrulata*, *Calopogon tuberosus*, *Glyceria grandis*, *Larix laricina*, *Pogonia ophioglossoides*, and *Toxicodendron vernix)* group on the left side of Figure 5.2 and are all negatively correlated with Axis 1. These species were thus more common at Schenob Brook North

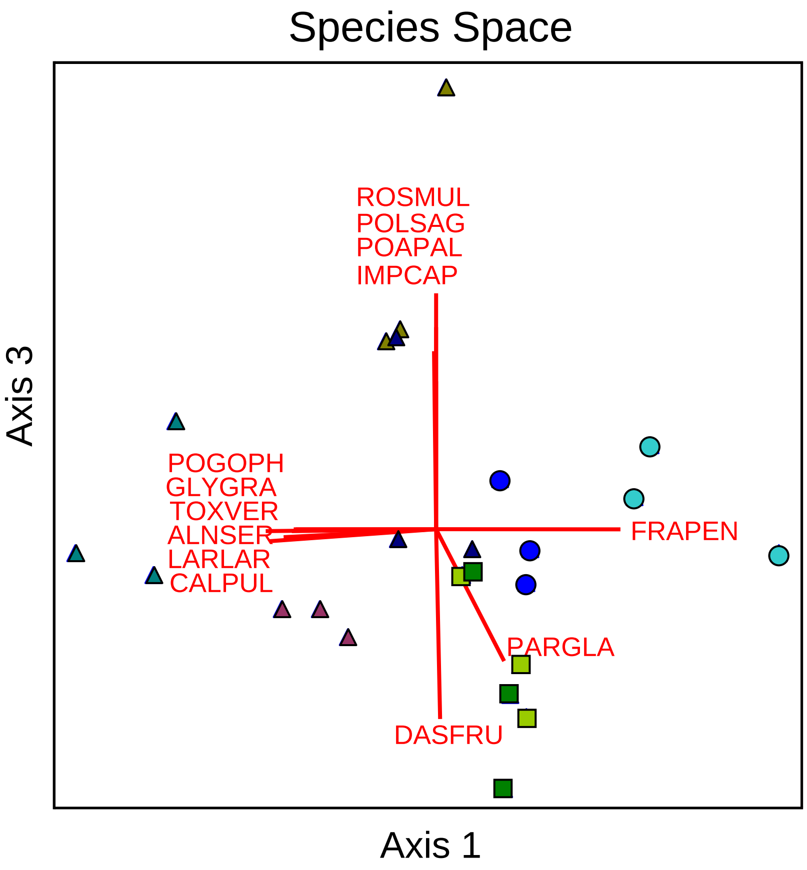

Figure 5.2. Joint plot of species with linear correlations ($r^2 > 0.4$) to species space (see Table 5.1). Symbols are coded by location (shapes), and site (colors). Refer to legend in Figure 5.1 for full clarification.

and less abundant at Shmulsky Shrub. By contrast, *Fraxinus pennsylvanica* is positively correlated with Axis 1 and has the opposite distribution to the previously discussed group.

The second largest group includes the four species, *Impatiens capensis*, *Poa palustris*, *Polygonum sagittatum*, and *Rosa multiflora*. These are all positively correlated with Axis 3 and were more common in Schenob Brook South and less abundant in the Jug End sites. Opposite these species are *Dasiphora fruticosa* and *Parnassia glauca*, which are

Table 5.1. Species with correlations of $r^2 > 0.4$ to the underlying vegetation matrix. This table corresponds to Figure 5.2.

Species	ID Code	Axis	Pearson's r
Alnus serrulata	ALNSER	1	-.659
Calopogon tuberosus var. *tuberosus*	CALTUB	1	-.639
Dasiphora fruticosa	DASFRU	3	-.736
Fraxinus pennsylvanica	FRAPEN	1	.724
Glyceria grandis	GLYGRA	1	-.633
Impatiens capensis	IMPCAP	3	.821
Larix laricina	LARLAR	1	-.698
Parnassia glauca	PARGLA	1, 3	.439, -.613
Poa palustris	POAPAL	3	.715
Pogonia ophioglossoides	POGOPH	1	-.688
Polygonum sagittatum	POLSAG	3	.758
Rosa multiflora	ROSMUL	3	.652
Toxicodendron vernix	TOXVER	1	-.638

negatively correlated with Axis 3 and were more abundant in the Jug End sites. *Parnassia glauca* is unique however because it is also positively correlated with Axis 1.

When the raw environmental data were overlaid on the underlying vegetation matrix (Figure 5.3, Table 5.2), four main groups of environmental correlations were observed. pH (water and soil), and minimum piezometric gradients were correlated negatively with Axis 3. A second group included numerous soil measurements (percent organic matter and soil nitrogen, phosphorus, iron, potassium and magnesium), which were positively correlated with Axis 3. These correlations were not surprising since an increase in pH usually results in a decrease in available iron and phosphorus. On Axis 1 a third group was correlated negatively and included median water potassium and weeks of constant saturation in the spring. In addition to Axis 3, a fourth trend was observed in which minimum water calcium was positively correlated with Axis 1 in addition to being negatively correlated to Axis 3.

Species and environmental properties that were correlated on the same axis suggest

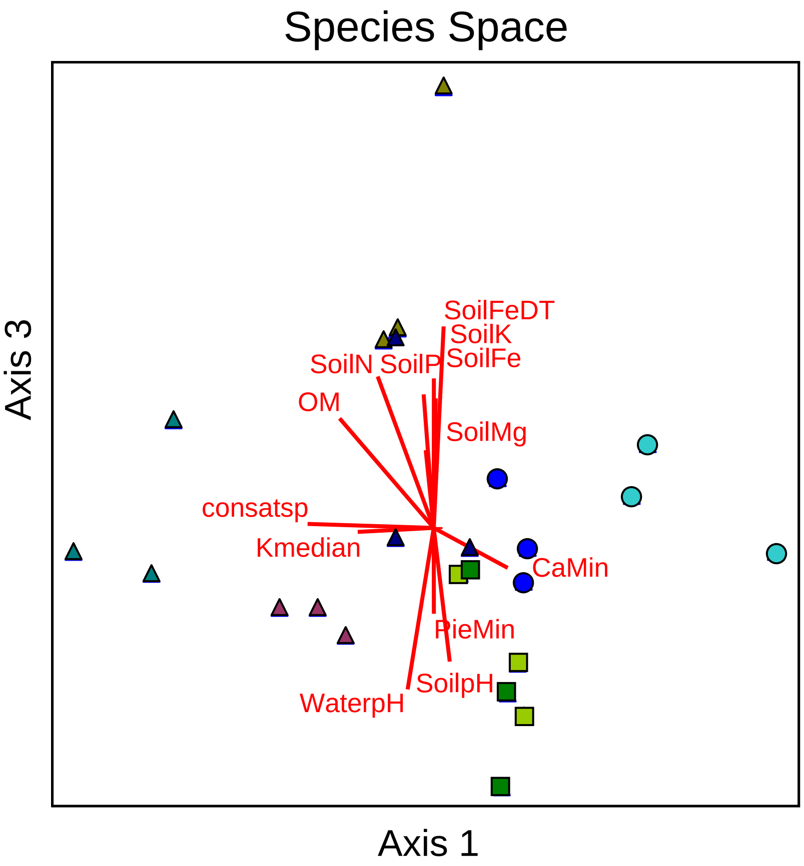

Figure 5.3. Joint plot of environmental variables with linear correlations ($r^2 > 0.2$) to species space (see Table 5.2). Symbols are coded by location (shapes), and site (colors) (see Figure 5.1).

that those species were responding to those particular environmental properties. *Alnus serrulata*, *Calopogon tuberosus*, *Glyceria grandis*, *Larix laricina*, *Pogonia ophioglossoides*, and *Toxicodendron vernix* were all negatively correlated with Axis 1, in which "weeks of constant saturation in the spring" was also. It is not surprising that these species would be responding to a hydrologic gradient as all of these plants are considered to be facultative wetland to obligate wetland species (USDA, 2007). What is more interesting however is that many of these species are associated with “bog vegetation”

Table 5.2. Ordination results for environmental variables with linear correlations with $r^2 > 0.2$ to the underlying vegetation matrix. This table corresponds to Figure 5.3.

Variable	ID Code	Axis	Pearson's r
Calcium Minimum – Water	CaMin	1, 3	.485, -.336
Groundwater gradients, minimum	PieMin	3	-.498
Iron – Soil	SoilFe	3	.618
Iron DTPA – Soil	SoilFeDT	3	.758
Magnesium - Soil	SoilMg	3	.475
Nitrogen - Soil	SoilN	3	.658
Organic matter – %	OM	1, 3	-.517, .558
pH – Soil	SoilpH	3	-.617
pH – Water[1]	WaterpH	3	-.681
Phosphorus - Soil	SoilP	3	.611
Potassium – Soil	SoilK	3	.654
Potassium Median – Water	Kmedian	1	-.466
Saturation – spring, Weeks of continuous	consatsp	1	-.601

(Weatherbee, 1996; Mitch and Gosselink, 1993). This association would be accurate as bog areas are continually inundated and species that occur in them are highly adapted to such wet and potentially anoxic conditions. In addition, the area where these species were more abundant, Schenob Brook North, (but also at Schenob Brook Central) is similar to the "lake basin" type fen (Chapter 4) that had a large amount of organic matter accumulation and where the water table never dropped below the surface (Chapter 3) and where it is not uncommon for bog vegetation to be present.

Impatiens capensis, *Poa palustris*, *Polygonum sagittatum*, and *Rosa multiflora* were all correlated positively with Axis 3. The same relationship was observed for the percent organic matter and the soil chemistry. These findings imply that plots where these species were more abundant had greater levels of these elements and also had lower pH as pH was negatively correlated with Axis 3. Upon further inspection, it is clear that

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[1] Mean, median, minimum and maximum water pH all showed similar correlations to Axis 3. For simplicity, water pH is shown as one line on the joint plot and the mean water pH correlation is reported.

these species were not abundant across the 24 plots, but all only occurred in three or fewer plots mainly in the Schenob Brook South fen. Schenob Brook South was the only fen in this study that was determined to be a "medium fen", indicating that it had a lower average pH and lower calcium levels. Thus these species are a reflection of a wetland with weaker calcareous qualities with elevated primary nutrient levels, elevated iron and lower pH. By contrast sites on the opposite side of Figures 5.2 and 5.3 represent species and environmental characteristics typical of a calcareous fen. Species correlated to this region were *Dasiphora fruticosa* and *Parnassia glauca*.

Dasiphora fruticosa was the only species to be negatively correlated with Axis 3, in which the ordination pattern is strongly controlled by soil and water pH. This suggests that pH may have a strong role in the abundance of this species and that it increases as pH does. *Parnassia glauca* was the only species that was positively correlated to Axis 1 and negatively correlated to Axis 3. These correlations correspond directly to only one environmental property: minimum water calcium. Thus it appears that this species may be dependent upon the minimum calcium value at each plot.

The ordination of the plots in species space revealed interesting relationships among the plots and their associations with species and environmental properties. Only 29.3% of the data were represented by the two axes. While this may appear low, separating 30% - 50% of the “signal” from the “noise” is considered adequate when conducting ordinations on data sets of 20 or more species (McCune and Grace, 2002).

It was anticipated that more species would be strongly associated with environmental calcium levels (the calciphiles), yet only one species (*Parnassia glauca*) showed this pattern and only one other showed a relationship with pH (*Dasiphora fruticosa).* Instead, species significantly correlated with this underlying ordination seemed to be more associated with hydrologic gradients or to lower pH levels. This result seems to imply that by viewing plots in terms of species abundance, a few very strong species respond to hydrologic gradients and the effects of calcium and pH on the vegetation patterns remain largely unexplained. Perhaps the low level of explanation of these underlying axes is unable to detect minor differences within plant distributions and only the larger more obvious differences (i.e., hydrology) are teased out. To circumvent this problem, the environmental properties were themselves ordinated to see how the vegetation compared when it was overlaid on the environmental ordination matrix. This approach allows finer scale differences in the environmental properties to become more apparent since the ordination was driven by those environmental properties.

Environmental Data Analysis

The axes of the ordination for the environmental properties explain quite a bit more variation among plots than did the vegetation ordination. The three-dimensional ordination in environmental space explains 79% of the variation in the data set. Figure 5.4 shows the distribution of the study plots in “environmental space”.

Across all three axes, Schenob Brook North, Jug End South, Shmulsky Shrub and Schenob Brook Bartholomew are clustered in the center of the diagram (Figure 5.4), indicating that those sites are more similar in overall environmental properties as compared to the more distant sites. Shmulsky Open plots 1 and 2 occur on the opposite end of Axis 1 from where the Schenob Brook Central plots occur. Thus on Axis 1 these plots represent extremes in environmental property differences. On Axis 2, Schenob Brook South-3 occurs at the opposite end to Jug End North-1. Axis 3 mainly represents

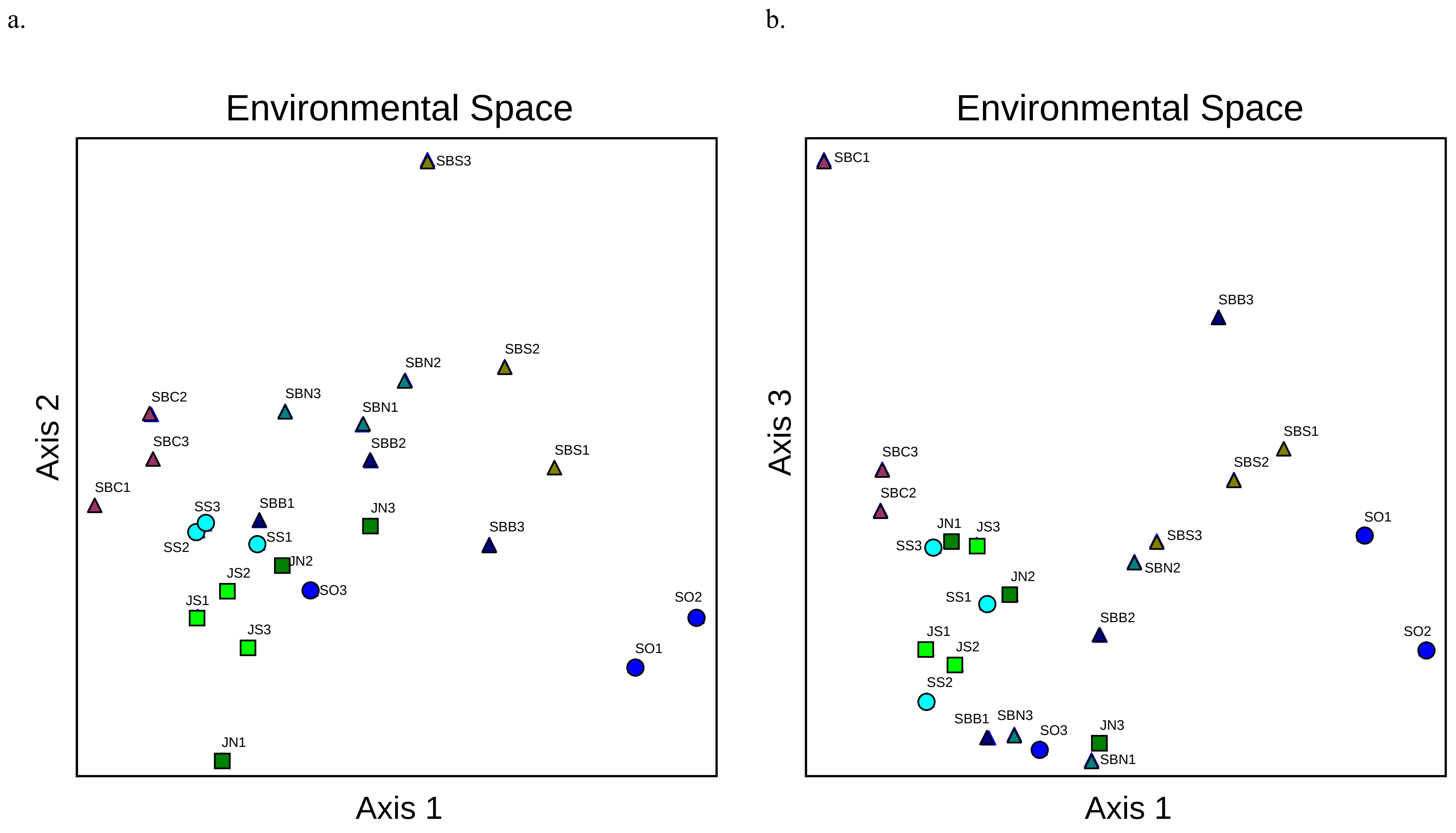

Figure 5.4. Three-dimensional ordination of study plots represented in environmental space. a) Axis 1: r^2=0.531 and Axis 2: r^2= 0.141, b) Axis 1: r^2=0.531 and Axis 3: r^2= 0.120. Cumulative r^2=0.792. Larger distances between plots represent greater dissimilarity in species composition at those plots. Symbols are coded by location (shapes), and site (colors); refer to legend in Figure 4.1.

the distinction of the Schenob Brook Central-1 and Schenob Brook Bartholomew-3 from the other plots.

It is notable that larger differences exist in environmental properties among plots within the same site than were observed by differences in vegetation. Although plots within sites mostly group together, their distribution is farther spread than with the vegetation ordination. Some plots (e.g. Shmulsky Open-3) stand alone far from where the other replicate plots at that site are distributed. In addition, sites that occur within the same geographic location are widely separated. Shmulsky Open and Shmulsky Shrub are at opposite ends of Axis 1. This is interesting as originally it was thought that sites in the same geographic location would have similar environmental properties. These differences are in agreement with findings in Chapter 4 where the overall community characterization indicated that sites within locations were different types of fen communities. These findings are most important in that they validate the concept that the environmental variables, to some degree, were independent of the observable vegetation patterns (as was a previously mentioned concern with analyzing pseudoreplicates).

The significance of the explanatory variables used to generate the environmental matrix are presented in Table 5.3. Individual environmental properties with linear correlations ($r^2 > 0.4$) to the underlying axes are presented in the joint plots in Figure 5.5. Because this ordination is in "Environmental Space" these strongly correlated environmental properties are largely responsible for the underlying plot distribution in the resulting ordination. On Axis 1 (albeit weakly), water iron and ammonium are correlated positively and water pH is negatively correlated. All water calcium and magnesium measurements are correlated positively with Axis 1, and these, along with maximum

Table 5.3. Environmental variables included in the main matrix of the ordination analysis with Pearson's r for axes of correlation (corresponds to Figure 5.5).

Variable	ID Code	Pearson's r for Axis:		
		1	2	3
Ammonium Mean – Water	NMean	.784		
Calcium – Soil	CaSoil	.515		-.630
Calcium Maximum – Water	CaMax	.411	-.707	-.416
Calcium Mean – Water	CaMean	.333	-.727	-.472
Calcium Median – Water	CaMedian	.333	-.706	-.492
Calcium Minimum – Water	CaMin		-.756	-.428
Groundwater gradients, maximum	PieMax	.603		-.410
Groundwater gradients, mean	PieMean		-.304	-.429
Groundwater gradients, median	PieMedian	.559		-.527
Groundwater gradients, minimum	PieMin	-.311	-.309	
Iron – Soil	FeSoil		.716	.309
Iron DTPA – Soil	FeDTPASo	.423	.534	
Iron Maximum – Water	FeMax	.904		
Iron Mean – Water	FeMean	.912		
Iron Median – Water	FeMedain	.897		
Iron Minimum – Water	FeMin	.735		
Magnesium – Soil	MgSoil	.580		-.511
Magnesium Maximum – Water	MgMax	.412	-.596	-.437
Magnesium Mean – Water	MgMean	.391	-.639	-.515
Magnesium Median – Water	MgMedian	.389	-.666	-.466
Magnesium Minimum – Water	MgMin		-.550	-.548
Nitrogen – Soil	NSoil	.524	.643	-.303
Organic Matter – %	OM	.459	.644	
pH – Soil	pHSoil	-.390	-.520	
pH Maximum – Water	pHMax	-.659		
pH Mean – Water	pHMean	-.634		
pH Median – Water	pHMedian	-.604		
pH Minimum – Water	pHMin	-.451	-.417	
Phosphorus – Soil	PSoil		.452	
Phosphorus Mean – Water	PMean		-.505	
Ponding, Total weeks of	Totpond			-.632
Ponding, Weeks of continuous	ConPond			-.589
Potassium – Soil	KSoil		.664	
Potassium Maximum – Water	KMax	.471		
Potassium Mean – Water	KMean	low correlation		
Potassium Median – Water	KMedian	low correlation		
Potassium Minimum – Water	KMin	low correlation		
Saturation – fall, Weeks of continuous	Consatfall	.309		-.562
Saturation – spring, Weeks of continuous	Consatsp		.342	-.437
Saturation, Total weeks of	Totsat		.300	-.571

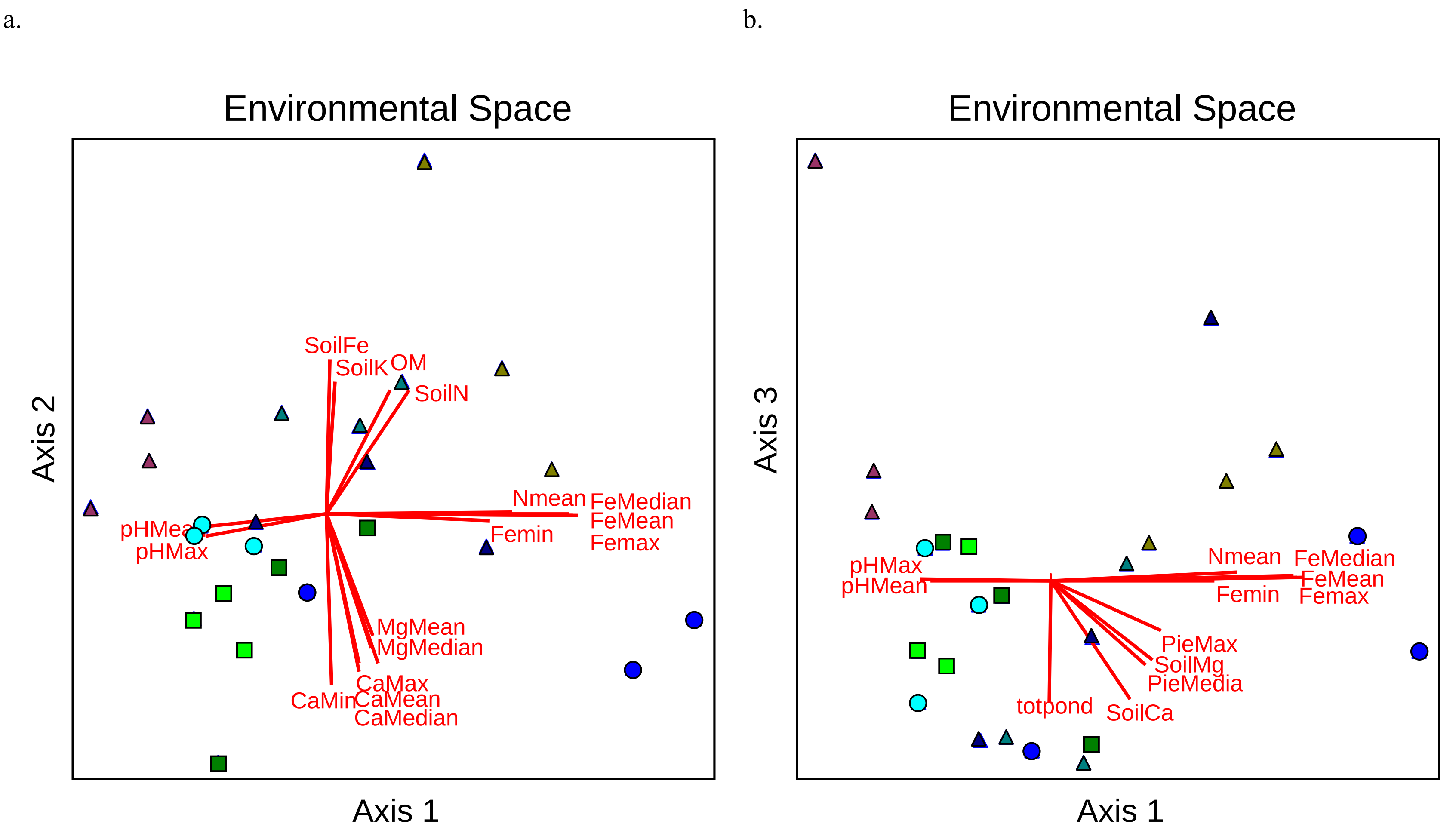

Figure 5.5. Joint plot of environmental properties with linear correlations ($r^2 > 0.4$) to environmental space on a) Axis 1 and Axis 2 and b) Axis 1 and Axis 3 (see Table 5.3). Symbols are coded by location (shapes), and site (colors). Refer to legend in Figure 5.4.

groundwater gradients, are negatively correlated to Axes 2 and 3. The variables strongly and positively correlated with only Axis 2 are soil iron and soil potassium but percent organic matter and soil nitrogen are strongly positively correlated to both Axes 1 and 2. Total weeks of ponding is negatively correlated with Axis 3.

In summary, two major groups of environmental differences appear to separate the plots. Plots that are distributed in either the positive direction on Axes 1 or 2 have higher levels of water and soil iron and nitrogen and by contrast, those in the negative direction on Axis 1 have higher water pH. Although the micaceous schist may be a significant contributor of iron in groundwater, it appears from Figure 5.5a that exchangeable soil iron was less where pH was greatest. This makes sense considering that elevated pH values associated with alkalinity from upwelling groundwater would drive iron into amorphous (non-exchangeable) precipitates. Plots that are distributed in the negative direction of Axis 1 and 3 appear to be strongly influenced by upwelling groundwater that has traveled through carbonate bedrock. This is evidenced by the correlation of these axes to various hydrogeochemical factors, i.e., water pH (Axis 1 only), groundwater gradients, total weeks of ponding and both the water (Axis 1 only) and soil calcium and magnesium.

Axis 1

Species that were correlated on the same axis as underlying environmental variables may be responding to those particular environmental properties (Table 5.4). In the positive direction on Axis 1, where water iron and ammonium cause separation of the plots, species such as *Thelypteris palustris*, *Equisetum fluviatile*, *Juncus effusus*, *Calamagrostis canadensis*, *Populus tremuloides*, and *Sium suave* (Figure 5.6) were more abundant. Although these species may be responding to increased water iron and

Table 5.4. Species correlations ($r^2 > 0.2$) to the underlying environmental matrix (corresponds to Figure 5.6).

Species	ID Code	Pearson's r for Axis:		
		1	2	3
Amphicarpaea bracteata	AMPBRA		.617	
Betula papyrifera	BETPAP		.617	
Calamagrostis canadensis	CALCAN	.492	-.306	
*Carex granularis**	CARGRA		-.494	
Carex stricta	CARSTR		.524	
Coptis groenlandica	COPGRO	-.318		.624
*Dasiphora fruticosa **	DASFRU	-.572	-.444	
*Equisetum fluviatile**	EQUFLU	.690		-.324
Equisetum hyemale	EQUHYE	-.318		.624
Gallium asprellum	GASASP		.617	
Impatiens capensis	IMPCAP		.655	
Juncus effusus	JUNEFF	.419	-.462	
Juniperus communis	JUNCOM	-.318		.624
Muhlenbergia glomerata	MUHGLO		-.448	
Osmunda cinnimonea	OSMCIN			.626
*Packera aurea**	PACAUR		-.462	
*Parnassia glauca**	PARGLA		-.742	
Parthenocissus quinquefolia	PARQUI		.617	
Poa palustris	POAPAL	.409	.574	
Polygonum sagittatum	POLSAG		.538	
Populus tremuloides	POPTRE	.498		
Rubus pubescens	RUBPUB		.500	
Sium suave	SIUSUA	.498		
Sphagnum sp.	SPHAG	-.399		.614
*Thelypteris palustris**	THEPAL	.478		-.307
Trientalis borealis	TRIBOR	-.318		.624
Vaccinium corymbosum	VACCOR	-.322		.523
Viburnum dentatum	VIBDEN		-.487	

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[*] Denotes species from *a priori* category groupings.

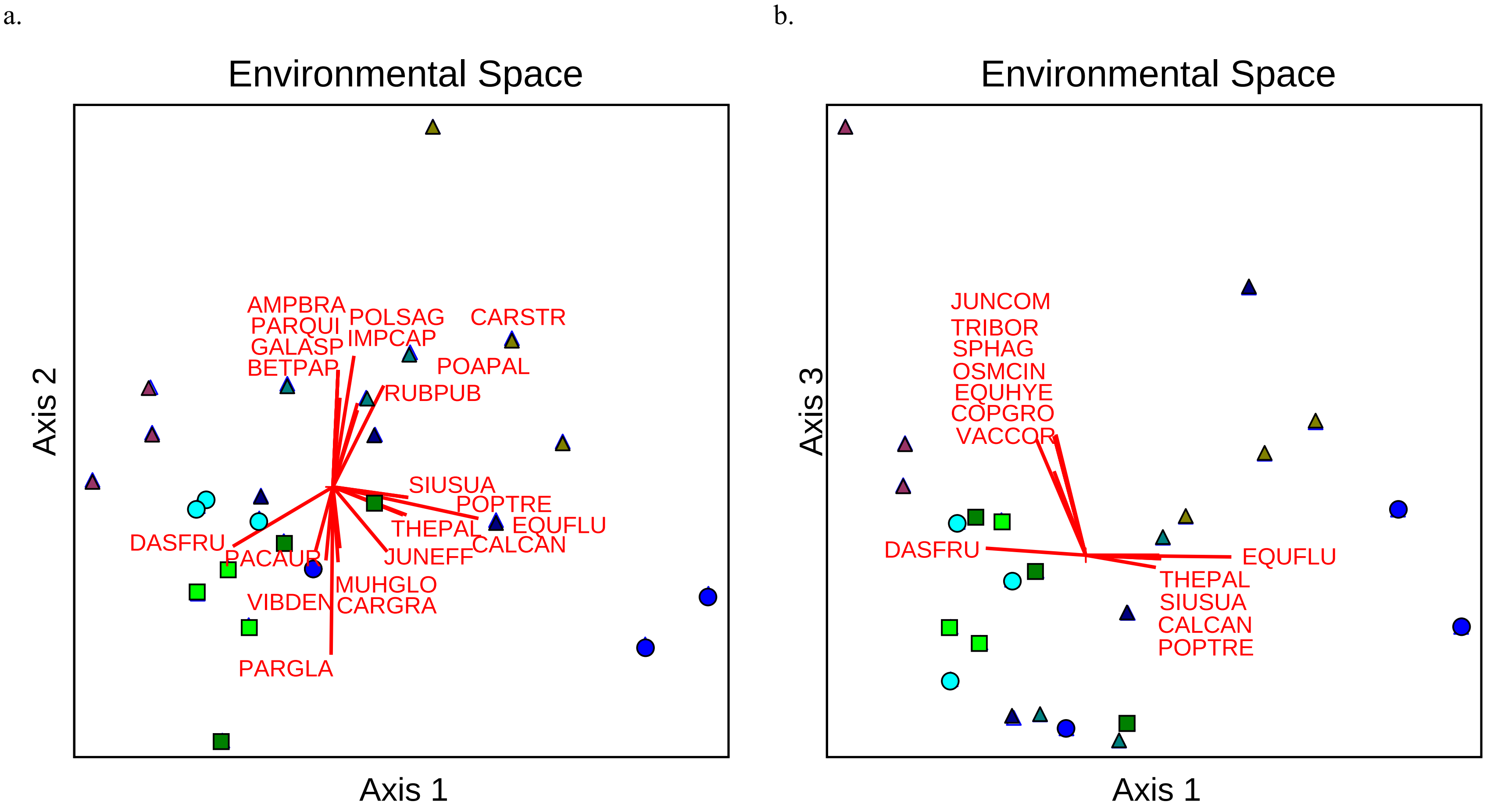

Figure 5.6. Joint plot of species with linear correlations ($r^2 > 0.2$) to environmental space on a) Axis 1 and Axis 2 and b) Axis 1 and Axis 3 (see Table 5.4). Symbols are coded by location (shapes), and site (colors). Refer to legend in Figure 5.4.

ammonium, they may also be responding to lower water pH levels, as water pH had a negative correlation (in the opposite direction) with Axis 1. Only *Dasiphora fruticosa* had a negative correlation with Axis 1, and thus appears to increase as water pH does. Except for *Equisetum fluviatile*, none of the species positively correlated with Axis 1 are established calciphiles but they are all species adapted to moist conditions, waste places, or to both (Weatherbee, 1996). This response reflects their distribution as generalists that are not selecting habitats with increased calcareous nature. Auclair (1979) reported that *Equisetum fluviatile* had elevated concentrations of iron in its tissues. Thus the correlation of this species to water iron may be directly related with its ability to tolerate elevated iron levels.

Axis 2

Parnassia glauca, *Packera aurea, Viburnum dentatum*, *Muhlenbergia glomerata*, and *Carex granularis* had strong negative correlations with Axis 2 where water and soil calcium and magnesium were correlated. By contrast *Carex stricta, Rubus pubescens*, *Polygonum sagittatum*, *Betula papyrifera*, *Gallium asprellum*, *Amphicarpaea bracteata*, *Parthenocissus quinquefolia*, *Poa palustris*, and *Impatiens capensis* were positively correlated with Axis 2 where percent organic matter and soil iron, nitrogen and potassium were positively correlated. Collectively, these findings imply that the former group (with *Parnassia glauca*, etc.) increases in abundance with increasing water calcium and magnesium and is less abundant where there is more organic matter and soil iron, nitrogen and potassium. This finding reinforces the nature of the three species, *Parnassia glauca, Packera aurea,* and *Carex granularis* as calciphiles. Although not included in the original category groupings, *Muhlenbergia glomerata* is also known to be common in calcareous fens (Weatherbee and Crow, 1992). By contrast, the latter group (with *Carex stricta*, etc.) increases in abundance with increasing organic matter and soil iron and decreases where water calcium and magnesium are greater. Many species in this group are common wetland species (USDA, 2007) and thus it was not expected for them to occur in areas with strong calcareous influences, but it was expected that they would occur in response to a hydrologic gradient. Their increase in response to increasing organic matter may reflect this as organic matter accumulation is indicative of long-term saturation.

Axis 3

Soil calcium, magnesium and groundwater gradients were strongly negatively correlated with Axis 3 and several species appeared to respond to the opposite trend, increasing where groundwater gradients and soil calcium and magnesium were lower. These species included *Vaccinium corymbosum, Coptis groenlandica, Juniperus communis, Trientalis borealis, Equisetum hyemale, Osmunda cinnamomea,* and *Sphagnum* sp. These findings suggest that these species occurred where calcareous inputs, e.g. soil calcium, magnesium, and groundwater input, were lower. *Vaccinium corymbosum*, *Trientalis borealis*, and *Coptis groenlandica* are reported by Weatherbee (1996) as occurring in acidic soils in Berkshire County, Mass. and sphagnum moss is a well-known inhabitant of acidic substrates (Robichaud and Anderson, 1994).

The Schenob Brook Central and Schenob Brook South sites are distributed in the same region on Axis 3 in the direction where these species are correlated. Schenob Brook South was characterized by having low calcium, magnesium, pH and low upwelling of groundwater as well as high iron levels. However, Schenob Brook Central shared those same characteristics except that it had the highest water pH of any of the sites and had hardly any iron. Because Schenob Brook Central was a "calcareous seep" it had many rivulets that separated ~0.35-m high hummocks where the plants were growing. The soil pH in these hummocks was much lower than the water pH (around 6.6 compared to 7.6) and may explain this distribution. These acidophiles (acid loving plants) could also be responding to components that were not measured in this study, such as alkalinity, which would be lower where there was less groundwater input and where calcium and magnesium levels were lower.

Summary

In general, the results of both the ordinations of species distributions and environmental parameters seem to indicate that one general theme describes plant distributions. Species seem to either be related to increases in calcareous inputs (calcium, magnesium, pH, and groundwater upwelling) or increases in iron, which may reflect a decrease in calcareous inputs. Six species from the *a priori* category groupings were observed, in which all except *Equisetum fluviatile* increased in abundance in relation to higher calcareous inputs. More variation in plots was explained by the environmental ordination than the vegetation ordination and this may be why more species appeared to be patterned with the underlying calcareous gradients. Calcareous properties were more detectable when viewed as a function of environmental properties than as a function of species compositions, which makes sense since they are environmental properties. Of the species from the a priori category groupings, both *Parnassia glauca and Dasiphora fruticosa* consistently showed relationships to calcareous gradients in both ordinations. This may reflect a strong dependence of these species to those gradients.

Determination of Gradient and Threshold Species

The species that occurred in the study plots that also were selected in the *a priori* category groupings were sorted based on their correlations in the ordinations. Species were grouped by 1) whether they only occurred at the extreme range of an axis of correlation related to calcareous groundwater inputs, 2) whether their abundance increased gradually along an axis of correlation related to calcareous groundwater inputs, or 3) whether neither of these patterns occurred. These three trends were the basis for the establishment of three new categories in which species were grouped as: Category 1, calcium threshold species; Category 2, calcium gradient species; and Category 3, calcium generalists.

Category 1 – Threshold Species

In the determination of threshold species, species were considered regardless of the strength of their correlations to the environmental axis. Even though these species did not show strong linear correlations with specific axes, this was expected of a species that occurred only at a gradient extreme. Table 5.5 presents the remaining species from the a priori category groupings with their Pearson's r values and axes of correlation. These were all weakly correlated; however, several appeared to be threshold species, and two appeared to still follow a trend indicative of a gradient species, albeit weakly. The latter will be discussed in the next section.

Figure 5.7 represents the output in PC-ORD Ver 5.0 (McCune and Mefford, 1999) for the correlation of mean water calcium to Axes 2 and 3. Water calcium was correlated to both these axes, although most strongly with Axis 2. *Carex granularis* (Figure 5.8a), *Carex sterilis* (Figure 5.8b), and *Geum rivale* (Figure 5.8c) appeared to mainly occur at the extremes of Axes 2 and this relationship indicates that these species only occurred once a certain environmental calcium threshold was reached.

Table 5.5. Remaining species from a priori category groupings as they correlate ($r^2 = <$ 0.2) to the environmental ordinations.

Species	Pearson's r for Axis: 1	2	3
Carex flava			.245
Carex hystericina	.278		
Carex interior	-.300		
Carex leptalea^{+}		-.285	
Carex sterilis*	.288	-.320	
Carex tetanica	.217		.390
Geum rivale*		-.142	
Juncus brachycephalus^{+}	-.349	-.257	
Juncus nodosus	-.271	-.209	
Lobelia kalmii		.294	
Solidago patula			-.252
Solidago uliginosa		-.152	
Symphyotrichum puniceum	.338		
Symplocarpus foetidus	.294	.296	

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[*] indicates suspected threshold species.
[+] indicates suspected gradient species.

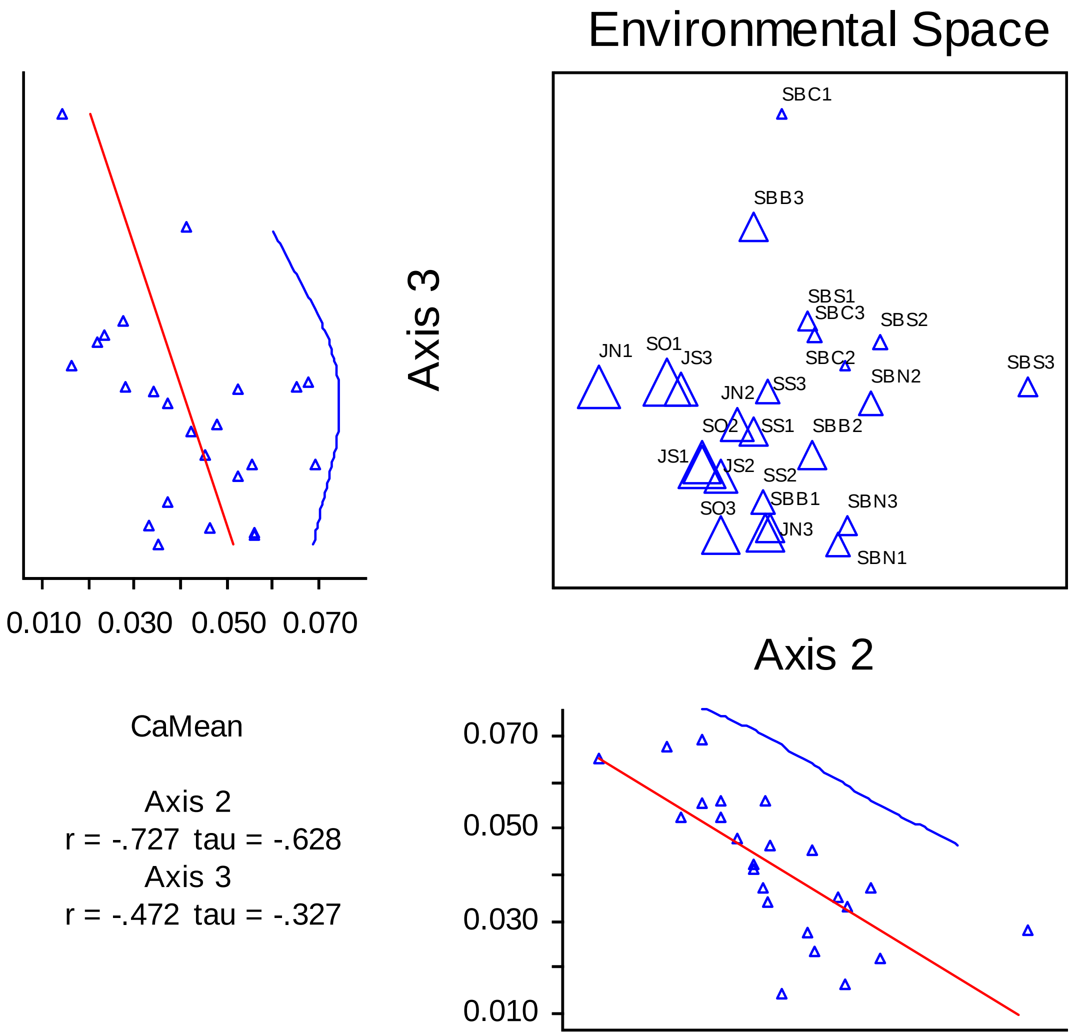


Figure 5.7. PC-ORD output representing the linear correlation between Axes 2 and 3 and mean water calcium.

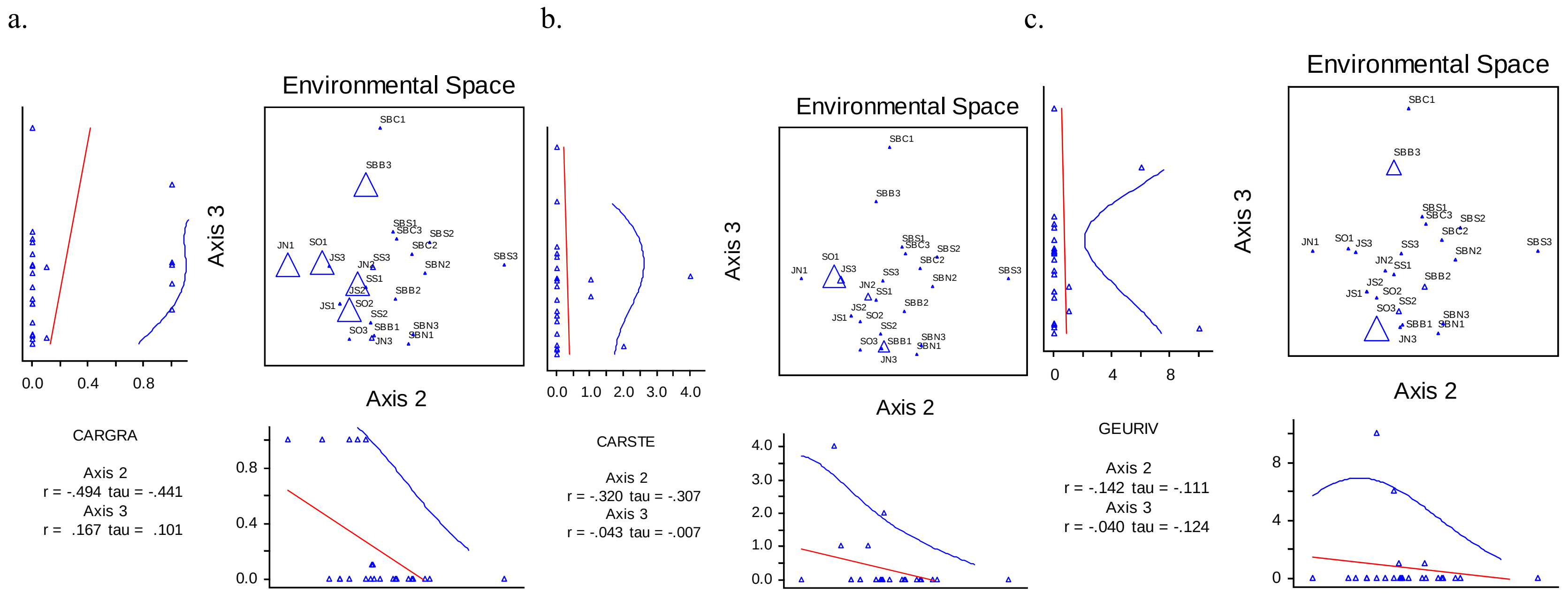


Figure 5.8. PC-ORD output for the overlay of the distribution of a) *Carex granularis*, b) *Carex sterilis,* and c) *Geum rivale* along the axes (2 and 3) where water calcium is the strongest explanatory variable.

To test whether the distributions presented in Figure 5.8 were actually related to water calcium, the abundance of those species was compared to actual mean water calcium levels. For selected species, its percent cover was averaged at each site and compared to the seasonal mean water calcium at that site. It is clear in Figures 5.9-5.11 that these three species occur mainly (or solely) once a threshold calcium level had been reached. *Carex granularis* (Figure 5.9) was not present until mean water calcium exceeded 50 mg·L^{-1} and *Carex sterilis* (Figure 5.10) was not present until mean water calcium exceeded 60 mg·L^{-1}. For *Geum rivale* (Figure 5.11), the relationship is not as clear, as there were sites where calcium values were high and yet it does not occur. This response however, may be related to other environmental properties that limit its growth. What is notable however, is that of places where *Geum rivale* does occur, all exceed 50 mg·L^{-1} calcium.

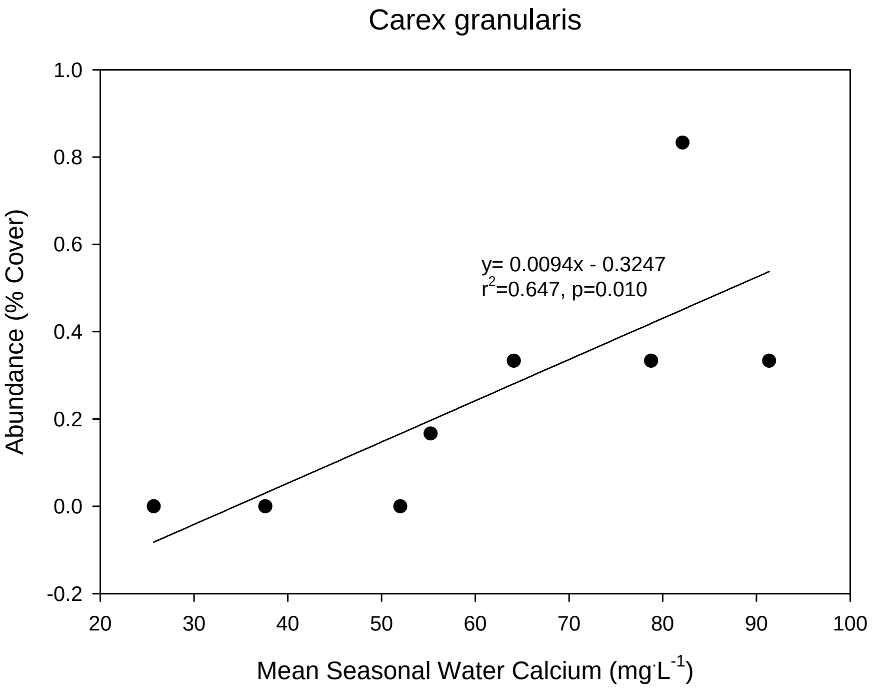


Figure 5.9. Abundance of *Carex granularis* as compared to mean water calcium. Statistics are based on log-transformed data; raw data are presented in the graph. Each point represents a mean for that site (n=3).

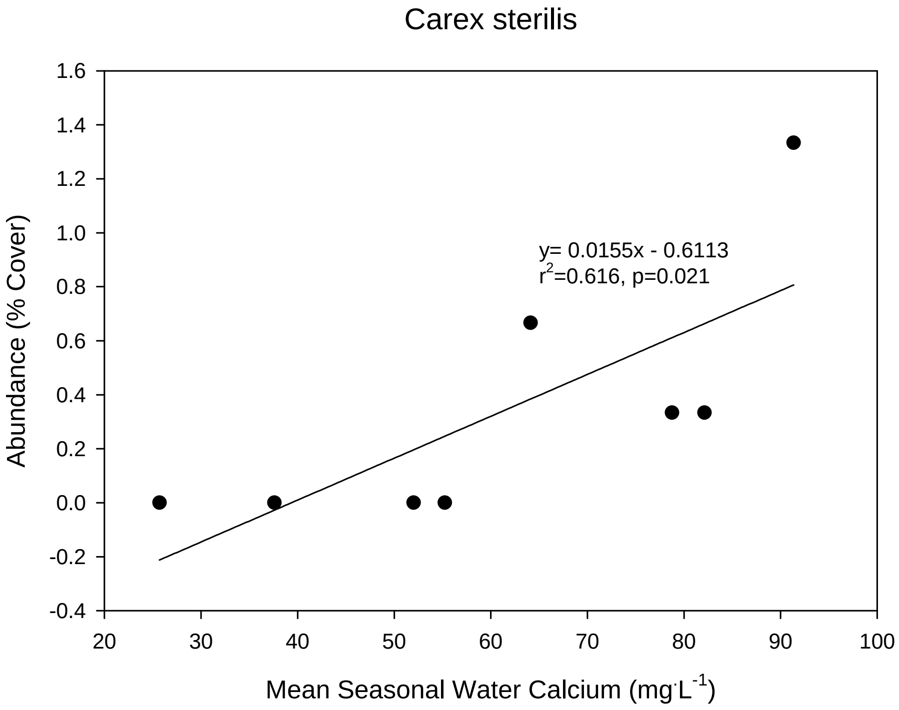


Figure 5.10. Abundance of *Carex sterilis* as compared to mean water calcium. Statistics are based on log-transformed data; raw data are presented in the graph. Each point represents a mean for that site (n=3).

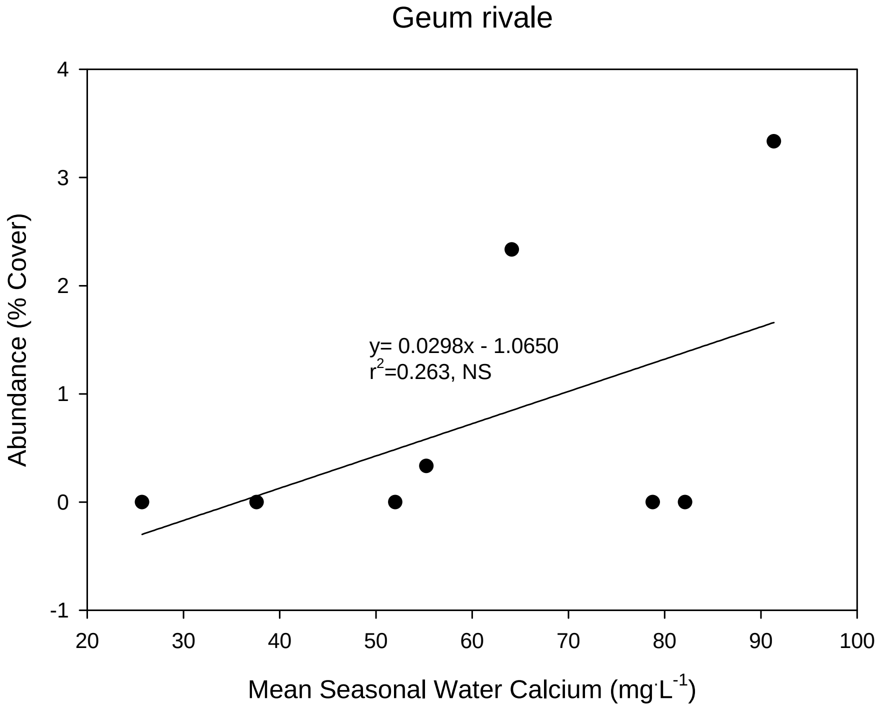


Figure 5.11. Abundance of *Geum rivale* as compared to mean water calcium. Statistics are based on square root-transformed data; raw data are presented in the graph. Each point represents a mean for that site (n=3). NS = not significant.

Category 2 – Gradient Species

Gradient species were identified where a species' distribution occurred linearly along an axis where calcium was the strongest explanatory variable. Of species in Table 5.5 that also were selected in the a priori categories, these included *Parnassia glauca*, *Packera aurea*, *Dasiphora fruticosa*, and *Thelypteris palustris*. Of these, *Parnassia glauca* and *Packera aurea* had correlations in abundance changes associated with elevated water calcium levels, and *Dasiphora fruticosa* had correlations in abundance changes in relation to water pH. Figure 5.12 represents the output in PC-ORD of the distribution of "gradient" species. One can clearly see in Figure 5.12a that abundance of *Parnassia glauca* decreases linearly along Axis 2 and 3, as did water calcium.

Of species listed in Table 5.6 that had weaker correlations ($r^2 = < 0.2$) to Axes 2 and 3, *Carex leptalea* seemed to show an increase in abundance in relation to water calcium levels (Figure 5.12b). In addition, *Juncus brachycephalus* had a weak negative correlation with Axis 1, where water pH was significantly correlated (Figure 5.13). Even though both of these species had weak correlations, it is clear that they increased in abundance as did water calcium or water pH, respectively. Finally, *Equisetum fluviatile* also was identified as a gradient species (with abundance patterns strongly correlated with an underlying environmental axis), but to water iron and ammonium and not calcareous inputs.

To validate the species-environmental correlations, the abundance of those species were regressed against the environmental variables. The abundance of *Equisetum fluviatile* did not show a significant correlation with water calcium (r^2=0.073, p=0.518) which was tested since it was reported to be a calciphile in previous literature (see

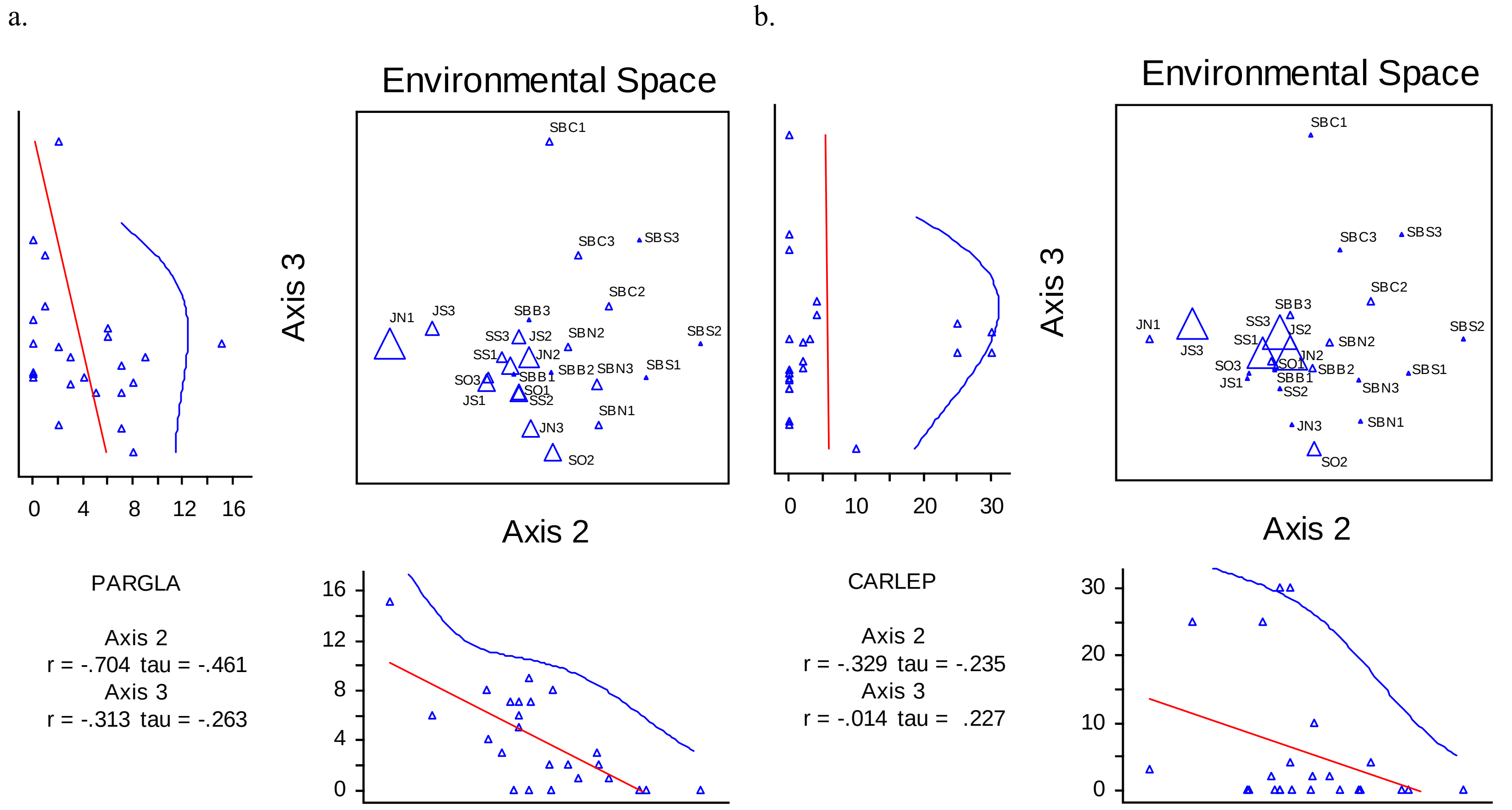


Figure 5.12. PC-ORD output for the overlay of the distribution a) *Parnassia glauca*, b) *Carex leptalea* along the axes (2 and 3) where water calcium is the strongest explanatory variable. This distribution indicates that these species increases in abundance as calcium levels increase and are thus considered gradient species.

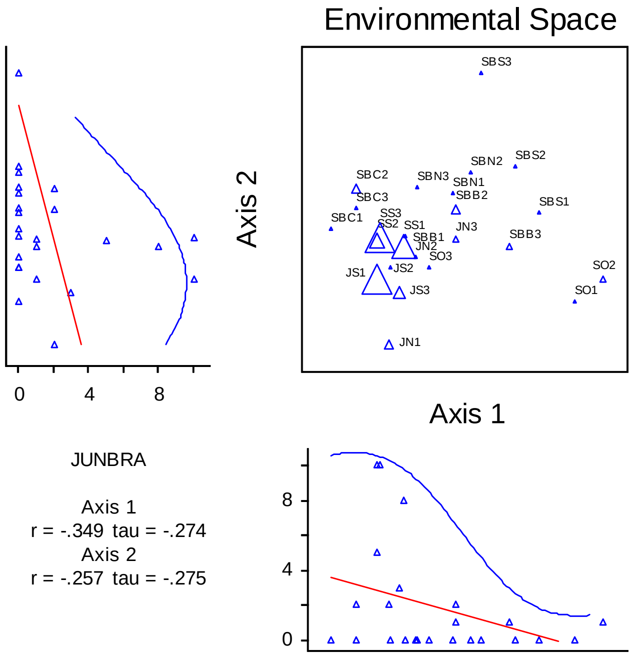


Figure 5.13. PC-ORD output for the overlay of the distribution of *Juncus brachycephalus* along Axis 1, where water pH is a strong explanatory variable.

Chapter 4). As the ordination results suggest, it did show a strong relationship with water iron. Its abundance was most strongly correlated with minimum total water iron at each site (Figure 5.14). This finding confirms *Equisetum fluviatile* as a gradient species, the abundance of which appears to be affected by iron, not calcium. Of species whose abundance was suspected to increase along a calcium gradient, only *Parnassia glauca* (Figure 5.15) had a significant linear relationship. Figure 5.15 shows clearly that this species becomes more abundant as water calcium levels increase.

Packera aurea, *Thelypteris palustris*, *Carex leptalea*, *Dasiphora fruticosa* and *Juncus brachycephalus* did not show significant linear correlations with water calcium when regressed against water calcium levels. However, *Packera aurea* (Figure 5.16), *Thelypteris palustris* (Figure 5.17) and *Carex leptalea* (Figure 5.18) appeared to be most abundant when calcium levels were elevated, still qualifying them as gradient species.

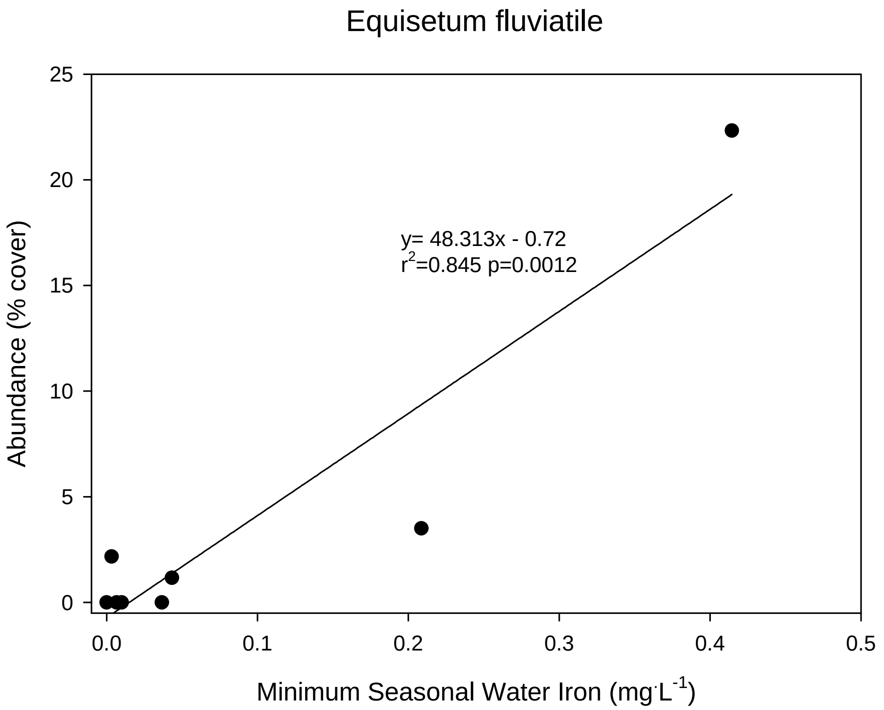


Figure 5.14. Abundance of *Equisetum fluviatile* as compared to minimum water iron. Statistics represent log-transformed data, raw data are presented. Each point represents a mean for that site (n=3).

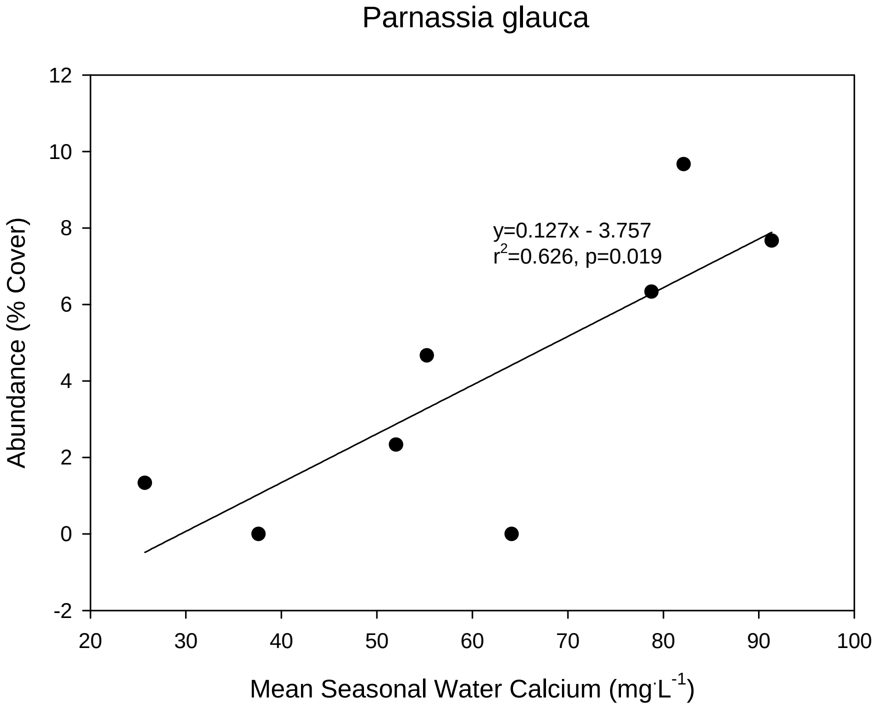


Figure 5.15. Abundance of *Parnassia glauca* as compared to mean water calcium. Raw data are presented. Each point represents a mean for that site (n=3).

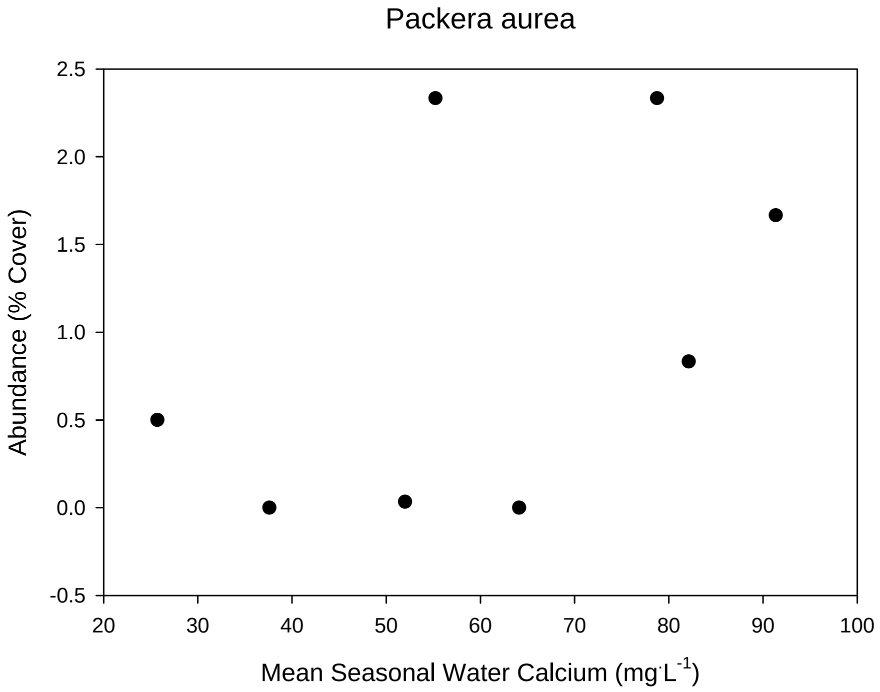


Figure 5.16. Abundance of *Packera aurea* as compared to mean water calcium. Raw data are presented. Each point represents a mean for that site (n=3). No significant relationship occurred between abundance and calcium concentrations in water.

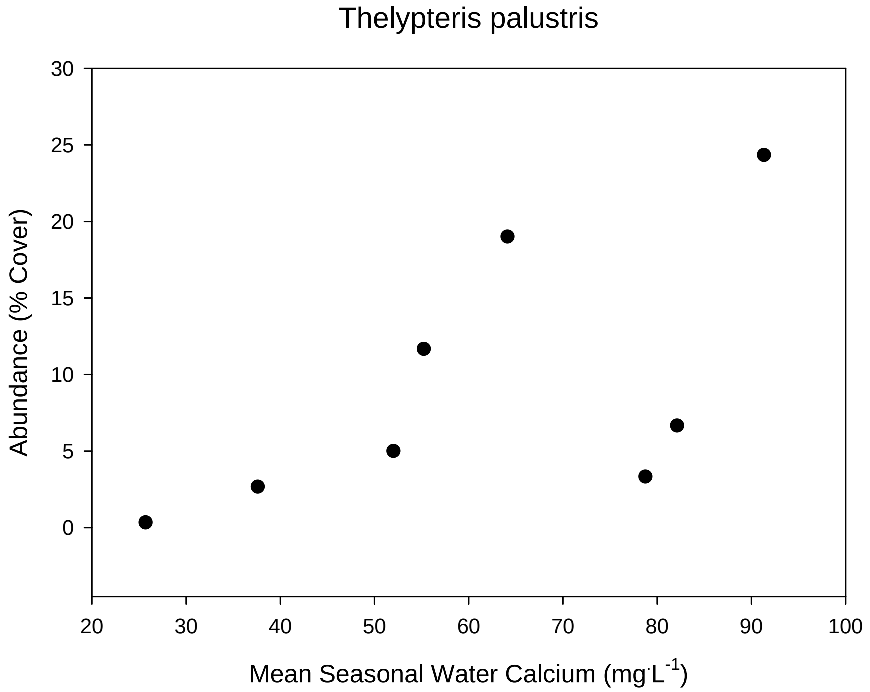


Figure 5.17. Abundance of *Thelypteris palustris* as compared to mean water calcium. Raw data are presented. Each point represents a mean for that site (n=3). No significant relationship occurred between abundance and mean concentrations of calcium in water.

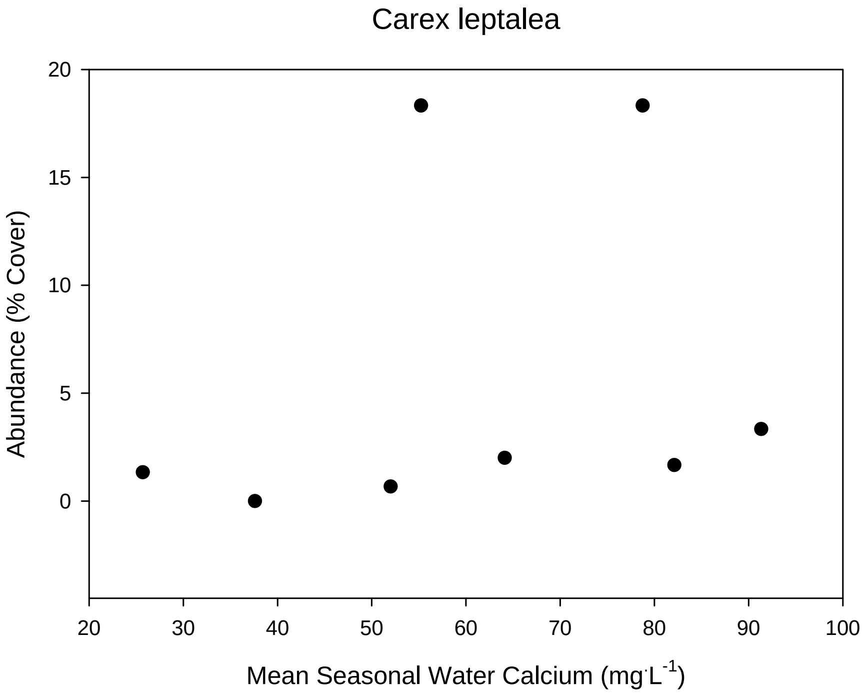


Figure 5.18. Abundance of *Carex leptalea* as compared to mean water calcium. Raw data are presented. Each point represents a mean for that site (n=3). No significant relationship occurred between abundance and mean concentrations of calcium in water.

They would not be considered true threshold species as they still did occur at lower calcium levels, just in lower abundance. For all three of these species, there seemed to be a pseudo-threshold above 50 $mg{\cdot}L^{-1}$, where their abundance markedly increased.

The abundance of *Dasiphora fruticosa* (r^2=0.015, p=0.777) and *Juncus brachycephalus* (r^2=0.053, p=0.585) were not correlated significantly to mean water calcium levels. However, abundance of *Dasiphora fruticosa* was correlated strongly to the pH of the water at each site (Figure 5.19). In a similar fashion to the weakly correlated calcium gradient species, *Juncus brachycephalus* was present in the largest amounts only above a pH of 6.8 (Figure 5.20).

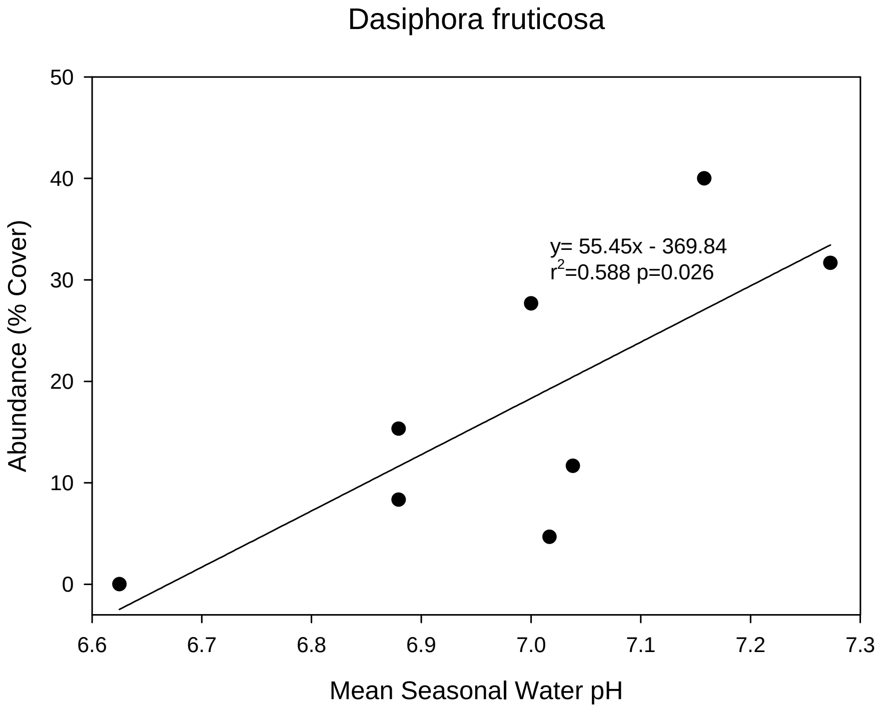


Figure 5.19. Abundance of *Dasiphora fruticosa* as compared to mean water pH. Raw data are presented. Each point represents a mean for that site (n=3).

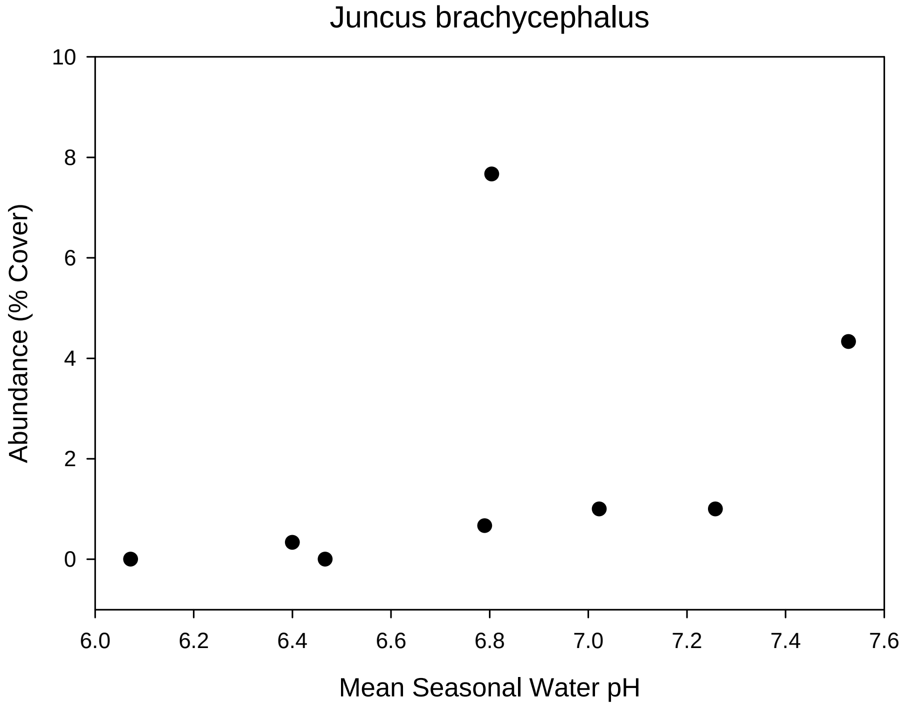


Figure 5.20. Abundance of *Juncus brachycephalus* as compared to mean water pH. Raw data are presented. Each point represents a mean for that site (n=3). No significant relationship occurred between abundance and mean water pH.

Category 3 - Generalists

Species from the a priori groupings that had no response to any particular gradient were *Carex hystericina, Symplocarpus foetidus, Juncus nodosus, Carex tetanica, Carex flava, Carex interior, Solidago uliginosa, Solidago patula, Symphyotrichum puniceum* and *Lobelia kalmii*. Of these, *Carex tetanica, Carex interior* and *Lobelia kalmii* had too few occurrences to make a valid assessment. Perhaps life-cycle traits of these species limit their distribution more than environmental substrate chemistry.

The assumption governing the generalists was that although there may have been a specific required amount of calcium before they would occur, that lower limit could have been so low that it was exceeded at every site in this study, as no sites were determined to be "poor fens". Thus, some of these species may grow in calcareous fens because they respond favorably to the elevated calcium; however, others may simply grow there because they are unaffected by the unique geochemical properties of these fens that might potentially exclude other species. The latter could be true of *Symplocarpus foetidus* as it commonly occurs in all types of wetlands. The same may also be true for *Thelypteris palustris* (a common wetland species) as the ordination results suggest that it increases in abundance as does water iron.

New Species Categories

Based on the findings of this research, new category groupings were established. The new category groupings (Table 5.6) differ from the original groupings in that species were not arranged by their rarity, but only by how their distribution seemed to be affected by environmental calcium and pH levels. Many of the very rare species listed in the original categories were not observed in this study, and it seemed less important to include those species as factors affecting rarity are not necessarily related to how selective they are to calcium in their environment. Table 5.7 represents the suspected numerical ranges and thresholds to which those species in Table 5.6 appear to be responding.

Literature Revisited

Although it cannot be said for certain that the ranges and thresholds determined in Table 5.7 would remain identical if similar analyses were repeated in other fen systems, it provides the first estimate of the range and minimum level for the suspected selectivity of environmental calcium for these individual species. These threshold values do correspond with Motzkin's (1994) study of calcareous fens in western New England in which *Geum rivale*, *Carex sterilis* and *Carex granularis* where classified into his "Group III" species and occurred where calcium values averaged within the range of 26 – 62 $mg \cdot L^{-1}$.

In Europe, *Geum rivale* occurs in mildly acidic to calcareous, wet soils (Taylor, 1997). More detailed studies on its relation to environmental geochemistry have not been conducted and perhaps even though it occurs in mildly acidic soils, calcium values could have been above the established threshold. Unpublished data referenced in Taylor (1997) suggest that *Geum rivale* has a low requirement of nitrogen and a high requirement of phosphorus. This requirement seems counterintuitive to its occurrence in calcareous fen ecosystems as they are often phosphorus limited and have readily available amounts of nitrogen. Thus some other aspect calcareous fens (e.g. pH or calcium) may facilitate its distribution there.

Table 5.6. New category groupings based on ordination results.

Category 1	**Category 2**		**Category 3**
Calcium Threshold Species	**Calcium Gradient Species**	**pH Gradient Species**	**Calcium Generalist Species**
Carex granularis *Carex sterilis* *Geum rivale*	*Carex leptalea* *Packera aurea* *Parnassia glauca*	*Dasiphora fruticosa* *Juncus brachycephalus*	*Carex flava* *Carex hystericina* *Equisetum fluviatile* *Juncus nodosus* *Solidago patula* *Solidago uliginosa* *Symphyotrichum puniceum* *Symplocarpus foetidus* *Thelypteris palustris* *Juncus brachycephalus* *Dasiphora fruticosa*
No determination: *Carex interior, Carex tetanica, Lobelia kalmii*			

Table 5.7. Ranges and thresholds of water calcium at which gradient and threshold species occurred.

Gradient Species		**Threshold Species**	
Calcium	**Range mg·L^{-1}**	**Calcium**	**Threshold mg·L^{-1}**
Parnassia glauca	20-100	*Carex granularis*	48
Packera aurea	20-97	*Geum rivale*	57
Carex leptalea	24-100	*Carex sterilis*	66
pH			
Juncus brachycephalus	6.8 – 7.2		
Dasiphora fruticosa	6.8 – 7.4		

Motzkin (1994) identified a vegetation association (Group III) that included *Carex leptalea* as a characteristic species in rich fens. The findings of this study agree with Motzkin's study that *Carex leptalea* increases in abundance as environmental calcium levels do, and that it is more abundant when calcium levels are higher. Motzkin (1994) studied rich fens where surface water calcium concentrations had an average of 46 mg/L and pH values were around 7.2. Motkin's calcium values were much lower than those observed in the current study and may provide insight as to why the correlation of *Carex leptalea* to the underlying environmental axes was weak. Perhaps this species reached a required calcium "threshold" at a much lower level and thus the range of calcium observed in this study exceeded the range where calcium would continue to influence its abundance.

Picking and Veneman (2004), in their study of vegetation and environmental gradients in a calcareous sloping fen, reported that *Solidago uliginosa* and *Equisetum fluviatile* appeared to be indicator species in the study plot that had the highest levels of dissolved calcium, iron and orthophosphate. Findings in the current study in relation to the occurrence of *Solidago uliginosa* were inconclusive. It occurred in five of the eight study sites, and no distinct environmental properties were correlated with its occurrence. Interestingly though, *Equisetum fluviatile* did show a correlative relationship to dissolved iron in the water. This association corresponds to the findings of Auclair (1979), who noted this species to have the highest levels of iron (in addition to calcium) in its tissues than any other species studied in a *Scirpus-Equisetum* wetland in Canada. In addition, a recent study in the Netherlands (Lucassen et al., 2006) determined *Equisetum fluviatile* to be one of the dominant species in seepage locations where dissolved calcium and iron were high (calcium= 82 $mg \cdot L^{-1}$; iron= 12 $mg \cdot L^{-1}$). These levels were comparable to some of the upper ranges of calcium and iron measured in the current study however an important distinction exists. Some locations that had high levels of calcium (in the current study) did not have high levels of iron, and in these locations, *Equisetum fluviatile* was absent. It seemed to occur only in the locations where iron levels were elevated, and this result leads one to conclude that the growth of *Equisetum fluviatile* may be in response to dissolved iron and not calcium.

Some of the early literature reports on *Dasiphora fruticosa* (Elkington and Woodell, 1963) refer to its preferred substratum as wet soils made of glacial drift underlain by limestone. The exchangeable calcium in the sites where this species occurred was extremely variable. Although the pH of the soils where it occurred was between 5.1 and 7.0, no assessments of the water pH were made. Since this early study, many researchers have identified *Dasiphora fruticosa* as a common species in all varieties of calcareous fens. Other than being associated with "rich fens," no specific environmental properties have been isolated that seem to strongly dictate its distribution. This research seems to imply that pH (instead of calcium) strongly affects its distribution. Schenob Brook Central had the lowest calcium observed in the eight study sites (mean Ca=25.7 $mg \cdot L^{-1}$), yet the percent cover of *Dasiphora fruticosa* was more abundant than at any other site. More interesting though, this site had the highest average water pH of any of the sites studied (mean water pH = 7.3). By contrast, at Schenob Brook South, the water calcium was slightly higher than at Schenob Brook Central and yet with the lowest average pH (6.6) had not a single occurrence of *Dasiphora fruticosa*. This site was the only one in which this species did not occur. Perhaps, there is a threshold calcium requirement for this species that was exceeded in this study and that above that threshold pH governs its distribution. Thus, the lower pH limit for *Dasiphora fruticosa* may be around 6.6.

There are no studies that have established specific environmental calcium or pH ranges for *Parnassia glauca, Packera aurea* and *Juncus brachycephalus*; however, all of these species have been well documented as being associated with growth in rich calcareous fens. *Parnassia glauca* and *Packera aurea* were grouped into Motzkin's (1994) Group III vegetation category. It was surprising that other species thought to be indicative of rich fens did not show any significance with the calcium gradients in the ordination. These included *Carex flava, Carex hystericina, Solidago patula, Juncus nodosus, Symphyotrichum puniceum* and *Solidago uliginosa*. Although all of these species have shown responsiveness along calcium gradients (Reschke, 1990; Motzkin, 1994; Picking, 2002) it is possible that distributions could be affected by other factors related to geochemistry, such as alkalinity, or that very low minimum calcium thresholds are required for these species, which all sites in this study exceeded.

Conclusions

The ordination analysis of the vegetation abundance explained little of the variation in the species community structure but indicated that overall, differences in vegetation composition may be due to duration of soil saturation and calcareous groundwater inputs. The ordination of the environmental variables explained more variation among sites in which distinctions between calcareous inputs and water and soil iron became apparent. Distributions of individual species were viewed in terms of those underlying gradients, and species were regrouped into categories based on their observed distributions in relation to environmental calcium levels. Determinations were made as to species which distributions appeared to be responding to a threshold of environmental calcium, a gradient of calcium or which appeared to be generalists with respect to calcium growth. Although once thought to be a calciphile, findings suggest that the abundance of *Equisetum fluviatile* increases with water iron. In addition, *Juncus brachycephalus* and *Dasiphora fruticosa* appear to increase in abundance as does pH, hence, to declining concentrations of soluble iron. Most importantly, *Carex granularis, Geum rivale*, and *Carex sterilis* appear to occur only at elevated calcium levels, and *Parnassia glauca, Packera aurea* and *Carex leptalea* appear to increase in abundance as calcium does.

CHAPTER VI

TISSUE ANALYSIS

Introduction

In agricultural systems, the lack or excess of a certain plant nutrient often can be mitigated by crop fertilization to satisfy the requirements of a specific crop. By contrast, variability in nutrient concentrations in natural systems (among other factors) governs the plant species that will grow there. This effect is particularly evidenced in environments with elevated calcium. Although calcium is required by all plants, studies have demonstrated that the amount of calcium in different plant species varies from 0.1% to 5% (by weight) and rarely up to 10% (White and Broadley, 2003). This variability is explained partly by species called "calciphiles" for which experiments have shown to absorb calcium into their tissues in greater amounts as environmental calcium levels increase (Clarkson, 1965; Lee, 1999). To date, this comparison has been carried out only in laboratory experiments with controlled nutrient solutions and has not been applied to calciphiles occurring in calcareous fens. Understanding the absorption of calcium in plants occurring in these fens will help to gain knowledge of these species and the role that calcium plays in their distributions and physiology. The previous chapters of this thesis have shown that the distributions of some species occurring in calcareous fens appear to be associated with underlying gradients of calcium. This chapter seeks to analyze whether those similar trends are evidenced in the amount of calcium stored in the tissues of those plants.

Materials and Methods

Tissue Analysis

After the vegetation survey was completed, herbaceous species that occurred in three or more of the eight sites were revisited, and leaf samples were collected for tissue analysis. Leaves of trees and shrubs (except for the calciphile *Dasiphora fruticosa*) were not collected as their long-term growth may be less related to environmental calcium levels than for herbaceous and graminoid species and also few are considered calciphiles. The upper-most recently mature leaves (Mills and Jones, 1991) were collected during the two-week vegetation sampling period of July 10 to 26, 2006, to ensure that calcium tissue levels could be compared across species without confounding the expected differences in calcium uptake across the growing season. No rare or threatened species were collected as special regulations govern their collection; however, tissue analysis of rare species may be an area of interest for future research. Leaves from several plants of the same species were combined to form a composite sample at each study plot. Leaf tissues were prepared by dry ashing, and extracts (Hamze et al., 1984) were analyzed by atomic absorption spectrophotometry (Jones, 2001). Calcium levels in the plants' tissues were recorded as a percent of plant dry weight.

A subset of the tissues for each species was analyzed for magnesium (following the same procedure as for calcium) as many of the environments studied were influenced by dolomitic limestone ($CaMg(CO_3)_2$). Knowledge of plant uptake of magnesium may provide an indication as to where underlying magnesium gradients may influence species distributions in addition to or instead of calcium and may elucidate the importance of calcium in relation to other dominant cations.

Plant tissues were collected and analyzed for sixteen species that occurred in three of more sites. The species analyzed included *Equisetum fluviatile, Parnassia glauca, Solidago patula, Packera aurea, Carex flava, Carex hystericina, Symplocarpus foetidus, Dasiphora fruticosa, Thelypteris palustris, Symphyotrichum puniceum, Fragaria vesca*, *Mentha arvensis*, *Eriophorum viridicarinatum, Carex granularis, Geum rivale* and *Scirpus atrovirens*. For *Geum rivale*, *Carex granularis* and *Scirpus atrovirens*, only a small number of plants (n<3) were sampled. *Carex granularis* and *Scirpus atrovirens* were documented earlier in the season when they were in bloom and by the time of tissue collection only two plants were positively identifiable. Only one sample of *Geum rivale* was collected as its low frequency made it difficult to locate when revisiting the site. For these species only, their absolute tissue calcium values were reported; however they were not compared to environmental levels. Should any of these species become interesting points of study, more samples should be taken to more accurately define the average concentration of calcium in these plants and comparisons should be made with environmental calcium levels.

Data Analysis

A regression analysis was conducted with statistical software (SAS Version 9.1.3, SAS Institute, 2004) to compare environmental calcium and magnesium levels to plant tissue calcium and magnesium levels. Although the plots occurred in triplicate at individual sites, it was assumed that the uptake of calcium into individual plants in relation to environmental levels was independent of the physical location where it occurred. The environmental ordination in Chapter 5 indicated that plots were largely independent with respect to environmental properties and that wide variability in environmental gradients existed within sites. Other researchers have noted that soil chemistry can vary greatly within only a few meters (Lee, 1999), and thus if plant tissue calcium and substrate calcium were averaged across a site, results would be less accurate than if averaged by the smaller plots as plants are responding only to the area immediately around them. Thus, the regression analysis compares plant tissue concentrations at each plot to environmental levels at those plots, even though they may be thought of as pseudoreplicates.

Plant tissue concentrations were compared against the soil calcium and magnesium as well as the mean, median, minimum, maximum, and each monthly water calcium and magnesium measurement. Since water calcium values did not remain constant across the growing season at all of the sites, simply comparing the tissue concentrations to the seasonal mean might not give a true relationship between the tissue calcium and the environmental levels at any one time. Evaluation of tissue concentrations against monthly water chemistry measurements was conducted to evaluate if plant tissue calcium was reflective of environmental conditions prior to the collection of the leaves (in May and June) or to the levels present at the time of collection in July. Data of tissue calcium for *Thelypteris palustris, Carex flava, Carex hystericina, Eriophorum viridicarinatum* were log-transformed prior to analyzing to meet the assumptions of normality. Tissue calcium data for *Parnassia glauca* and *Mentha arvensis* were transformed by square root as the log transformation was insufficient (Wilk-Shapiro <0.05). Data for tissue magnesium for *Solidago patula* and *Thelypteris palustris* were log-transformed. Graphs of these results present raw values; however, the statistics reported are for the normalized data. ANOVA was conducted to see if uptake of calcium was significantly different among species. Tukey's test (to be conservative) was used to separate these means where appropriate.

Results and Discussion

Calcium

Calcium concentrations in plant tissues

The tissue calcium for all species was generally below 2% (Table 6.1). Monocots tend to have lower concentrations of calcium in their tissues than dicots (Broadley et al., 2003), so comparisons of tissue calcium were made within monocots and within dicots and not between them. Although the concentration of calcium in the tissues of *Parnassia glauca* varied widely depending on substrate levels, it had the overall highest mean accumulation at 4.0%. It was interesting to observe that the dicot species for which abundance appeared to be correlated to environmental calcium (as presented in Chapter 5: *Parnassia glauca, Packera aurea*, *Geum rivale and Dasiphora fruticosa*), along with other species suspected of being calciphiles in Chapter 4 (*Equisetum fluviatile* and *Symphyotrichum puniceum*) had higher average calcium levels in their tissues than did the calcium generalists (as determined in Chapter 5). Tissue analyses also were conducted on several species not mentioned in the a priori category groupings established in Chapter 4 as they occurred in three or more sites. These were *Fragaria vesca*, *Mentha arvensis,* and *Scirpus atrovirens*. Although *Mentha arvensis* was not considered originally to be a calciphile, it was interesting to observe that this species had an elevated level of calcium in its tissues with a mean of 2.4%. This value exceeds the accumulation mean for many of the dicot calciphiles studied.

Table 6.1. Mean percent calcium and percent calcium range in the tissues of the thirteen species collected from the eight study sites. Means are presented ± standard deviation. Means with different letters are significantly different by Tukey's Test (p=0.05). Composite samples (n) = number of plots where that species occurred. Number of sites where these samples were collected are included for reference.

Species	**% Calcium**	**% Calcium Range**	**Composite samples**	**Sites**
Parnassia glauca	4.0 ± 0.9 a	1.3 – 5.1	18	6
Mentha arvensis	2.4 ± 1.4 b	1.2 – 5.9	13	6
Symplocarpus foetidus	2.1 ± 0.5 b	1.5 – 3.2	13	8
Equisetum fluviatile	1.9 ± 0.2 bc	1.6 – 2.2	11	4
Geum rivale	1.9	1.9	1	1
Packera aurea	1.5 ± 0.3 c	1.1 – 2.0	11	6
Dasiphora fruticosa	1.3 ± 0.1 cd	1.0 – 1.6	22	7
Carex granularis	1.2 ± 0.6 cde	0.8 – 1.6	2	2
Fragaria vesca	1.0 ± 0.2 de	0.7 – 1.5	10	7
Symphyotrichum puniceum	1.0 ± 0.2 de	0.7 – 1.5	18	6
Solidago patula	0.8 ± 0.1 ef	0.6 – 1.0	12	7
Eriophorum viridicarinatum	0.8 ± 0.3 ef	0.4 – 1.5	9	5
Carex hystericina	0.8 ± 0.2 ef	0.6 – 1.3	10	6
Carex flava	0.8 ± 0.2 ef	0.6 – 1.3	16	7
Thelypteris palustris	0.6 ± 0.1 ef	0.4 – 0.8	20	8
Scirpus atrovirens	0.6 ± 0.1 ef	0.5 – 0.6	2	1

Of the monocots, *Carex granularis* had the highest tissue calcium value. Interestingly, this value was so high that it even exceeded the tissue concentration of several of the generalist dicots. This species was the only monocot that was determined to only occur at a threshold level of environmental calcium (as determined in Chapter 5), even though in Chapter 4 several of the other monocots were assumed to be calciphiles based on literature. This elevated accumulation of calcium further confirms this species' association with calcium.

Relationships of tissue calcium to environmental calcium

Of the thirteen species for which tissue calcium concentrations were compared to environmental calcium, five showed a significant linear relationship ($p=<0.05$) between the two variables. These species were *Equisetum fluviatile, Parnassia glauca, Symphyotrichum puniceum, Packera aurea* and *Symplocarpus foetidus*. Of these, *Parnassia glauca*, *Equisetum fluviatile*, and *Symphyotrichum puniceum* exhibited a significant relationship ($p=<0.05$) between tissue calcium and water calcium only. *Symplocarpus foetidus* showed a relationship between tissue calcium and soil calcium only, while *Packera aurea* showed a relationship to both forms of calcium.

After exploring the correlations between tissue calcium and environmental calcium, it was apparent that plants that were significantly correlated to mean water calcium seemed to exhibit the strongest correlation with samples collected in the month of June. This relationship would correspond to the month proceeding the collection of the plant leaves in July. Although one would expect the calcium in the leaves to reflect the environmental calcium during the time in which the leaves were sampled, this trend may be an indication that the most recently mature leaves had spent most of their growing time during the month proceeding their collection, i.e., in June. For this reason, the correlations of tissue calcium will be reported as compared to the water calcium levels in June.

Parnassia glauca had the strongest relationship between its tissue calcium and the water calcium in June ($r^2=0.47$ $p=0.002$; Figure 6.1), yet its tissue calcium did not follow a similar trend compared to the soil calcium levels ($r^2=0.01$, $p=0.691$). Interestingly, calcium in the tissue of *Parnassia glauca* showed a significant linear relationship to

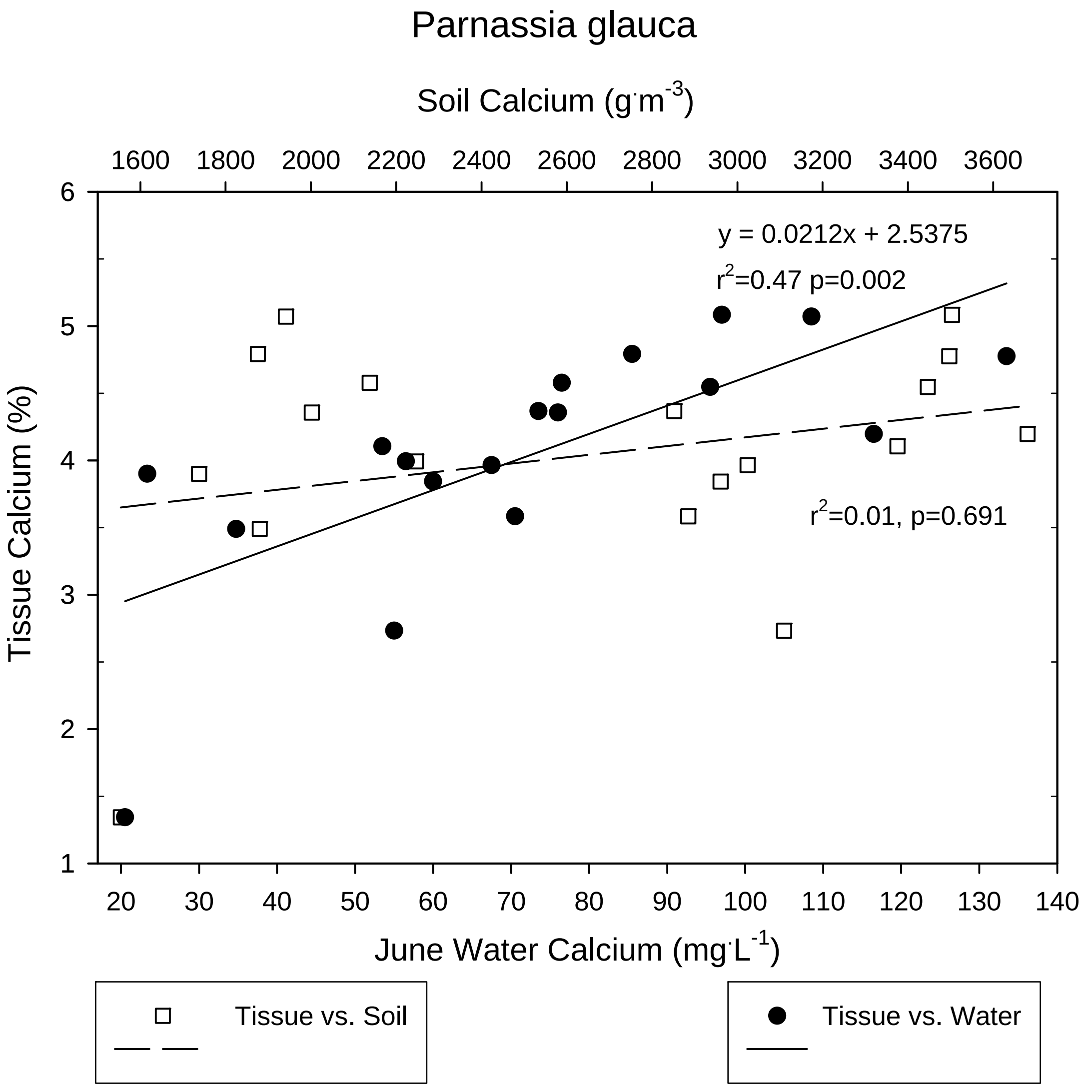


Figure 6.1. Percent calcium in the tissues of *Parnassia glauca* as a function of mean water calcium in June and soil calcium. Equations and statistics refer to the line that they are in closest proximity to.

calcium at all months when water calcium was measured, although these comparisons may not be very meaningful. This species is the only one for which the best-fit curve was non-linear (Figure 6.2). The tissue concentration appeared to reach an asymptote once it reached about 4.5%. The tissue concentration increased linearly as water calcium did until it reached a critical concentration around $80 mg \cdot L^{-1}$. At this point, there was little change in tissue concentrations as water calcium levels continued to increase. It appears that this species reached a "saturation" level of environmental calcium and tissue calcium only increased very slightly as environmental levels did. This result is not surprising considering that the actual percent of calcium in its tissues already exceeds that of most dicots. Thus, this species does exclude calcium from its tissues; however, only after it has accumulated more than most plants would.

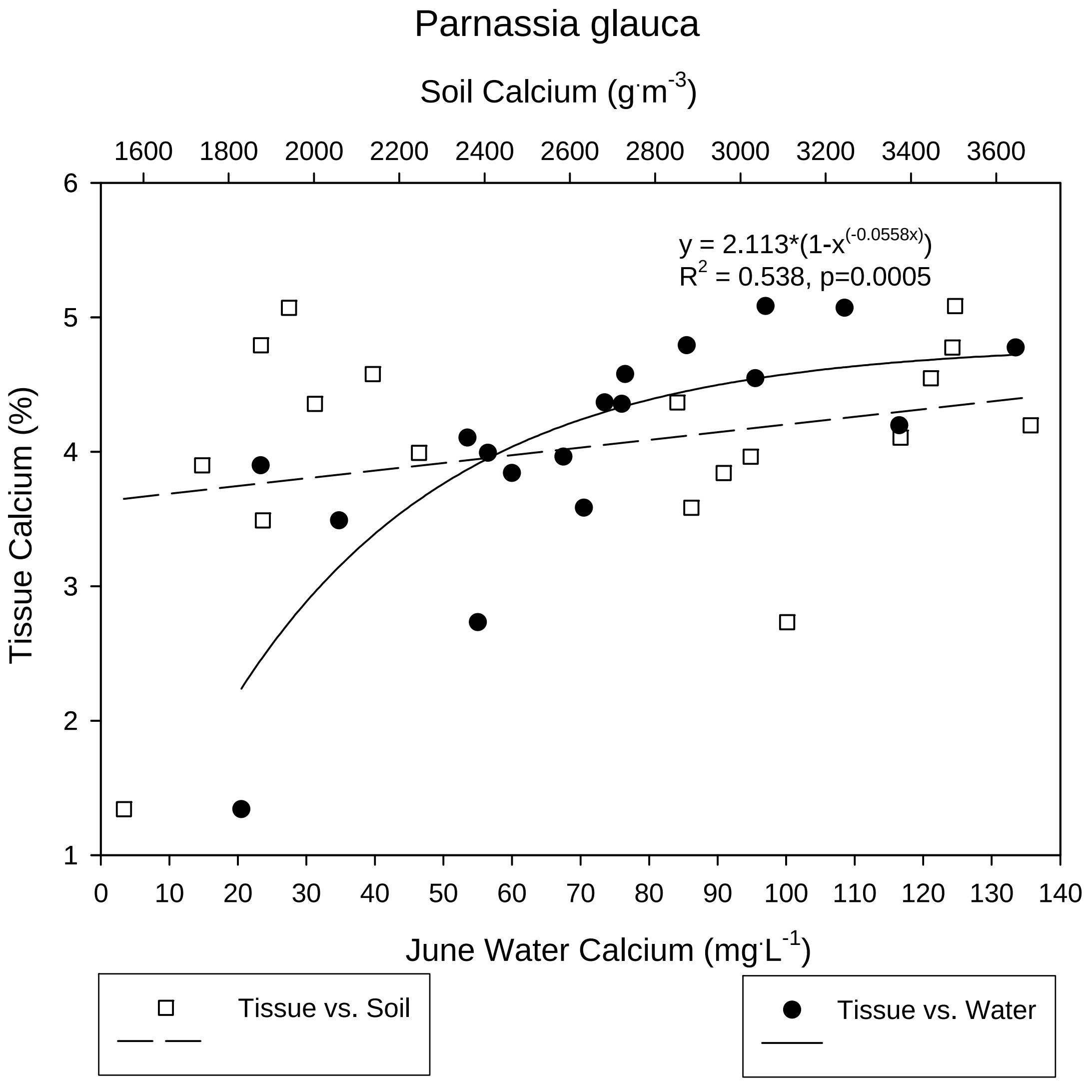


Figure 6.2. Best fit line (exponential rise, single, 2 parameter) for relationship between percent calcium in the tissues of *Parnassia glauca* as a function of mean water calcium in June and soil calcium. Equations and statistics refer to the line that they are in closest proximity to.

Calcium in the tissues of *Equisetum fluviatile* showed a significant correlation to the water calcium in June (r^2=0.40 p=0.036; Figure 6.3) and did not show a significant correlation with the soil calcium (r^2=0.02, p=0.700). *Symphyotrichum puniceum* and *Packera aurea* showed significant correlations of tissue calcium to the water calcium in months proceeding their collection in May and June. Calcium in the tissues of

Symphyotrichum puniceum was correlated to the water calcium in May (r^2=0.23, p=0.047; not shown) slightly less strongly than it was to June water calcium (r^2=0.27, p=0.027; Figure 6.4) and was not correlated to soil calcium (r^2=0.05 p=0.393; Figure 6.4).

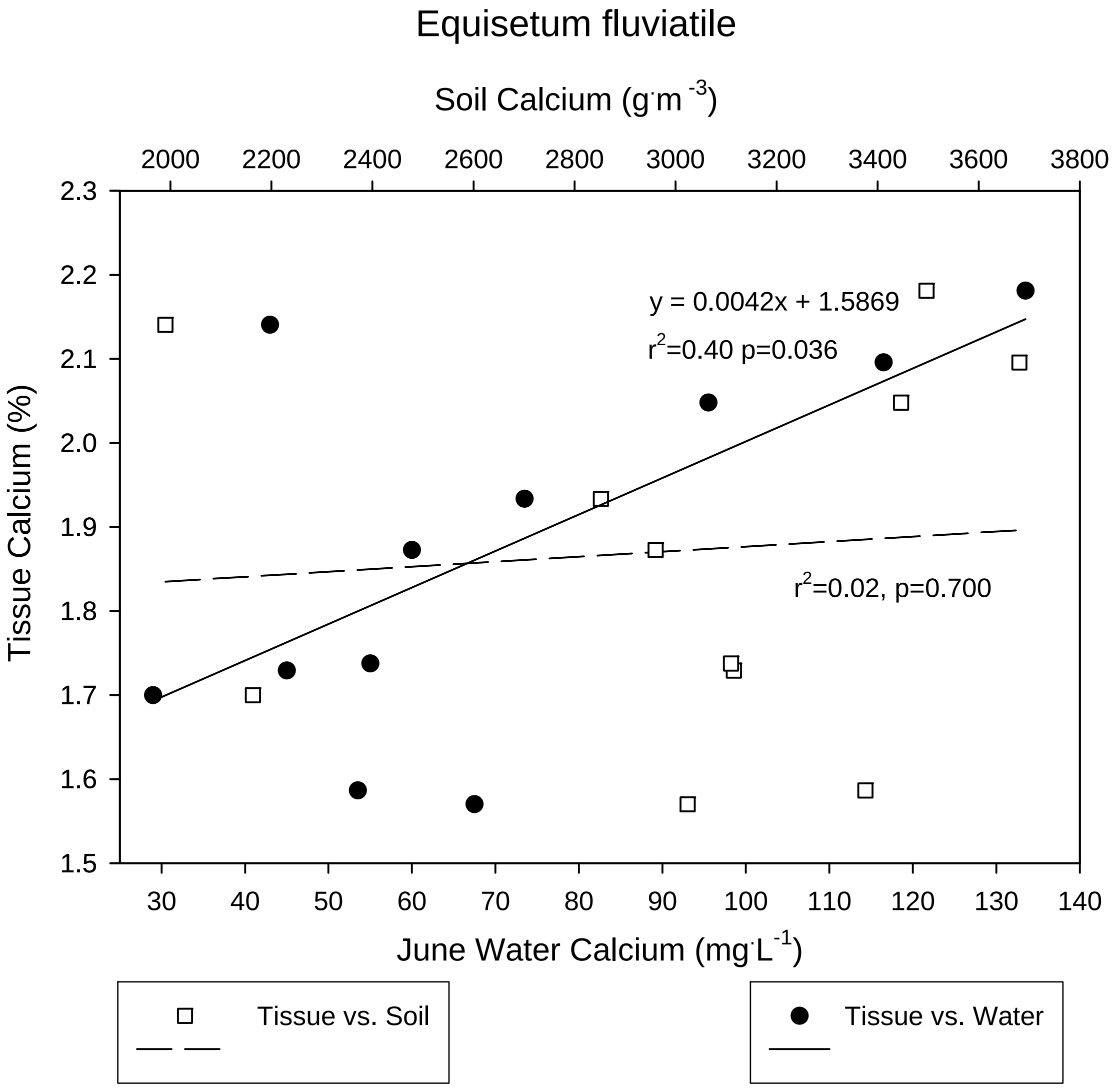


Figure 6.3. Percent calcium in the tissues of *Equisetum fluviatile* as compared to mean water calcium in June and soil calcium. Equations and statistics refer to the line that they are in closest proximity to.

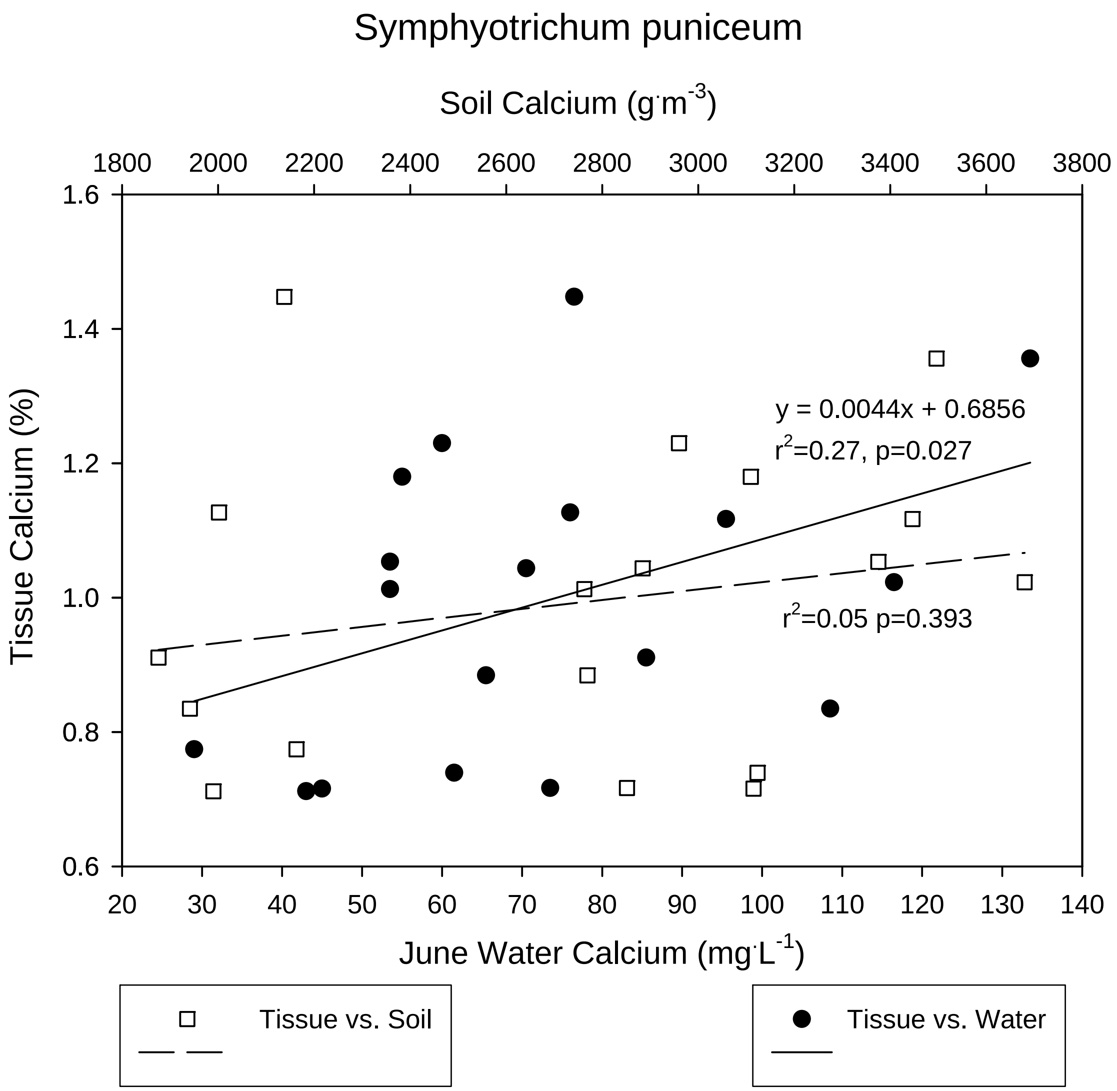


Figure 6.4. Percent calcium in the tissues of *Symphyotrichum puniceum* as compared to mean water calcium in June and soil calcium. Equations and statistics refer to the line that they are in closest proximity to.

Tissue calcium of *Packera aurea* was correlated to the water calcium in May (r^2=0.50, p=0.016; not shown) somewhat stronger than it was in June (r^2=0.41, p=0.034; Figure 6.5) and exhibited an almost identical trend in June as it did with soil calcium (r^2=0.57, p=0.007; Figure 6.5). It is interesting that *Packera aurea* is correlated to soil and water calcium, as no other species showed this relationship. Perhaps this response suggests that in sites where this species occurred, the soil and water calcium were correlated with each other. This correlation would exist if *Packera aurea* was only found at sites that had steady inputs of groundwater and fairly unchanging calcium levels. Upon further inspection, no relationship was detected as this species occurred at sites where calcium remained fairly constant (e.g., Shmulsky Shrub) and where large differences were observed (e.g. Shmulsky Open). Thus, the relationship between the tissue calcium of this species to both environmental calcium forms (in May, and June) remains unexplained.

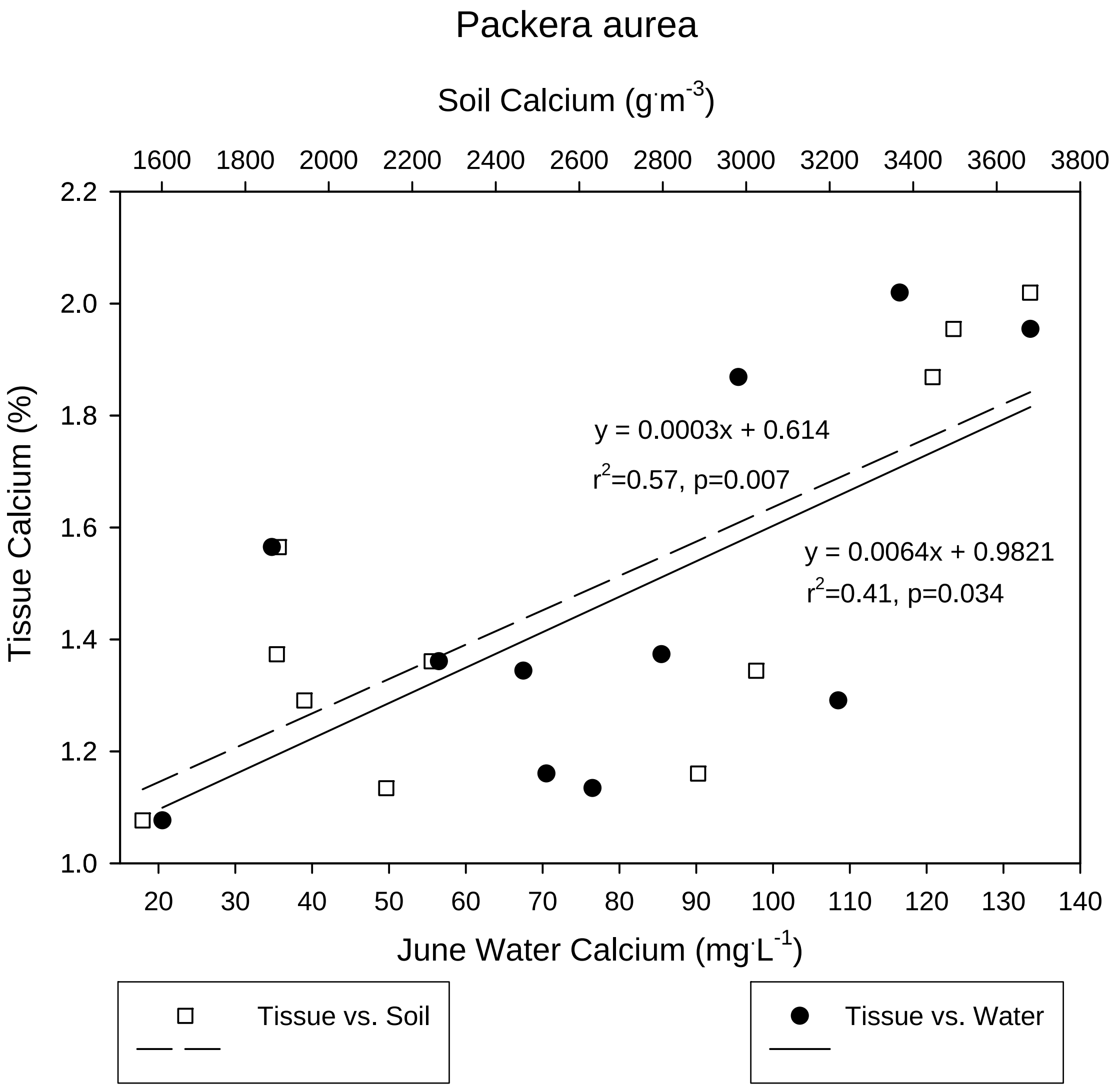


Figure 6.5. Percent calcium in the tissues of *Packera aurea* as compared to mean water calcium in June and soil calcium. Equations and statistics refer to the line that they are in closest proximity to.

The calcium in the tissues of *Symplocarpus foetidus* was not correlated significantly with water calcium (for June r^2=0.0273 p=0.589) but showed a significant relationship to the soil calcium (r^2=0.33, p=0.040; Figure 6.6). This species is the only one that correlates solely to the soil calcium. This correlation may be related to the fact that this

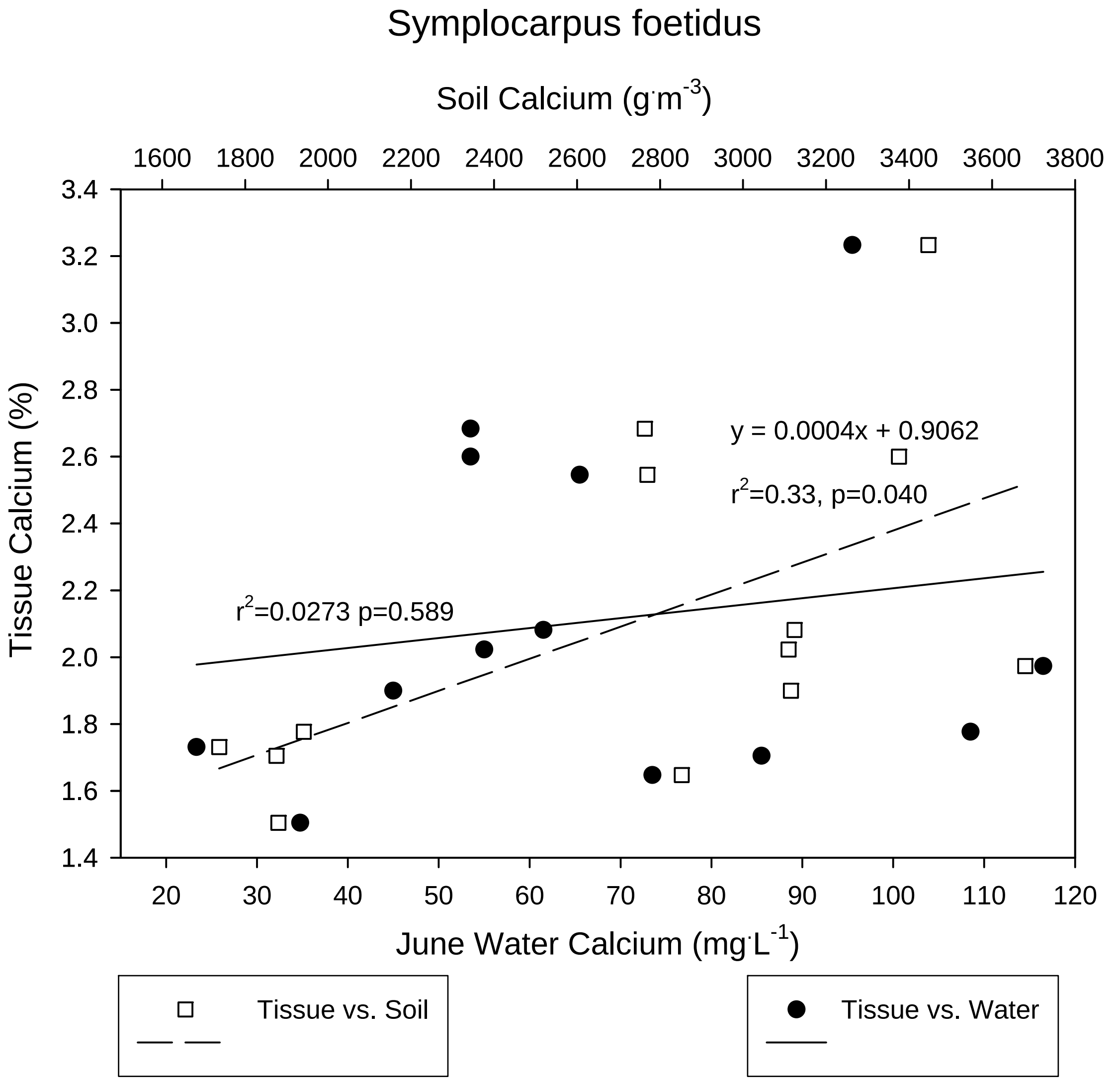


Figure 6.6. Percent calcium in the tissues of *Symplocarpus foetidus* as compared to mean water calcium in June and soil calcium. Equations and statistics refer to the line that they are in closest proximity to.

species has the ability to uncouple electron transport and ATP production and release energy as heat (Solomon et al., 1996). This gives this species the ability to begin growth early in the spring when snow is still on the ground. Even though the leaves collected for analysis were the most recently mature leaves, this process may have resulted in their development far earlier in the season than the other species sampled. Thus, water measurements taken may not have been reflective of the growing conditions when the leaves formed. The soil calcium could more accurately reflect the average pool of available calcium released into the system and thus reflect this earlier period of uptake.

No significant correlations were observed between tissue and soil/water calcium for *Carex flava*, *Carex hystericina*, *Dasiphora fruticosa, Eriophorum viridicarinatum, Fragaria vesca, Mentha arvensis, Solidago patula,* and *Thelypteris palustris* (Table 6.2).

Table 6.2. Statistics for species with no correlation between tissue calcium and soil and mean water calcium in June.

Species	water		soil	
	r^2	p	r^2	p
Carex flava	0.044	0.434	0.059	0.365
Carex hystericina	0.196	0.200	0.013	0.756
Dasiphora fruticosa	0.060	0.272	0.0002	0.950
Eriophorum viridicarinatum	0.038	0.616	0.084	0.449
Fragaria vesca	0.038	0.591	0.192	0.205
Mentha arvensis	0.176	0.154	0.002	0.888
Solidago patula	0.002	0.895	0.014	0.715
Thelypteris palustris	0.004	0.793	0.049	0.350

Summary

Parnassia glauca, Equisetum fluviatile, Symphyotrichum puniceum, Dasiphora fruticosa and *Packera aurea* are recognized as calciphiles in the literature as discussed in Chapter 4, but of these only *Parnassia glauca* and *Packera aurea* were shown in Chapter 5 to respond in distribution and abundance to environmental calcium levels. All of these species except *Dasiphora fruticosa,* do however, display a significant correlation between levels of calcium in their tissues and calcium in the environment.

The occurrence of *Equisetum fluviatile* and *Symphyotrichum puniceum* in calcareous fens indicates that these plants are obviously tolerant of elevated calcium levels even though they may not need those high levels for increased growth or health. Since they are accumulating this element, they must have a mechanism for keeping it from affecting vital processes.

It was interesting to observe that neither *Dasiphora fruticosa*, *Carex flava*, nor *Carex hystericina* showed correlations in calcium accumulation to calcium availability since these species were originally deemed calciphiles in Chapter 4. This study, however, showed that although these three species were present in calcareous environments, their distribution and abundance did not change in relation to variations in environmental calcium levels. Thus, these species may be excluding calcium actively from their tissues beyond a certain point of accumulation. Perhaps, these plants are found once a slightly higher level of calcium exists, as compared to 'non-calciphiles', which is why they occur where they do. Thus, all of the sites in this study may have exceeded the minimum calcium threshold for these species. In addition, abundance of *Dasiphora fruticosa* was determined in Chapter 5 to be correlated strongly with gradients in pH, more so than calcium, and thus it makes sense for this species to exclude calcium if pH affects its distribution most.

Similar calcium-independent relationships can be concluded for the species that also had no abundance changes in relation to calcium as determined in Chapter 5, i.e., *Mentha arvensis*, *Solidago patula*, *Eriophorum viridicarinatum*, *Thelypteris palustris*, and *Fragaria vesca*. This finding is consistent with the generalist nature of *Thelypteris palustris*, but presents an interesting situation for *Solidago patula* and *Eriophorum viridicarinatum* who appeared to be calciphiles from early literature reports. These findings suggest that these species have a weak relationship with calcium, even though they are found mostly in calcareous environments.

It was very curious to observe that both *Mentha arvensis* and *Symplocarpus foetidus,* common wetland species, showed substantial levels of calcium accumulation that correlated with water or soil calcium levels. The precipitation of calcium oxalate crystals has been observed in species in the Lamiaceae (as is *Mentha arvensis*) (Kinzel, 1983) and in *Symplocarpus foetidus* (Higley, 1880) and this process may relate to why these plants can absorb large quantities of calcium. This action does not mean that these species are calciphiles as they exist in areas without high calcium levels but suggests that their needs for calcium are low and that they can grow in environments where calcium is elevated as they have a successful mechanism by which to tolerate it.

Magnesium

Plants absorbed 0.1 to 0.9 % magnesium into their tissues; however, the average was less than 0.5%. *Parnassia glauca* had the greatest average tissue magnesium at 0.7% (Table 6.3). Only two species, *Thelypteris palustris* and *Carex flava*, showed significant correlations between the magnesium in their tissues to the magnesium in the water or soil. Tissue magnesium in *Thelypteris palustris* had a highly significant correlation to the water magnesium in June (the month proceeding its leaf collection) (r^2=0.73 p=0.007) (Figure 6.7). *Carex flava* (Figure 6.8) had a significant correlation with the water magnesium for all months proceeding and during its collection, and thus the water magnesium for June is reported (r^2=0.75 p=0.012). In addition, this species showed a very strong correlation to the soil magnesium as well (r^2=0.77, p=0.009).

Table 6.3. Mean percent magnesium and percent magnesium range in the tissues of the thirteen species collected from the eight study sites. Means are presented ± standard deviation. Means with different letters are significantly different by Tukey's Test (p=0.05). Composite samples (n) = number of plots where that species occurred. Number of sites where these samples were collected are included for reference.

Species Code	% Magnesium	% Magnesium Range	Composite samples	Sites
Parnassia glauca	0.7 ± 0.1 a	0.5 – 0.9	8	6
Mentha arvensis	0.6 ± 0.3 ab	0.2 – 1.2	8	6
Packera aurea	0.4 ± 0.1 abc	0.3 – 0.6	5	5
Equisetum fluviatile	0.4 ± 0.04 abc	0.4 – 0.5	5	4
Symplocarpus foetidus	0.4 ± 0.1 abcd	0.4 – 0.6	4	4
Solidago patula	0.3 ± 0.1 bcd	0.2 – 0.4	5	4
Thelypteris palustris	0.3 ± 0.1 cd	0.3 – 0.5	8	8
Dasiphora fruticosa	0.3 ± 0.03 cd	0.2 – 0.3	6	6
Carex flava	0.3 ± 0.1 cd	0.2 – 0.4	7	7
Fragaria vesca	0.3 ± 0.1 cd	0.1 – 0.5	6	6
Symphyotrichum puniceum	0.2 ± 0.1 cde	0.1 – 0.4	9	6
Carex hystericina	0.2 ± 0.1 cde	0.1 – 0.4	7	6
Eriophorum viridicarinatum	0.1 ± 0.1 de	0.1 – 0.2	7	5
Carex granularis	0.09 ± 0.1 de	0.0 - 0.18	2	2

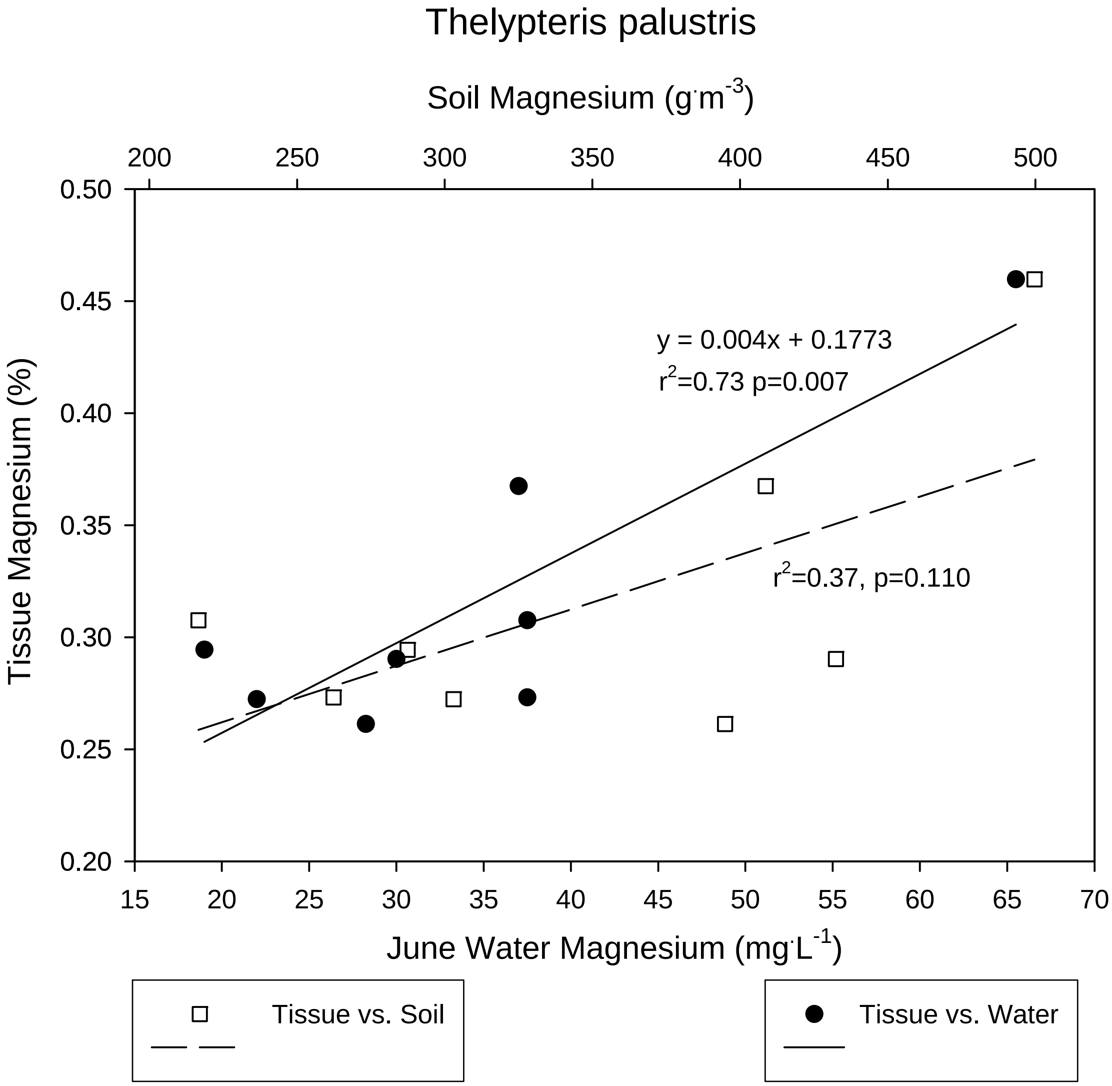


Figure 6.7. Percent magnesium in the tissues of *Thelypteris palustris* as compared to mean water magnesium in June and soil magnesium. Equations and statistics refer to the line that they are in closest proximity to.

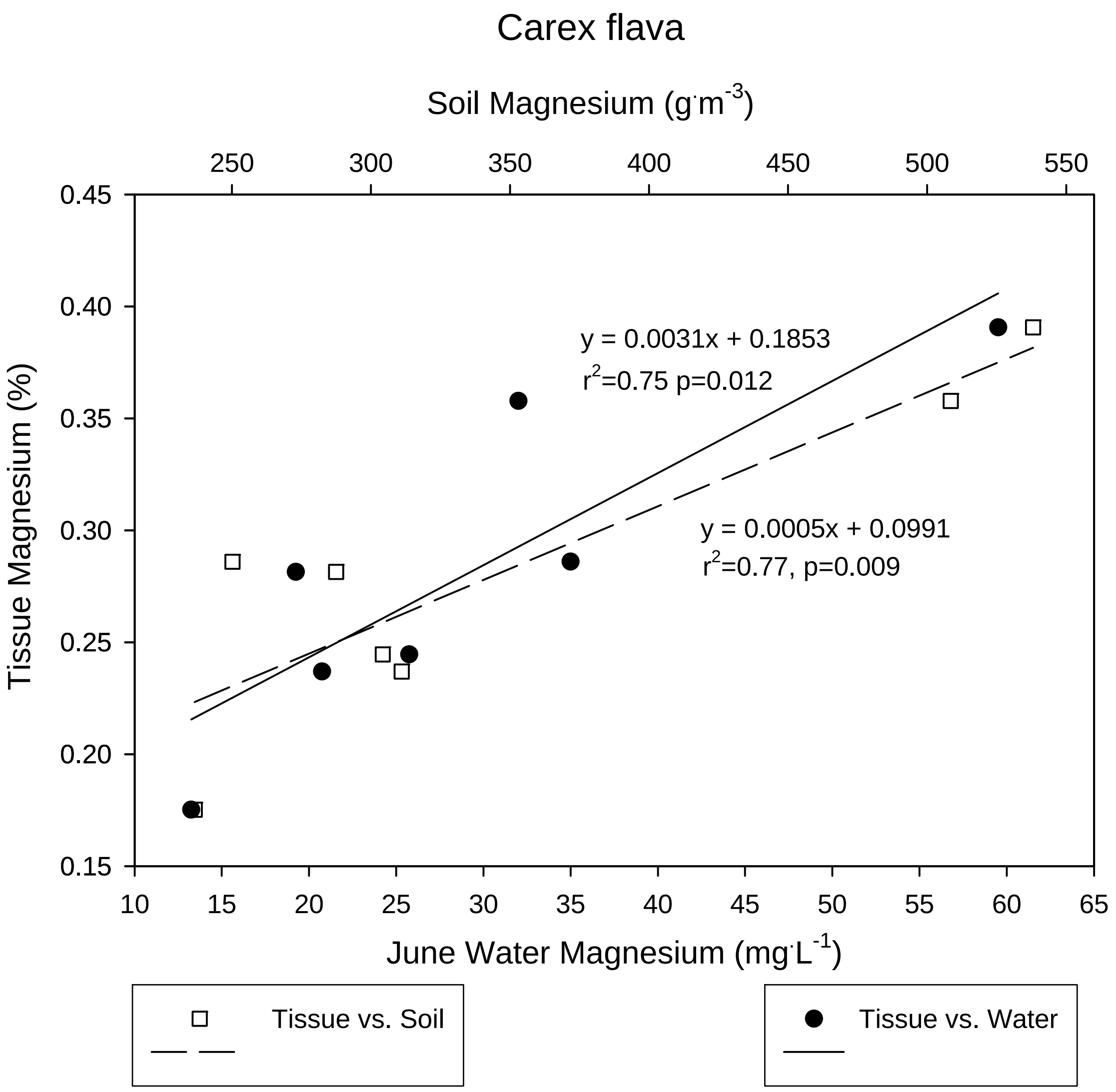


Figure 6.8. Percent magnesium in the tissues of *Carex flava* as compared to mean water magnesium in June and soil magnesium. Equations and statistics refer to the line that they are in closest proximity to.

Carex flava and *Thelypteris palustris* present interesting comparisons to the species that responded physiologically to environmental calcium. *Carex flava* was considered to be a calciphile, but appeared to restrict calcium accumulation in its tissues. This response was interesting since this species did not appear to increase in abundance in response to environmental calcium as described in Chapter 5. This accumulation of magnesium in relation to the available supply would seem to suggest that the environmental magnesium concentration, rather than calcium, is somehow important to the growth of this species. Perhaps this species is considered to be a calciphile because it occurs in calcium-enriched areas, but in actuality is selecting calcareous fen habitats for the elevated magnesium levels associated with them. This may particularly be where dolomitic limestone dominates.

Thelypteris palustris, which was considered to be a common wetland fern, exhibited a strong relationship to the magnesium it absorbs in relation to water magnesium levels (Figure 6.7). This activity is particularly interesting as it did not absorb calcium in proportion to the available calcium supply. The reasons as to why this species would accumulate greater of amounts of magnesium into its tissues is unknown; however, the accumulation may be associated with either a need for or tolerance to magnesium, explaining why it is also common in calcareous fens.

Magnesium is essential for plants as it is the central atom of the chlorophyll molecule (Walker and Weinstien, 1991) and is involved in regulation of cellular pH and ionic balance (Marschner, 1995) among other processes. The accumulation of magnesium in the current study was higher than the amount of magnesium generally required by plants (0.15 - 0.35%; Marschner, 1995) and is indicative of an abundant supply (Marschner, 1995). It is interesting that *Thelypteris palustris* and *Carex flava* did not have the highest levels of tissue magnesium in this study even as they appeared to accumulate more magnesium as more was available. Interestingly, the other calciphiles had higher tissue magnesium levels than these species but did not exhibit continuously enhanced accumulation as the supply increased.

Since in calcareous fens, water calcium and water magnesium often are correlated closely (Chapter 3), changes in plant distributions associated with calcium could just as easily be related to magnesium. Although similar plant communities often occur in nondolomitic-influenced fens, these results convey the importance of calcium to the calciphiles. At most of the sites, calcium and magnesium absorption was competitive as values exceeded 0.001M (Marschner, 1995). All of the species that showed significant tissue-environmental calcium correlations did not show the same relationship to magnesium. This finding indicates that while the calciphiles selectively allowed calcium into their tissues, they actively excluded magnesium. Thus the magnesium supply appears to be less important for most calciphiles as compared to the calcium supply. By contrast, *Carex flava* (a calciphile) and *Thelypteris palustris* (a generalist), may have a greater physiological need for magnesium, which explains their increased uptake of it (to the exclusion of calcium) and details possible reasons that they are found in calcareous fens.

Literature Revisited

In general, species tissue calcium fell within the expected range of 0.1% – 5.0% dry weight (White and Broadley, 2003), and *Parnassia glauca* showed the highest levels and most remarkable changes in accumulation as water concentrations increased. Monocots had generally lower values (<1%) than dicots (>1%). The general conclusions of this study validate the findings of Clarkson (1965), who reported that calciphiles absorb calcium in larger amounts than non-calciphiles. It is interesting however that not all species originally suspected to be calciphiles showed this relationship. All of the species that showed a response to environmental calcium in their abundance did show increased levels of calcium in their tissues in response to increased calcium in the environment. However, it appears that among species for which abundance was not affected by environmental calcium levels, some restrict its absorption and some do not.

It is difficult to compare the results of the exact concentrations of calcium and magnesium in the plants studied with previous research since for many of these species these data have never been reported. For *Dasiphora fruticosa*, however, Marosz (2004) measured the accumulation of calcium and other elements along an experimental soil salinity gradient. He reported that calcium values in leaves varied between 0.71 and 0.89 % dry weight in a solution that had an undisclosed amount of calcium in it. The calcium concentration of the leaves of *Dasiphora fruticosa* analyzed in this study were higher (1.0 - 1.6 %). This result may indicate that in calcareous fens, the substrate calcium levels are elevated beyond the minimum threshold required for this species. Thus, the plants may have higher average tissue concentrations, but that accumulation is not related to environmental levels. Another explanation for why the values were lower in Marosz's study could be that he studied a different variety of *Dasiphora fruticosa* in which its calcium uptake differed from the native North American varieties. Marosz observed changes in the accumulation of calcium in the tissues, but this response was related to increased soil salinity. It was proposed that this response had to do with the changes in osmotic potential as a result of increased sodium chloride (NaCl); however, other research has indicated that the uptake of calcium decreases when NaCl concentrations are high (Lynch and Lauchli, 1985) as the NaCl might inhibit calcium transport from the external medium into the root xylem under certain circumstances.

Previous research on the calcium accumulation in *Geum rivale* had reported that it had

3.4 % calcium in its leaves (Waldren et al., 1987). Although the value measured the current study was higher than many of the dicots (1.9%), it was not nearly as high as observed by Waldren et al. (1987). Since this species occurred only at an elevated calcium threshold, a very strong relationship may exist between its tissue calcium and environmental calcium. Since only one sample was collected and analyzed for calcium, relationships to the environment were not established, and if perhaps, this one sample was collected where environmental levels were lower than in the current study, its tissue level may also be lower. Waldren et al. (1987) does not report the substrate calcium conditions for comparison.

Auclair (1979) found that *Equisetum fluviatile* had elevated concentrations of iron and calcium in its tissues as compared to other elements. Calcium values measured 1.8%; however, the water calcium and iron were not measured. This value corresponds to the mean tissue calcium of *Equisetum fluviatile* measured in this study (1.9%). This high accumulation of calcium (in addition to iron as mentioned in Chapter 5) indicates the strong ability for this species to sequester ions in general and that this species has a unique response to calcium and iron in the environment.

Conclusions

Although this study has not analyzed how calcium is used in plants of calcareous fens, it has established that five species common to calcareous fens (*Parnassia glauca, Packera aurea, Symphyotrichum puniceum, Equisetum fluviatile,* and *Symplocarpus foetidus*) accumulate calcium in response to amounts available in their environment. It is evident that these species grow well in calcareous environments because they are either protected from the toxicity of excess calcium, or they actually need the calcium for various aspects of their growth and development. In addition, not all plants maintain the same levels of calcium in their tissues, and there are large differences among species. The most important finding in relation to these trends is the confirmation that the species that showed the strongest correlation of their tissue calcium to environmental calcium levels are species that are associated with calcareous environments. Some of these species showed changes in distribution and abundance with changes in environmental calcium (*Parnassia glauca, Packera aurea)*, and some did not *(Symphyotrichum puniceum* and *Equisetum fluviatile).* On the contrary, most of the non-calciphiles were restrictive in the amount of calcium they accumulated and maintained steady levels as calcium supply increased. Species originally deemed calciphiles (based on the literature) such as *Dasiphora fruticosa, Carex hystericina*, and *Carex flava* did not show a significant relationship between their tissue calcium and environmental calcium levels, and this observation may be related to the lack of relationship between environmental calcium and their abundance as was previously determined in Chapter 5. The subsample analysis of tissue magnesium highlighted the relationship of *Carex flava* and *Thelypteris palustris* to environmental magnesium levels as their tissue magnesium was correlated to the environmental magnesium levels and not to the calcium supply.

CHAPTER VII

CONCLUSIONS

This study has reinforced current knowledge that calcareous fens are unique and fascinating ecosystems. Although these wetlands exhibit the quality of being enriched with calcium, the degree to which it accumulates in plants varies widely. In addition, processes bringing groundwater to the surface are not necessarily similar among fens in close proximity to each other, as site geochemistry showed significant variation. As a result, patterns of species distribution vary among and within fen ecosystems. This thesis identified individual species for which distributions and physiological responses appear to be linked strongly to the chemical properties of calcareous fens. However, some original hypotheses on the relationships between specific plants and the chemical factors were rejected: some calciphiles did not appear to show any response to environmental calcium other than being tolerant to elevated levels. The following section will revisit the original hypotheses and reevaluate them based on the new knowledge of plant selectivity in calcareous fens.

Hypotheses Revisited

The original hypotheses suggested that:

1) Specific environmental substrate calcium levels can be identified and a minimum and maximum *range* can be established for each calciphile as well as a *threshold* of calcium that must be met for a given species to occur.

This study successfully monitored the chemical and hydrologic environment of eight calcareous fens in the Berkshire-Taconic region of Western Massachusetts over the course of one growing season. Sites varied greatly in their soil and water calcium levels, and for some species, these differences appeared to influence their abundance. For species that occurred only above a specific calcium threshold, that level was noted. Likewise, for species for which abundance appeared to increase as environmental levels did, that range was noted. Although these values may not represent all absolute possibilities as to the geochemical environment in which these species occur, they provide an estimate (based on 24 sampling locations) as to the environmental calcium selectivity of these species as this had not been accomplished previously. Future research should expand the number of study sites and focus on these particular species to try to determine if this trend holds in general or across different climate regimes.

2) The most selective calciphile species (calcium specialists) that grow in calcareous fens will have the highest calcium concentrations in their tissues and these levels will correspond to environmental substrate calcium levels.

The species that appeared to be most responsive to environmental calcium levels were *Parnassia glauca, Geum rivale, Carex sterilis, Carex granularis, Carex leptalea*, and *Packera aurea* (the threshold and gradient species). Of these, calcium tissue concentrations in *Parnassia glauca* and *Packera aurea* were compared to environmental calcium levels. Both of these species showed significantly higher accumulation of calcium as environmental levels increased indicating that these species do not selectively exclude calcium from their tissues.

Although several non-calciphile species had calcium tissue concentrations greater than *Packera aurea*, *Parnassia glauca* had unusually high levels, at times above 5%. Analysis of *Geum rivale* and *Carex granularis* showed that they too had elevated calcium levels compared to the non-calciphiles, particularly *Carex granularis*, which had the highest concentration of the monocots. It appears that in general, plants that increase in abundance in relation to environmental calcium levels also have high concentrations of calcium in their tissues. In addition, these tissue levels increase as environmental levels increase; indicating that these plants' adaptations are extremely efficient at facilitating growth in calcareous environments.

3) The least selective species (calcium generalists) that grow in calcareous fens will maintain lower tissue calcium levels that will appear to be absorbed independent of environmental substrate levels.

The calcium generalists were determined to be *Carex flava*, *Carex hystericina*, *Equisetum fluviatile*, *Juncus nodosus, Solidago patula, Solidago uliginosa, Symphyotrichum puniceum, Symplocarpus foetidus, Thelypteris palustris, Dasiphora fruticosa*, and *Juncus brachycephalus*. Of these, tissue analyses were conducted on *Symphyotrichum puniceum, Symplocarpus foetidus, Thelypteris palustris, Dasiphora fruticosa, Solidago patula, Carex flava, Carex hystericina*, and *Equisetum fluviatile*. Most of these species did not show a significant relationship between the amount of calcium in their tissues and the environmental calcium levels; however, *Symphyotrichum puniceum, Equisetum fluviatile* and *Symplocarpus foetidus* did. Even more fascinating is that two of these species (*Symplocarpus foetidus* and *Equisetum fluviatile*) had the third and fourth (respectively) highest calcium in their tissues. Therefore, it does not appear that all generalists exclude calcium as environmental levels increase.

Modes of Growth in Calcareous Fens

In conclusion, there appear to be several factors that regulate a plant's occurrence and growth in a calcareous environment.

1) Certain plant species increase in abundance as calcium does and have elevated levels of tissue calcium that increase as environmental levels do. These plants are ecological calciphiles but without knowledge of the amount of water soluble calcium in their tissues, it cannot be said whether they are also physiological calciphiles. Nevertheless, these species have strong adaptations to dealing with large amounts of calcium in their systems and to the other limitations associated with calcium-rich environments. Although speculative, these plants may have a physiological need for calcium that is satisfied when they grow in a calcareous fen. These plants can be said to be calciphile specialists and include *Parnassia glauca, Packera aurea, Geum rivale, Carex granularis* and possibly *Carex leptalea*.

2) Certain calciphiles do not increase in abundance relative to environmental calcium levels and exclude calcium from their tissues. These plants are thus tolerant of elevated calcium levels and are perhaps better competitors in those environments because they have well-suited mechanisms to avoid the toxicity that calcium can impose and/or benefit from non-calcium related aspects of fen geochemistry. The distribution of these species cannot be linked directly to environmental calcium levels. Thus, plants of this sort may have more than one physiological response to dealing with the excess calcium. It is likely that they either A) exclude the calcium from their tissues and maintain constantly lower levels, or they B) have an ability to absorb higher calcium levels without it affecting physiological processes. Condition A) is represented by species, such as *Carex*

hystericina, Eriophorum viridicarinatum, and *Solidago patula,* which exhibited no relationship to environmental calcium other than growing in elevated levels of it. Condition B) is represented by *Symphyotrichum puniceum, Equisetum fluviatile* and *Symplocarpus foetidus*, which all had higher concentrations of calcium and accumulated calcium in greater amounts as environmental calcium levels increased. Both of these conditions could include a third response, in which growth in a calcareous fen appears to be associated with accessory benefits such as elevated pH or increased magnesium levels. Species that appeared to fall into this growth regime were *Dasiphora fruticosa* and *Juncus brachycephalus*, which appeared to respond to pH gradients and *Carex flava* and *Thelypteris palustris*, which may have a physiological response to magnesium. For species of this sort that are ecological calciphiles, it can be said that they are calciphile generalists. However, while behaving as calcium generalists, *Thelypteris palustris* and *Symplocarpus foetidus* would not be considered ecological calciphiles due to their occurrence elsewhere.

Future Research

This project has analyzed many new and interesting aspects of plant relationships and environmental substrate gradients in calcareous fens. However these new answers have fueled new questions. The following section will list research that needs to be considered in this area to strengthen the investigations presented in this thesis.

1) The current study could be expanded to determine if the same species-environmental gradient relationships hold true for a larger geographic or climatic region. Investigations of the responses of *Juncus brachycephalus* and *Dasiphora fruticosa* to pH gradients are needed to verify and explain them. The response of *Equisetum fluviatile* to

an iron gradient instead of a calcium gradient needs further study. Whether *Geum rivale*, *Carex sterilis* and *Carex granularis* ever occur below the calcium threshold established in this study needs further verification as does whether *Parnassia glauca* and *Packera aurea* continue to show distribution increases as environmental calcium increases.

2) A similar study as to this one could be repeated in which more leaf samples of *Geum rivale*, *Carex granularis* and *Carex leptalea* are collected from multiple sites and analyzed for calcium. Currently it is still not clear whether these three species absorb calcium in proportion to environmental availability.

Overall, only subsets of the plants collected were analyzed for tissue magnesium. A similar study should be conducted in which the tissues of a suite of species growing across an environmental magnesium gradient are analyzed for magnesium and compared to the environmental levels. It would be important to focus on *Carex flava* and *Thelypteris palustris*, as they showed very strong correlations to the magnesium supply in the current study. In addition, sites where *Carex flava* occurs across the country should be compared with geologic information to see if its distribution relates to calcareous fens that are fed by groundwater that has passed through dolomite limestone and would thus have a significant magnesium input.

No rare species were analyzed in this study and thus it would be interesting to see if the tissues of some of the endangered and threatened species of calcareous fens also absorb elevated levels of calcium.

3) Although perhaps a difficult question to answer, it would be interesting to understand what plants in calcareous fens do with the extra calcium that they absorb. This would include an analysis of the forms of calcium that it is stored in and where it is

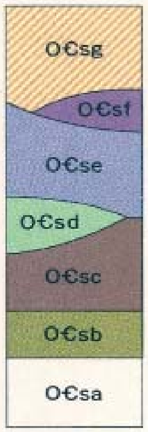
stored. If calcium in the tissues of the ecological calciphiles is soluble as calcium malate, then these species would also be physiological calciphiles. It would be interesting to determine which species, if any, are of this nature. In addition, it is still unclear if the ecological calciphiles need more calcium and if so, for what purpose. For the generalists species (and physiological calcifuges), it would be interesting to understand the dominant forms that calcium is stored in and/or how they may exclude it.

4) All study sites shared typical characteristics of calcareous fens. They all had elevated calcium levels, elevated pH, extended periods of saturation, and plant communities that included many calciphiles. There were, however, large differences in all of these characteristics among sites, even among those that were adjacent. It would be interesting to understand the sources and reasons of this variability and provide more detailed characterizations of the hydrogeochemical settings. This investigation would involve tracking groundwater movement and determining the specific bedrock types to which upwelling groundwater in each site was exposed and how known atomic ratios of elements in the surrounding bedrock relate to those measured in the sites. It will be important to determine the proximity of the point of delivery of groundwater to the site and verify whether sites are supplied direct upwelling groundwater or how long that water may have traveled through the soil as subsurface flow. In addition, it will be important to monitor if the waters are diluted by other sources (i.e. nearby streams or non-calcareous groundwater flow paths) and how these and other factors may influence the pore-water geochemistry.

APPENDICES

APPENDIX A

STOCKBRIDGE FORMATION



Stockbridge Formation

Lower Cambrian to Lower Ordovician

CAMBRIAN AND ORDOVICIAN

O€sg, *massive, white to gray mottled marble, weathering into smooth, glistening surfaces; interbeds of uniform, compact, homogeneous, pale yellow-gray dolostone a few feet thick showing boudinage structure and weathering cream colored and with craggy surfaces.*

O€sf, *a discontinuous, heterogeneous unit, whose thickness varies rapidly along strike. Lithic types include (1) calcareous sandstone showing prominent cross-stratification on 6-inch scale; (2) silty, gray, fine-grained limestone; (3) massive calcareous quartzite; and (4) massive gray dolostone.*

O€se, *massive gray, white, or gray-mottled white calcite marble weathering into smooth, glistening surfaces. Locally rock is mottled by irregular, gray, rough-weathering dolomite masses about 1 cm across. Rare beds of cream-weathering massive dolostone. The unit on the whole resembles* O€sg *and stratigraphic association is essential to its proper assignment.*

O€sd, *heterogeneous, discontinuous unit of widely ranging thickness, consisting of several rock types: (1) silty, impure limestone; (2) massive, cream-gray fine-grained dolostone; (3) calcareous siltstone; and (4) calcareous sandstone showing prominent cross-stratification on 6-inch scale on weathered surfaces. Unit as a whole resembles* O€sf, *and assignment of isolated outcrops to either unit may be uncertain.*

O€sc, *massive to sheared, iron gray to white dolostone; weathers pale gray; forms outcrops of rounded blocks. Individual beds are as much as 20 feet thick. Brownish-orange weathering dolostone with phyllitic partings locally present near the base of the unit, grading into* O€sb *below. Stratification on mm scale, defined by floating rounded quartz sand grains, manifest on weathered surfaces and is especially common near the top of the unit; cross-stratification rarely seen. Upper dolostone beds may be calcite-cemented and upon slight weathering turn into a soggy, sandy, flexible rock that readily soaks up water.*

O€sb, *pale gray, salmon, and white dolostone, massive, compact, and uniform; weathers pale cream to gray. Individual beds a few inches to several feet thick, interbedded with gray silty to green micaceous phyllite a few inches thick, or calcareous siltstone 10 feet thick or more. Beds of calcareous and feldspathic sandstone which weather dull green gray, as well as sparse dolomite quartzite and rusty-weathering siltstone, are diagnostic of the unit.*

O€sa, *(not exposed in area but shown at depth in section B–B′ and known from area to the east). Massive, uniform, smooth-weathering, gray, white, and mottled dolostone*

Figure A.1. The Stockbridge Formation (Zen and Ratcliffe, 1971)

APPENDIX B

SITE PHOTOS

Figure B.1. Vegetation community at Jug End North.

Figure B.2. Vegetation community and water monitoring instruments at Jug End South.

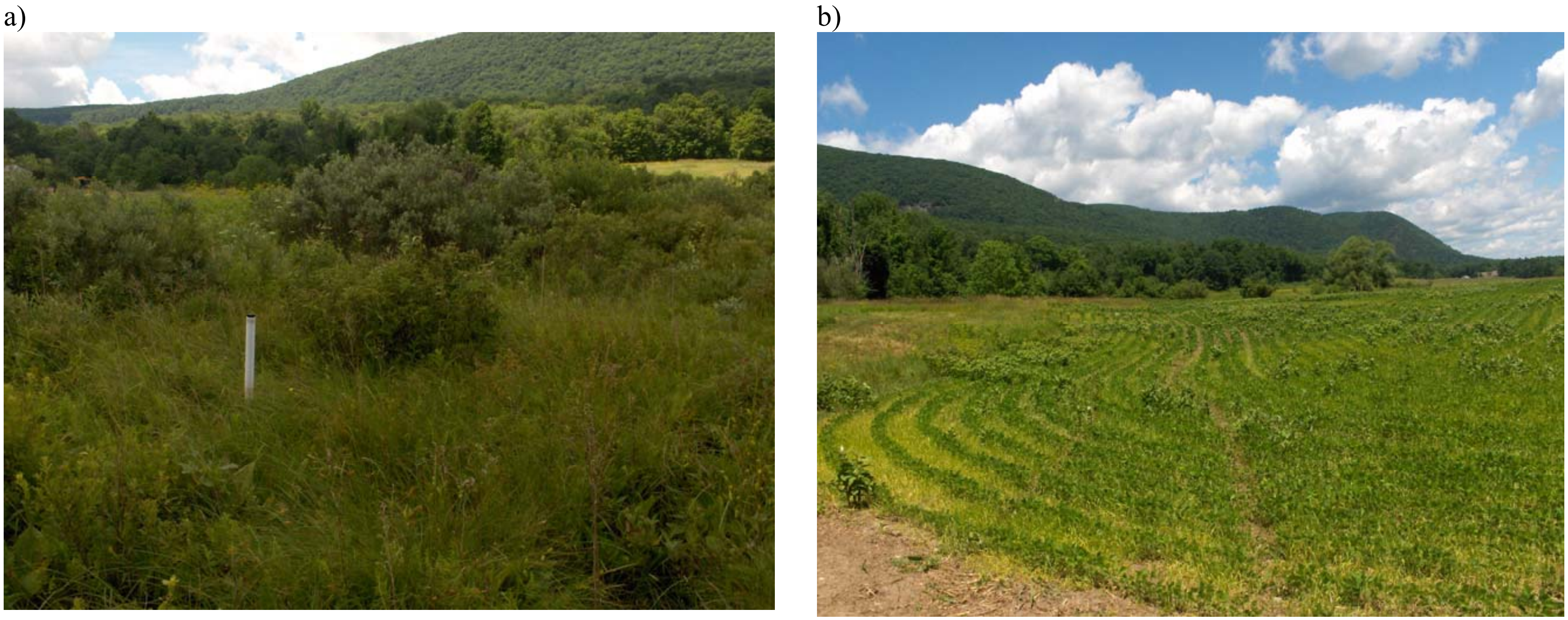


Figure B.3. General vegetation at of Schenob Brook Bartholomew: a) plot SBB3, b) adjacent agricultural fields.

Figure B.4. General view of vegetation at Schenob Brook North.

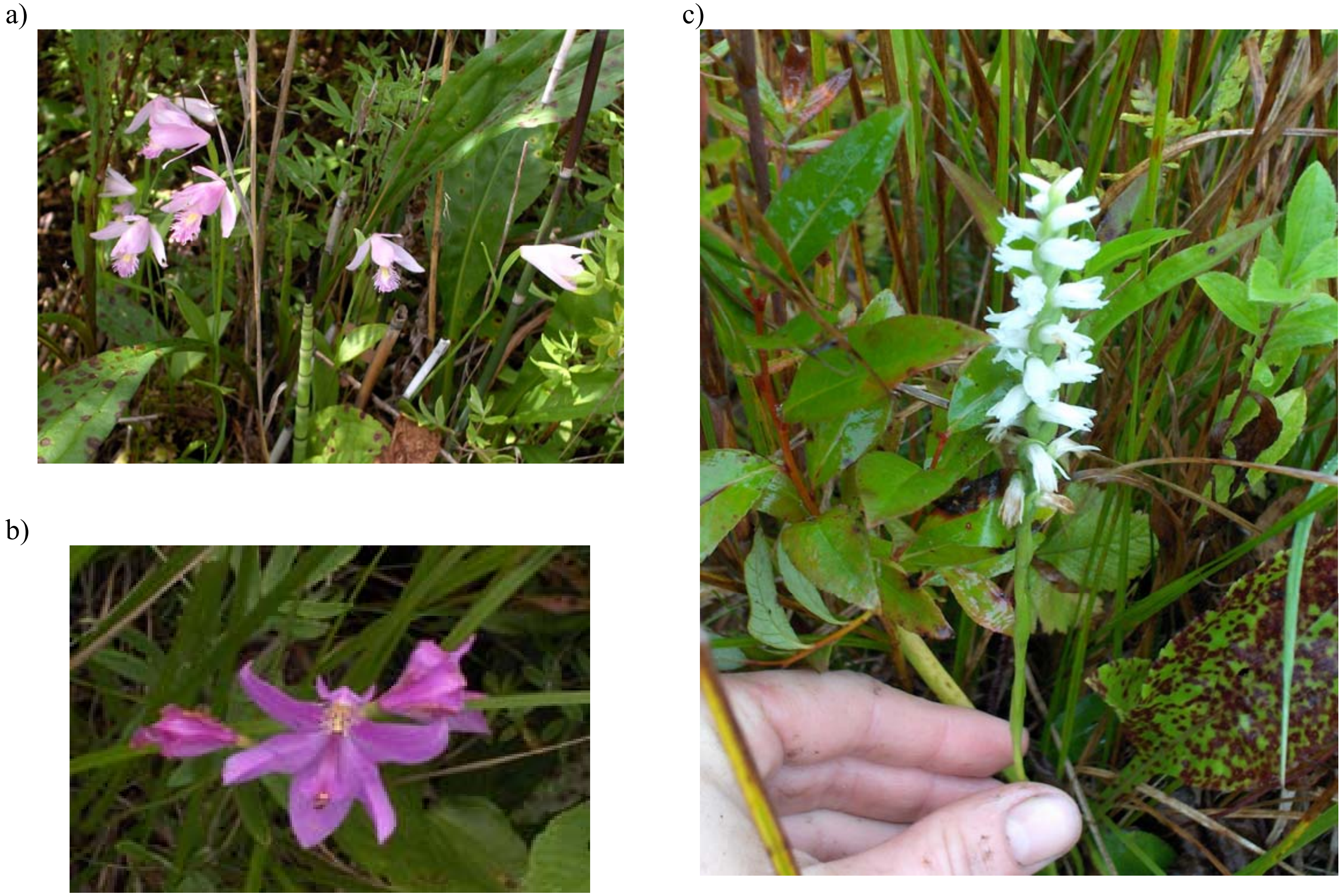


Figure B.5. Unique species at Schenob Brook North: a) *Pogonia ophioglossoides*, b) *Calopogon tuberosus*, c) *Spiranthes cernua*.

Figure B.6. Vegetation community at Schenob Brook Central in autumn of 2006.

Figure B.7. Hummock landscape at Schenob Brook South before the emergence of vegetation in March, 2006.

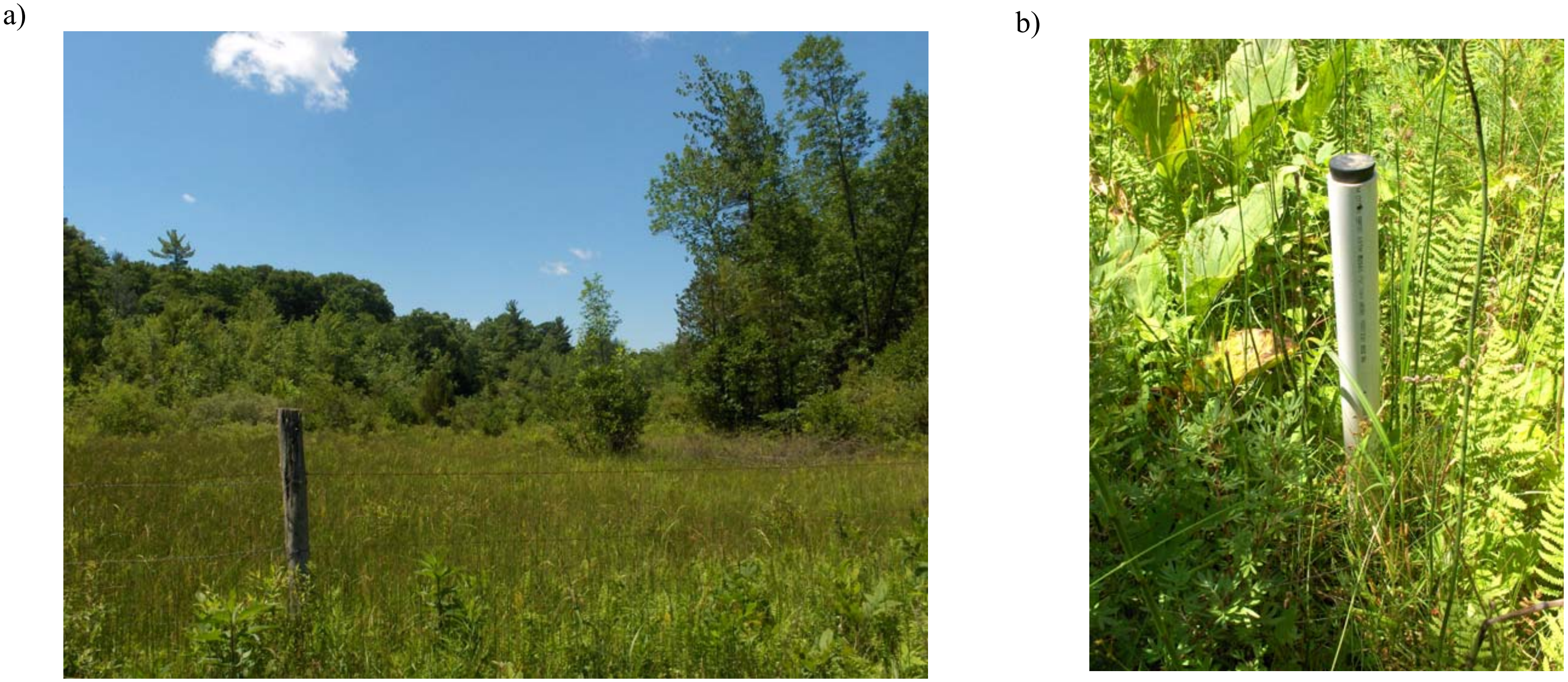


Figure B.8. Vegetation at Shmulsky Open: a) general fen view, and b) plant composition and sampling well at SO1.

Figure B.9. Vegetation cover at Shmulsky Shrub, plot 1.

APPENDIX C

QUALITATIVE ASSESSMENT OF REDUCED IRON

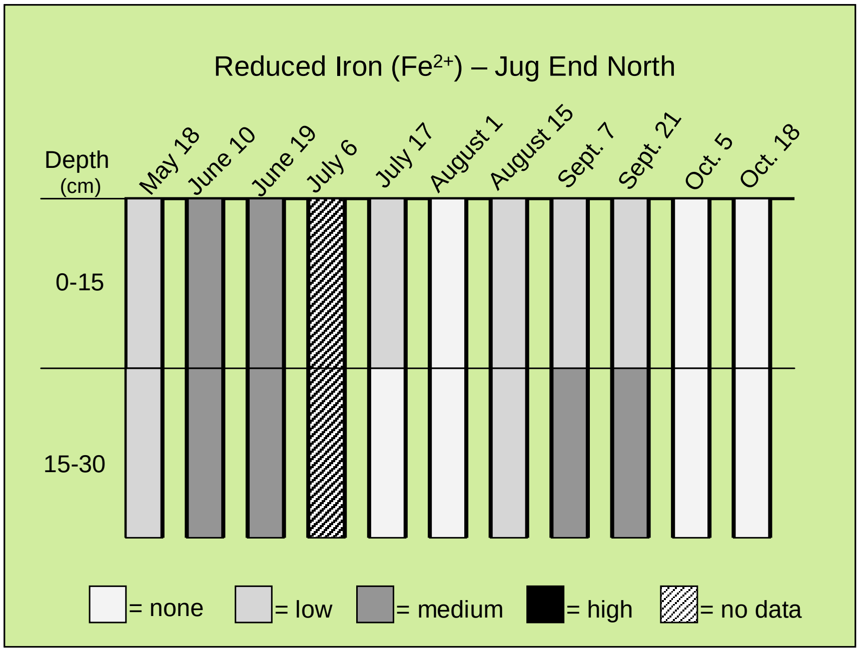


Figure C.1. Reduced iron at Jug End North (JN).

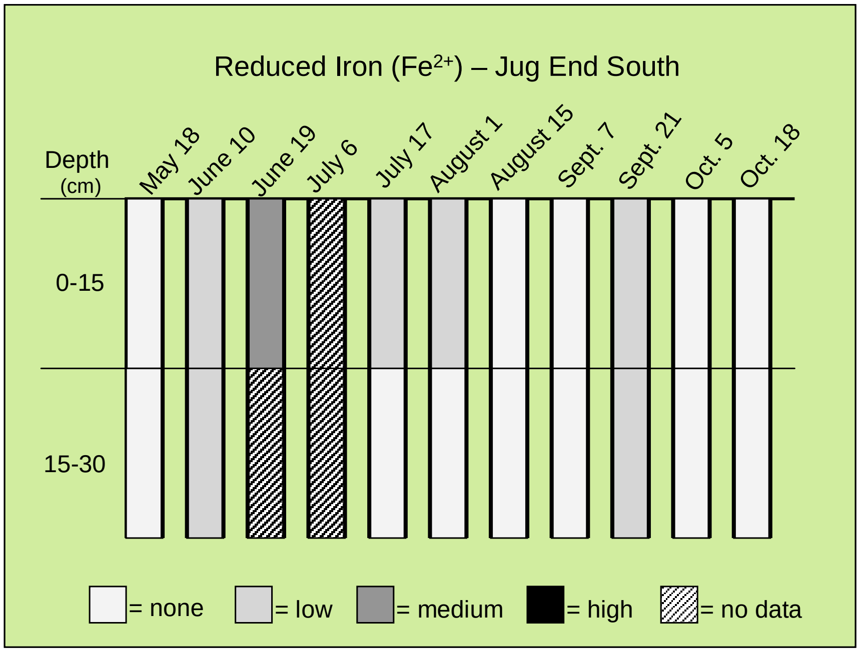


Figure C.2. Reduced iron at Jug End South (JS).

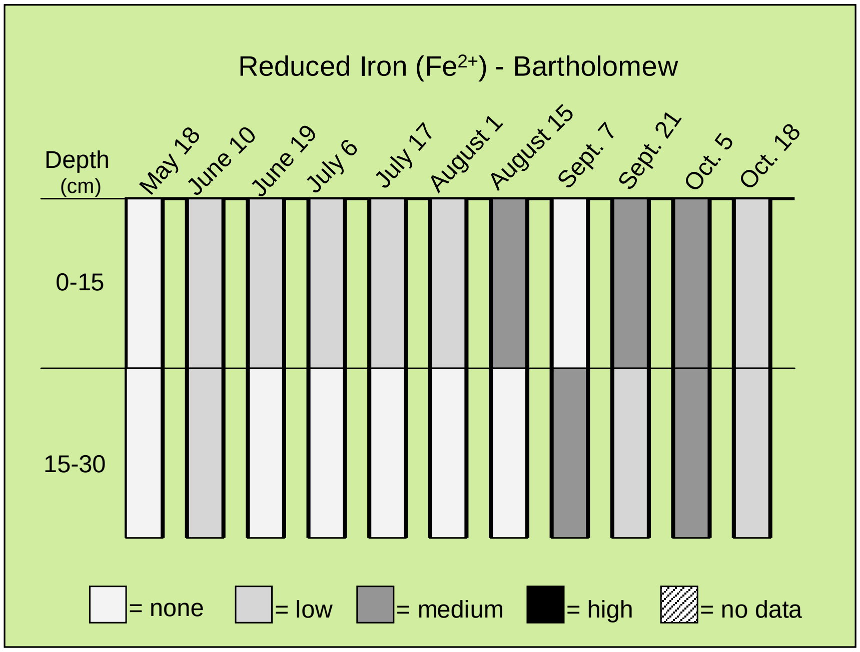


Figure C.3. Reduced iron at Bartholomew (SBB).

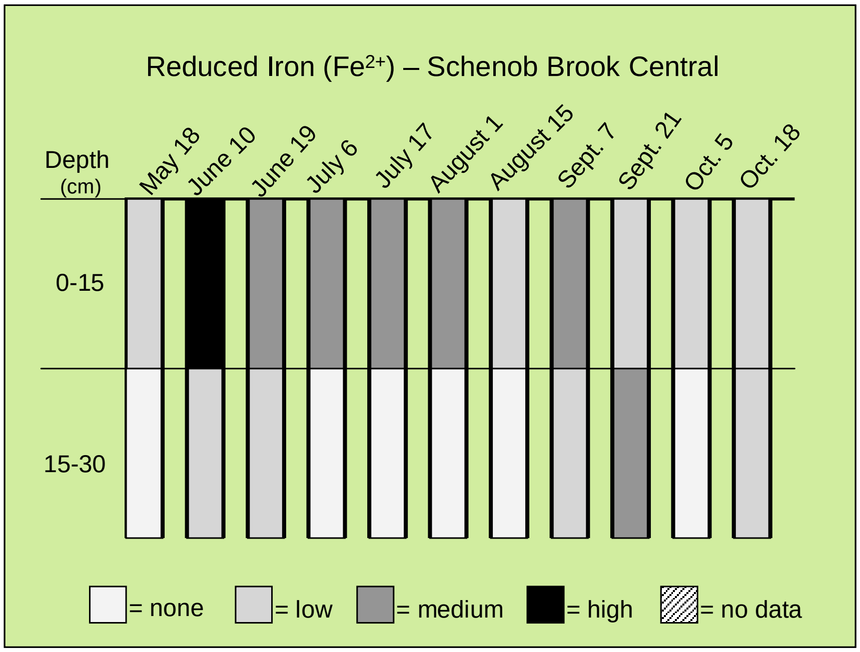


Figure C.4. Reduced iron at Schenob Brook Central (SBC).

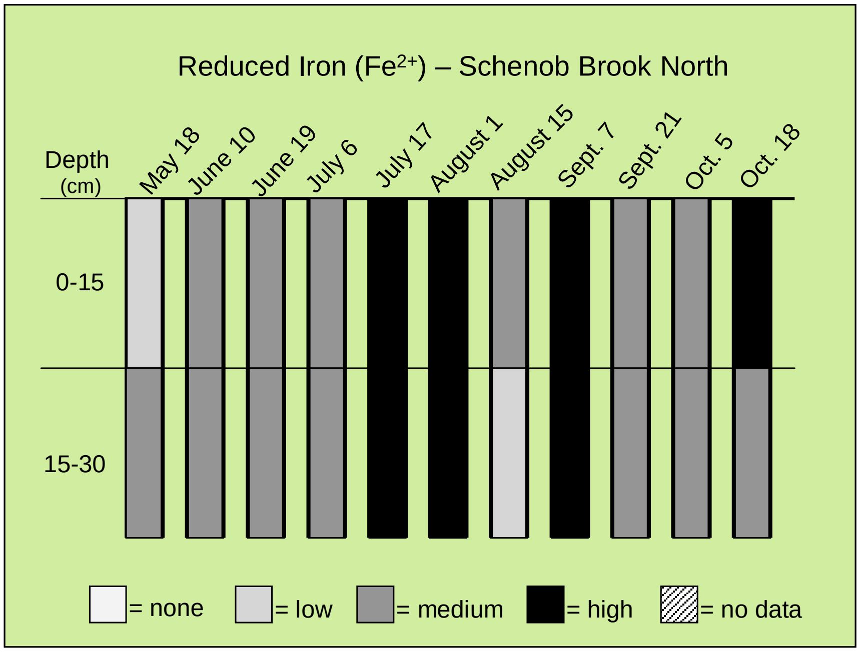


Figure C.5. Reduced iron at Schenob Brook North (SBN)

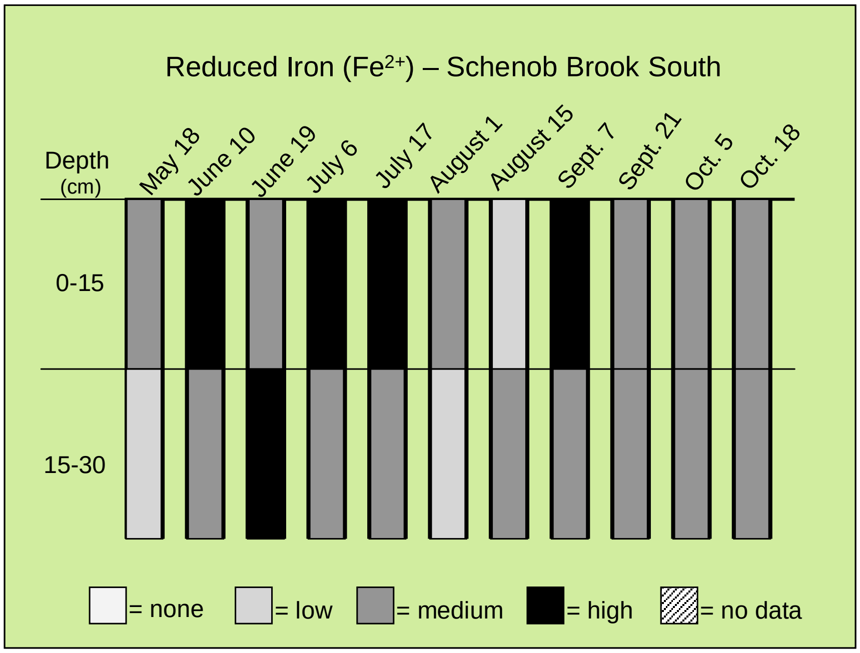


Figure C.6. Reduced iron at Schenob Brook South (SBS).

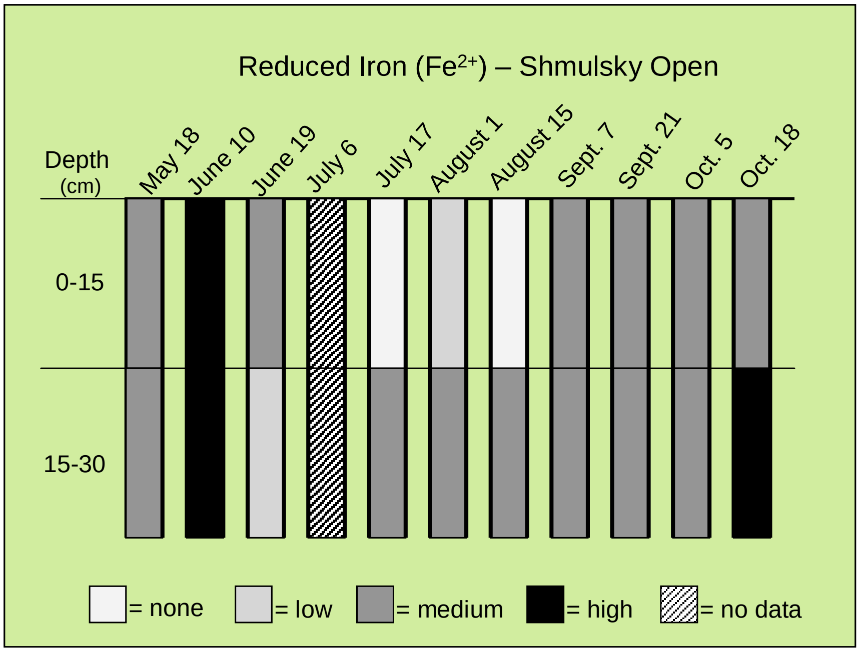


Figure C.7. Reduced iron at Shmulsky Open (SO)

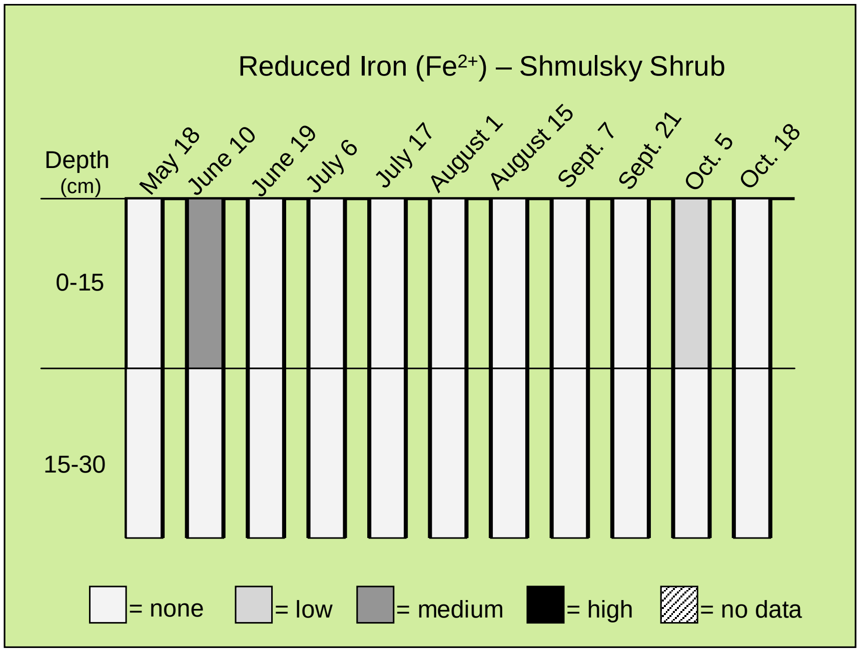


Figure C.8. Reduced iron at Shmulsky Shrub (SS)

APPENDIX D

SEASONAL VARIATION IN PH

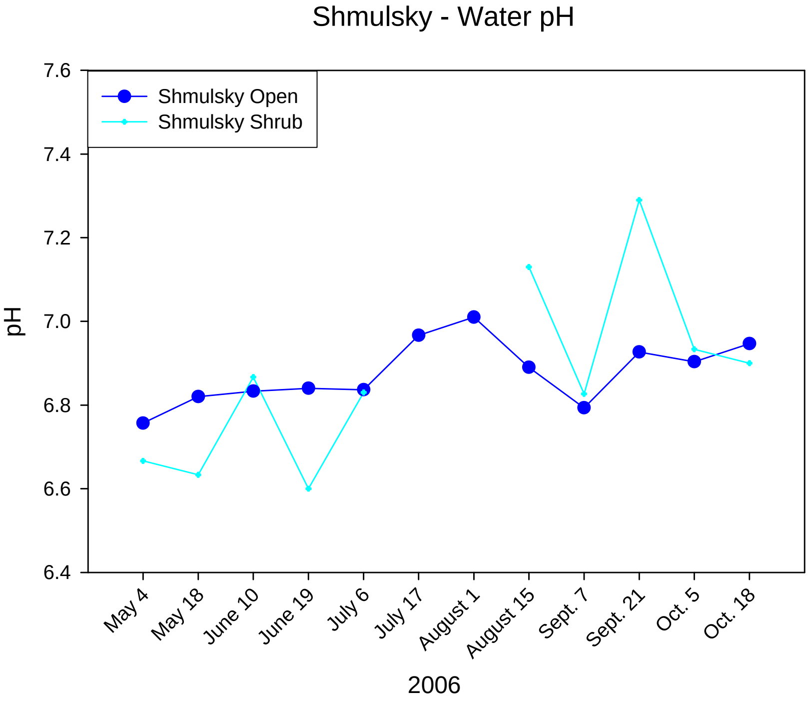


Figure D.1. Seasonal variation in water pH at the two sites in the Shmulsky location. Each point represents a mean of three replicate samples.

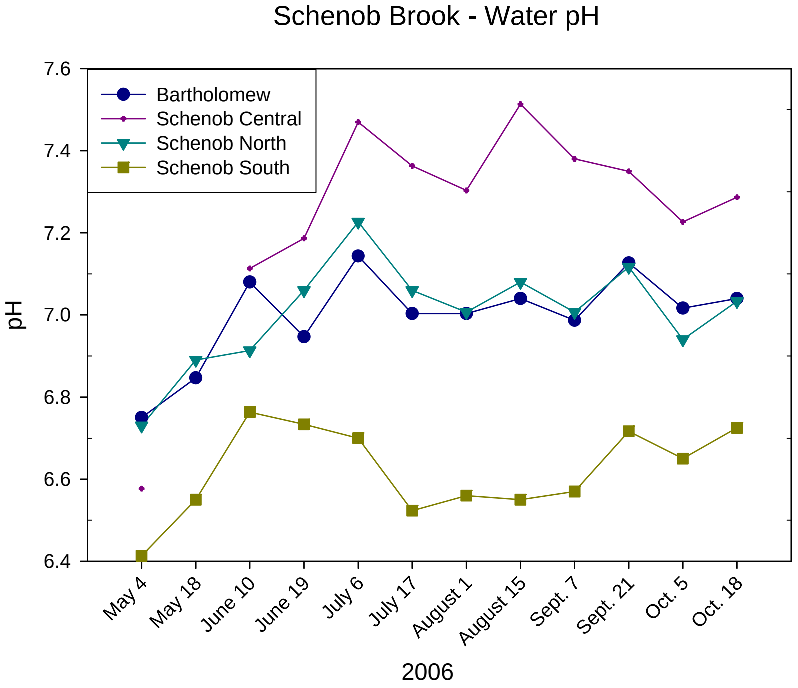


Figure D.2. Seasonal variation in water pH at the four sites in the Schenob Brook location. Each point represents a mean of three replicate samples.

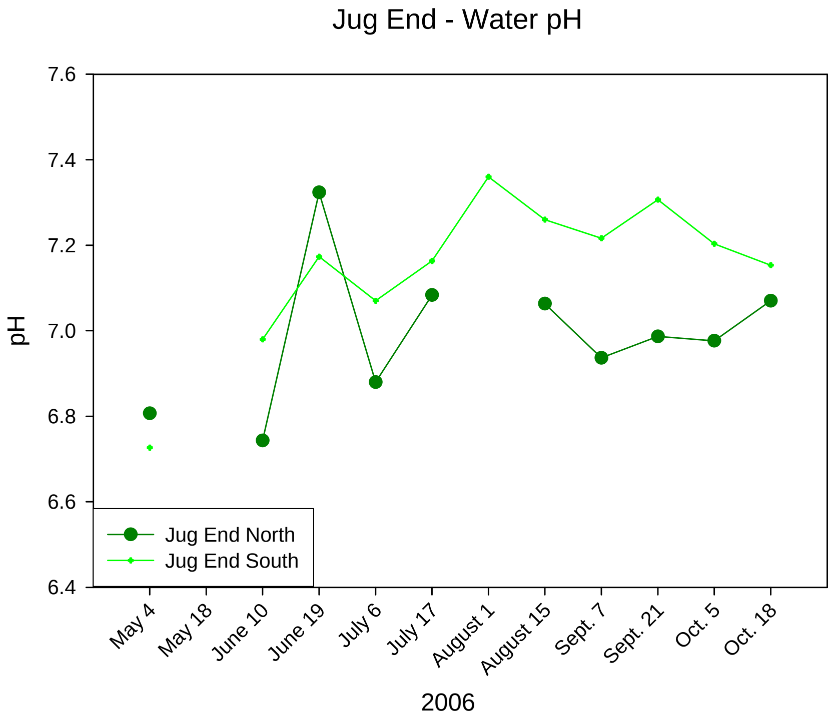


Figure D.3. Seasonal variation in water pH at the two sites in the Jug End location. Each point represents a mean of three replicate samples.

APPENDIX E

SOIL PROFILE DATA AND CHEMICAL CHARACTERISTICS

Table E.1. Soil profile descriptions for Jug End North. Soil chemical measurements are expressed by volume $(g\cdot m^{-3})$.

Site:	**Jug End North**			**Soil Taxonomic Classification:** Histic Humaquept									
Plot:	JN1												
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A	0-31	Silt Loam	2.5Y 5/2	10YR 4/6 45%	7.3	10.5	2226	248	25	0.34	76	0.81	4320
B_{eut}	31-40+	Loam	5Y 5/2	N1/ 1% 5Y8/4 1% 5YR6/3 1% 10YR 8/6 1%	8.2	2.4	1241	85	14	0.03			
Plot:	JN2	**Soil Taxonomic Classification:** Histic Humaquept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A	0-32	Silt Loam	2.5 Y 3/1		7.1	11.0	2360	276	33	0.26	118	0.71	4020
B_g	32-50+	Sandy Loam	2.5Y 4/2	2.5Y 4/4 20% 7.5YR 5/1 1%	7.4	7.8	2299	256	26	0.07			
Plot:	JN3	**Soil Taxonomic Classification:** Histic Humaquept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
O_a	0-22	Muck	10YR 2/1		7.4	20.4	4813	483	61	1.07	104	1.70	8600
A	22-37	Loam	2.5Y 3/4		7.1	18.6	4216	482	33	0.35	106	1.13	8340
B_g	37-44+	Sandy Loam	5Y 5/2		7.4	5.9	1616	163	16	0.04			

Note: Profiles could not be described to 60 cm due to gravel at and below ~40 cm depth.

Table E.2. Soil profile descriptions for Jug End South. Soil chemical measurements are expressed by volume ($g \cdot m^{-3}$). nd= not detectable.

Site:	**Jug End South**	**Soil Taxonomic Classification:** Oxyaquic Eutrudept											
Plot:	JS1												
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A	0-28	Loam	10YR 3/1		7.2	11.3	2388	313	29	0.79	65	1.55	4910
C	28-35+	Sandy Loam	2.5Y 4/2		7.8	4.7	1366	167	18	nd			
Plot:	JS2	**Soil Taxonomic Classification:** Oxyaquic Eutrudept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A	0-21	Sandy Loam	10YR 2/2		8.2	13.0	3512	309	29	0.89	41	1.68	5430
C	21-30+	Loamy sand	7.5YR 4/1	7.5YR 5/8 10%	8.5	4.2	1393	117	12	0.15			
Plot:	JS3	**Soil Taxonomic Classification:** Oxyaquic Eutrudept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A	0-13	Sandy Loam	7.5 YR 4/2		7.2	9.1	2046	247	36	0.29	86	0.67	3620
C1	13-45	Sandy Loam	2.5 Y 5/3		7.9	3.5	965	126	21	0.52			
C2	45-60	Sandy Loam	2.5Y 5/3	7.5YR 5/8 5%	8.3	1.5	483	62	9	0.12			

Note: Profiles could not be described to 60 cm at plots JS1 and JS2 due to gravel at and below 30 cm depth.

Table E.3. Soil profile descriptions for Bartholomew. Soil chemical measurements are expressed by volume (g·m^{-3}). nd = not detectable.

Site:	**Bartholomew**			**Soil Taxonomic Classification:** Histic Humaquept									
Plot:	SBB1												
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
O_a	0-23	Muck	2.5YR 2.5/1		7.2	21.5	4887	618	67	0.94	165	3.19	9960
B_g	23-60	Sandy Loam	2.5Y 4/2		7.6	7.6	2114	257	40	0.15			
Plot:	SBB2	**Soil Taxonomic Classification:** Histic Humaquept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A_p	0-23	Silt Loam	2.5Y 2.5/1		6.7	17.6	3784	416	68	0.83	185	1.38	7900
B_g	23-37+	Loam	5Y 5/1	10YR 5/6 2%	7.6	3.2	1088	137	43	0.95			
Plot:	SBB3	**Soil Taxonomic Classification:** Histic Humaquept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A_p	0-27	Loam	10YR 3/1		7.2	12.1	3072	346	42	1.46	63	1.33	5030
B_g	27-30+	Sandy Loam	2.5Y 5/2	10YR 6/4 40%	7.5	4.5	1227	149	26	nd			

Note: Profiles could not be described to 60 cm at plots SBB2 and SBB3 due to a layer of schistose flags underlying the profiles at ~30 cm.

Table E.4. Soil profile descriptions for Schenob Brook Central. Soil chemical measurements are expressed by volume (g·m^{-3}).

Site:	Schenob Brook Central			**Soil Taxonomic Classification:** Aeric Endoaquept									
Plot:	SBC1												
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A	0-5	Loamy sand	7.5YR 2.5/1		6.6	8.7	2403	283	54	2.63	81	2.05	2910
BC	5-11	Sandy Loam	7.5YR 3/1	5YR 3/3 10%	6.8	4.8	1089	139	17	1.63			
C	11-60	Loamy sand	2.5Y 3/3		7.2	2.5	696	86	14	0.16			
Plot:	SBC2	**Soil Taxonomic Classification:** Humic Endoaquept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A	0-8	Sandy Loam	7.5YR 2.5/1		6.5	13.8	2946	401	43	1.25	120	3.34	4900
BC	8-21	Loamy sand	10YR 2/1		6.6	6.9	1198	164	13	0.77			
C	21-60	Sand	mixed gravel		7.0	2.8	793	106	9	0.75			
Plot:	SBC3	**Soil Taxonomic Classification:** Oxyaquic Eutrudepts											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A	0-18	Loamy sand	7.5YR 2.5/1		7.3	9.4	2101	321	28	1.92	67	0.87	3050
B_w	18-60	Sand	2.5Y4/3	10YR 3/6 10%	8.1	0.6	566	57	6	0.26			

Note: Horizon A at SBC1 is a soil sample from the hummock where vegetation was growing.

Table E.5. Soil profile descriptions for Schenob Brook North. Soil chemical measurements are expressed by volume ($g \cdot m^{-3}$).

Site:	Schenob Brook North			**Soil Taxonomic Classification:** Typic Haplosaprist									
Plot:	SBN1												
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
O_a	0-28	Muck	5YR 2.5/1	5YR 5/1 10%	5.6	32.5	7157	1082	56	1.26	202	1.72	11470
B_g	28-60	Loamy Sand	5Y 5/2	7.5YR 3/2 20% 5 PB7/1 1%	7.8	3.3	860	183	9	0.22			
Plot:	SBN2	**Soil Taxonomic Classification:** Typic Haplosaprist											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
O_i	4-0	Fibric	5YR 2.5/1		6.9	45.9	8110	1230	152	2.57	332	3.42	16450
O_a	0-54	Muck	10YR 2/1		6.0	31.3	5494	853	91	0.91			11690
B_g	54-56	Sandy Loam	5Y 5/2		7.5	3.1	558	96	10	0.36			
Plot:	SBN3	**Soil Taxonomic Classification:** Typic Haplosaprist											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
O_i	6-0	Fibric	5YR 2.5/1		6.6	74.9	12589	1949	240	2.65	268	2.29	16610
O_a	0-54	Muck	5YR 2.5/1		6.2	52.5	8637	1300	95	0.40			

Table E.6. Soil profile descriptions for Schenob Brook South. Soil chemical measurements are expressed by volume ($g\cdot m^{-3}$). nd = not detectable.

Site:	**Schenob Brook South**			**Soil Taxonomic Classification:** Histic Humaquept									
Plot:	SBS1												
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A1	0-12	Silt Loam	10YR 2/1		6.3	18.7	3419	521	56	1.24	162	2.16	7230
A2	12-45	Silt Loam	10YR 2/1		6.3	19.1	3330	547	80	0.82	160	2.76	7670
C_g	45-59	Silt Loam	N5/	10YR 5/6 15%	7.0	2.8	754	160	19	nd			
Plot:	SBS2	**Soil Taxonomic Classification:** Typic Haplosaprist											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
O_{a1}	0-14	Muck	5YR 5/1		6.1	33.7	5386	823	79	1.18	518	4.03	14020
O_{a2}	14-55	Muck	5YR 5/1	5YR 4/6 5%	5.9	27.1	3839	669	57	1.58	365	1.36	10900
C_g	55-60+	Silt Loam	10Y 5/1	10YR 6/6 20%	7.0	3.7	957	437	52	0.13			
Plot:	SBS3	**Soil Taxonomic Classification:** Typic Haplosaprist											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
O_{a1}	0-10	Muck	5YR 2.5/1		5.9	52.7	6892	999	515	6.95	524	6.52	21720
O_{a2}	10-60+	Muck	5YR 2.5/1		6.2	38.5	6067	1006	65	0.93	295	2.45	15360

Table E.7. Soil profile descriptions for Shmulsky Open. Soil chemical measurements are expressed by volume ($g \cdot m^{-3}$). nd = not detectable.

Site:	**Shmulsky Open**			**Soil Taxonomic Class:** Histic humaquept									
Plot:	SO1												
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
O_a	0-33	Muck	5YR 2.5/1		6.4	25.5	4970	710	48	1.31	154	2.75	11010
B_g	33-47	Sand	5Y 4/2	7.5YR 5/8 5%	6.8	1.5	476	83	7	nd			
B_w	47-60	Sand	2.5Y 5/4		8.3	0.9	425	56	5	0.53			
Plot:	SO2	**Soil Taxonomic Class:** Histic humaquept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
O_a	0-30	Muck	10YR 2/1		6.7	30.5	5555	812	38	0.97	166	2.25	12170
B_g	30-60+	Loamy Sand	2.5Y 4/2	10YR 5/8 5%	7.0	2.5	665	103	6	0.15			
Plot:	SO3	**Soil Taxonomic Class:** Histic humaquept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
O_a	0-30	Muck	10YR 2/1		6.2	22.3	4195	523	23	0.52	168	0.42	8360
B_g	30-60+	Sand	2.5Y 5/2	2.5Y 6/6 15%	7.5	1.0	331	72	5	0.09			

Table E.8. Soil profile descriptions for Shmulsky Shrub. Soil chemical measurements are expressed on by volume ($g \cdot m^{-3}$). nd = not detectable.

Site:	**Shmulsky Shrub**			**Soil Taxonomic Class:** Histic humaquept									
Plot:	SS1												
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A_p	0-22	Sandy Loam	10YR 3/1		6.8	16.0	3474	470	53	0.90	137	1.67	7070
B_w	22-60+	Loamy Sand	2.5Y 4/3	2.5 Y 5/6 50%	7.4	2.6	843	142	12	nd			
Plot:	SS2	**Soil Taxonomic Class:** Histic humaquept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
A_p	0-35	Sandy Loam	7.5YR 2.5/1		6.8	15.1	3205	446	31	1.01	125	1.26	7080
B_w1	35-50	Sandy Loam	2.5Y 5/3	2.5Y 5/6 5%	7.4	3.1	870	131	11	0.03			
B_w2	50-62	Loamy Sand	7.5YR 6/8	2.5Y 3/3 10%	7.5	1.8	531	93	6	nd			
C	62+		2.5Y 4/2										
Plot:	SS3	**Soil Taxonomic Class:** Histic humaquept											
Horizon	**Depth (cm)**	**Texture**	**Color (moist)**	**Soil Mottling**	**pH**	**% OM**	**Ex-Ca**	**Ex-Mg**	**Ex-K**	**Ex-Fe**	**Fe-DTPA**	**Extr-P**	**Total N**
Ap	0-30	Sandy Loam	7.5YR 3/1		6.7	10.6	2101	289	21	1.32	120	0.92	4590
B_w	30-42	Loamy Sand	2.5Y 5/4	2.5Y 5/6 25%	7.4	2.1	636	103	6	0.30			
BC	42-60	Sandy Loam	10Y 6/1	10YR 6/6 20%	7.4	1.3	432	80	6	0.29			

APPENDIX F

SPECIES LIST

Table F.1. Species observed in the 24 study plots. Nomenclature is derived from the most current reports at the USDA Plants Database (USDA, 2007). A "*" in the boxes below the site codes indicates the species was found at that site. A "y" in the column titled "85" indicates it was one of the 85 species used in the vegetation ordination.

Common Name	**Scientific Name**	**Species Code**	**85**	**Site Code**							
				JN	**JS**	**SBB**	**SBC**	**SBN**	**SBS**	**SO**	**SS**
Red Maple	*Acer rubrum* L.	ACERUB	y	*			*	*	*	*	*
Redtop	*Agrostis gigantea* Roth	Grass4	y	*	*		*	*		*	*
Creeping Bentgrass	*Agrostis stolonifera* L.	Grass3	y	*	*	*				*	*
Speckled Alder	*Alnus incana* (L.) Moench ssp. *rugosa* (Du Roi) Clausen	ALNRUG									*
Hazel Alder	*Alnus serrulata* (Ait.) Willd.	ALNSER	y	*			*	*			
American Hogpeanut	*Amphicarpaea bracteata* (L.) Fern.	AMPBRA							*		
Paper Birch	*Betula papyrifera* Marsh.	BETPAP							*		
Bog Birch	*Betula pumila* L.	BETPUM						*			
Fringed Brome	*Bromus ciliatus* L.	Grass9		*		*					
Bluejoint	*Calamagrostis canadensis* (Michx.) Beauv.	Grass11								*	*
Tuberous Grasspink	*Calopogon tuberosus* (L.) B.S.P. var. *tuberosus*	CALPUL	y	*			*	*			
Yellow Sedge	*Carex flava* L.	CARFLA	y	*	*	*	*	*		*	*
Limestone Meadow Sedge	*Carex granularis* Muhl. ex Willd.	CARGRA	y	*	*	*				*	*
Bottlebrush Sedge	*Carex hystericina* Muhl. ex Willd.	CARHYS	y	*	*	*	*	*		*	
Inland Sedge	*Carex interior* Bailey	CARINT	y	*	*	*	*			*	
Hairy Sedge	*Carex lacustris* Willd.	CARLAC	y	*							

Common Name	Scientific Name	Species Code	85	Site Code							
				JN	JS	SBB	SBC	SBN	SBS	SO	SS
Bristlystalked Sedge	*Carex leptalea* Wahlenb.	CARLEP	y	*	*	*	*	*		*	*
Shallow Sedge	*Carex lurida* Wahlenb.	CARLUR	y							*	*
	Carex sp.1	CARunk1					*				
	Carex sp.2	CARunk2					*				
Dioecious Sedge	*Carex sterilis* Willd.	CARSTE	y	*	*	*				*	
Upright Sedge	*Carex stricta* Lam.	CARSTR	y	*			*	*	*		
Rigid Sedge	*Carex tetanica* Schkuhr	CARTET				*					
Fox Sedge	*Carex vulpinoidea* Michx.	CARVUL	y	*	*	*				*	*
Spotted Water Hemlock	*Cicuta maculata* L.	CICMAC		*		*					
Threeleaf Goldthread	*Coptis trifolia* (L.) Salisb.	COPGRO					*				
Silky Dogwood	*Cornus amomum* P. Mill.	CORAMO	y	*		*				*	
Redosier Dogwood	*Cornus sericea* L.	CORSER	y	*	*	*		*			
Shrubby Cinquefoil	*Dasiphora fruticosa* (L.) Rydb. ssp. *floribunda* (Pursh) Kartesz	DASFRU	y	*	*	*	*	*		*	*
Queen Anne's Lace	*Daucus carota* L.	DAUCAR								*	
Roundleaf Sundew	*Drosera rotundifolia* L.	DROROT	y	*			*	*			
Spikerush	*Eleocharis* R. Br.	ELEOCH	y	*		*	*	*		*	
Field Horsetail	*Equisetum arvense* L.	EQUARV	y	*	*	*	*	*			*
Water Horsetail	*Equisetum fluviatile* L.	EQUFLU	y	*				*	*	*	*
Scouringrush Horsetail	*Equisetum hyemale* L.	EQUHYE					*				
Tawny Cottongrass	*Eriophorum virginicum* L.	ERIVIR	y	*	*	*	*	*		*	

Common Name	**Scientific Name**	**Species Code**	**85**	**Site Code**							
				JN	**JS**	**SBB**	**SBC**	**SBN**	**SBS**	**SO**	**SS**
Spotted Trumpetweed	*Eupatoriadelphus maculatus* (L.) King & H.E. Robins.	EUPMAC	y	*	*		*	*	*	*	*
Common Boneset	*Eupatorium perfoliatum* L.	EUPPER	y	*					*	*	*
Low Rough Aster	*Eurybia radula* (Ait.) Nesom	ASTRAD	y	*				*	*	*	*
Nodding Fescue	*Festuca subverticillata* (Pers.) Alexeev	Grass12		*							*
Woodland Strawberry	*Fragaria vesca* L.	FRAVES	y	*	*	*	*	*		*	*
Green Ash	*Fraxinus pennsylvanica* Marsh.	FRAPEN	y	*	*	*			*	*	*
Rough Bedstraw	*Galium asprellum* Michx.	GALASP							*		
False Baby's Breath	*Galium mollugo* L.	GALMOL	y	*	*	*		*	*	*	
Common Marsh Bedstraw	*Galium palustre* L.	GALPAL	y						*	*	*
Purple Avens	*Geum rivale* L.	GEURIV	y			*				*	*
American Mannagrass	*Glyceria grandis* S. Wats	Grass15	y	*				*			
Fowl Mannagrass	*Glyceria striata* (Lam.) A.S. Hitchc.	Grass8	y	*		*				*	*
American Marshpennywort	*Hydrocotyle americana* L.	HYDAME	y							*	
Jewelweed	*Impatiens capensis* Meerb.	IMPCAP	y			*			*		
Harlequin Blueflag	*Iris versicolor* L.	IRIVER	y						*		*
Smallhead Rush	*Juncus brachycephalus* (Engelm.) Buch.	JUNBRA	y	*	*	*	*			*	*
Narrowpanicle Rush	*Juncus brevicaudatus* (Engelm.) Fern.	JUNBRE	y	*	*		*	*		*	*
Common Rush	*Juncus effusus* L.	JUNEFF	y	*						*	*
Knotted Rush	*Juncus nodosus* L.	JUNNOD	y	*			*			*	*

Common Name	Scientific Name	Species Code	85	Site Code							
				JN	JS	SBB	SBC	SBN	SBS	SO	SS
	Juncus sp.1	JUNunk1		*				*			
	Juncus sp.2	JUNunk2					*				
	Juncus sp.3	JUNunk3					*				
Common Juniper	*Juniperus communis* L.	JUNCOM					*				
Tamarack	*Larix laricina* (Du Roi) K. Koch	LARLAR	y	*			*	*			
Canada Lily	*Lilium canadense* L.	LILCAN				*					
Yellow False Pimpernell	*Lindernia dubia* (L.) Pennell	LINDUB									*
Ontario Lobelia	*Lobelia kalmii* L.	LOBKAL		*				*			
Morrow's Honeysuckle	*Lonicera morrowii* Gray	LONMOR		*							
American Water Horehound	*Lycopus americanus* Muhl. ex W. Bart.	LYCAME	y	*	*	*		*		*	*
Fringed Loosestrife	*Lysimachia ciliata* L.	LYSCIL	y			*				*	*
Earth loosestrife	*Lysimachia terrestris* (L.) B.S.P.	LYSTER	y							*	*
Winged Lythrum	*Lythrum alatum* Pursh	LYTALA									*
Purple Loosestrife	*Lythrum salicaria* L.	LYTSAL	y		*				*		
Peppermint	*Mentha ×piperita* L. (pro sp.) [*aquatica* × *spicata*]	MENPIP		*	*						
Wild Mint	*Mentha arvensis* L.	MENARV	y	*	*	*			*	*	*
Spiked Muhly	*Muhlenbergia glomerata* (Willd.) Trin.	Grass1	y	*	*	*	*			*	*
Watermilfoil	*Myriophyllum* L.	MYRIO		*				*			
Sensitive Fern	*Onoclea sensibilis* L.	ONOSEN	y	*					*		*
Cinnamon Fern	*Osmunda cinnamomea* L.	OSMCIN					*	*			
Royal Fern	*Osmunda regalis* L.	OSMREG		*				*			
Common Yellow Oxalis	*Oxalis stricta* L.	OXAEUR	y						*	*	*

Common Name	Scientific Name	Species Code	85	Site Code							
				JN	JS	SBB	SBC	SBN	SBS	SO	SS
Golden Ragwort	*Packera aurea* (L.) A.& D. Löve	PACAUR	y	*	*		*	*		*	*
Panicgrass	*Panicum sp.* L.	Grass6	y	*		*				*	*
Fen Grass of Parnassus	*Parnassia glauca* Raf.	PARGLA	y	*	*		*	*		*	*
Virinia Creeper	*Parthenocissus quinquefolia* (L.) Planch.	PARQUI							*		
Wild Parsnip	*Pastinaca sativa* L.	PASSAT				*					
Reed Canarygrass	*Phalaris arundinaea* L.	Grass14							*		
Eastern White Pine	*Pinus strobus* L.	PINSTR	y							*	*
Common Plantain	*Plantago major* L.	PLAMAJ			*						
Northern Green Orchid	*Platanthera aquilonis* Sheviak	HABHYP	y			*	*				
Fowl Bluegrass	*Poa palustris* L.	Grass13	y						*		
Snakemouth Orchid	*Pogonia ophioglossoides* (L.) Ker-Gawl.	POGOPH	y	*			*	*			
Arrowleaf Tearthumb	*Polygonum sagittatum* L.	POLSAG	y			*			*	*	
Quaking Aspen	*Populus tremuloides* Michx.	POPTRE								*	
Common Cinquefoil	*Potentilla simplex* Michx.	POTSIM	y		*					*	*
Common Selfheal	*Prunella vulgaris* L.	PRUVUL								*	
Black Cherry	*Prunus serotina* Ehrh.	PRUSER						*			
Virgina Mountainmint	*Pycnanthemum virginianum* (L.) T. Dur. & B.D. Jackson ex B.L. Robins. & Fern.	PYCVIR	y								*
Tall Buttercup	*Ranunculus acris* L.	RANACR	y	*	*	*				*	*
Bristly Buttercup	*Ranunculus hispidus* Michx.	RANHIS	y						*	*	*

Common Name	**Scientific Name**	**Species Code**	**85**	**Site Code**							
				JN	**JS**	**SBB**	**SBC**	**SBN**	**SBS**	**SO**	**SS**
Alderleaf Buckthorn	*Rhamnus alnifolia* L'Hér.	RHAALN	y	*		*					*
Multiflora Rose	*Rosa multiflora* Thunb. ex Murr.	ROSMUL	y			*			*	*	
American Red Raspberry	*Rubus idaeus* L.	RUBIDA	y						*	*	
Dwarf Red Blackberry	*Rubus pubescens* Raf.	RUBPUB	y	*		*		*	*		
Sageleaf Willow	*Salix candida* Flueggé ex Willd.	SALCAN	y	*		*		*			
Silky Willow	*Salix sericea* Marsh.	SALSERIC	y	*		*		*	*		*
Autumn Willow	*Salix serissima* (Bailey) Fern.	SALSERIS	y	*		*				*	*
Green Bulrush	*Scirpus atrovirens* Willd.	SCIATR	y	*	*	*				*	
Rufous Bulrush	*Scirpus pendulus* Muhl.	SCIPEN		*							*
Marsh Skullcap	*Scutellaria galericulata* L.	SCUEPI		*				*			
Strict Blue-eyed Grass	*Sisyrinchium montanum* Greene	SISMON		*							*
Hemlock Waterparsnip	*Sium suave* Walt.	SIUSUA								*	
Giant Goldenrod	*Solidago gigantea* Ait.	SOLGIG	y	*	*	*	*	*	*	*	*
Roundleaf Goldenrod	*Solidago patula* Muhl. ex Willd.	SOLPAT	y	*	*	*	*	*	*	*	
Bog Goldenrod	*Solidago uliginosa* Nutt.	SOLULI	y	*			*	*		*	*
Sphagnum Moss	*Sphagnum* L.	SPHAG					*				
White Meadowsweet	*Spiraea alba* Du Roi var. *latifolia* (Ait.) Dippel	SPILAT	y			*		*	*	*	*
Nodding Lady's Tresses	*Spiranthes cernua* (L.) L.C. Rich.	SPICER	y	*	*			*		*	*
Purplestem Aster	*Symphyotrichum puniceum* (L.) A.& D. Löve var. *puniceum*	SYMPUN	y	*	*	*		*	*	*	
Skunk Cabbage	*Symplocarpus foetidus* (L.) Salisb. ex Nutt.	SYMFOE	y	*	*	*	*	*	*	*	*

Common Name	**Scientific Name**	**Species Code**	**85**	**Site Code**							
				JN	**JS**	**SBB**	**SBC**	**SBN**	**SBS**	**SO**	**SS**
King of the Meadow	*Thalictrum pubescens* Pursh.	THAPOL	y					*	*	*	
Eastern Marsh Fern	*Thelypteris palustris* Schott	THEPAL	y	*	*	*	*	*	*	*	*
Eastern Poison Ivy	*Toxicodendron radicans* (L.) Kuntze	TOXRAD							*	*	
Poison Sumac	*Toxicodendron vernix* (L.) Kuntze	TOXVER	y	*				*			
Virginia Marsh St. Johnswort	*Triadenum virginicum* (L.) Raf.	HYPVIR	y	*				*	*	*	
Starflower	*Trientalis borealis* Raf.	TRIBOR					*				
Broadleaf Cattail	*Typha latifolia* L.	TYPLAT	y	*	*	*	*	*			
American Elm	*Ulmus americana* L.	ULMAME									*
	Unknown 1	UNK1		*							
	Unknown 2	UNK2					*				
	Unknown Graminoid10	Grass10				*					*
	Unknown Graminoid2	Grass2	y	*	*			*		*	
	Unknown Graminoid5	Grass5								*	
	Unknown Graminoid7	Grass7	y	*						*	
Highbush Blueberry	*Vaccinium corymbosum* L.	VACCOR	y	*			*	*			
Southern Arrowwood	*Viburnum dentatum* L.	VIBDEN	y	*	*						
Violet	*Viola* Sp. L.	VIOLA				*					
Species Richness				79	39	50	43	52	36	67	57

LITERATURE CITED

Aldrige, L. P. and G. J. Churchman. 1991. The role of iron in the weathering of a climosequence of soils derived from schist. Australian journal of soil research 29:387-398.

Auclair, A. D. 1979. Factors affecting tissue nutrient concentrations in a *Scirpus-Equisetum* wetland. Ecology 60:337-348.

Atkinson, C. J. 1991. The flux and distribution of xylem sap calcium to adaxial and abaxial epidermal tissue in relation to stomatal behaviour. Journal of Experimental Botany 42:987-993.

Beals, E. W. 1984. Bray-Curtis ordination: an effective strategy for analysis of multivariate ecological data. Advances in Ecological Research 4:1-55.

Bedford, B. L., M. R. Walbridge, and A. Aldous. 1999. Patterns in nutrient availability and plant diversity of temperate North American wetlands. Ecology 80:2151-2169.

Bedford, B. L. and K. S. Godwin. 2003. Fens of the United States: Distribution, characteristics, and scientific connection versus legal isolation. Wetlands 23:608-629.

Blake, G. R. and K. H. Hartge. 1986. Bulk Density. Pages 363-382 in A. Klute, editor. Methods of Soil Analysis Part 1. Soil Science Society of America, Inc., Madison, Wisconsin, USA.

Bonnier, G. 1894. Remarques sur les différences que presénte l'Ononis Natrix cultivé sur un sol calcaire on sur un sol sans calcaire (Comments on the differences presented by natrix cultivars in l'Ononis on a calcareous soil or on a soil without limestone). Bulletin of the Botanical Society of France xli.

Bowden, W. B. 1987. The biogeochemistry of nitrogen in freshwater wetlands. Biogeochemistry 4:313-348.

Bowles, M. L., P. D. Kelsey, and J. L. McBride. 2005. Relationships among environmental factors, vegetation zones, and species richness in a North American calcareous prairie fen. Wetlands 25:685-696.

Boyd, C. E. 1966. The elemental composition of several freshwater algae. Ph.D. Dissertation, Auburn University, Auburn, Alabama, USA.

Boyd, C. E. and L. W. Hess. 1970. Factors influencing shoot production and mineral nutrient levels in *Typha latifolia*. Ecology 51:296-300.

Boyer, M. H. L. and B. D. Wheeler. 1989. Vegetation patterns in spring-fed calcareous fens: calcite precipitation and constraints on fertility. Journal of Ecology 77:597-609.

Brady, N. C. and R. R. Weil. 2000. The nature and properties of soils. Prentice-Hall Inc. Upper Saddle River, New Jersey, USA.

Bremmer, J. M. and C. S. Mulvaney. 1986. Nitrogen-Total. Pages 595-624 in A. Klute, editor. Methods of Soil Analysis Part 2. Soil Science Society of America, Inc., Madison, Wisconsin, USA.

Bremmer, J. M. and K. Shaw. 1958. Denitrification in soil. I. Methods of investigation. Journal of Agricultural Science 51:22-39.

Broadley, M. R., H.C. Bowen, H.C. Cotterlii, J. P. Hammond, M. C. Meacham, A. Mead, and P. J. White. 2003. Variation in the shoot calcium content of angiosperms. Journal of Experimental Botany 54:1431-1446.

Brooks, K. N., P. F. Ffolliott, H. M. Gregersen, and L. F. DeBano. 1997. Hydrology and the management of watersheds, 2nd Edition. Iowa State University Press, Ames, Iowa, USA.

Clarkson, D. T. 1965. Calcium uptake by calcicole and calcifuge species in the genus *Agrostis* L. Journal of Ecology 53:427-435.

Clesceri, L. S., A. E. Greenberg, R. R. Trussell (editors). 1989. Standard methods for the examination of water and wastewater, 20th edition. American Public Health Association, American Water Works and the Water Pollution Control Federation, Washington, D.C., USA.

Cobbaert, D., L. Rochefort, and J. S. Price. 2004. Experimental restoration of a fen plant community after peat mining. Applied Vegetation Science 7:209-220.

Contejean, C. 1881. Géographie botanique, influence du terrain sur la vegetation. (Geographic botany, The influence of the ground on vegetation). 8vo. Paris, France.

Da Silva, D. L. R., L. P. Ruiz, C. J Atkinson, and T. A. Mansfield. 1994. Physiological disturbance caused by high rhizosphere calcium in the calcifuge, *Lupinus luteus*. Journal of Experimental Botany 45:585-590.

Dean, W. E. 1974. Determination of carbonate and organic matter in calcareous sediments and sedimentary rocks by loss on ignition: comparison with other methods. Journal of Sedimentary Petrology 44:242-248.

Douglas, M. E. & J. A. Endler. 1982. Quantitative matrix comparisons in ecological and evolutionary investigations. Journal of Theoretical Biology 99:777-795.

Drexler, J. Z., and B. L. Bedford. 2002. Pathways of nutrient loading and impacts on plant diversity in a New York peatland. Wetlands 22:263-281.

Dyck, A. W. J and R. R. McKibbin. 1935. The non-protein nature of a fraction of soil organic nigtrogen. Canadian Journal of Research 13B:264-298.

Eggers, S. D. and D. M. Reed, 1987. Wetland plants and plant communities of Minnesota and Wisconsin. US Army Corps of Engineers, St. Paul, Minnesota., USA.

Ehrenberg, P. 1919. Das Kalk-Kali-Gesetz. Parey, Berlin, Germany.

Ehrenfeld, J.G. and J. P. Schneider. 1993. Responses of forested wetland vegetation to perturbations of water chemistry and hydrology. Wetlands 13:122-129.

Elkington T. T., and S. Woodell. 1963. *Potentilla-fruticosa* L. **(***Dasiphora fruticosa* (L). Rydb.)**.** Journal of Ecology 51: 769-781.

Gauch, H.G. 1972. Inorganic Plant Nutrition. Dowden, Hutchinson & Ross, Stroudsburg, Pennsylvania, USA.

Gee, G. W. and J. W. Bauder. 1986. Particle-size Analysis. Pages 383-411 in A. Klute, editor. Methods of Soil Analysis Part 1. Soil Science Society of America, Inc., Madison, Wisconsin, USA.

Gleason, H. A. and A. Cronquist. 1991. Manual of vascular plants of northeastern United States and adjacent Canada, second edition. New York Botanical Garden, Bronx, New York, USA.

Godwin, K.S., J.P. Shallenberger, D. J. Leopold, B. L. Bedford. 2002. Linking landscape properties to local hydrogeologic gradients and plant species occurrence in minerotrophic fens of New York State, USA: A hydrogeologic setting (HGS) framework. Wetlands 22:722-737.

Grootjans, A. P., R. van Diggelen, M. J. Wassen, and W.A. Wiersinga. 1988. The effects of drainage on groundwater quality and plant species distribution in stream valley meadows. Vegetatio 75:37-48.

Hach Company, 1992. DR/2000 Spectrophotometer Procedures Manual. Hach Company World Headquarters, Loveland, Colorado, USA.

Hamze M, M. Nimah, and M. Zaabout. 1984. Effectiveness of six digestion procedures to evaluate the status of major elements (Ca, K, Mg, and Na) in citrus leaves. Communications in Soil Science and Plant Analysis 15:1135-1145.

Higley, W. K. 1880. On the microscopic crystals contained in plants. The American Naturalist 14:720-725.

Hope-Simpson, J. 1938. The chalk flora of the Lower Greensand: its uses in interpreting the calciole habit. Journal of Ecology 26:218-235.

Iljin, W. S. 1936. Zur Physiologie der kalkfeindlichen Pflanzen. Beihefte zum Botanischen Centralblatt 54:569-598.

Jones, J. B., Jr. 2001. Laboratory guide for conducting soil tests and plant analysis. CRC Press, New York, New York, USA.

Kearsley, J. B. 1999. The natural communities of Massachusetts: Palustrine section. Unpublished draft. Massachusetts Natural Heritage and Endangered Species Program, Westborough, Massachusetts, USA.

Kerner von Marilaun, A. 1895. The Natural History of Plants. Blackie and Son, London, England.

Kinzel, H. 1963. Zellsaftanalysen zum pflanzlichen Calcium- und Säurestoffwechsel und zum Problem der Kalk- und Silikatpflanzen. Protoplasma 57:522-555.

Kinzel, H. 1983. Physiological Plant Ecology III: Responses of the chemical and biological environment. Chapter 6: Influence of Limestone, Silicates and Soil pH on Vegetation. Springer-Verlag, New York, New York, USA.

Komor, S. C. 1994. Geochemistry and hydrology of a calcareous fen with the Savage Fen wetlands complex, Minnesota, USA. Geochimica et Cosmochimica Acta 58:3353-3367.

Le Gales, Y., A. Lamart, and R. Heller. 1980. Fixation due calcium par des fractions macromoleculaires solubles iolees a partir de vegetaux superieurs (Calcium fixing protiens in plants). Physiologie Vegetale 18:431-441.

Lee, J. A. 1999. The calcicole-calcifuge problem revisited. Advances in Botanical Research. 29:1-29

Loneragan, J. F., K. Snowball, W. J. Simmons. 1968. Response of plants to calcium concentration in solution culture. Australian Journal of Agricultural Research 19:845-857.

Lucassen, E. C. H. E. T., A. J. P. Smolders, G. Boedeltje, P. J. J. van den Munckhof, J. G. M. Roelofs. 2006. Groundwater input affecting plant distribution by controlling ammonium and iron availability. Journal of Vegetation Science 17:425-43.

Lynch, J. and A. Lauchli. 1985. Salt stress disturbs the calcium nutrition of barley (*Hordeum vulgare* L.). New Phytologist 99:345-354.

Marosz, A. 2004. Effect of soil salinity on nutrient uptake, growth, and decorative value of four ground cover shrubs. Journal of Plant Nutrition 27:977-989.

Marschner, H. 1995. Mineral Nutrition of Higher Plants, 2nd Edition. Academic Press: New York, New York, USA.

McLean, E. O. 1986. Soil pH and Lime Requriement. Pages 199-224 in A. Klute, editor. Methods of Soil Analysis Part 2. Soil Science Society of America, Inc., Madison, Wisconsin, USA.

McCune, B. and J. B. Grace. 2002. Analysis of Ecological Communities. MjM Software Design, Gleneden Beach, Oregon, U.S.A.

McCune, B. and M. J. Mefford. 1999. PC-ORD. Multivariate Analysis of Ecological Data. Version 5.0. MjM Software Design, Gleneden Beach, Oregon, U.S.A.

Miller, E. L., A. P. Bimbo, S. M. Barlow and B. Sheridan. 2007. Repeatability and reproducibility of determination of the nitrogen content of fishmeal by the combustion (Dumas) method and comparison with the Kjeldahl method: Interlaboratory study. Journal of AOAC International 90:6-20.

Mills, H. A. and J. B. Jones, Jr. 1991. Plant Analysis Handbook II. Micromacro Publishing, Athens, Georgia, USA.

Mitsch, W. J. and J. G. Gosselink. 1993. Wetlands, 2nd Edition. John Wiley and Sons, Inc. New York, New York, USA.

MNHESP (Massachusetts Natural Heritage and Endangered Species Program), 2004. List of Endangered, Threatened, & Special Concern Species, in Code of Massachusetts Regulations, 321 CMR 10.90. Massachusetts Division of Fisheries & Wildlife, Westborough, Massachusetts, USA.

Molisch, H. 1918. Über den mikrochemischen Nachweis und die Verbreitung gelöster Oxalate im Pflanzenreiche. Flora 11:60-70.

Motzkin, G. 1994. Calcareous fens of western New England and adjacent New York State. Rhodora 96:44-68.

Nägeli, C. W. von, 1865. Ueber die Bedingungen des Vorkommens von Arten und Varietaten innerhalb ihres Verbreitungsbezirkes (About the conditions of the occurence of kinds and varieties within a spreading district). Sitzungsber. d. Konigl. Bayrischen Akademie.

National Atlas. 2006. National Atlas of the United States. http://www.nationalatlas.gov/index.html. Last modified: May 22, 2006

Nekola, J. C. 2004. Vascular plant compositional gradients within and between Iowa fens. Journal of Vegetation Science 15:771-780.

NOAA, National Oceanic and Atmospheric Administration. 2006. Local climatological data: Annual summary with comparative data for Albany, New York. NCDC Asheville, North Carolina.

NRCS, Natural Resources Conservation Service. 2003. Climate Analysis for Wetlands by State: http://www.wcc.nrcs.usda.gov/climate/wetstate.html. Accessed 10/2006.

Olbe, M. and M. Sommarin. 1991. ATP-dependent calcium transport in wheat root plasma membrane vesicles. Physiologia Plantarum 83:535-543.

Palta, J. P. and E. J. Stadelmann. 1980. On simultaneous transport of water and solute through plant cell membranes: Evidence for the absence of solvent drag effect and insensitivity of the reflection coefficient. Physiologia Plantarum 50:83-91.

Picking, D.J. 2002. Vegetation patterns and associated hydrogeochemical cycles in a calcareous sloping fen of southwestern Massachusetts. Ph.D. Dissertation. University of Massachusetts, Amherst, Massachusetts, USA.

Picking, D. J. and P. L. M. Veneman. 2004. Vegetation patterns in a calcareous sloping fen of southwestern Massachusetts, USA. Wetlands 24:514-528.

Portis, A. R., C. Ja Chon, A. Mosbach, and H. W. Heldt. 1977. Fructose- and Sedoheptulosebis-phosphatase. The sites of possible control of CO_2 fixation by light dependent changes of the stromal Mg^{2+} concentration. Biochimica et Biophysica Acta 461:313-325.

Reader, R. J. 1990. Competition constrained by low nutrient supply: an example involving *Hieracium floribundum* Wimm and Grab. Compositaceae. Functional Ecology 4:573-577.

Reschke, C. 1990. Ecological Communities of New York State. New York Natural Heritage Program, N.Y.S. Department of Environmental Conservation, Latham, New York, USA.

Richardson, C. J., and P. E. Marshall. 1986. Processes controlling movement, storage, and export of phosphorus in a fen peatland. Ecological Monographs 56:279–302.

Robichaud, B. and K. H. Anderson. 1994. Plant communities of New Jersey – A study in landscape diversity. Rutgers University Press, New Brunswick, New Jersey, USA.

Römheld, V. 1987. Different strategies for iron acquisition in higher plants. Physiologia Plantarum 70:231-234.

Rorison, I. H. 1960. Some experimental aspects of the calcicole-calcifuge problem. I. The effects of competition and mineral nutrition upon seedling growth in the field. Journal of Ecology 48:585-599.

Ruiz, L.P., C. J. Atkinson, T. A. Mansfield. 1993. Calcium in the xylem and its influence on the behavior of stomata. Philosophical Transactions of the Royal Society of London B 341:67-74.

Salisbury, E. J. 1920. The significance of the calcicolous habit. Journal of Ecology 8:202.

Saltonstall K., 2002. Cryptic invasion by a non-native genotype of the common reed, Phragmites australis, into North America. Proceedings of the National Academy of Sciences of the United States of America 99:2445-2449

SAS Institute. 2004. SAS Version 9.1.3. SAS Institute, Cary, North Carolina, USA.

Scanu, R. J., 1988. Soil Survey of Berkshire County Massachusetts. United States Department of Agriculture, Soil Conservation Service. In cooperation with Massachusetts Agricultural Experiment Station.

Schot. P. P. and M. J. Wassen, 1993. Calcium concentration in wetland groundwater in relation to water sources and soil conditions in the recharge area. Journal of Hydrology 141:197-217.

Sendtner. O. 1860. Die Vegetationsverhaltnisse des Baverischen Waldes. Munchen. Germany.

Shaw, S. C. 2003. Do pollen assemblages in surface samples reflect community composition? A study of a Massachusetts fen. M.S. Thesis. University of Massachusetts, Amherst, Massachsetts, USA.

Shrimper, A. F. W. 1898. Plant Geography upon a Physiological Basis. Engl. Ed., Oxford, 1903.

Sjörs, H. 1950. On the relation between vegetation and electrolytes in North Swedish mire water. Oikos 2:242-258.

Slack, N. G., D. H. Vitt, and D. G. Horton. 1980. Vegetation gradients of minerotrophically rich fens in western Alberta. Canadian Journal of Botany 58:330-350.

Solomon, E. P., L. R. Berg, D. W. Martin, and C. Villee. 1996. Biology 4th Edition. Saunders college publishing, New York, New York, USA.

Soil Survey Staff, 1993. Soil Survey Manual. USDA Handbook No. 18. United States Government Printing Office, Washington, D. C., USA.

Soltanpour, P. N. and A. P. Schwab. 1977. A new soil test for simultaneous extraction of macro- and micronutrients in alkaline soils. Communications in soil science and plant analysis. 8:195-207.

Stewart, B. A., L. K. Porter, and F. E. Clark. 1963. The reliability of a micro-Dumas procedure for determining total nitrogen in soil. Soil Science Society of America Proceedings. 28:366-368.

Tansley A. G. 1917. On competition between *Galium Saxatile* L. (*G. Hercynicum* Weig.) and *Galium Sylvestre* Poll. (*G. Asperum* Schreb.) on different types of soil. The Journal of Ecology 5:173-179.

Taylor, K. 1997. *Geum rivale* L. Journal of Ecology 85:721-731.

Thomas, G. W. 1986. Exchangeable Cations. Pages 159-165 in A. Klute, editor. Methods of Soil Analysis Part 2. Soil Science Society of America, Inc., Madison, Wisconsin, USA.

Thompson, R., J. A. Parkinson, S. R. Band, R. E. Spencer. 1997. A comparative study of leaf nutrient concentrations in a regional herbaceous flora. New Phytologist 136:679-689.

Tyler, G. and L. Ström. 1995. Differing organic acid exudation patterns explain calcifuge and acidifuge behaviour of plants. Annals of Botany 75:75-78.

USDA, NRCS. 2007. The PLANTS Database (http://plants.usda.gov, 3 October 2007). National Plant Data Center, Baton Rouge, Louisianna, USA.

Vitt, D. H., P. Achuff, and R. E. Andrus. 1975. The vegetation and chemical properties of patterned fens in the Swan Hills, north central Alberta. Canadian Journal of Botany 53: 2776-2795.

Vitt, D. H. and W. Chee. 1990. The relationship of vegetation to surface water chemistry and peat chemistry in fens of Alberta, Canada. Vegetatio 89:87-106.

Waldren, S., J. R. Etherington, and M. S. Davies. 1987. Comparative Studies of Plant Growth and Distribution in Relation to Waterlogging. XIV. Iron, Manganese, Calcium and Phosphorus Concentrations in Leaves and Roots of *Geum rivale* L. and *G. urbanum* L. Grown in Waterlogged Soil. New Phytologist 106:689-696.

Walker, C. J. and J. D. Weinstien, 1991. Further characterization of the magnesium chelatase in isolated developing cucumber chloroplasts - substrate-specificity, regulation, intactness, and ATP requirements. Plant Physiology 95:1189-1196

Watson, M. E. and T. L. Galliher. 2001. Comparison of Dumas and Kjeldahl methods with automatic analyzers on agricultural samples under routine rapid analysis conditions. Communications in Soil Science and Plant Analysis 32:2007-2019.

Weatherbee, P.B. 1996. Flora of Berkshire County Massachusetts. The Studley Press, Inc., Dalton, Massachusetts, USA.

Weatherbee, P.B. and G. E. Crow. 1992. Natural plant communities of Berkshire county, Massachusetts. Rhodora 94:171-209.

White, P. J. 1998. Calcium channels in the plasma membrane of root cells. Annals of Botany 81:173-183.

White, P. J. 2001. The pathways of calcium movement to the xylem. Journal of Experimental Botany 52:891-899.

White, P. J. and M. R. Broadley, 2003. Calcium in plants. Annals of Botany 92:487-511.

Woolhouse, H. W. 1966. The effect of bicarbonate on the uptake of iron in four related grasses. New Phytologist 65:372-375.

Zampella, R. A. and K. J. Laidig. 1997. Effect of watershed disturbance on Pinelands stream vegetation. Journal of the Torrey Botanical Society 124:52-66.

Zen, E. and J. H. Hartshorn. 1966. Geologic map of the bashbish quadrangle, Massachusetts, Connecticut, and New York. Department of the Interior, United States Geological Survey. Washington, D.C., USA.

Zen, E. and N. M. Ratcliffe. 1971. Bedrock geologic map of the Egremont quadrangle and adjacent areas, Berkshire County, Massachusetts, and Colombia County, New York. Department of the Interior, United States Geological Survey. Washington, D.C., USA.