Abstract

Ecophysiological and morphological responses of balsam fir and red spruce to elevation and the canopy light gradient in the mountains of the northeastern United States

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Results did, however, support the hypothesis that plasticity is reduced in a harsh growth environment, as both species exhibited significantly less sun/shade plasticity at the highest elevation sites than at either the mid or low elevation sites.

The physiological response to elevation and crown position was comparable for the two species, which suggests that they share surprisingly similar ecophysiologies when grown together in an unfavorable environment. In contrast to the morphological and anatomical results, however, chlorophyll fluorescence and spectral reflectance suggested a physiological divergence of sun and shade needles with increasing elevation. Sun needles became progressively more stressed with increasing elevation, whereas shade needles did not.

Many other studies commonly focus on just the full-sun response to environment. These results demonstrate, however, that studies of foliage from only one extreme crown position cannot hope to capture the whole-plant response to environment, because foliage from the other extreme may reveal very different patterns.

Ecophysiological and morphological responses of balsam fir and red spruce to elevation and the canopy light gradient in the mountains of the northeastern United States

A Dissertation
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By
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This dissertation would not have got off the ground were it not for two Dartmouth

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Chapter 1:

Introduction

The relationship between an organism and its environment is of fundamental importance in almost all branches of biology and ecology. Even non-specialists cannot help but notice some of the most general patterns, such as how community structure is related to environment. For example, there is a clear difference in the species composition between the highly diverse rainforest of the tropics, and the comparatively minimalist spruce-fir-aspen-birch mix of Canada’s vast boreal forests. Similarly, the plant communities under a dense and dark forest canopy are obviously different from those in an open and bright old field.

However, as illustrated in Figure 1.1, it is not only the species mixture that changes with environment. There are a variety of different modes of response, and these responses can occur across a range of scales. For example, plants have the capacity for phenotypic plasticity, which means that the same genotype can express different phenotypes in different environments: in other words, genetically identical individuals may look different, depending on where they were grown. Across generations, natural selection acts on the expressed phenotype, and those that are more competitive in a given

Phenotypic plasticity or
Specialized ecotype?

Anatomy
Morphology
Physiology

can give rise to ecotypes, or locally adapted populations. Thus there can be a genotypic response to environment, as well as a phenotypic response (Figure 1.2).

Environmental responses can also be classified as morphological, anatomical, or physiological. Most studies of plastic responses to environment focus on the morphological response, since it is usually the easiest response to quantify. For example, leaf size and shape (i.e. morphology) change depending on the light environment in which a plant grows (see Chapter 4). However, frequently there are accompanying anatomical and physiological changes as well: leaves grown in high light often have more layers of palisade tissue, and often a thicker cuticle (Chapter 4), as well as higher rates of dark respiration and light-saturated photosynthesis (Chapter 7), compared to leaves grown in low light. To a degree, these responses are all interconnected: leaf structure, for example, has a direct influence on the concentrations of CO_2 and water vapor within the leaf, and thus on physiological processes like photosynthesis (Smith et al. 1997) and transpiration (Chapter 7). Relationships between structure and function are a main theme throughout this dissertation.

Finally, environmental responses can occur at a range of scales that spans several orders of magnitude, from molecular and sub-cellular to organ, organism, and even larger. For example, there are biochemical differences between sun and shade leaves (e.g. sun leaves have a higher carotenoid:chlorophyll ratio), and microscopic structures may differ, too (e.g. sun leaf chloroplasts usually have more grana stacks but fewer thylakoids per granum). At larger scales, plants respond to the light environment by altering leaf morphology (size and shape) or crown architecture (branching patterns and leaf display). Although many of the examples I have just used all refer to plant responses to the light

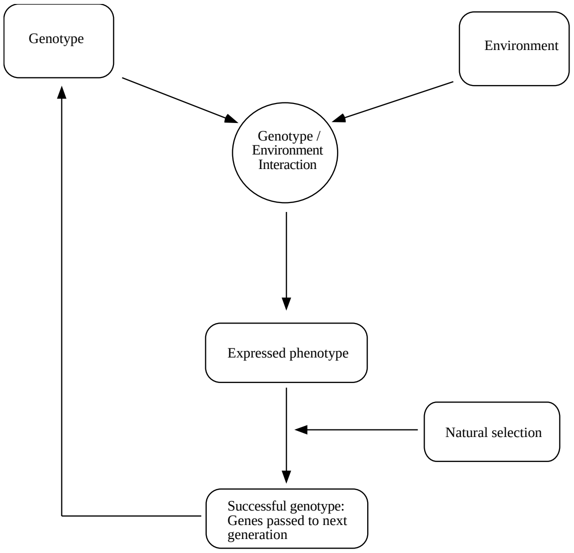

Figure 1.2. A simple model illustrating how the expressed phenotype is the product of the interaction between genotype and environment. Natural selection acts on the expressed phenotype, and may result in that phenotype being selected for, or against. If the phenotype is successful, it passes some of its genes on to the next generation.

environment, a similar range of responses can be imagined in response to many other environmental factors.

This dissertation is a comparative study of how two co-occurring conifer species, red spruce and balsam fir, respond to their growth environments. These species dominate the montane spruce-fir forests that are found at high elevations throughout the eastern United States. In the 19th and early 20th centuries, loggers harvested red spruce trees growing at all but the highest elevations and in all but the most remote locations in New England and adjacent New York state. Now these forests are valued more for their recreation potential—hiking, snow-shoeing, cross-country and alpine skiing, and backpacking— rather than their harvestable timber.

The land area actually occupied by montane spruce-fir forests is quite small. For example, according to Miller et al. (1993), even within the Adirondacks “High Peaks” area, only 30% of the land area is above 600 m in elevation, and slightly more than 3% is above 1000 m elevation (Miller et al. 1993). There is concern that, as a consequence of global change, the structure and composition of high elevation forests may change dramatically, and that their aesthetic and recreational value may be significantly reduced (Bloomfield and Hamburg 1997, New England Regional Assessment Group 2001). For this reason, studies of how these two species respond to their growth environment should provide valuable data which may help scientists to accurately predict not whether the competitive balance between red spruce and balsam fir (and between hardwoods and conifers) may be expected to change in the coming decades. Trees at high-elevation are at the limit of existence: they are carbon limited, and therefore can be considered “indicator ecosystems” (Berlyn et al. 1993) which may be more sensitive to climatic change than, for example, low elevation trees.

The two environmental gradients I use, the elevational gradient (Figure 1.3) and the canopy light gradient (Figure 1.4), have both been extensively studied in the past by other authors (e.g. Wardle 1971, Boardman 1977, Tranquillini 1979, Lichtenthaler et al. 1981, DeLucia and Berlyn 1984, Lichtenthaler 1985, Oleksyn et al. 1998). The primary reason for this is that they are both excellent model systems for studying plant responses to the environment. However, my main question, “Does the response to the canopy light gradient change along the elevational gradient?” (or, phrased differently, “Does the response to elevation differ for sun and shade leaves?”), has not, to the best of my knowledge, been investigated before.

The elevational gradient provides the opportunity to gain insight into the long-term response of populations to an environment that becomes progressively less favorable (i.e. more stressful) with increasing elevation. Because the transition from favorable to unfavorable growth environment occurs quite quickly with increasing elevation (one only has to hike from valley floor to treeline on a summer’s day to appreciate this), confounding biogeographic differences among populations, such as might occur across larger spatial scales, can be minimized.

The canopy light gradient is somewhat unique, in that it provides the opportunity to study how a single individual responds to two environmental extremes: sun at the top of the crown, and shade at the bottom of the crown. In this way, the capacity for phenotypic plasticity can be assessed, without genotypic differences between “sun” and “shade” sample trees being a confounding influence.

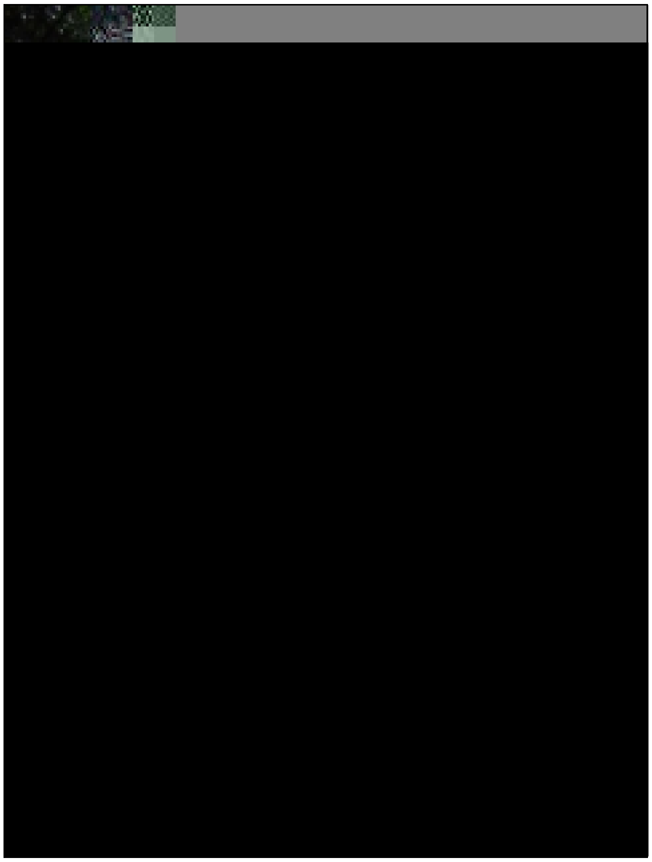

Figure 1.4. The forest canopy defines a significant light gradient. Leaves at the top of the canopy are exposed to full sunlight, whereas lower-crown leaves, and the leaves of understory plants, are exposed to only a fraction of full sunlight. The chlorophyll in leaves absorbs most incident light at red wavelengths (680 nm). However, leaves reflect about half of the incident light at far-red wavelengths (730 nm). Thus the quality of light, as measured by the red:far red ratio, is also different in the lower canopy compared to the upper canopy.

This method of using “natural experiments” to investigate how plants respond to their environment had its roots in a number of previous projects with which I was involved. Preliminary work on red spruce and balsam fir (Richardson et al. 2001a), as well as mountain paper birch (Richardson and Berlyn 2002), suggested that the species exhibited quantifiable physiological and morphological responses to the elevational gradient. These results indicated that the physiological response to increasing elevation is similar for red spruce and balsam fir, but that these two conifers are much less sensitive to increasing elevation than the broadleaf mountain paper birch. However, in those studies, our focus was exclusively on sun foliage, and questions remained about whether shade foliage would show parallel patterns. It is, for example, difficult to say anything about the whole-plant response when only sun foliage has been sampled.

At the same time, I was also involved with studies of how within-crown plasticity to the canopy light gradient varied among conifer species and also along a chronosequence of stand development (Richardson et al. 2000, Richardson et al. 2001b). These results suggested that not only does the capacity for plasticity differ between hybrid spruce and western hemlock, but also that the capacity for plasticity might change with ontogeny. This then raised the question of whether the capacity for plasticity might change across different growth environments. To date, there has been only limited research into this question (see Chapter 4), especially with mature trees.

I had a three primary questions I wanted to try to answer with my research.

First, I was interested in determining whether red spruce and balsam fir exhibited similar responses to the canopy light gradient. Although both species are considered shade tolerant, I hypothesized that balsam fir, with its greater geographical range and broader ecological niche, would have a greater capacity for sun/shade plasticity than red spruce.

Second, I hoped to be able to assess whether the capacity for sun/shade plasticity changed with elevation. I hypothesized that plasticity would be reduced in the harshest growth environment, i.e. at the highest elevation sites.

Third, I wanted to examine whether red spruce and balsam fir exhibited similar responses to the elevation gradient. Given the tendency for red spruce to be reach peak abundance at a lower elevation than balsam fir, it could be hypothesized that red spruce might show a larger stress response at the highest elevations. However, this hypothesis had not been borne out by the preliminary study (Richardson et al. 2001a), and so I wanted to see whether what the result would be when a much wider range of traits was considered.

To investigate these questions, I collected samples from “sun” and “shade” canopy positions of red spruce and balsam fir trees at three different elevations. The

In Chapter 3, I present the results of my micrometeorological studies on the different mountains. These results define the environmental gradient one find moving up the side of a mountain. I compare air temperature lapse rates on the different mountains, and investigate the hypothesis (Richardson and Berlyn 2002) that light levels at the highest elevations may be dramatically reduced compared to those at lower elevations, due to the increased cloud frequency on mountain summits.

In Chapter 4, I describe how needle anatomy and morphology of red spruce and balsam fir changes both across the canopy light gradient and along the elevational gradient. I focus my discussion on the first two hypotheses presented above. A measure of within-crown sun/shade plasticity is proposed, and plasticity is compared across traits, between species, and in relation to elevation.

In Chapter 5, I compare the foliar chemistry of balsam fir and red spruce, and examine whether there is evidence that this chemistry changes in response to crown position or elevation. I discuss micro- and macro-nutrients, as well fiber constituents (hemicellulose, cellulose and lignin). To the best of my knowledge, there are no previously published data on foliar fiber content in relation to elevation.

In Chapter 6, I consider a range of physiological methods, which I arrange along a spectrum from “dynamic” (photosynthesis and chlorophyll fluorescence) to “integrated (spectral reflectance and stable carbon isotope ratios). Focusing on the third hypothesis described above, I try to assess the degree of ecophysiological similarity between red spruce and balsam fir. In my discussion, I integrate results from Chapters 4 and 5 in order to emphasize the connection between structure and function.

A brief summary of the important results from Chapters 3 through 6 is presented in Chapter 7.

<u>**References**</u>

Berlyn GP, Anoruo AO, Johnson AH, Vann DR, Strimbeck GR, Boyce RL, Silver WL. 1993. Effects of filtered air and misting treatment on cuticles of red spruce needles on Whiteface Mountain, NY.

New England Regional Assessment Group. 2001. *Preparing for a changing climate: The potential consequences of climate variability and change. New England regional overview.* Durham, NH: University of New Hampshire.

Oleksyn J, Modrzynski J, Tjoelker MG, Zytkowiak R, Reich PB, Karolewski P. 1998. Growth and physiology of *Picea abies* populations from elevational transects: common garden evidence for altitudinal ecotypes and cold adaptation. *Functional Ecology* **12**: 573-590.

Richardson AD, Berlyn GP, Ashton PMS, Thadani R, Cameron IR. 2000. Foliar plasticity of hybrid spruce in relation to crown position and stand age. *Canadian Journal of Botany* **78**: 305-317.

Richardson AD, Berlyn GP, Gregoire TG. 2001a. Spectral reflectance of *Picea rubens* (Pinaceae) and *Abies balsamea* (Pinaceae) needles along an elevational gradient, Mt. Moosilauke, New Hampshire, USA. *American Journal of Botany* **88**: 667-676.

Richardson AD, Ashton PMS, Berlyn GP, McGroddy ME, Cameron IR. 2001b. Within-crown foliar plasticity of western hemlock, *Tsuga heterophylla*, in relation to stand age. *Annals of Botany* **88**: 1007-1015.

Richardson AD, Berlyn GP. 2002. Spectral reflectance and photosynthetic properties of *Betula papyrifera* (Betulaceae) leaves along an elevational gradient on Mt. Mansfield, Vermont, USA. *American Journal of Botany* **89**: 88-94.

Smith WK, Vogelmann TC, DeLucia EH, Bell DT, Shepherd KA. 1997. Leaf form and photosynthesis. *BioScience* **47**: 785-793.

Tranquillini W. 1979. *Physiological ecology of the alpine timberline*. New York: Springer-Verlag.

Wardle P. 1971. An explanation for alpine timberline. *New Zealand Journal of Botany* **9**: 371-402.

Chapter 2:

Study species and site description

Introduction

This chapter begins with a basic description of “the spruce-fir zone” as this term applies to the mountains I studied in the northeastern United States. These research sites, located in the Adirondack Mountains (New York), Green Mountains (Vermont), and

The montane spruce-fir zone

As is the case around much of the World, the most abundant tree species at higher elevations in the montane forests of eastern North America are members of the family Pinaceae (Wardle 1974). Here the dominant genera are *Picea* spp. (spruce) and

hardwood to coniferous spruce-fir is often demarcated by a comparatively narrow band of yellow birch, which quickly gives way to a spruce-dominated zone and then a fir-dominated zone (White and Cogbill 1992). Canopy tree height generally decreases with increasing elevation; at the treeline, there is a transition from a closed canopy of vertically erect stems (about 2.0 m or less in height, though some individual stems may be as tall as 3.0 m), to an open canopy of scattered, prostrate (" 1.0 m in height) shrub-like *krummholz* (or "elfin wood") of both red spruce and balsam fir mixed with alpine tundra species and bare rock. At the highest elevations, balsam fir is considerably more abundant than red spruce, although on some (but not all) mountains there may be black spruce (or hybrid black × red spruce, see Berlyn et al. 1990) at the treeline and continuing into the *krummholz*.

In the Adirondacks, Green Mountains, and White Mountains, the transition from hardwoods to spruce-fir occurs at an elevation of between 700 and 800 m, with the transition from spruce-dominated to fir-dominated spruce-fir typically occurring between 1000 and 1200 m (Cogbill and White 1991). In my experience, treeline generally occurs between 1200 and 1400 m, with only *krummholz* and tundra above these elevations.[5] However, there is at least one 1500 m peak in the White Mountains (South Twin Mt., eighth highest summit in the range) without any alpine tundra zone in spite of the mountain's considerable height (Cogbill and White 1991). Clearly, there is some variation among mountains in the elevations at which these ecotones occur: in the

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Presidential Range of the White Mountains, for example, the standard deviation of treeline elevation is over 100 m (Cogbill and White 1991).

Cogbill and White (1991) determined latitude-elevation relationships for the spruce-fir forest along the length of the Appalachians. They found that the spruce-fir/deciduous ecotone elevation decreased from 1,680 m at 35°N to 150 m at 49°N (-100 m elevation per degree of latitude), while treeline elevation decreased from 1,480 m at 44°N to 550 m at 55°N (-83 m elevation per degree of latitude). Furthermore, the spruce-fir/deciduous ecotone occurred at the elevation with a mean July temperature of roughly 17°C, whereas treeline occurred at roughly 13°C. In the northeast, at least, the position of the spruce-fir/deciduous ecotone is thought to be determined by the cloud base height. One proposed mechanism suggests that frequent rime-icing events, which occur in autumn and winter when the cloud base is frequently low enough to intersect with the montane forest, prevent the upslope movement of less frost-hardy deciduous species (Siccama 1974). Recent evidence (Richardson et al. in press) indicates that over the last three decades, the cloud ceiling in the northeastern United States has been rising at a rate of 6.3 ± 0.9 m/y. One implication of this is that we may begin to see an upward shift in the base of the montane spruce-fir zone. However, such a shift is likely to occur only very slowly, as the existing trees would have to die off before a deciduous invasion could begin. This raises a very important point, namely that the ecotones I have discussed are not static boundaries. For example, pollen and plant macrofossil analysis has been used to document long-term changes (over the last 10,000 years) in community structure along the elevational gradient in both the Adirondack (Jackson and Whitehead 1991) and White Mountains (Spear 1989, Spear et al. 1994). It is thought that zonation patterns approaching what is seen at present have developed only during the last three millennia (Jackson and Whitehead 1991). To some degree, climatic oscillation, at both regional and global scales, has always, and will always, occur. In systems where zonation patterns are climatically determined, temporal change in the associated ecotones is, therefore, inevitable.

Biology and ecology of red spruce and balsam fir

The biology and ecology of red spruce and balsam fir are reviewed by Burns and Honkala (1990), Uchytil (1991), White and Cogbill (1992), and Sullivan (1993), and the following summary is based largely on these sources. Although red spruce and balsam fir share a number of key ecological traits, they also differ in quite a few ways. Perhaps most importantly, balsam fir is faster growing and more of a mid-successional species, whereas red spruce is slower growing and more of a late-successional species. Secondarily, balsam fir has both a wide geographical and ecological distribution, whereas red spruce has a much narrower native range, and much more restricted habitat range.

Balsam fir: *Abies balsamea* [L.] Mill.

Balsam fir is one of 40 species in the genus *Abies,* of which nine species are native to the United States and Canada. The continuous native range of balsam fir stretches from Newfoundland and Labrador across to northwestern Alberta, south into the lake states of Minnesota, Wisconsin and northern Michigan, and east through New York and most of New England. Isolated populations are found in Iowa, Pennsylvania, West Virginia, and Virginia. The closely-related Fraser fir is restricted to isolated populations in the mountains of Virginia, North Carolina and eastern Tennessee. Balsam fir can be subdivided into two varieties, var. *balsamea* and var. *phanerolepis*; the varieties are distinguished by cone measurements, similar to the way in which balsam fir is typically differentiated from Fraser fir. Indeed, the variety *phanerolepis* is morphologically most similar to Fraser fir. Myers and Bormann (1963) studied different provenances of balsam fir over a wide geographic and altitudinal range. In the White Mountains, trees at high elevation on Mt. Washington (NH) were generally var. *phanerolepis* or intermediate between the two varieties. At low elevation, trees were generally intermediate between the two varieties, although some individuals were clearly var. *balsamea*. On Mts. Katahdin (ME) and Moosilauke (NH), trees were generally more similar to var. *phanerolepis*. Myers and Bormann concluded that the species is not comprised of two independent varieties: rather there is continuous variation between the two extremes, and the variation can be related to altitudinal and geographic gradients, with var. *phanerolepis* dominant at high elevations and in the east, and var. *balsamea* in the midwest.

Balsam fir is thought to hybridize with Fraser fir at the southern edge of its range, in the southern Appalachians (the hybrids sometimes referred to as *Abies intermedia* Full., though this is not universally accepted), and with subalpine fir (*Abies lasiocarpa*) at the western edge of its range, in the Canadian Rockies. In carefully controlled experiments, Hawley and DeHayes (1985) found that viable seed was produced in the following hybrids: *A. balsamea* var. *balsamea* × *A. fraseri* and reciprocals, *A. fraseri* × *A. balsamea* var. *phanerolepis* and reciprocals, and *A. balsamea* var. *phanerolepis* × *A. lasiocarpa*. Because these taxa were completely crossable, Hawley and DeHayes suggested the taxa are more separated by geographic, rather than genetic, isolation.

Furthermore, because interspecific crosses within the genus *Abies* appear to be more successful than within other Pinaceae genera (e.g. *Pinus*, *Picea*, *Pseudotsuga* or *Larix*), Hawley and DeHayes suggested that *Abies* species are more genetically similar, and perhaps more recently differentiated, than say the *Picea*. In particular, balsam and Fraser fir have likely been separated (and thus evolved independently) only since the retreat of the most recent glaciation (Myers and Bormann 1963). Clark et al. (2000) used molecular genetic markers (chloroplast microsatellites) to study genetic discontinuities among var. *balsamea*, var. *phanerolepis,* and Fraser fir; in spite of the recent separation, there was clear genetic evidence that these taxa have in fact diverged genetically, and results suggested that recent gene flow among taxa has been very limited.

Balsam fir is a component of 30 different SAF (Society of American Foresters) cover types (see Table 2.1). Balsam fir typically grows in mixed stands (especially mixed boreal stands with paper birch, aspen, black spruce and white spruce) but pure stands can be found in Newfoundland, Ontario, Quebec and Maine. Balsam fir can tolerate a wide variety of sites (e.g. steep mountain slopes, alluvial flats, peat bogs and swamps), and it seems to be little affected by soil parent material, for it is found growing on soils derived from all types of rock (e.g., gneiss and schist, anorthosite and granite, sandstone and limestone). Balsam fir also can tolerate a wide range of soil textures (from clay to rocky) and acidities (from the acid soils of the northeast to the more neutral soils found on limestone outcrops in Wisconsin).

The wood of balsam fir is light weight and soft, but it is not as strong as that of spruce or pine, and it is subject to rapid decay. Heart rot is especially a problem in older trees. In high elevation forests, winter wind damage to balsam fir is common; red spruce,

Table 2.1

with its higher quality wood, is much less susceptible. Balsam fir is also highly susceptible to infestation by the spruce budworm and the balsam wooly adelgid; both pests have posed serious problems for management of commercial stands. Pest-killed stands are a serious fire threat, because the fuel is easily ignited (due to the abundant resin in both trunk and foliage) and burns with intense heat.

Balsam fir grows to a height of 27 m and DBH of 86 cm, but trees are more commonly in the range of 12-18 m high, with DBH of 30-45 cm. Trees often die by 100 years of age, and rarely exceed 200 years. Because the life span of balsam fir is so much shorter than that of red spruce, stand dynamics in montane spruce-fir forests are strongly influenced by the fact that canopy turnover of balsam fir is much more rapid than that of red spruce.

When grown in the open, balsam typically has a narrow, conical profile, with a live crown that reaches almost to the ground. In dense forest settings, however, lower branches are often dead but retained on the tree. Root systems are often shallow (penetrating less than 75 cm), and restricted to the organic and upper mineral soil horizons. However, root development in seedlings is more rapid than in other conifer species. As a result, balsam fir can seedlings become established more quickly than red spruce seedlings, which often suffer heavy mortality before they can become established. Once established, however, red spruce is better suited to understory survival than balsam fir.

Balsam fir starts to produce seeds at about 20 years of age. Although seeds are produced every year, large crops generally occur only every 2 to 4 years. The winged seeds are dispersed primarily by wind (to a lesser degree by small mammals), and mostly during the autumn. Wind-transported seeds generally land within 60 m of the parent tree, though some can travel more than twice as far. Seeds are usually viable for less than one year, and germination rates are low (20-50%). Germination occurs in late spring and early summer. With adequate moisture, seedlings can become established on most substrates, though seedlings are usually most successful on mineral soil. At high elevations, *krummholz* balsam fir reproduce by layering.

The very shade tolerant balsam seedlings can survive in the understory (which, because it is usually moist and experiences less extreme temperatures, is an ideal microsite for seedling establishment) for many years, awaiting a disturbance that will promote their release. Balsam fir is therefore a mid- or late-successional species. In the boreal forest, for example, balsam fir is usually not present in the first few decades following a fire; it establishes later, and then assumes canopy dominance only when pioneer trees (e.g. pines and paper birch) begin to die. Because balsam fir is more short-lived than red spruce, it cannot dominate really old stands in quite the same way. Rather, old red spruce trees are likely to have witnessed several generations of balsam fir regeneration, maturity and senescence. In this regard, differences in life history between species are key.

Red spruce—*Picea rubens* Sarg.

Red spruce is one of the 40 species in the genus *Picea*, of which seven species are native to the United States and Canada. Compared to the transcontinental distribution of the congeneric white spruce and black spruce (or the associated balsam fir), the range of red spruce is very limited. The continuous range includes Prince Edward Island, Nova

Scotia and New Brunswick, southern Quebec (along both sides of the St. Lawrence River), north-eastern New York, and most of northern New England. Isolated stands are found near Algonquin Park in southern Ontario, and in the southern Appalachians of West Virginia, Virginia, North Carolina and Tennessee. Its ecological range is more restricted than that of balsam fir, and it is a component species in 21 different SAF cover types (compared to 30 for balsam fir, see Table 2.1).

As described earlier in this chapter, red spruce is a characteristic species of the montane spruce-fir forests in the eastern United States. Red spruce can be found all the way to treeline (and in the *krummholz*) but on some mountains there is evidence that the highest elevation spruce are actually black spruce (*Picea mariana* [Mill.] B.S.P.) or black × red spruce hybrids, and not pure red spruce. Red spruce can out-compete black spruce under mesic conditions. The reverse is true when conditions are extreme. Recent work by Perron and Bousquet (1997) used genetic markers to show that hybridization between red and black spruce is extensive. Although it has been suggested that both ecological isolation and hybrid adaptive inferiority may act as barriers to hybridization (Manley and Ledig, 1979), Berlyn et al. (1990) reported that the proportion of black spruce genes in putative red spruce trees increased with increasing elevation in the mountains of New England. Thus cold climate may favor selection for black spruce's characteristic hardiness at the highest elevations. Berlyn et al. (1990) suggested that hybrids could be more competitive than either pure red spruce or pure black spruce.[6]

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[6] The matter has been complicated by the recent suggestion (Perron et al. 2000), that there is a progenitor-derivative relationship between black spruce and red spruce. Molecular evidence supports the claim that that red spruce might have evolved as recently as the last glaciation, when black spruce could have been fragmented into several different populations, one of which then remained isolated and evolved into the derivative species red spruce. Derivative species generally have lower genetic diversity and a more

Based on my own field observations in the summer of 1999, I believe that the spruce on Mt. Moosilauke (at the far western edge of the White Mountains) are pure red spruce; I have not observed any black spruce there. Farther east (e.g. in the peaks of the Carter Range, White Mountains, NH), there are high-elevation spruce with morphological traits characteristic of black spruce, and some with intermediate traits (i.e. short, grayish needles of black spruce, but the longer cones of red spruce), suggesting hybridization. The trees from which I collected samples for this study were, based on their morphological traits, definitely more red spruce-like than black spruce-like.

The light weight, straight-grained, resilient wood of red spruce makes it an important timber species. Its uses range from paper making to soundboards for musical instruments. Extensive harvesting of red spruce during the last century and a half has had long-lasting effects on forest composition; for example, it is estimated that in the mountains of West Virginia there were 200,000 ha of red spruce in the late 19th century, whereas now there may be only 7,000 ha.

Red spruce reproduce only from seed; cones are first borne at about 15 or 20 years. Most years, only a small seed crop is produced; bumper crops are somewhat more rare (3-8 years) than for balsam fir. Seeds are dispersed primarily by wind, and even 100 m from the edge of a stand there can be enough wind-transported seeds for good regeneration. Germination can occur in the same autumn as the seeds are dropped, though it is more common for germination to occur the following spring. Seeds have a very low viability after one year. Adequate advance regeneration is essential if a harvested area is to regenerate as red spruce; otherwise, hardwood regeneration will quickly out-compete

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restricted ecological range than the progenitor species, but can usually hybridize with the progenitor species, which is exactly what we see with red and black spruce.

new red spruce seedlings. Seedlings establish best on shallow, infertile soils that are unsuitable for hardwood growth. Unlike balsam fir, red spruce seedlings do not develop a vigorous root system (and mature red spruce have an even shallower rooting habit than balsam fir). Rather, seedling roots are usually confined to the organic soil horizon, which can make them very susceptible to summer droughts; adequate moisture is therefore essential for seedlings to become established. Frost heaving during the winter is a leading cause of seedling mortality.

Mature red spruce are susceptible to a number of pests and diseases, including spruce budworm and eastern spruce beetle, but younger trees are usually not affected. Overall, red spruce is much less susceptible to pest infestations than balsam fir. However, 1991, Battles et al. 2003). Recent work, which documented reduced mortality, increased recruitment, and increased growth rates of canopy spruce trees, suggests that red spruce may be recovering from this period of decline, at least at some sites in the Adirondacks (Battles et al. 2003).

Red spruce grows to a maximum height of 35 m. Mature trees more typically are in the range of 18-23 m, with a DBH of 30 to 60 cm. Red spruce therefore grows to be somewhat larger than balsam fir. The lifespan of red spruce can be extremely long, up to about 400 years, which is about twice as old as the maximum for balsam fir.

Red spruce is normally found growing on soil derived from glacial drift and till, and typically these soils are acid (pH between 4.0 and 5.5). Midslope sites slope s ford tto al. 2003).

subalpine fir forests of Colorado, the species are characterized by similar differences in life history, and it has been suggested that, combined with the frequent small-scale divides the states of Vermont and New Hampshire, and thus, to some degree, the Green and White Mountains.

Whiteface Mt., at 1485 m the fifth highest peak in the Adirondacks, stands somewhat isolated at the northern edge of the range, about 15 km distant from the main “High Peaks” region. Whiteface is a large, triangular massif that towers almost 1,000 m above Mirror Lake to the south; a minor sub-peak, Mt. Esther (1292 m), protrudes from the mountain’s northern flank. The cirque on Whiteface’s south face, and the knife-edge west ridge which descends from the summit, is evidence of previous glaciation. Due in part to the thin soil, shallow rooting habit of red spruce and balsam fir, as well as the impermeable bedrock, landslides have exposed long, narrow strips of bedrock on the north, east, and south faces. On the south and east faces, treeline (transition to prostrate *krummholz*) occurs at about 1360-1380 m, whereas on the north face, small but still erect stems continue growing almost all the way to the summit.

Mt. Mansfield, with a summit elevation of 1339 m, is the highest peak in the Green Mountains, and the most northerly of Vermont’s “four thousand foot” (i.e. greater than 1220 m) summits. The Green Mountains are different from the Adirondacks to the west or the White Mountains to the east in that they are characterized by a linear, north-south orientation, which is more or less defined by a single ridgeline, whereas the other two ranges are complex, irregularly aggregated masses of peaks and ridges. The west slope of Mt. Mansfield falls away in a smooth slope towards Lake Champlain, which is about 15 to 20 km distant. To the east lies the valley of Waterville Brook and then the smaller peaks (Mt. Hunger, 1083 m) of the Worcester Mountains. Treeline on Mt. Mansfield is at about 1190 m, but *krummholz* red spruce and balsam fir grow almost all the way to the main summit, known as the Chin, which is separated from the southern summit, known as the Nose (1238 m), by a two kilometer-long narrow ridge of exposed bedrock and scatterd *krummholz* (Figure 2.2).

The tenth-highest peak in the White Mountains is Mt. Moosilauke (1463 m). Moosilauke stands at the western edge of that range, and, like Whiteface, is somewhat isolated from the highest peaks of its range. For example, Mts. Lafayette (1603 m) and Lincoln (1551 m) lie roughly 20 km to the northeast, whereas Mt. Washington (1916 m) and the other peaks of the Presidential Range lie 50 km to the northeast. Moosilauke is a complex mountain, with a long south ridge connecting to the minor South Peak (1390 m),

Figure 2.2. Summit ridge on Mt. Mansfield, at about 1190 m, looking north towards the main summit ("the Chin", 1339 m). Note the exposed, highly metamorphosed, bedrock and flagged *krummholz* red spruce and balsam fir.

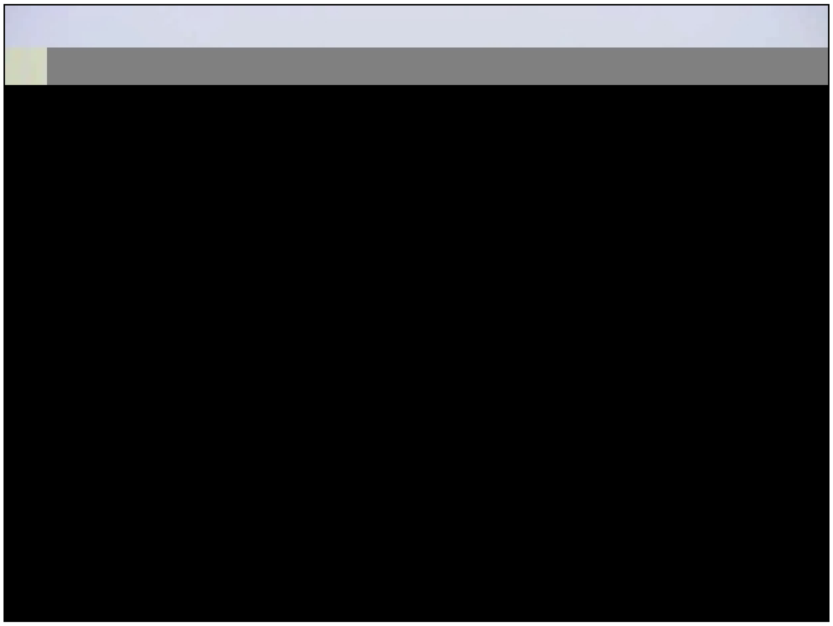


Figure 2.3. Near the East Peak of Mt. Moosilauke, at about 1425 m, about 40 m above the treeline transition from a closed-canopy forest with vertical stems to prostrate, flagged *krummholz*.

however, it is not clear whether basal area increases or decreases with elevation (White and Cogbill 1992).

Siccama (1974) surveyed the northern hardwood and spruce-fir forests on the west slope of Camels Hump, in the Green Mountains of Vermont. In the northern mountain paper birch than red spruce. At the “Esther” site (1000-1063 m elevation), canopy tree density was 904 stems/ha, of which balsam fir accounted for 86%, and red spruce 6%, of the total stem count. Sub-canopy tree (2.0-9.5 cm DBH) density was 4300 stems/ha. 64% of canopy tree basal area (26.7 m^2/ha) came from balsam fir, whereas 18% came from red spruce.

Although Battles et al. (2003) did not conduct a regeneration survey, some data are available for Whiteface Mt. from the 1985 study of Fitzgerald and Raynal (1991). At 900 m, they found small seedling (defined as ! 10 cm in height) densities of 225,000 stems/ha for balsam fir, and 4,150 stems/ha for red spruce; at 1100 m, the corresponding figures were 90,000 stems/ha and 1,500 stems/ha, respectively. Large seedling (> 10 cm, but < 1.4 m, in height) densities were 17,500 stems/ha (balsam fir) and 1,500 stems/ha (red spruce) at 900 m, and 50,000 stems/ha (balsam fir) and 800 stems/ha (red spruce) at 1100 m.

An interesting phenomenon in many subalpine fir forests, not only in North America but also in Japan, is the presence of “fir waves”. Reiners and Lang (1979) estimate that 15% of the forested land area above 1220 m in the White Mountains is occupied by fir waves, and so they merit mention in any discussion of forest structure. From a distance, fir waves are clearly visible as parallel lines of mortality that often run upslope or along ridgetops. On closer inspection it is noted that the “waves” are actually defined by zones of growth, senescence, and death. The “wave front” is a band of standing dead fir trees, which suggests that the pattern is not generated by blowdowns, because if that were the case, the dead trees would be no longer standing. Moving away the next wave front is reached. Tree-ring analyses suggest that the waves move across the landscape at a rate of several meters per year. The actual mechanism producing this pattern is still not clear (Reiners and Lang 1979), although the results of Boyce (1988) were consistent with the idea that winter winds and rime-ice buildup could play a role.

Geology and forest soils

The Adirondacks, Green Mountains and White Mountains have all followed somewhat similar developmental paths (uplifting and erosion, then perhaps more uplifting and erosion, and, most recently, glaciation), although the ranges are products of distinctly different geological events. It is important to note that the neither the Adirondacks, nor the Green Mountains, are in any way an extension of the Appalachians, contrary to what is commonly believed. The geological history of the northeastern United States is well described by Raymo and Raymo (2001); I will give only a brief overview here.

The Adirondacks are perhaps the most interesting of the three main mountain ranges in the northeast. They are a comparatively young range, and are thought to be still rising. More or less circular in shape, their dome-like form is due to uplifting far below the Earth’s surface, rather than the more typical surface collision of tectonic plates. The exposed bedrock in the Adirondacks is Precambrian (over 1 billion years old) in origin, and is part of the Grenville Province. During the Grenville Orogeny (1.1 billion years ago), when the Baltic Shield collided with the proto-North American continent, a mountain range comparable to today’s Himalaya was formed. Over the following 400 million years, this range was eroded to sea level, leaving exposed metamorphosed

Precambrian rock. The Adirondacks are comprised mainly of metamorphosed granite and anorthosite, which is overlain in places by the eroded remains of ancient metamorphosed sedimentary rock (sandstones and shales).

The bedrock of the Green Mountains is younger than that of the Adirondacks, and is derived mainly from deposits in basins along the edge of the Grenville Province, when it formed the shore of the Iapetus Ocean. Some 500 million years ago, sand, carbonates and mud were deposited to form the sedimentary rock, which was subsequently metamorphosed when the Iapetus Ocean closed up and the region was violently compressed during the Taconic Orogeny. During this time, strata were re-oriented vertically and the metamorphosis resulted in gneisses, marbles and schists. The rocks of both the Berkshire Hills of western Massachusetts and the Green Mountains of Vermont thus had their origin during the Cambrian and Ordovician periods; subsequent erosion, followed by more uplifting, resulted in the mountains we see today.

The White Mountains of New Hampshire, which lie towards the northern end of the Appalachians, were formed primarily during the Acadian Orogeny of the Devonian Period (380 million years ago). This mountain-building event was the result of the collision of Baltica (Northern Europe) and Laurentia (North America), which caused the crumpling of the Gander and Avalonia Terranes (most of present-day New England). Once again, a huge mountain chain was created across the northeast, and the resulting deformation of the Earth’s crust led to massive intrusions of granite that would ultimately form the White Mountains of New Hampshire. First, however, many thousands of vertical meters of overlying sedimentary rock had to be worn away; this erosional debris would later form the sedimentary rocks of the Catskill Mountains of southeastern New York.

A subsequent mountain-building event, the Alleghenian Orogeny (290 million years ago), led to the formation of the southern Appalachians (but had less of an impact on the northern Appalachians), as Gondwanaland (in particular, what would later be north-west Africa) collided with the eastern margin of present-day North America. Soon after this occurred, Sibera collided with Europe, and assembly of the supercontinent Pangaea was completed around 250 million years ago.

During the Triassic and Jurassic Periods (250-140 million years ago), Pangaea broke apart, forming what is now the Atlantic Ocean. During the Cretaceous Period (140-65 million years ago), extensive erosion occurred across the entire northeast, as the towering mountains were slowly worn down. The end of the Cretaceous was marked by a catastrophic extinction and the end of the dinosaurs.

During the Cenozoic era (the last 65 million years), the Earth’s climate has slowly cooled, and for most of the Quaternary Period (the most recent 2 million years), the northeastern United States has been covered by glacial ice up to 2 km in thickness. The continental ice sheets originated in central Canada, in the vicinity of Hudson Bay, and as they slowly advanced across the land (as far south as the Ohio and Missouri River valleys of today) they bulldozed sediments, and scraped and polished bedrock surfaces. The most recent glaciation began about 100,000 years ago. As the climate cooled, the forests of New England were replaced by tundra, which was then pushed aside by the advancing ice sheet; only the highest summits of the northeast were left exposed above the ice. What had been V-shaped valleys, typical of erosion by water, were scoured and rounded, leading to the characteristic U-shaped valleys of glacial erosion. Massive amounts of debris (including house-sized boulders) were re-located by the ice. For example, the southern moraines left by the receding glaciers became Long Island, Block Island, Martha's Vineyard, Nantucket and Cape Cod.

The reason that the soils of the northeast are typically so sandy, rocky, and poorly developed is that they have formed from the meager layer of glacial till that was left spread across the landscape by the retreating ice sheets about 14,000 years ago. In the mountains, in particular, where subsequent alluvial deposition did not occur, and where temperatures have remained cool, these soils are a less than ideal growth medium for plants. However, plants have managed to re-colonize the barren ground left behind by the glaciers—first sedges and tundra plants, and then later coniferous forest trees, such as *Picea* spp. and balsam fir (a balsam-fir dominated treeline very similar to that of the present day existed at least 10,000 years ago, see Spear 1989, Spear et al. 1994). Larch (*Larix laricina* Du Roi [K. Koch.]), poplar (*Populus* spp.) and paper birch (*Betula papyrifera*) preceded the arrival of *Pinus* spp. (pine) and *Quercus* spp. (oak); by 7,000 years ago, mixed-hardwood forests similar to those of the present day were established, and as spruce increased in abundance about 2,000 years ago, montane spruce-fir forests were developed (Spear et al. 1994).

In the northeastern United States, spruce-fir zone soils differ from those farther south in that they have developed relatively recently from glacial till, whereas more southern soils are the product of bedrock weathered over a longer time period. In both north and south, however, parent material is usually nutrient-poor and quite resistant to erosion, and the vegetation has had a positive feedback effect on soil formation (Siccama

1974). Generally, spruce-fir zone soils have both extremely low pH and low base saturation. High precipitation enhances nutrient leaching (White and Cogbill 1992), and combined with cool temperatures and organic acids present in coniferous litterfall, results in podzolization being a key soil forming process (Fernandez 1992). These Spodosols have subsurface accumulations (the spodic horizon) of organic matter and Al (possibly also Fe) oxides (Brady and Weil 1999). Histosols (organic soils) and Inceptisols (soils with minimal profile development) also commonly underlie spruce-fir forests (Fernandez 1992). Because of slow rates of litter decomposition (Fernandez 1992), the forest floor is often quite thick. Forest floor depth increases, whereas base saturation decreases, with increasing elevation (Siccama 1974, Johnson et al. 1994). The upper organic (O) horizons are generally more acidic than the deeper mineral soil horizons, but base cation concentrations are also frequently highest in the upper horizons (Fernandez 1992). Although Ca is often abundant ("10 cmol$_c$/kg) in the O horizons, concentrations of Al are also typically high (" 5 cmol$_c$/kg); in the mineral horizons below, Ca concentrations decline rapidly with depth (to less than 1 cmol$_c$/kg), and Al becomes the dominant exchangeable cation, possibly to the point of having a detrimental e m(re)t at either the Whiteface Mt. (July-September, 13.2°C; January-March, –8.5°C; annual 1.9°C) or Mt. Moosilauke (July-September, 12.9°C; January-March, –8.7°C; annual 1.8°C) stations. My data indicate that temperature decreases with increasing elevation by high frequency with which “flagged” crowns (uneven crown development on the side opposite the prevailing wind direction) are observed in the *krummholz* (Tranquillini 1979). During the winter, mechanical abrasion (blowing snow and ice crystals) can remove virtually the entire epicuticular wax layer from exposed high-elevation shoots (Hadley and Smith 1986), which may reduce cuticular resistance to water loss and lead to severe desiccation (or death).

Extremely high (# 90%) relative humidities on Mt. Moosilauke were more than twice as common at 1425 m (75% of time during January-March, 50% of the time during July-September) than at 247 m (27% of the time during January-March, 35% of the time during July-September). Other researchers have documented the fact that the spruce-fir forests of the northeastern United States are frequently engulfed in clouds. For example, it is estimated the summit of Whiteface Mt. in the Adirondacks is immersed in cloud for 40-50% of all hours, whereas the summit of Mt. Moosilauke in the White Mountains is immersed in cloud for roughly 40% of all hours; at 1000 m elevation, the frequencies are thought to be more on the order of 15-19% (Mohnen 1992). Throughout the Appalachians, the probability of summit cloud immersion is lowest in the afternoon and significant chemical input to high-elevation forests (Mohnen 1992), and the effect of cloud drip on precipitation totals has already been discussed.

Site details and sampling design

My objective was to study foliar differences between red spruce and balsam fir both along the elevational gradient and across the canopy light gradient. To do this, I used a split-split plot design: the main plot (low, mid or high elevation) was split first by species (red spruce or balsam fir) and then crown position (sun or shade needles). On each mountain, I had two transects, one on each of the south and east sides of the peak. The protocol for selecting individual trees (3 of each species per site; a total of 108 trees for the whole study), as well as collecting samples and conducting measurements, is described in greatest detail in Chapter 4, and supplemented where necessary in Chapters 5 and 6. As should be apparent from the preceding discussion of forest structure, balsam fir is generally far more abundant than red spruce in these forests, and thus it was generally much more difficult to find suitable individuals of red spruce than balsam fir. I looked for individuals that were representative of those growing at each elevation, with healthy, well-developed crowns. I used trees that were growing on the edge of a gap or trail, so that leaves growing in full sun could be easily obtained, but I was limited in what I could reach by the length of the pole pruner we carried (8 m). Trees were selected in pairs, which usually meant that once an appropriate red spruce had been located, I collected samples from that tree and the nearest balsam fir trees which met my criteria.

In Table 2.2, I briefly describe the individual sites on each mountain. I sampled red spruce and balsam fir trees at three different elevations: 1) near the bottom edge of the spruce-fir forest (“low elevation”); 2) at the treeline, or transition from forest to *krummholz* (“mid elevation”); and 3) within the highest possible patches of *krummholz* (“high elevation”). Sites were centered around treeline because this was a physiognomic boundary that could be used to standardize across different mountain ranges. Furthermore, these trees are at the very limit of existence, and I expected that this would result in the most intense selective pressures for traits that are truly adaptive. There were approximately 300 vertical meters between low and mid elevation sites, and 100 vertical meters between mid and high elevation sites. Across this elevational range, the growth form of both red spruce and balsam fir changes from a tall tree, 30 cm or more in diameter, and ten or more meters in height, to a prostrate shrub, generally 1.0 m or less in height. Tree heights in Table 2.2 are estimated from regressions developed by Battles et al. (1995), based on measured DBH. Although these regressions include a term to take into account the fact that trees become more stout with increasing elevation, they appeared to over-estimate tree heights at the mid-elevation sites. Hence, tree heights are reported only for the low elevation sites.

The statistical model appropriate for the analysis of this design is described in detail in Chapter 4.

Table 2.2. Description of study sites, arranged by mountain. Air temperatures (October 2001-September 2002) are based my own measurements; further details are in Chapter 3 (Micrometeorology). “Ann.” is the annual mean, JAS is July-September mean. Tree heights at low elevation are estimated from measured DBH using regressions of Battles et al. (1995). Reported tree dimensions are the mean of three individuals of each species at each site.

a) Whiteface Mt. (summit 1485 m, 44°22’ N 73°54’ W)

	Low Elevation	Mid Elevation	High Elevation
South Transect			
Site elevation	1120 m	1377 m	1475 m

b) Mt. Mansfield (summit 1339 m, 44°33’ N 72°49’ W)

Low Elevation Mid Elevation High Elevation

c) Mt. Moosilauke (summit 1463 m, 44°01’N 71°51’W)

	Low Elevation	Mid Elevation	High Elevation
South Transect			
Site elevation	1070 m	1380 m	1460 m
Aspect	SE	flat	S
Air Temp. (Ann./JAS)	n/a	n/a	n/a
DBH (fir/spruce)	22.6/19.6 cm	16.4/11.3 cm	n/a
Tree height (fir/spruce)	13.5/11.2 m	n/a	1.5/1.2 m
Notes	along old Carriage Road on ridge running to S. Peak	along hiking trail	immediately below summit *krummholz* mixed with sedge
East Transect			
Site elevation	1070 m	1390 m	1460 m
Aspect	E	SE	flat
Air Temp. (Ann./JAS)	n/a	1.8/12.6°C	n/a
DBH (fir/spruce)	14.0/15.6 cm	14.9/12.5 cm	n/a
Tree height (fir/spruce)	9.2/9.4 m	n/a	0.7/0.8 m
Notes	along abandoned hiking trail steep, rocky spruce rare	just below East Peak	north of summit flat, open area mostly sedge

References

Aplet GH, Laven RD, Smith FW. 1988. Patterns of community dynamics in Colorado Engelmann spruce-subalpine fir forests. *Ecology* **69**: 312-319.

Battles JJ, Fahey TJ, Harney EMB. 1995. Spatial patterning in the canopy gap regime of a subalpine *Abies-Picea* forest in the Northeastern United States. *Journal of Vegetation Science* **6**: 807-814.

Battles JJ, Fahey TJ, Siccama TG, Johnson AH. 2003. Community and population dynamics of spruce-fir forests on Whiteface Mountain, New York: recent trends, 1985-2000. *Canadian Journal of Forest Research* **33**: 54-63.

Berlyn GP, Royte JL, Anoruo AO. 1990. Cytophotometric differentiation of high elevation spruces: Physiological and ecological implcations. *Stain Technology* **65**: 1-14.

Boyce RL. 1988. Wind direction and fir wave motion. *Canadian Journal of Forest Research* **18**: 461-466.

Boyce RL. 1990. Canopy water dynamics of red spruce and balsam fir. Unpublished Ph.D. dissertation, School of Forestry & Environmental Studies, Yale University, New Haven, CT.

Brady NC, Weil RR. 1999. *The nature and properties of soils, 12th ed.* Upper Saddle River, NJ: Prentice-Hall.

Burns RM, Honkala BH. 1990. *Silvics of North America: Volume 1, Conifers*. Washington, DC: U.S. Department of Agriculture Forest Service.

Clark CM, Wentworth TR, O'Malley DM. 2000. Genetic discontinuity revealed by chloroplast microsatellites in eastern North American *Abies* (Pinaceae). *American Journal of Botany* **87**: 774-782.

Cogbill CV, White PS. 1991. The latitude-elevation relationship for spruce-fir forest and treeline along the Appalachian mountain chain. *Vegetatio* **94**: 153-175.

Cook ER, Zedaker SM. 1992.

Fitzgerald BT, Raynal DJ. 1991. Population dynamics and growth of balsam fir on Whiteface Mountain, New York. *Bulletin of the Torrey Botanical Club* **118**: 255-264.

Hadley JL, Smith WK. 1986. Wind effects of needles on timberline conifers: Seasonal influence on mortality. *Ecology* **67**: 12-19.

Hawley GJ, DeHayes DH. 1985. Hybridization among several North American firs. I. Crossability. *Canadian Journal of Forest Research* **15**: 42-49.

Jackson ST, Whitehead DR. 1991. Holocene vegetation patterns in the Adirondack Mountains. *Ecology* **72**: 641-653.

Johnson AH, Schwartzman TN, Battles JJ, Miller R, Miller EK, Friedland AJ, Vann DR. 1994. Acid rain and soils of the Adirondacks. II. Evaluation of calcium and aluminum as causes of red spruce decline at Whiteface Mountain, New York. *Canadian Journal of Forest Research* **24**: 654-662.

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

Manley SAM, Ledig FT. 1979. Photosynthesis in black and red spruce and their hybrid derivatives: ecological isolation and hybrid adaptive inferiority. *Canadian Journal of Botany* **57**: 305-314.

Markus MJ, Bailey BH, Stewart R, Samson PJ. 1991. Low-level cloudiness in the Appalachian region. *Journal of Applied Meteorology* **30**: 1147-1162.

Mohnen VA. 1992. Atmospheric deposition and pollutant exposure of eastern U.S. forests. In: Eagar C and Adams MB, eds. *Ecology and decline of red spruce in the eastern United States*. New York: Springer-Verlag, 64-124.

Myers O, Bormann FH. 1963. Phenotypic variation in *Abies balsamea* in response to altitudinal and geographic gradients. *Ecology* **44**: 429-436.

Peart DR, Nicholas NS, Zedaker SM, Miller-Weeks MM, Siccama TG. 1992. Condition and recent trends in high-elevation red spruce populations. In: Eagar C and Adams MB, eds. *Ecology and decline of red spruce in the eastern United States*. New York: Springer-Verlag, 125-191.

Perron M, Bousquet J. 1997. Natural hybridization between black spruce and red spruce. *Molecular Ecology* **6**: 725-734.

Perron M, Perry DJ, Andalo C, Bousquet J. 2000. Evidence from sequence-tagged-site markers of a recent progenitor-derivative species pair in conifers. *Proceedings of the National Academy of Sciences, USA* **97**: 11331-11336.

Raymo C, Raymo ME. 2001. *Written in stone*. Hensonville, NY: Black Dome Press.

Reiners WA, Hollinger DY, Lang GE. 1984. Temperature and evapotranspiration gradients of the White Mountains, New Hampshire, U.S.A. *Arctic and Alpine Research* **16**: 31-36.

Reiners WA, Lang GE. 1979. Vegetational patterns and processes in the balsam fir zone, White Mountains, New Hampshire. *Ecology* **60**: 403-417.

Richardson AD, Denny EG, Siccama TG, Lee X. 2003.

Wardle P. 1974. Alpine timberlines. In: Ives JD and Barry RG, eds. *Arctic and alpine environments*. London: Methuen, 371-402.

White PS, Cogbill CV. 1992. Spruce-fir forests of eastern North America. In: Eagar C and Adams MB, eds. *Ecology and decline of red spruce in the eastern United States*. New York: Springer-Verlag, 3-39.

Chapter 3:

Micrometeorology of montane spruce-fir forests

Abstract

Mircometeorological stations were installed on three different mountains, representing the major ranges of the northeastern United States: Adirondacks (New York), Green Mountains (Vermont), and White Mountains (New Hampshire). Vegetation patterns in these mountains are strictly controlled by the steep environmental gradient from valley to summit.

Although mean lapse rates of air temperature were comparable to those previously reported for this region, there was clear evidence of a previously unreported east-west lapse rate gradient. The data also document considerable variability in the lapse rate, which was mostly related to diurnal (and, to a lesser degree, seasonal) effects. For some applications, it may be necessary to take this variability into consideration. The diurnal lapse rate pattern was more pronounced on Mt. Moosilauke than either of the other two mountains, and this is likely related to topography. Mean annual soil temperatures and soil temperature heat sums did not show a consistent pattern with regard to elevation.

In these mountains, it has been suggested that frequent cloud immersion at high elevation results in radiation fluxes at that are dramatically reduced compared to those at mid and low elevation. However, results of this study did not support this hypothesis. Clear-sky fluxes of photosynthetically active radiation increased moderately with increasing elevation, but mean (including cloudy days) mid-day fluxes during the growing season were almost identical between mid and high elevation on Mt. Moosilauke.

Introduction

Microclimatology of montane landscapes is dependent on latitude, continentality and topography. The only climatic or meteorological traits generally characteristic of montane environments are altitude-dependent decreases in atmospheric pressure (and is collection of real data through field instrumentation. To this end, two fully-instrumented stations were established on Mt. Moosilauke, in the White Mountains of New Hampshire, and supplementary stations were established on Whiteface Mt. in the Adirondacks of New York, and Mt. Mansfield in the Green Mountains of Vermont (Figure 3.1).

This study improves on previous efforts in this geographic region in two regards. First, in past studies where multiple stations have been installed on a single mountain, either the research objectives were different and the elevational difference between stations was small (e.g. Friedland et al. 1992, 2003), or chart-recording thermographs (rather than electronic data loggers) were used (e.g. Siccama 1974, Reiners et al. 1984). Second, no published studies for the northeast have included extensive data from more than one mountain. The three mountains used here represent the three main ranges in the northeast, and offer an east-west gradient over 200 km in length.

Data and Methods

On Mt. Moosilauke, stations were installed at mid (just below the transition from deciduous forest to spruce-fir) and high elevation (just above the treeline, defined as the transition from a forest of erect stems to prostrate *krummholz*, see Figure 3.2). Hourly data (air temperature, relative humidity, solar radiation and wind speed) from a station at the Hubbard Brook Experimental Forest (an LTER site managed by the United States Forest Service, located approximately 10 km south-east of the Mt. Moosilauke mid elevation station) were used to as a low elevation station for that mountain. On Whiteface Mt. and Mt. Mansfield, stations were installed at three different elevations on each

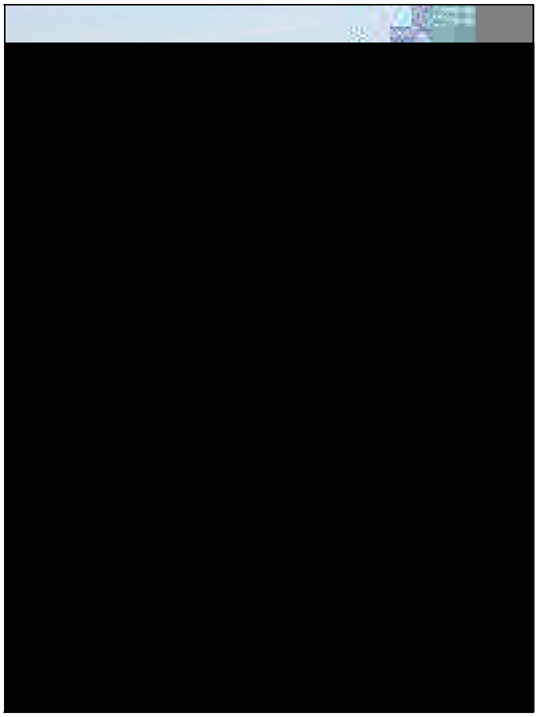


Figure 3.2. High elevation site (1425 m) on Mt. Moosilauke, early October 2001. Accumulated rime ice can be seen on the support pole and anemometer cup. Mt. Washington is the large, snow-capped peak seen in the far distance.

mountain. Site locations and descriptions are given in Table 3.1, and instrumentation details are given in Table 3.2.

Table 3.1. Meteorological station locations and abbreviated site descriptions.

	Low	Mid	High
Adirondack Mountains:			
Whiteface Mt.	• 44.3582°N	• 44.3633°N	• 44.3650°N
summit 1485 m ASL	• 73.8961°W	• 73.9029°W	• 73.9033°W
	• 1095 m	• 1377 m (+282 m)	• 1475 m (+380 m)
	• SE aspect	• S aspect	• S aspect
	• edge of ski trail	• transition from closed forest	• scattered *krummholz*
	• open to S, closed forest to N	• trees " 2-3 m in height	• flagged stems " 1 m in height
Green Mountains:			
Mt. Mansfield	• 44.5200°N	• 44.5288°N	• 44.5412°N
summit 1339 m ASL	• 72.8003°W	• 72.8165°W	• 72.8160°W
	• 917 m	• 1197 m (+280 m)	• 1317 m (+400 m)
	• SE aspect	• flat, ridgetop	• flat, ridgetop
	• edge of road	• just below transition to *krummholz*	• scattered *krummholz*
	• closed forest to N and S	• trees " 4 m in height	• flagged stems " 1 m in height
White Mountains:			
Mt. Moosilauke	• 43.9435°N	• 43.9933°N	• 44.0190°N
summit 1463 m ASL	• 71.7021°W	• 71.8160°W	• 71.8263°W
	• 247 m	• 748 m (+501 m)	• 1425 m (+1178 m)
	• flat, valley	• flat to moderately SW aspect	• flat to moderately E aspect
	• mowed clearing	• fire escape on side of lodge	• 40 m above treeline
	• Hubbard Brook research station	• forest and ridge directly to east	• near top of Moosilauke East Peak

Note: Elevations given in parentheses (e.g. Whiteface mid, +282 m) denote elevation difference between “mid” or “high” elevation stations and “low” elevation stations.

Table 3.2. Instrumentation details of meteorological stations installed for this study.

Whiteface Mt. (June 2001–September 2002): 1095 m, 1377 m, 1475 m

- Hobo H8 Pro (Onset Computer Corp., Bourne, MA) data logger with integral thermistor for air temperature, and external probe for soil temperature (15 cm depth)
- 30 min sampling interval

Mt. Mansfield (October 2000–September 2002): 917 m, 1197 m, 1317 m

- Hobo H8 Pro data logger with integral thermistor for air temperature and capacitive sensor for relative humidity
- 30 min sampling interval

Mt. Moosilauke (September 2001–September 2002): 748 m, 1425 m

- multi-channel data logger (CR-10, Campbell Scientific Inc. [CSI], Logan, UT)
- copper-constantan thermocouples (in conjunction with CR10XTCR (CSI) reference thermocouple) for air temperature and soil temperature (15 cm depth)
- capacitive sensor for relative humidity (CS 500 at 748 m, HMP35C at 1425 m; both CSI)
- quantum sensor (190SZ, Li-Cor Inc., Lincoln, NE) and pyranometer (Model 50, Eppley Laboratory, Newport, RI; 748 m elevation only)
- wind speed and direction (03001 Wind Sentry, R.M. Young Co., Traverse City, MI)
- 10 s sampling interval, 15 minute means output to storage

Air temperature

The location of the alpine treeline is generally considered to be driven by growing-season temperatures, although small-scale variation in treeline elevation may be due to a multitude of other factors, including topography, aspect, wind, winter snow accumulation, and so forth (Griggs 1938, Daubenmire 1954, Körner 1999). The mid elevation sites on Whiteface Mt. and Mt. Mansfield were both located at treeline, and the high elevation site on Mt. Moosilauke was located about 40 vertical m above treeline. Treeline on Mt. Mansfield was located at an elevation almost 200 m lower than on either of the other two mountains. Results suggested that air temperature at treeline was slightly higher on Mt. Mansfield (July-September, 13.7°C; January-March, –8.1°C; annual 2.5°C) than either Whiteface Mt. (July-September, 13.2°C; January-March, –8.5°C; annual 1.9°C) or Mt. Moosilauke (July-September, 12.9°C; January-March, –8.7°C; annual 1.8°C). On this basis it is hypothesized that some factor other than temperature must contribute to the low elevation treeline on Mt. Mansfield.

Mean annual temperatures (October 2001-September 2002 data for both stations installed for this study and NOAA stations) exhibited a very direct relationship with elevation ($R^2 = 0.985$, Figure 3.3), despite the fact that stations are spread out across an east-west distance of approximately 200 km and presumably have different climatic influences. Multiple regression analysis indicated that the mean annual temperature decreased at a lapse rate of –0.57±0.02°C per 100 m elevation (coefficient significantly different from 0 at P ! 0.001), whereas it increased (from east to west) at a rate of 0.29±11°C per degree of longitude ($P = 0.03$). Temperature was not correlated with

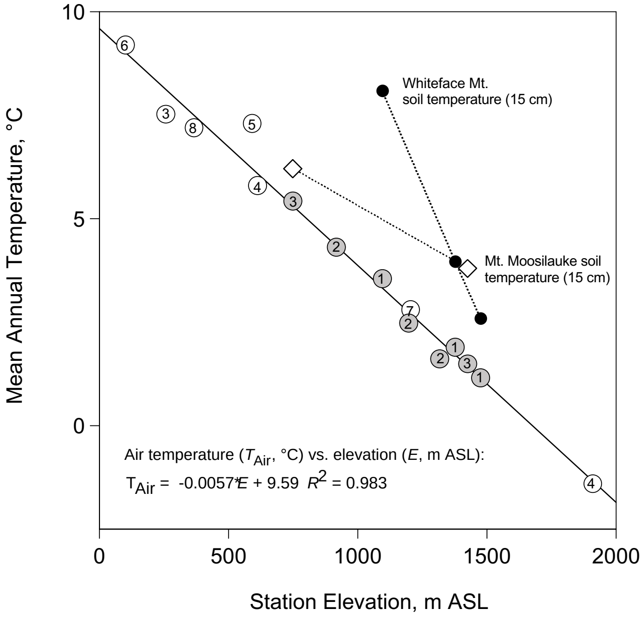


Figure 3.3. Mean annual temperature (based on October 2001-September 2002 measurements) plotted against station elevation for selected weather stations in the northeastern United States. Numbers inside circles refer to stations: 1) Whiteface Mt., NY; 2) Mt. Mansfield, VT; 3) Mt. Moosilauke, NH; 4) Mt. Washington, NH and Pinkham Notch, NH; 5) Lake Placid, NY; 6) Burlington Airport, VT; 7) Mt. Mansfield, VT; and 8) Benton, NH. Shaded symbols for sites 1-3 indicated data collected by the author. NOAA data for other stations obtained from the National Climatic Data Center (see text for further details). Soil temperature gradients on Whiteface Mt. and Mt. Moosilauke are shown for comparative purposes. The linear regression line is based only on air temperature data.

latitude, presumably because the narrow latitudinal band across which the stations were located (less than 1°) makes such patterns difficult to detect (Leffler 1981 calculated that mean annual temperature decreases by 1.08°C per degree of latitude along the Appalachian Mts.). Although Leffler (1981) and Schmidlin (1982) demonstrated similar elevation–temperature relationships for mountaintop, or “crest” stations, the present data set includes a number of stations located either mid-slope or in valleys, and thus validates a much more general relationship across a greater variety of topographical situations.

Monthly mean air temperatures at different elevations on the same mountain were very well correlated with each other, and seasonal patterns were similar across the three different mountains (Figure 3.4). Mean annual lapse rates on each mountain were intermediate between the dry adiabatic lapse rate (–0.98°C/100 m) and the saturated adiabatic lapse rate (–0.50°C/100 m at 20°C) (Barry 1992), and compared favorably with those previously reported for the eastern United States (Table 3.3), as well as mountain ranges in Japan and Europe (Barry 1992) and the tropics (Körner 1999). These figures were also in keeping with (though slightly steeper than) those calculated using NOAA data (Table 3.3). There was a longitudinal trend to the lapse rates at these stations, with the most easterly mountain (Mt. Moosilauke) having the least steep lapse rate, and the most westerly mountain (Whiteface Mt.) having the most steep lapse rate. This pattern is at least consistent with the idea that the Adirondacks have a drier, more continental climate than the White Mountains, which are more humid and maritime (Miller et al. 1993b). However, there was no evidence that the seasonal temperature amplitude was larger on Whiteface Mt. than either of the mountains farther east.

Table 3.3. Comparison of lapse rates calculated in this study with those previously published for the mountains of the eastern United States.

Lapse rate	Location	Source
–0.34	Mt. Ascutney, VT (winter only)	Friedland et al. (2003)
–0.41	Great Smoky Mountains, NC-TN	Shanks (1954)
–0.5	White Mountains, NH	Sabo (1980)
–0.53	Mt. Mansfield, VT	NOAA data (1957-2001)
–0.56	Mt. Washington, NH	NOAA data (1950-2001)
–0.57	Whiteface Mt., NY	NOAA data (1985-1988)
–0.57	NY, VT & NH data	Figure 3, this study
–0.58	Mt. Moosilauke, NH	this study
–0.60	22 NOAA sites, VT & NH	Dingman (1981)
–0.6	Camels Hump, VT	Siccama (1974)
–0.62	Mt. Mansfield, VT	this study
–0.64	Mt. Moosilauke, NH (Apr.-Nov. only)	Reiners et al. (1984)
–0.64	Whiteface Mt., NY	this study
–0.70	Whiteface Mt., NY	Miller et al. (1993b)

Both Figure 3.3 and Table 3.3 are based on annual means, and therefore obscure the considerable variation in lapse rates which occurred at shorter temporal (or spatial) scales. For example, the standard deviation of the mean annual lapse rate calculated using NOAA data was ±0.04°C/100 m, and the standard deviation of the mean monthly lapse rate was ±0.10°C/100 m. There was a general tendency for lapse rates to be least steep during the autumn (October-December) and winter (January-March), and most steep during the spring (April-June) and summer (July-September) (Table 3.4). Although these results fit in with what is generally observed in temperate zone mountains (Körner 1999), the pattern was by no means universal. For example, Whiteface Mt. data from this study

Table 3.4. Quarterly mean lapse rates of air temperature (°C/100 m elevation) on mountains in the northeastern United States.

	JFM	**AMJ**	**JAS**	**OND**
*Calculated using hourly or quarter-hourly data (*Author's own stations)*:*				
Whiteface Mt. (10/01-10/02)	-0.64	-0.63	-0.65	-0.63
Mt. Mansfield (10/00-10/02)	-0.56	-0.65	-0.64	-0.64
Mt. Moosilauke (9/01-10/02)	-0.60	-0.63	-0.58	-0.52
Calculated using monthly mean data (NOAA/ASRC stations)*:*				
Mt. Washington (1950-2001)	-0.49±0.06	-0.63±0.03	-0.59±0.03	-0.51±0.05
Mt. Mansfield (1957-2001)	-0.45±0.10	-0.57±0.07	-0.56±0.05	-0.52±0.06
Whiteface Mt. (1985-1988)	-0.54±0.06	-0.56±0.02	-0.60±0.10	-0.58±0.06

Note: JFM, January–March; AMJ, April–June; JAS, July-September; OND, October–December. Values are mean ± 1 S.D. for lapse rates calculated using monthly data.

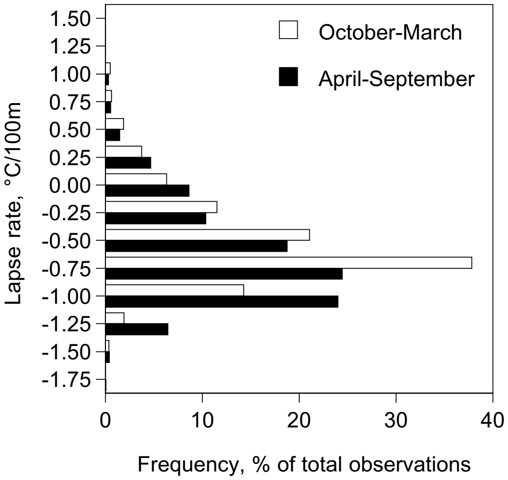
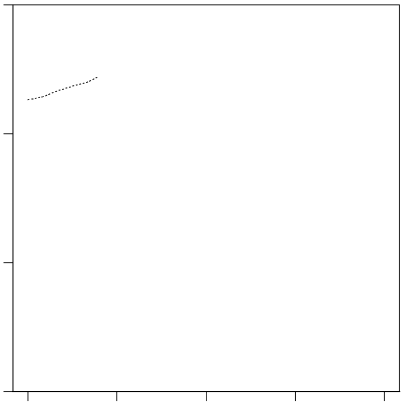


Figure 3.5. Histogram of lapse rates calculated between mid and high elevation station on Mt. Moosilauke (September 2001-September 2002).

There was a clear diurnal lapse rate pattern, with lapse rates during the day generally being much stronger than those at night (Figure 3.6). The diurnal variation in lapse rate was stronger on Mt. Moosilauke than the other two peaks. This can be attributed to differences in the mean diurnal temperature range (mean[daily maximum – daily minimum]), which was larger at low (10.7 ± 4.8°C) and mid (9.4 ± 4.1°C) elevation than at high elevation (6.9 ± 3.1°C) on Mt. Moosilauke. Although this fits the expected pattern (Körner 1999), on both Whiteface Mt. and Mt. Mansfield, there was no elevation pattern in the mean diurnal temperature range, which was fairly constant at about 7.5°C. The phase or timing of the diurnal lapse rate pattern also differed among mountains. For example, on Mt. Moosilauke, lapse rates reached a minimum at 0600 h and peaked at 1500 h. In contrast, on Whiteface Mt. and Mt. Mansfield, lapse rates were quite stable from 1800-0600 h, but reached a maximum in mid-morning. Both magnitude and phase differences in the diurnal pattern can likely be explained by the different topographies of the three mountains. For all three of the mountains, high elevation sites were located in exposed positions where strong winds and good mixing were probably common. The low elevation sites on Whiteface Mt. and Mt. Mansfield were located mid-slope, which contrasts with the mid elevation site on Mt. Moosilauke, which was located in a valley. Valley sites, which typically have stronger diurnal patterns, warm more during the day, and cool more during the night, than mid-slope sites (Barry 1992). The diurnal mountain/valley breeze pattern (described below), as well as differences in aspect, may further enhance this effect and contribute to the diurnal lapse rate pattern.



Soil temperatures

In the Great Smoky Mountains of the Southern Appalachians, Shanks (1956) found that the elevational gradient for mean soil temperature (at 15 cm depth) was similar to that for air temperature from May to October. Similarly, Siccama (1974) reported that mean annual soil temperature (15 cm) on Vermont's Camels Hump decreased linearly with elevation, from 7.2°C at 549 m to 3.9°C at 1158 m. In that study, soils were frozen for only a few weeks of the year, even at the highest elevation sites. The results of the present study give a somewhat different view. Not only were soils frozen for much of the year (10 weeks of the year at mid elevation, and 28 weeks of the year at high elevation, on Whiteface Mt., and 17 weeks of the year at high elevation on Mt. Moosilauke), but the low elevation site on Whiteface Mt. stands as an outlier in Figure 3.3. Thus, compared to air temperatures, soil temperatures did not vary consistently with elevation, and there were clear differences in the elevation pattern on different mountains (Figure 3.4). At mid elevation on Whiteface Mt., soil temperatures were generally warmer than those at high elevation, except during June, July and August, when they were cooler. This is probably related to the open spacing and sparse crowns of trees at high elevation, which allows more solar radiation to reach the soil surface compared to mid elevation (Körner 1999). A contributing factor may be that windswept *krummholz* sites can have surprisingly thin snow cover compared to lower elevation sites. A thin layer of snow provides less insulation against extreme cold in the winter, and melts more quickly in the spring, compared to a thick layer. At high elevation, monthly mean soil temperature dropped well below freezing during the winter months, whereas at mid elevation, monthly mean soil temperature remained right at freezing, and at low elevation, monthly mean soil temperature never dropped below 3°C. On Mt. Moosilauke, high elevation soil temperatures remained right at freezing throughout the winter months, whereas this threshold was never reached at mid elevation.

On Mt. Moosilauke, additional soil temperature probes were installed at sites approximately 50-100 m from each the main weather stations, thus providing a measure of the variation in soil temperature at a given elevation. At both high and mid elevation sites, mean annual soil temperature measured by the supplementary probe was within 0.2°C of that measured by the primary station. These data suggest that soil temperatures measured on Mt. Moosilauke are representative of their sampling elevation.

Air and soil temperature thresholds: Heat sums

Many physiological processes show a temperature response (e.g. photosynthesis and respiration, see Tranquillini 1979), and various temperature thresholds have been suggested for different aspects of plant function. For example, soils must be unfrozen if water uptake by roots is to occur, whereas air temperatures in the range of 5-7°C are thought necessary for the development of new tissues (this is the basis of Körner's 1999 "sink oriented hypothesis" for treeline location, which was recently supported by the work of Hoch et al. 2002), and for a long time it was thought that treeline was determined by the point at which the mean July air temperature reached 10°C (e.g. Daubenmire 1954, Tranquillini 1979). In this study, mean air temperatures were above freezing for about 25-30 weeks of the year at all elevations and on all mountains. However, the length of the frost-free period decreased steadily with increasing elevation, and as a result there was only a very short time period suitable for tree growth and development at the highest elevation sites (this suggests a “tissue ripening hypothesis” to explain treeline location, Wardle 1971, Tranquillini 1979). That high-elevation systems are energy limited is illustrated by the heat sums given in Table 3.5. There was an near-perfect linear relationship between station elevation and heat sums[1] at all three reference temperatures used (0°C, 5°C and 10°C). For example, with a reference temperature of 0°C, the annual heat sum (in degree-days) could be predicted for a station at elevation E using the equation:

$$\text{Heat sum} = 3595 - 1.25 \times E \ (R^2 = 0.994)$$

This relationship compares quite favorably with that derived by Reiners et al. (1984), who reported that heat sums (0°C reference temperature) decreased at a rate of 1.3 degree-days per m elevation. The present heat sum–elevation relationship became less steep as the reference temperature increased. For example, 5°C heat sums decreased at a rate of 0.95 degree-days/m ($R^2 = 0.993$), whereas 10°C heat sums decreased at a rate of 0.71 degree-days/m ($R^2 = 0.992$). Forest stand modeling at Hubbard Brook by Botkin et al. (1972) has suggested that heat sums are a critical factor in determining the vegetational zonation along elevational gradients in the northeastern United States.

Soil temperature heat sums did not follow the same linear pattern (Table 3.5). For example, on Whiteface Mt., the heat sums calculated with reference temperatures of 5°C and 10°C were higher at the high elevation site (636 and 127 degree-days) than the mid elevation site (571 and 54 degree-days), which is the opposite of the expected pattern. Coupled with the somewhat anomalous soil temperature data from the low elevation

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[1] Heat sums, measured in degree-days, are calculated by taking the difference (daily mean temperature – reference temperature) and doing a summation across the entire year for all positive differences.

Table 3.5. Temperature sums (in degree-days) computed for different reference temperatures at stations located along the elevational gradient on three mountains of the northeastern United States.

Reference temperature:	**0°C**	**5°C**	**10°C**
Air temperature:			
Whiteface Mt.			
Low (1095 m)	2317	1362	629
Mid (1377 m)	1879	1011	360
High (1475 m)	1746	907	291
Mt. Mansfield			
Low (917 m)	2453	1482	712
Mid (1197 m)	2080	1178	496
High (1317 m)	1926	1065	426
Mt. Moosilauke			
Low (247 m)	3279	2086	1187
Mid (748 m)	2668	1639	833
High (1425 m)	1869	1017	391
Soil temperature:			
Whiteface Mt.			
Low (1095 m)	2983	1359	553
Mid (1377 m)	1420	571	54
High (1475 m)	1320	636	127
Mt. Moosilauke			
Mid (748 m)	2280	1130	400
High (1425 m)	1404	553	55

station on Whiteface Mt. (Figure 3.3), these data from the mid and high elevation sites further indicate that strict correlations between elevation and soil temperature do not always occur.

Relative humidity

On Mt. Moosilauke, the frequency of very high relative humidities (RH # 90%) increased with station elevation (Figure 3.7), and similar results were obtained for Mt. Mansfield (data not shown). Across the whole year, RH # 90% were more than twice as common at high elevation as at low elevation (Figure 3.7). However, patterns were somewhat different in winter (January-March) and summer (July-September); the difference between low and high elevation was less extreme during the summer months. For example, in the winter, RH was 90% or greater for 26.7% of observations at the low station, 64.9% of observations at the mid station, and 74.8% of the observations at the high station. In the summer, RH was 90% or greater for 35.7% of observations at the low station, 40.0% of the observations at the mid station, and 49.7% of the observations at the high station.

Wind speed and direction

At high elevation on Mt. Moosilauke, measured wind gusts reached a maximum of 22.0 m/s during the period June-September 2002 (the only period for which consistently valid data were obtained, due to rime icing of the anemometer cup). In contrast, at mid elevation, gusts reached only 12.0 m/s. It should be noted that actual gusts may have been higher, as both the inertia of the cup anemometer and the 15 s

sampling frequency could result in some damping of the measured peak. Mean wind speeds decreased with decreasing elevation, from 4.8 m/s at high elevation to less than 1.0 m/s at mid and low elevations (Figure 3.8). A reasonable estimate of winter wind speeds can be obtained by doubling the above figures: the mean wind speed on the summit of Mt. Washington is 12 m/s during the summer and nearly twice that (23 m/s) during the winter (Barry 1992).

In addition to higher gust and mean wind speeds at high elevations, the standard deviation of wind speeds was much larger (2.7 m/s) at high elevation than at either mid or low elevation (both ! 1.0 m/s). From the distribution of mean wind speeds across the three elevations (Figure 3.8), it is clear that extreme wind events were much less common at both low and mid elevation than at high elevation. For example, at high elevation, 19% of 15-minute mean wind speeds were greater than 6 m/s, whereas at mid and low elevations, the wind was never as consistently strong. Topography (i.e. the exposed position of the high elevation station on the East Peak of Mt. Moosilauke), rather than elevation, is thought to be the key factor that determines mountain wind patterns (Barry 1992). This hypothesis is supported by the patterns in wind direction observed on Mt. Moosilauke. At the high elevation station, there were no diurnal patterns in wind direction (Figure 3.9), as the prevailing winds were from the SW and NW, regardless of time of day. In contrast, at the mid elevation station there was a classic, clearly defined valley wind (upslope or anabatic) during the day and mountain wind (downslope or katabatic) at night (Oke 1978). At the mid elevation site, the Baker River valley runs in a south-west to north-east direction, which almost perfectly matches the observed

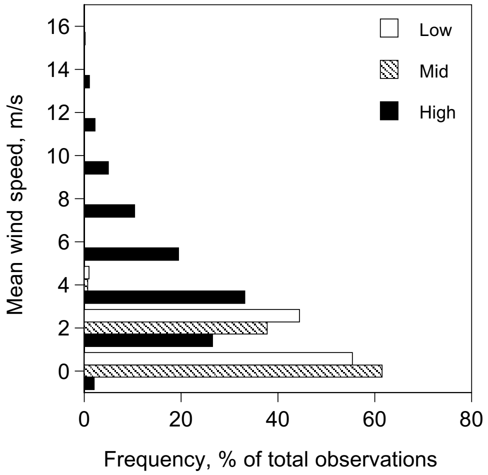
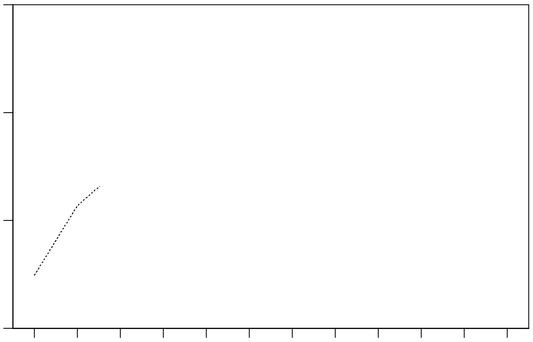


Figure 3.8. Histogram depicting frequency distribution of mean wind speed observations at three elevations on Mt. Moosilauke (June 2002-September 2002).

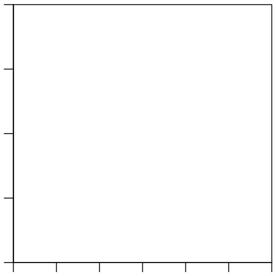
directional pattern. During the day, winds blew from southwest, up the valley to the northeast. The pattern was reversed at night.

Solar radiation and PAR fluxes

Solar radiation generally increases with increasing elevation (with the effect most pronounced below 2000 m) because the shorter atmospheric path length reduces molecular scattering and absorption by gases (Barry 1992). On clear days at Mt. Moosilauke, mid-day total solar radiation fluxes (R) at mid elevation were about 5% higher than those at low elevation, and PAR fluxes (Q) at high elevation were about 4% higher than those at mid elevation. However, when cloudy days are included and data are averaged on a monthly basis, results were somewhat different. Monthly mean R was somewhat higher at low elevation on Mt. Moosilauke compared to mid elevation (Figure 3.10A). This pattern was more or less consistent across the calendar year, and can probably be attributed to the greater frequency of local cloud immersion (which blocks incoming radiation) at mid, compared to low, elevation. In the northern Appalachians, Markus et al. (1991) found that elevations between 900 and 1300 m were more likely to experience cloud impaction (typically stratus clouds) than elevations above or below this range, and Miller et al. (1993a, b) determined that spruce-fir forests on Whiteface Mt. were immersed in cloud for 10% of the year at 1050 m but perhaps as much as 35% of the year at 1350 m. In comparison, monthly mean Q (during the growing season, at least) was quite similar between mid and high elevation sites (Figure 3.10B), which suggests that any increased frequency of high-elevation clouds is offset by the increased Q flux under clear-sky conditions at high elevation. Therefore, these data do not support the hypothesis that mean PAR fluxes at higher elevations are dramatically reduced compared to those at lower elevations, as has been suggested previously (e.g. Richardson and Berlyn 2002). However, since the frequency of cloud immersion appears to differ among



September, between the hours of 1000 and 1700 h, were retained. This yielded a data set (15 minute means) with 3538 observations. The mean Q/R ratio was 2.11 ± 0.12, whereas a linear regression of Q against R (with no intercept) suggested a ratio of 2.07 ($R^2 = 0.998$). There were only a handful of points with unusually low (21 points with Q/R ! 1.85) or high (80 points with $Q/R > 2.45$) Q/R ratios. The Q/R ratio was, to a large degree, a function of R (Figure 3.11A). For example, high (> 2.2) values of Q/R were rare except when R was less than 250 W/m^2. R



The dewpoint temperature is a measure of how much precipitable water there is in the atmosphere, which will affect the *Q*/*R* ratio in two ways, as described by Alados et al. (1996). First, because infrared radiation is strongly absorbed by water, an increase in dewpoint will cause greater reduction of *R* than *Q*. Second, scattering by aerosols will be enhanced by water vapor, and an increase in scattering will cause a greater reduction of *Q* than *R*. These results suggest that an increase in dewpoint temperature causes an increase in *Q*/*R*, and so the first effect seems to outweigh the second. Alados et al. (1996) also found a positive correlation between dewpoint temperature and *Q*/*R*.

Summary

In the mountains of the northeastern United States, vegetation patterns are driven by climate. Results of the present study confirm a general elevation–mean temperature relationship that holds across the area of study. At shorter time periods, there was significant variation in the lapse rate up the side of each mountain. Topography seems to play a major role in the diurnal lapse rate pattern. Energy limitation becomes progressively more pronounced with increasing elevation, and the heat sum of degree-days above 0°C decreased linearly with elevation at a rate of 1.25 degree-days per m elevation. Other variables, such as soil temperature, were not as closely correlated with elevation as has been previously suggested. Solar radiation flux, which had been hypothesized to decrease with increasing elevation (because of increased cloud frequency) was found to be similar at both summit and mid-slope stations.

Acknowledgements

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References

Alados I, Foyo-Moreno I, Alados-Arboledas L. 1996. Photosynthetically active radiation: measurements and modelling. *Agricultural and Forest Meteorology* **78**: 121-131.

Alados I, Alados-Arboledas L. 1999. Direct and diffuse photosynthetically active radiation: measurements and modelling. *Agricultural and Forest Meteorology* **93**: 27-38.

Barry RG. 1992. *Mountain weather and climate, 2nd ed.* New York: Routledge.

Botkin DB, Janak JF, Wallis JR. 1972. Some ecological consequences of a computer model of forest growth. *Journal of Ecology* **60**: 849-872.

Daubenmire R. 1954. Alpine timberlines in the Americas and their interpretation. *Butler University Bot. Stud.* **11**: 119-136.

Dingman SL. 1981. Elevation: a major influence on the hydrology of New Hampshire and Vermont, USA. *Hydrological Sciences Bulletin* **26**: 399-413.

Eagar C, Adams MB. 1992. *Ecology and decline of red spruce in the eastern United States*. New York: Springer-Verlag.

Friedland AJ, Boyce RL, Vostral CB, Herrick GT. 2003. Winter and early spring microclimate within a mid-elevation conifer forest canopy. *Agricultural and Forest Meteorology* **In press**.

Friedland AJ, Boyce RL, Webb ET. 1992. Winter and early spring microclimate of a subalpine spruce-fir forest canopy in central New Hampshire. *Atmospheric Environment* **268**: 1361-1369.

Griggs RF. 1938. T9cARf-f-1236.-2.16 TD249A857 r! .66 ir gradient at Whiteface Mountain, NY, U.S.A. *Atmospheric Environment* **27A**: 2121-2136.

Miller EK, Panek JA, Friedland AJ, Kadlecek J, Mohnen VA. 1993a. Atmospheric deposition to a high-elevation forest at Whiteface Mountain, New York, USA. *Tellus* **45B**: 209-227.

Mohnen VA. 1992. Atmospheric deposition and pollutant exposure of eastern U.S. forests. In: Eagar C and Adams MB, eds. *Ecology and decline of red spruce in the eastern United States*. New York: Springer-Verlag, 64-124.

Oke TR. 1978. *Boundary layer climates*. London: Routledge.

Reiners WA, Hollinger DY, Lang GE. 1984. Temperature and evapotranspiration gradients of the White Mountains, New Hampshire, U.S.A. *Arctic and Alpine Research* **16**: 31-36.

Reiners WA, Lang GE. 1979. Vegetational patterns and processes in the balsam fir zone, White Mountains, New Hampshire. *Ecology* **60**: 403-417.

Sabo SR. 1980. Niche and habitat relations in subalpine bird communities of the White Mountains of New Hampshire. *Ecological Monographs* **50**: 241-259.

Schmidlin TW. 1982. Leffler's method of estimating average temperatures of Appalachian summits: Evaluation in New York. *Journal of Applied Meteorology* **21**: 745-747.

Shanks RE. 1954. Climates of the Great Smoky Mountains. *Ecology* **35**: 354-361.

Shanks RE. 1956. Altitudinal and microclimatic relationships of soil temperature under natural vegetation. *Ecology* **37**: 1-7.

Siccama TG. 1974. Vegetation, soil and climate on the Green Mountains of Vermont. *Ecological Monographs* **44**: 325-349.

Tranquillini W. 1979. *Physiological ecology of the alpine timberline*. New York: Springer-Verlag.

Wardle P. 1971. An explanation for alpine timberline. *New Zealand Journal of Botany* **9**: 371-402.

Chapter 4:

Foliar morphology and anatomy

Abstract

Red spruce (*Picea rubens* Sarg.) and balsam fir (*Abies balsamea* [L.] Mill) are the dominant conifers at treeline in the mountains of the northeastern United States. Balsam fir, with its nearly transcontinental distribution and wide ecological range, is considered to have a much broader niche than red spruce, with its narrow geographic distribution and more specific site requirements. The objective of this study was to investigate changes in needle morphology and anatomy of these species along elevational and canopy light gradients.

Measured traits included needle mass, area, length, width, thickness, cuticle thickness, epidermis thickness, the cross-sectional area of the vascular cylinder (VC_{CSA}), and, at the shoot level, the density of needle packing. Statistical analysis of these traits revealed few species × elevation interactions, suggesting that balsam fir and red spruce respond similarly to the elevational gradient. More common were species × crown interactions, indicating that the species respond differently (either in magnitude or direction) to the canopy light gradient.

A plasticity index, Π, was constructed to measure the relative difference in measured trait values between sun and shade needles. Shoot morphology (i.e. needle packing), was more plastic than any of the needle-level anatomical or morphological traits measured.

Results only weakly supported the hypothesis that the capacity for plasticity is correlated with ecological breadth: across the sun/shade gradient, balsam fir was marginally more plastic than red spruce, but the difference between species was not significant ($P = 0.30$).

Results gave strong support for the hypothesis that plasticity is reduced in a harsh growth environment: in both species, there was less sun/shade plasticity at the highest elevation sites compared to either the low- or mid-elevation sites (P ! 0.05). Above treeline, survival may depend more on stress tolerance, and less on plasticity to the light environment, than at lower elevations.

Introduction

Leaves are the major interface between plants and their environment, and must therefore be constructed in such a way as to withstand significant environmental stresses. However, as the key photosynthetic organs, leaves are also essential for primary production. There is a direct connection between structure and function because foliar anatomical features control the internal gradients of light and carbon dioxide, which are the two key inputs for photosynthesis (Smith and Knapp 1990, Smith et al. 1997). Thus, optimal leaf design must balance these potentially conflicting requirements.

For a century or more, botanists and ecologists have studied the ways in which leaf structure and function vary with environment. Early observers (e.g. Clements 1905, Hanson 1917, McDougall and Penfound 1928, Büsgen and Münch 1929, Stover 1944, Wylie 1951) recognized differences not only among functional groups (e.g. mesophytes and xerophytes), but also among individuals of the same species, such as those growing in different light (i.e. sun vs. shade) and temperature (i.e. alpine vs. lowland) regimes.

The capacity for a response to environment is limited by both phylogenetic constraints and structural/physiological constraints. Recent research demonstrates that there are universal relationships among fundamental leaf traits that hold across different biomes (Reich et al. 1999; Ackerly and Reich 1999). Thus, evolutionary trade-offs place strong limitations on foliar form and function, and limit the ways in which leaf traits can be combined (Reich et al. 1999). Nevertheless, within these bounds, leaves can still be highly specialized in their adaptations to different environments (Hanson 1917, Larsen 1927, Gutschick 1999). From an ecological perspective, quantifying differences in leaf structure and function along environmental gradients may reveal the adaptations necessary to survive in extreme conditions (e.g. high elevation, Körner et al. 1989, Smith and Knapp 1990), and also gives insight into how the capacity to respond to environment varies among species. Studies along elevational gradients are just one example of natural experiments from which it may be possible to determine long-term species responses to a range of environmental conditions. Such knowledge is crucial for the accurate prediction of the effects of climate change on terrestrial ecosystems. However, there have been few studies of the relationships between elevation and leaf structure, especially in conifers at the limits of tree growth (Smith and Knapp 1990).

The primary objective of this paper is to compare the foliar response to light and elevation of two montane conifers, balsam fir (*Abies balsamea* [L.] Mill.) and red spruce (*Picea rubens* Sarg.). The concept of phenotypic plasticity, which refers to the ability of a genotype to express different phenotypes in response to the growth environment (Schlichting 1986, West-Eberhard 1989, de Jong and Stearns 1990, Via 1994) is a central theme. Plasticity itself may be thought of as a functional trait with ecological significance, because it enables long-lived, sessile individuals to function across changing or heterogeneous environments, or environments to which they may not be perfectly adapted (Bradshaw 1965, Schlichting 1986, Chapin et al. 1993). This can help maintain the competitiveness of a particular genotype; it also provides a species with a means of achieving broad ecological distribution even when genetic diversity is low.

The response to light is considered the classic example of phenotypic plasticity in plants. From gap to understory, or upper canopy to lower canopy, plants can experience a light environment that varies in intensity over several orders of magnitude. A key trait of plants, distinguishing them from animals, is their capacity for meristematic growth. This trait, combined with branching architecture, results in a modularity that enables single organisms to simultaneously express multiple phenotypes. For example, variation in leaf structure and function can occur not only among plants growing in different environments, but also within an individual. Therefore, analogous to sun and shade plants are sun and shade leaves on the same plant (Boardman 1977, Lichtenthaler et al. 1981, Lichtenthaler 1985).

In trees, sun and shade leaves are typically displayed as a response to the canopy light gradient. Photomorphogenetic responses are a product of both light quality, as measured by the red:far red (R:FR) ratio (van Hinsberg and van Tienderen 1997), and the total integrated amount of light energy (rather than the peak flux density—Chabot et al. 1979) received during bud development and leaf expansion. Direct shading reduces the quantity of light available in the lower crown positions, and the quality of light is altered by the strong absorption of red wavelengths (" 680 nm) and strong reflectance of far red wavelengths (" 730-1000 nm) by foliage. Other abiotic factors (e.g. evaporative demand, air and leaf temperatures, wind) may vary along the canopy gradient, but light is considered the main environmental cue (Fitter and Hay 1987). In mature trees of most deciduous species, sun leaves are thicker, but smaller in area, than shade leaves, and have more palisade mesophyll relative to spongy mesophyll, as well as a higher stomatal density but smaller guard cells (Boardman 1977, Lichtenthaler 1985). In conifers, sun needles are often thicker, wider, and larger in projected area than shade needles, and they also typically have thicker cuticles, thicker epidermal walls, more vascular tissue, and increased transverse cross-sectional area (Richardson et al. 2000, 2001). Sun leaves usually have high light-saturated photosynthetic rates, but are less efficient at low light levels than shade leaves. This is because shade leaves have lower rates of dark respiration (Boardman 1977). The plastic response to light has been shown to vary among species, and may be related to shade tolerance (Jackson 1967), successional status (Ashton and Berlyn 1992), or ecological niche (Ashton and Berlyn 1994). This leads to the first of two hypotheses to be explicitly tested in the following paper, *Hypothesis A*: The capacity for plasticity is correlated with ecological breadth. *Prediction*: generalists are more plastic than specialists, and so the species with the broader niche (balsam fir: mid-successional, shade tolerant, nearly transcontinental range) will display greater plasticity than the

These peaks represent an east-west transect from 71°50’W to 73°45’W at 44°N across a latitudinal range of one-half degree. These three ranges are characterized by similar climates and patterns of vegetational zonation (Cogbill and White 1991), with hardwood forests which extend to roughly 700-800 m elevation, spruce-fir forests to roughly 1100-1200 m, stunted *krummholz* above 1100-1200 m, and alpine tundra at the summits (1400+ m).

Data from weather stations at two elevations (valley station " 800 m and summit station " 1400 m) on Mt. Moosilauke indicate a mean temperature lapse rate of between -0.52°C/100 m (October-December) and -0.63°C/100 m (April-June) (see Chapter 3). Mean summer (July-September) air/soil temperatures were 12.6°C/10.2°C at 1400 m and modestly increased flux intensity at high elevation is balanced out by a greater frequency of clouds.

Relative humidity at 1400 m was above 90% for 65% of the time, compared to 50% of the time at 800 m. Precipitation and soil moisture were not monitored, but Reiners et al. (1984) used soil moisture data combined with models of potential and actual evapotranspiration to demonstrate that water limitation decreases with increasing elevation, and is typically rare above elevations of about 800 m.

Sampling procedure

A split-split plot experimental design was used, with the main plot (site, defined by elevation), split first by species (red spruce vs. balsam fir) and then by crown position (sun vs. shade). Conifer foliage was sampled at three different elevations: 1) near the bottom edge of the spruce-fir forest; 2) at the tree line (or transition from forest to *krummholz*); 3) and within the highest patches of *krummholz*. These elevations are denoted henceforth as low (L), mid (M), and high (H), respectively. Two transects, one on the east side and one on the south side, were run on each mountain. The actual elevations varied somewhat among transects, depending on physiognomy and forest structure. There was typically 300 m elevation between L and M sites, and 100 m elevation between M and H (see Chapter 2 for more details, including station locations and elevations). The sampling design is illustrated in Figure 4.1.

At each elevation on each transect (i.e. at each plot), three trees of each of the two study species were identified. Mean tree dimensions are given in Table 4.1. Diameter at breast height (DBH) of sampled trees was recorded at L and M, whereas the height of

trees at H was measured directly. Bole diameters at L were used to estimate approximate tree heights using species-specific allometric equations from Whiteface Mt. (Battles et al. 1995). These equations include an elevation term to account for the fact that trees become

Table 4.1. Mean (± 1 S.D.) tree dimensions and canopy light transmittance of red spruce and balsam fir sampled at three different elevations. DBH, diameter at breast height; T_R, relative canopy transmittance of red (660 nm) light; T_{FR}, relative canopy transmittance of far red (730 nm) light; R:FR, red:far red ratio under canopy relative to red:far red ratio above canopy (calculated as T_R/T_{FR}). Tree height at low elevation was estimated from DBH using equations calibrated at Whiteface Mt. by Battles et al. (1995). Tree height at high elevation was measured directly but because trees did not reach breast height no DBH was recorded. Means are based on a total of 18 individuals of each species sampled at each elevation.

	Red spruce			Balsam fir		
	Low	Mid	High	Low	Mid	High
DBH (cm)	19.5±8.2	8.8±4.3	n/a	18.6±7.5	10.4±5.8	n/a
Height (m)	11.1±3.4	n/a	1.0±0.4	11.5±3.7	n/a	1.1±0.5
T_R	0.14±0.11	0.14±0.04	0.14±0.04	0.10±0.05	0.12±0.05	0.13±0.04
T_{FR}	0.20±0.13	0.20±0.06	0.21±0.05	0.16±0.07	0.17±0.05	0.18±0.05
R:FR	0.67±0.08	0.70±0.10	0.63±0.08	0.65±0.10	0.68±0.08	0.64±0.10

environment compared to the shade needle environment; the relative R:FR ratio of shade needles was only two-thirds (between 0.63–0.70) that of sun needles (Table 4.1). However, neither the quantity nor the quality of light received by shade needles varied significantly between species or among elevations (all *P* objectives of 2×, 3.5×, 10×, and 20×, depending on the cellular or histological attributes to be measured. The camera-computer setup provided an additional 8× magnification. Thus, the total magnification ranged from 80× (e.g. needle width measurements) to 800× (e.g. cuticle measurements).

Needle width, needle thickness, total transverse needle cross-sectional area (T_{CSA}) measured perpendicular to the long axis, cross-sectional area inside the epidermis (IE_{CSA}), and vascular cylinder cross-sectional area (VC_{CSA}) were all measured once on each mounted needle. Cuticle thickness and epidermal thickness were measured 10 times each on each mounted needle (5 on each of the adaxial and abaxial sides). Measurements from the three needles from each sample were averaged to generate a single mean value for each tree × crown position combination.

Needle and shoot morphology

Thirty needles were arranged in a 5×6 grid on the glass of a flatbed scanner (model Expression 636, Epson America, Torrance, California, USA). Needles were scanned as black and white images at 59 pixels / cm (150 dpi), using a threshold setting selected to minimize edge shadows and glare. Image analysis using particle recognition routines (NIH Image, in the public domain and available free over the Internet at http://rsb.info.nih.gov/ nih-image/) was conducted to measure the projected area of each individual needle. Dry mass of the 30 needles were measured to 0.0001 g using an electronic balance (model ER 182 A, A+D Company, Tokyo, Japan).

To quantify shoot morphology in terms of needle packing (mass of needles per cm of branch, or total projected needle area per cm of branch), the needles were removed from one 6 cm length of the previous-year's growth, and both projected area and dry mass were measured as described above.

Calculations

From the above measurements, additional morphological and anatomical attributes were calculated as follows. Mesophyll cross-sectional area (M_{CSA}) was calculated as $IE_{CSA} – VC_{CSA}$, and the ratio of vascular tissue to mesophyll tissue (vascular:mesophyll ratio, VMR) was calculated as VC_{CSA}/M_{CSA}. The needle width to thickness (NWT) ratio was calculated as (needle width)/(needle thickness). Needle length was calculated as (projected needle area)/(needle width). Needle tissue density was calculated as (needle mass)/(needle length $\times$ T_{CSA}), and the needle mass to area (NMA) ratio was calculated as (needle mass)/(projected needle area). NMA represents a measure of construction cost (i.e. leaf mass) relative to the potential for light collection (i.e. leaf area).

To quantify the relative size difference in measured traits between sun and shade leaves, a plasticity index, Π, was used. For each tree, Π was calculated as (mean sun leaf trait measurement)/(mean shade leaf trait measurement).[1] The mean Π across the three trees of each species at each plot was then calculated arithmetically: if Π was less than 1.0, then the reciprocal of Π, 1/Π, was taken to be the index value. In this way, Π always ranged from 1.0 upwards. Values of Π close to 1.0 indicate low plasticity (i.e. little

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[1] Two other plasticity indices were calculated for each species. Index 1 used the coefficient of variation (CV%) of each trait measurement as a relative measure of variability across all samples. Index 2 was calculated as (min trait measurement – max trait measurement)/(min trait measurement) where min and max refer to the minimum and maximum values of each trait across all samples. Consistent results were obtained regardless of the measure used, and both indices correlated well with Π.

difference in measured trait between sun and shade leaves), whereas more extreme values (e.g. greater than 1.5), indicate higher plasticity. The symbol $\overline{\Pi}$ is used to denote the mean plasticity across multiple traits.

Statistical analysis

Data were analyzed using a mixed model (see Table 4.2) to properly account for the split-split plot design, as well as the combination of fixed factors (elevation [E], species [S], crown position [C], and interactions of these factors) and random factors (transect, pairs of trees, and all interactions with these factors). Analysis was conducted using PROC MIXED (SAS 6.12, SAS Institute, Cary, North Carolina, USA). Where necessary, data were log-transformed to improve error term normality and variance homogeneity, but all reported values have been back-transformed. A significance level of $\alpha = 0.05$ was used for all tests.

Results

Foliar anatomy and morphology

Anatomical and morphological traits of both species varied somewhat among mountains (Table 4.3), but this variation was typically small compared to the pronounced differences between species (S effect significant for all traits except tissue density and cuticle thickness) and crown positions (C effect significant for all traits) (Table 4.4). Measured needle mass, NMA, tissue density, needle width, needle thickness, epidermis thickness, cuticle thickness, VC_{CSA}, M_{CSA}, and VMR were all consistently larger for sun needles than shade needles; however, shade needles were greater in length than sun

Table 4.2. Analysis of variance (ANOVA) table for split-split plot experimental design used in this study. The letters "r" and "f" identify random and fixed factors, respectively. There were six transects (two on each of three mountains), three elevations (low, mid, and high), two species (red spruce and balsam fir), and two crown positions (sun and shade), for a total of 216 samples collected.

r/f	Source of Variation		df	Error term
Main plot:				
r	Transect	T	5	P(T×E)
f	Elevation	E	2	T×E
r	Error 1	T×E	10	P(T×E)
r	Pairs of trees	P(T×E)	36	—
Split-plot:				
f	Species	S	1	Error 2
f	Elevation × Species	E×S	2	Error 2
r	Split-plot error, Error 2: (T×S + T×E×S)	T×E×S	15	S×P(T×E)
r	Species×P(T×E)	S×P(T×E)	36	—
Split-split-plot:				
f	Crown position	C	1	Error 3
f	Crown×Elevation	C×E	2	Error 3
f	Crown×Species	C×S	1	Error 3
f	Crown×E×S	C×E×S	2	Error 3
r	Split-split-plot error, Error 3: (T×C+T×C×E+T×C×S+T×C×E×S)	T×C×E×S	30	C×S×P(T×E)
r	(C×P(T×E) + C×S×P(T×E))	C×S×P(T×E)	72	—
			215	

Table 4.3. Arithmetic means (±1 S.D.), by mountain, for morphology and anatomy of red spruce and balsam fir foliage collected at three different elevations. Results are for sun needles only. NMA, needle mass to area ratio; NWT, needle width to thickness ratio; VC_{CSA}, vascular cylinder cross-sectional area; M_{CSA}, mesophyll cross-sectional area; VMR, vascular:mesophyll ratio.

	Balsam fir			Red spruce		
	Whiteface	Mansfield	Moosilauke	Whiteface	Mansfield	Moosilauke
Needle morphology						
Needle mass (mg)	4.5±0.9	4.1±1.0	4.3±0.6	2.7±0.6	2.6±0.5	2.8±0.6
Projected needle area (cm^2)	0.192±0.028	0.193±0.034	0.190±0.030	0.082±0.017	0.090±0.014	0.083±0.012
NMA (g/m^2)	236±26	211±28	226±16	330±34	289±22	340±33
Needle tissue density (g/mm^3)	0.54±0.05	0.50±0.06	0.50±0.05	0.54±0.05	0.52±0.05	0.58±0.06
Needle shape						
Needle length (cm)	1.11±0.16	1.20±0.19	1.22±0.20	0.79±0.13	0.88±0.12	0.81±0.12
Needle width (mm)	1.72±0.14	1.61±0.17	1.58±0.17	1.04±0.15	1.03±0.16	1.02±0.15
Needle thickness (mm)	0.61±0.11	0.58±0.10	0.60±0.08	0.96±0.09	0.93±0.10	0.92±0.08
NWT (mm/mm)	2.91±0.44	2.82±0.37	2.64±0.26	1.09±0.11	1.10±0.15	1.10±0.11
Needle anatomy						
Epidermis thickness (μm)	19.6±1.9	16.9±1.9	17.7±1.6	14.9±2.1	13.6±1.3	15.0±1.7
Cuticle thickness (μm)	3.4±0.4	4.5±0.4	3.7±0.3	3.4±0.3	3.8±0.4	3.7±0.4
VC_{CSA} ($\mu m^2 \times 10^3$)	91±17	81±20	79±18	70±19	59±17	64±15
M_{CSA} ($\mu m^2 \times 10^3$)	569±105	519±120	537±96	472±109	425±102	449±104
VMR ($\mu m^2/\mu m^2$)	0.12±0.01	0.12±0.01	0.11±0.02	0.11±0.01	0.10±0.02	0.11±0.01
Branch morphology						
Needle packing density (mg/cm)	80±18	80±21	80±18	45±13	53±17	55±19
Needle packing density (cm^2/cm)	3.64±0.70	4.03±0.81	3.46±0.70	1.67±0.47	2.10±0.58	1.73±0.55

Table 4.4. Results from statistical analysis of morphology and anatomy data for red spruce and balsam fir foliage. Samples were collected from two crown positions (sun vs. shade) and three elevations (low, mid and high) in the mountains of the northeastern United States. NDF and DDF indicate numerator and denominator degrees of freedom, respectively, for *F*-tests. *P*-values determined by mixed model analysis of split-split plot design; those significant at $\alpha = 0.050$ are shown in bold type. All variables were log transformed to improve error term normality and homogeneity of variance. NMA, needle mass to area ratio; NWT, needle width to thickness ratio; VC_{CSA}, vascular cylinder cross-sectional area; M_{CSA}, mesophyll cross-sectional area; VMR, vascular:mesophyll ratio.

	Main Factor Effects			Interaction Effects			
	Elevation (E)	Species (S)	Crown (C)	E×S	E×C	S×C	E×S×C
NDF, DDF	2, 10	1, 15	1, 30	2, 15	2, 30	1, 30	2, 30
Needle morphology							
Needle mass (mg)	0.19	!**0.001**	!**0.001**	0.09	!**0.01**	0.57	0.52
Projected needle area (cm^2)	**0.03**	!**0.001**	!**0.01**	0.21	0.74	!**0.001**	0.34
NMA (g/m^2)	0.28	!**0.001**	!**0.001**	0.08	!**0.01**	!**0.001**	0.37
Needle tissue density (g/mm^3)	0.06	0.05	!**0.001**	0.68	0.44	0.16	0.84
Needle shape							
Needle length (cm)	0.19	!**0.001**	!**0.001**	0.49	0.53	0.58	0.78
Needle width (mm)	0.22	!**0.001**	!**0.001**	0.29	0.07	!**0.001**	0.34
Needle thickness (mm)	0.14	!**0.001**	!**0.001**	**0.03**	**0.02**	!**0.001**	0.31
NWT (mm/mm)	0.79	!**0.001**	!**0.001**	!**0.01**	0.06	!**0.001**	**0.02**
Needle anatomy							
Epidermis thickness (µm)	0.21	!**0.001**	!**0.001**	0.35	**0.02**	0.69	0.42
Cuticle thickness (µm)	0.10	0.36	!**0.001**	0.41	0.84	0.06	0.84
VC_{CSA} ($\mu m^2 \times 10^3$)	0.08	!**0.001**	!**0.001**	0.21	0.06	0.78	0.20
M_{CSA} ($\mu m^2 \times 10^3$)	0.16	!**0.001**	!**0.001**	0.29	0.06	0.20	0.84
VMR ($\mu m^2/\mu m^2$)	0.27	!**0.01**	!**0.001**	0.70	0.97	**0.05**	**0.03**
Branch morphology							
Needle packing density (g/cm)	0.16	!**0.001**	!**0.001**	0.95	**0.02**	**0.03**	0.89
Needle packing density (cm^2/cm)	0.22	!**0.001**	!**0.001**	0.59	**0.03**	0.69	0.94

needles. Needle mass, projected needle area, needle length, needle width, NWT, epidermis thickness, M_{CSA}, VC_{CSA} and VMR were all larger in balsam fir than red spruce. Red spruce needles were thicker, and had higher NMA than balsam fir needles (Figure 4.2). Some of these differences in anatomical structure are visible in the photomicrographs shown in Figure 4.3.

Significant S×C interaction effects suggest that the two species differ in their capacity for a plastic sun/shade response for that trait. The significant S×C interaction effect for projected needle area and NWT (Table 4.4) can be attributed to the fact that either the direction or magnitude of the plastic response differed between species. Projected needle area was larger in sun needles than shade needles for red spruce (P ! 0.001), but was larger in shade needles than sun needles for balsam fir ($P = 0.05$, Figure 4.2). NWT was higher in balsam fir shade needles than sun needles (P ! 0.001), but the reverse was true for red spruce (P ! 0.01, Figure 4.2). Other significant S×C interaction effects (e.g. NMA, needle width, needle thickness, VMR; all P ! 0.05, Table 4.4) were due to the larger relative sun/shade differential of one species compared to the other. Balsam fir was more plastic than red spruce for NMA and needle thickness, but the reverse was true for needle width and VMR.

Only for projected needle area ($P = 0.03$, Table 4.4) was there an overall elevation effect.[2] Projected needle area generally decreased with increasing elevation, although only the difference between L and H was significant (P ! 0.01, Figure 4.2). There were

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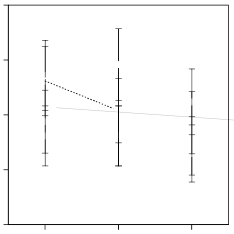
[2] From Figure 4.2 it can be seen that many traits varied somewhat with elevation. For the elevation effect to be significant, however, the overall mean (across all samples) must differ among elevations, and the difference must be significant. Thus, the effect must be reasonably consistent across both species and crown positions. See Appendix for more details on the interpretation of main and interaction effects.



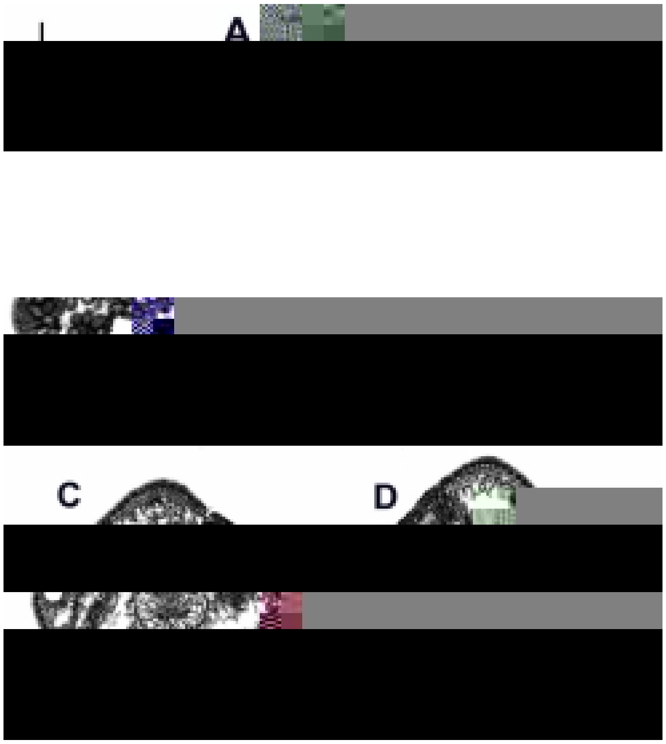


Figure 4.3. Photomicrographs of low-elevation needles of balsam fir and red spruce from two crown positions. Scale bar denotes 500 μm. A) Balsam fir, shade needle; B) balsam fir, sun needle; C) red spruce, shade needle, D) red spruce, sun needle. Note that the width and thickness of needles (A) and (D) are representative of the mean dimensions for those needle types, but (B) is 23% thicker and 11% wider than the “typical” low elevation balsam fir sun needle, whereas (C) is 7% thicker and 12% wider than the “typical” low elevation red spruce shade needle.

significant E×S or E×C interactions for a number of variables (Table 4.4), which indicate that the species response to elevation differed between red spruce and balsam fir (E×S), or that sun/shade plasticity differed along the elevational gradient (E×C). The E×S interaction was significant only for needle thickness ($P = 0.03$) and NWT (P ! 0.01). Spruce needle thickness did not change with elevation, but fir needles became thinner at high elevations (Figure 4.2). Partially as a consequence of changes in needle thickness, NWT increased in balsam fir but decreased slightly in red spruce, between low and high elevation (Figure 4.2). Thus balsam fir needles became flatter at high elevation, whereas red spruce needles became more cylindrical.

The significant E×C interactions for needle mass (P ! 0.01), NMA (P ! 0.01), needle thickness ($P = 0.02$), and epidermis thickness ($P = 0.02$) can all be attributed to the fact that the relative sun/shade difference varied among elevations (Table 4.4, Figure 4.2). In all four cases, there was less of a difference between sun and shade needles at H than at L. In other words, there was a tendency for sun and shade needle anatomy to converge at H.

Shoot morphology

Balsam fir shoots had more leaf tissue (on both a needle dry mass and projected

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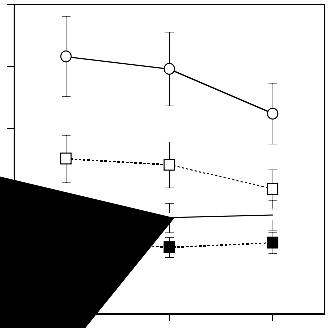

the sun shoots of both species. Thus for both mass- and area-based measures, there was a significant E×C interaction (Table 4.4).

Plasticity analysis

A significant C effect indicates trait differences between sun and shade needles, suggesting that there was sun/shade plasticity for this trait. As mentioned above, S×C interactions can indicate that sun/shade plasticity for a given trait varied between species, whereas E×C interactions indicate that sun/shade plasticity for a given trait varied among elevations.

The mean value of the plasticity index Π differed greatly among traits. Needle packing density was the most plastic trait measured, with $\Pi > 2.00$ for both species. $\mathrm{VC}_{\mathrm{CSA}}$ and $\mathrm{M}_{\mathrm{CSA}}$ were also both highly plastic traits, with $\Pi > 1.50$ for both species. On the other hand, epidermis thickness was not very plastic in either red spruce or balsam fir; both had Π " 1.13 (Figure 4.5). Sun needles had thicker cuticles than shade needles, but the difference was slight: cuticle plasticity was fairly low in both red spruce ($\Pi = 1.09$) and balsam fir ($\Pi = 1.19$).

S×C interactions indicated higher sun/shade plasticity in balsam fir than red spruce for some traits (NMA, needle thickness, NWT, needle packing density), but lower plasticity for other traits (needle area, needle width, VMR) (Figure 4.5). When considered across eight key leaf-level traits (needle mass, needle length, needle thickness, needle width, $\mathrm{VC}_{\mathrm{CSA}}$, $\mathrm{M}_{\mathrm{CSA}}$, epidermis thickness, cuticle thickness)[3], and one measure of shoot

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[3] These are the traits that might be considered fully independent of each other. Needle area is excluded, for example, because it can be calculated as needle length × needle width.

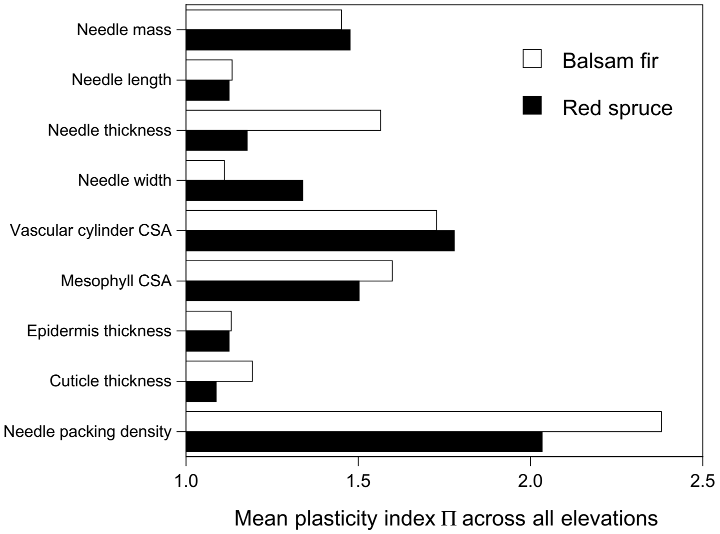


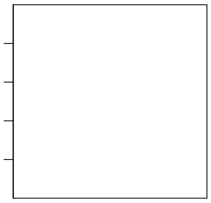
Figure 4.5. Differences in sun/shade plasticity between red spruce and balsam fir for different anatomical and morphological traits. A larger value of the index value Π indicates greater sun/shade plasticity, i.e. a larger difference in the measured trait between sun and shade shoots. VC CSA, vascular cylinder cross-sectional area, measured perpendicular to the main axis of the needle.

morphology (needle packing density), the mean plasticity index $\overline{\Pi}$ was somewhat higher for balsam fir ($\overline{\Pi}$ = 1.48) than red spruce ($\overline{\Pi}$ = 1.40). However, a paired *t*-test (paired by trait) suggested that the difference in plasticity between the two species was not significant ($P = 0.30$).

E×C interactions for needle mass, NMA, needle thickness, epidermis thickness, and needle packing density all suggested reduced sun/shade plasticity at high elevation. Using the same key traits listed in the previous paragraph, paired *t*-tests were again used to test for an overall elevation effect on plasticity. In balsam fir, mean plasticity was highest at M ($\overline{\Pi}_M = 1.54$) and L ($\overline{\Pi}_L = 1.51$), and lowest at H ($\overline{\Pi}_H = 1.38$). The difference between M and L was not significant ($P = 0.20$), but $\overline{\Pi}_H$ was significantly lower than either $\overline{\Pi}_M$ ($P = 0.01$) or $\overline{\Pi}_L$ ($P = 0.05$). Similarly, in red spruce, plasticity at H ($\overline{\Pi}_H = 1.31$) was significantly lower (both P ! 0.05) than that at either M ($\overline{\Pi}_M = 1.46$) or L ($\overline{\Pi}_L = 1.44$). As with balsam fir, the difference between M and L plasticity was not significant for red spruce.

Correlation analysis

Scatter plots of paired variables can be used to identify relationships among variables, and highlight structural differences between species, as the species are readily separated in two-dimensional space by most variable pairs (Figure 4.6). Lee et al. (2000) argued that correlations among different traits suggest, but do not prove, functional relationships. Among the major anatomical traits (needle width, needle thickness, cuticle thickness, epidermis thickness, and VC_{CSA}), there were generally good or excellent linear correlations among all variables within each species, but not between species (Figure



4.6). This suggests that suites of traits vary together in response to environmental stimuli. In all cases, correlation coefficients were positive. A positive correlation between two traits may indicate that the traits are complementary. A negative correlation could indicate that the traits are substitutes for each other. For seven of the 10 different paired combinations of these five traits, correlations were higher in red spruce than balsam fir. For red spruce, correlations were highest for the pairs needle width–VC_{CSA} ($r = 0.93$), needle thickness–VC_{CSA} ($r = 0.86$), and needle thickness–needle width ($r = 0.83$), and lowest for epidermis thickness–cuticle thickness ($r = 0.32$). For balsam fir, correlations were highest for the pairs needle thickness–VC_{CSA} ($r = 0.96$), epidermis thickness–VC_{CSA} ($r = 0.81$), and needle width–VC_{CSA} ($r = 0.75$), and lowest for epidermis thickness-cuticle thickness ($r = 0.22$).

According to the model of Parkhurst and Loucks (1972), which is based on optimal design principles and the balancing of photosynthetic gain with the regulation of leaf temperature and water loss, the ideal leaf size decreases as temperature decreases. This prediction is supported by the literature. High-elevation herbaceous species characteristically have small, thick leaves and high mass to area ratio, and it is hypothesized that low temperatures, and, to a lesser degree, higher light intensities (not a factor here—see Chapter 3), are responsible for these morphological changes (Körner et al. 1989). For conifers, Tranquillini (1979) showed that needle dimensions, and projected needle area, generally decrease with increasing elevation, though thickness may increase. To some extent, the results of the present study are in agreement with these other studies. For example, projected needle area was lower at H than at L for both species, and needle length and width tended to be smaller (though not significantly) at H than L.

Overall, however, elevation-related trends in needle structure were not as obvious (or significant) as expected, given that changes in leaf structure with elevation are thought to play a pivotal role in determining the altitudinal limits of tree growth. For example, Wardle (1971) proposed that at high elevations, newly formed leaf tissues (in particular, the cuticle) might be unable to completely “ripen” during the short growing season, and suggested that the location of treeline was determined by the point at which the failure to ripen resulted in inadequate plant resistance to climatic stress, especially winter frost drought, or *Frosttrocknis*. A similar theory was later advanced by Tranquillini (1979). Many other authors have provided evidence that cuticle thickness is sharply reduced at high elevations, and this may render trees susceptible to high levels of cuticular transpiration (Baig and Tranquillini 1976, 1980, Tranquillini 1979, DeLucia and

Berlyn 1984, Hansen-Bristow 1985, Berlyn et al. 1993). In the present study, cuticle thickness did not differ significantly among elevations, although cuticles at H tended to be thinner than those at L, and there was a general trend towards decreasing thickness with increasing elevation (Figure 4.2). It is hypothesized that the elevation effect was not significant for cuticle thickness because temperature-limited development at M and H was minimized by September 1999 temperatures that were roughly 3° warmer than the 25 year mean (data from the National Climatic Data Center, http://lwf.ncdc.noaa.gov/). In other words, the onset of autumn appears to have been significantly delayed, and cuticles at the highest elevations may have continued to ripen far beyond the date when this process normally ceases. This hypothesis is supported by evidence that the reduction in cuticle thickness with elevation is more pronounced in a cold summer compared to a warm summer (Baig and Tranquillini 1976). However, it should be noted that not all published research supports the idea that cuticles become thinner with increasing elevation. For example, Grace (1990) showed that in the mountains of Scotland, there was no evidence of poor cuticle development in *Pinus sylvestris*. In a variety of species, including conifers, Bonnier (cited in Tranquillini 1979) found that both the epidermis cell wall and cuticle were thicker in mountain plants compared to lowland plants. Stover (1944) compared needle morphology and anatomy of three conifer species across different growth environments and determined that the cuticle was thickest in *krummholz* and xeric habitats.

Sun/shade patterns

The observed differences between sun and shade needles are generally consistent with those that have been previously reported in the literature for other conifers. Sun needles have a higher dry mass, tissue density, and NMA, and are both wider and thicker than shade needles (Richardson et al. 2000, 2001, Sellin 2001). Anatomically, sun needles consistently have a thicker cuticle and epidermis, as well as more vascular tissue and more mesophyll, than shade needles (Richardson et al. 2000, 2001). A key difference between coniferous and broadleaf species with regard to sun/shade dimorphism is that whereas most conifers have larger (in projected area) sun needles than shade needles (with balsam fir a notable exception), sun leaves of broadleaf species are usually smaller than shade leaves (Lichtenthaler 1985). For broadleaf species, this response results in increased boundary-layer conductance and helps to keep sun leaf temperatures closer to air temperatures (Gutschick 1999), thereby improving water use efficiency (Parkhurst and Loucks 1972). In addition to the different direction of response, leaf area plasticity is much lower in conifers (e.g. area $\Pi = 1.17$ in red spruce, 1.04 in balsam fir) compared to that of some broadleaf species. Balaguer et al. (2001) reported that leaf area was among the most plastic trait in response to light in *Quercus coccifera* ($\Pi = 2.7$), and Lichtenthaler (1985) reported $\Pi = 3.7$ in *Fagus sylvatica*. Other factors, such as plasticity in shoot geometry (of which needle packing is just one aspect), may help to explain this apparent divergence among functional groups. Conifers may have low needle-level plasticity because they acclimate to the light environment through a combination of needle-level and shoot-level responses, where as broadleaf species may rely more on leaf-level responses.

Shoot morphology was consistent with that reported for closely related species (e.g.

hybrid *Picea* sp. (*P. engelmannii* × *glauca* × *sitchensis*) and *Tsuga heterophylla* in British Columbia (Richardson et al. 2000, 2001), VC_{CSA}, M_{CSA} and needle mass were found to be the most plastic anatomical traits in response to the canopy light gradient, in agreement with the results here. Overall, the very shade tolerant *T. heterophylla* was less plastic ($\overline{\Pi}$ " 1.2, mean across three age classes) than red spruce or balsam fir, whereas hybrid *Picea* was similarly plastic ($\overline{\Pi}$ " 1.4, mean across three age classes).

Plasticity for needle thickness differed between balsam fir ($\Pi = 1.56$) and red spruce ($\Pi = 1.18$). The pattern for both hybrid *Picea* and *T. heterophylla* varied with stand age, but ranged from 1.05 to 1.23 and from 1.07 to 1.62, respectively. Epidermis thickness was about twice as plastic in hybrid *Picea* as in either species in the present study, but there was almost no plasticity for epidermis thickness in *T. heterophylla*. Cuticle thickness was relatively unplastic in the present study (Π " 1.10-1.20), and in the *Picea* ($\Pi = 1.16$ to 1.24) and *T. heterophylla* ($\Pi = 1.04$ to 1.24) studies, compared to what has been reported for the broadleaf species *Betula papyrifera* (Π " 1.40), *Shorea* spp. (Π " 1.40), or *Quercus* spp. (Π " 2.00) (Ashton and Berlyn 1992, 1994, Ashton et al. 1998). Attributing a functional significance to these differences in plasticity among traits and species is difficult, since these studies have been conducted in entirely different ecosystems. However, plasticity differences may be a manifestation of alternative strategies to achieve a similar end (e.g. the “functional equivalency” of Press 1999; see also Gutschick 1999), or they may represent necessary responses to the environmental challenges of different ecosystems.

Hypothesis A: The capacity for plasticity is correlated with ecological breadth

Ecological breadth is often explained using two competing hypotheses (Sultan 1995, 2000). The first hypothesis (equivalent to *Hypothesis A*) states that a high capacity for phenotypic plasticity confers the ability to tolerate, and remain competitive across, a wide range of environments (the physiological stability of Sultan et al. 1998). This hypothesis therefore predicts that generalists will have high plasticity, whereas specialists will have more limited plasticity. The second hypothesis states that generalist species with wide ecological breadth are comprised of many distinct, locally-adapted populations or ecotypes, each of which is specialized for a different environment (Bradshaw 1965, Fryer and Ledig 1972). Specialist species, on the other hand, are thought to be more limited in their genetic diversity, and this then restricts them to a narrower range of habitats. It is, of course, possible that both phenotypic plasticity and ecotypic differentiation jointly contribute to ecological breadth (Abrams 1994, Sultan 1995, Cordell et al. 1998). Although Sultan (1987) suggested that it is unwise to attempt to infer any direct relationship between genetic diversity, capacity for phenotypic plasticity and ecological breadth, results of numerous studies are consistent with the prediction that generalists are more plastic than specialists (Cook and Johnson 1968, Carpenter and Smith 1981, Ashton and Berlyn 1992, 1994, Cordell et al. 1998, Ashton et al. 1999, but see Greer and McCarthy 1999 for an exception).

The results of the present study were generally consistent with *Hypothesis A*, in that in these high-elevation forests, where the two species coexist, balsam fir was marginally more plastic overall ($\overline{\Pi} = 1.48$) than red spruce ($\overline{\Pi} = 1.40$). However, the difference between species was not significant, and to a large degree this difference was caused by the fact that for one trait, needle packing density, balsam fir was considerably more plastic ($\Pi = 2.38$) than red spruce ($\Pi = 2.03$). With needle packing density excluded, the difference between species was negligible ($\overline{\Pi} = 1.36$ for balsam fir, $\overline{\Pi} = 1.33$ for red spruce). The modest difference in plasticity between balsam fir and red spruce, at least in this growth environment, appears far too small to explain the differences in ecological distribution of the two species (Chapter 2). Similarly, Valladares et al. (2000b) found little difference in plasticity between two *Quercus* species despite large differences in both geographical and ecological ranges.

Although it is not clear whether ecotypic differentiation is more prevalent in balsam fir than red spruce, this may be an explanation for balsam fir's much broader ecological range. Fryer and Ledig (1972) demonstrated that balsam fir had evolved "temperature races" at different elevations on Mt. Moosilauke, while a more recent study suggested that low genetic variability resulted in little physiological differentiation among red spruce provenances (Alexander et al. 1995). Other conifers of somewhat different ecologies seem to have similarly low plasticities (Richardson et al. 2000, 2001), which suggests that low leaf-level plasticity is simply a characteristic trait of these species (all Pineaceae), and may not be related to ecological breadth.

Hypothesis B: Plasticity is reduced in a harsh growth environment

The results from the present study were consistent with *Hypothesis B*: plants growing in a harsh growth environment are less plastic than plants growing in more favorable growth environments. For both red spruce and balsam fir, mean plasticity at H ($\overline{\Pi}_H = 1.38$ in balsam fir, $\overline{\Pi}_H = 1.31$ in red spruce) was significantly lower (P ! 0.05) than at L ($\overline{\Pi}_{\mathrm{L}}$ = 1.51 in balsam fir, $\overline{\Pi}_{\mathrm{L}}$ = 1.44 in red spruce). There have been few studies of plasticity in relation to the elevational gradient, although alpine *Potentilla glandulosa* (Clausen et al., reviewed in Bradshaw 1965) and *Stellaria longipes* (Emery et al. 1994) have both been shown to be morphologically less plastic than their lowland relatives.

In studying the plasticity of species, populations, or genotypes from contrasting environments, comparisons are often drawn between favorable/unfavorable, or resource-rich/resource-poor, growth environments (clearly higher elevation is a less favorable growth environment than lower elevation). The extent that these contrasts can be considered analogues of each other is not entirely clear, but the overall pattern (including studies that focus on plasticity in response to resources other than light) appears to offer further support for *Hypothesis B*. Valladares et al. (2000a) demonstrated that within the tropical rainforest genus *Psychotria*, gap species (more favorable light environment) were more plastic than their understory (less favorable light environment) relatives. Lee et al. (2000) found that *Hopea odorata*, which occupies mesic, shaded river margins (more favorable environment), was more plastic than *Hopea helferi*, which occupies drought-prone slopes (less favorable environment). Bennington and McGraw (1995) demonstrated, using transplant experiments, that floodplain (mesic, more favorable) populations of *Impatiens pallida* were morphologically more plastic than hillside (xeric, less favorable) populations. Crick and Grime (1987), showed high plasticity of root morphology in *Agrostis stolonifera*, a species common to fertile (resource-rich) sites, and low plasticity of root morphology in *Scirpus sylvaticus*, a species adapted to infertile (resource-poor) sites.

Models by Grime (1977, 1986) predict that species grown in stressful environments should be morphologically less plastic because this is a conservative, less risky, trait. Similarly, Chapin et al. (1993) suggested that reduced plasticity across a wide range of traits is part of a “stress response syndrome” that characterizes plants adapted to resource-poor environments. The stress tolerator strategy is thought to involve a tradeoff between reduced competitiveness and increased likelihood of survival. Grime (1977) proposed that in stressful environments, plasticity is not needed because competition is excluded by stress: plasticity is only valuable when competition and disturbance are factors. Thus, I suggest that in the *krummholz* above treeline, there is low sun/shade plasticity because there is little competition for light there. Survival hinges on tolerating the harsh environment, not out-competing neighbors. At lower elevation, survival

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Bennington CC, McGraw JB. 1995. Natural selection and ecotypic differentiation in *Impatiens pallida*. *Ecological Monographs* **65**: 303-323.

Berlyn GP, Anoruo AO, Johnson AH, Vann DR, Strimbeck GR, Boyce RL, Silver WL. 1993. Effects of filtered air and misting treatment on cuticles of red spruce needles on Whiteface Mountain, NY. *Journal of Sustainable Forestry* **1**: 25-47.

Berlyn GP, Ashton PMS. 1998. Forests and the ecosystem paradigm. *Journal of Sustainable Forestry* **7**: 141-157.

Berlyn GP, Miksche JP. 1976. *Botanical microtechnique and cytochemistry*. Ames, IA: Iowa State University Press.

Berlyn GP, Royte JL, Anoruo AO. 1990. Cytophotometric differentiation of high elevation spruces: Physiological and ecological implications. *Stain Technology* **65**: 1-14.

Boardman NK. 1977. Comparative photosynthesis of sun and shade plants. *Annual Review of Plant Physiology* **28**: 355-377.

Bradshaw AD. 1965. Evolutionary significance of phenotypic plasticity in plants. *Advances in Genetics* **13**: 115-154.

Büsgen M, Münch E. 1929. *The structure and life of forest trees*. London: Chapman & Hall.

Carpenter SB, Smith ND. 1981. A comparative study of leaf thickness among southern Appalachian hardwoods. *Canadian Journal of Botany* **59**: 1393-1396.

Chabot BF, Jurik TW, Chabot JF. 1979. Influence of instantaneous and integrated light-flux density on leaf anatomy and photosynthesis. *American Journal of Botany* **66**: 940-945.

Chapin FS, Autumn K, Pugnaire F. 1993. Evolutuion of suites of traits in response to environmental stress. *American Naturalist* **142**: S78-S92.

Clements ES. 1905. The relation of leaf structure to physical factors. *Trans. Amer. Micr. Soc.* **26**: 19-102.

Cogbill CV, White PS. 1991. The latitude-elevation relationship for spruce-fir forest and treeline along the Appalachian mountain chain. *Vegetatio* **94**: 153-175.

Hansen-Bristow K. 1985. Influence of increasing elevation on growth characteristics at timberline. *Canadian Journal of Botany* **64**: 2517-2523.

Hanson HC. 1917. Leaf-structure as related to environment. *American Journal of Botany* **4**: 533-560.

Jackson LWR. 1967. Effect of shade on leaf structure of deciduous tree species. *Ecology* **48**: 498-499.

Körner C, Neumayer M, Menendez-Riedl SP, Smeets-Scheel A. 1989. Functional morphology of mountain plants. *Flora* **182**: 353-383.

Larsen JA. 1927. Relation of leaf structure of conifers to light and moisture. *Ecology* **8**: 371-377.

Lee DW, Oberbauer SF, Johnson P, Krishnapilay B, Mansor M, Mohamad H, Yap SK. 2000. Effects of irradiance and spectral quality on leaf structure and function in seedlings of two Southeast Asian *Hopea* (Dipterocarpaceae) species. *American Journal of Botany* **87**: 447-445.

Lichtenthaler HK. 1985. Differences in morphology and chemical composition of leaves grown at different light intensities and qualities. In: Baker NR, Davies WJ, and Ong CK, eds. *Control of leaf growth*. New York: Cambridge UP, 201-221.

Lichtenthaler HK, Buschmann C, Döll M, Fietz HJ, Bach T, Kozel U, Meier D, Rahmsdorf U. 1981. Photosynthetic activity, chloroplast ultrastructure and leaf characteristics of high-light and low-light plants and of sun and shade leaves. *Photosynthesis Research* **2**: 115-141.

McDougall WB, Penfound WT. 1928. Ecological anatomy of some deciduous forest plants. *Ecology* **9**: 349-353.

Myers O, Bormann FH. 1963. Phenotypic variation in *Abies balsamea* in response to altitudinal and geographic gradients. *Ecology* **44**: 429-436.

Oke TR. 1978. *Boundary layer climates*. London: Routledge.

Oleksyn J, Modrzynski J, Tjoelker MG, Zytkowiak R, Reich PB, Karolewski P. 1998. Growth and physiology of *Picea abies* populations from elevational transects: common garden evidence for altitudinal ecotypes and cold adaptation. *Functional Ecology* **12**: 573-590.

Parkhurst DF, Loucks OL. 1972. Optimal leaf size in relation to environment. *Journal of Ecology* **60**: 505-537.

Press MC. 1999. The functional significance of leaf structure: a search for generalizations. *New Phytologist* **143**: 213-219.

Reich PB, Ellsworth DS, Walters MB, Vose JM, Gresham C, Volin JC, Bowman WD. 1999. Generality of leaf trait relationships: A test across six biomes. *Ecology* **80**: 1955-1969.

Reiners WA, Hollinger DY, Lang GE. 1984. Temperature and evapotranspiration gradients of the White Mountains, New Hampshire, U.S.A. *Arctic and Alpine Research* **16**: 31-36.

Richardson AD, Berlyn GP, Ashton PMS, Thadani R, Cameron IR. 2000. Foliar plasticity of hybrid spruce in relation to crown position and stand age. *Canadian Journal of Botany* **78**: 305-317.

Richardson AD, Ashton PMS, Berlyn GP, McGroddy ME, Cameron IR. 2001. Within-crown foliar plasticity of western hemlock, *Tsuga heterophylla*, in relation to stand age. *Annals of Botany* **88**: 1007-1015.

Schlichting CD. 1986. The evolution of phenotypic plasticity in plants. *Annual Review of Ecology and Systematics* **17**: 667-693.

Sellin A. 2001. Morphological and stomatal responses of Norway spruce foliage to irradiance with a canopy depending on shoot age. *Environmental and Experimental Botany* **45**: 115-131.

Siccama TG. 1974. Vegetation, soil and climate on the Green Mountains of Vermont. *Ecological Monographs* **44**: 325-349.

Smith WK, Knapp AK. 1990. Ecophysiology of high elevation forests. In: Osmond CB, Pitelka LF, and Hidy CM, eds. *Plant biology of the basin and range*. Berlin: Springer-Verlag,

Smith WK, Vogelmann TC, DeLucia EH, Bell DT, Shepherd KA. 1997. Leaf form and photosynthesis. *Bioscience* **47**: 785-793.

Stenberg P, Palmroth S, Bond BJ, Sprugel DG, Smolander H. 2001. Shoot structure and photosynthetic efficiency along the light gradient in a Scots pine canopy. *Tree Physiology* **21**: 805-814.

Stenberg P, Smolander H, Sprugel D, Smolander S. 1998. Shoot structure, light interception, and distribution of nitrogen in an *Abies amabilis* canopy. *Tree Physiology* **18**: 759-767.

Stover EL. 1944. Varying structure of conifer leaves in different habitats. *Botanical Gazette* **106**: 12-25.

Sultan SE. 1987. Evolutionary implications of phenotypic plasticity in plants. *Evolutionary Biology* **21**: 127-178.

Sultan SE. 1995. Phenotypic plasticity and plant adaptation. *Acta Botanica Neerlandica* **44**: 363-383.

Sultan SE. 2000. Phenotypic plasticity for plant development, function and life history. *Trends in Plant Science* **5**: 537-542.

Sultan SE, Wilczek AM, Bell DL, Hand G. 1998. Physiological response to complex environments in annual *Polygonum* species of contrasting ecological breadth. *Oecologia* **115**: 564-578.

Tranquillini W. 1979. *Physiological ecology of the alpine timberline*. New York: Springer-Verlag.

Valladares F, Martinez-Ferri E, Balaguer L, Perez-Corona E, Manrique E. 2000b. Low leaf-level response to light and nutrients in Mediterranean evergreen oaks: a conservative resource-use strategy? *New Phytologist* **148**: 79-91.

Valldares F, Wright SJ, Lasso E, Kitajima K, Pearcy RW. 2000a. Plastic phenotypic response to light of 16 congeneric shrubs from a Panamanian rainforest. *Ecology* **81**: 1925-1936.

van Hinsberg A, van Tienderen P. 1997. Variation in growth form in relation to spectral light quality (red/far-red ratio) in *Plantago lanceolata* L. in sun and shade populations. *Oecologia* **111**: 452-459.

Via S. 1994. The evolution of phenotypic plasticity: What do we really know? In: Real LA, ed. *Ecological Genetics*. Princeton, NJ: Princeton U.P., 35-57.

Wardle P. 1971. An explanation for alpine timberline. *New Zealand Journal of Botany* **9**: 371-402.

West-Eberhard MJ. 1989. Phenotypic plasticity and the origins of diversity. *Annual Review of Ecology and Systematics* **20**: 249-278.

Wylie RB. 1951. Principles of foliar organization shown by sun-shade leaves from ten different species of deciduous dicotyledonous trees. *American Journal of Botany* **36**: 355-361.

Chapter 5:

Foliar chemistry: nutrients and fiber

Abstract

Red spruce (*Picea rubens* Sarg.) and balsam fir (*Abies balsamea* [L.] Mill) are the dominant conifer species at treeline in the mountains of the northeastern United States. The objective of this study was to investigate changes in foliar chemistry of these species along both elevational (below, at, and above treeline) and canopy light (sun vs. shade leaves) gradients.

None of the nutrients showed any significant (all $P > 0.05$) differences among elevations, although mean concentrations of all macronutrients (N, P, K, Ca, Mg) tended to be higher at low elevation sites compared to high elevation. This result contradicts the traditional view that plants in cold growth environments are adapted to maintain high foliar nutrient concentrations, and it gives only weak support for the hypothesis that nutrient limitation plays a role in determining treeline location.

Foliar concentrations of lignin (both sun and shade needles) and cellulose (sun needles only) decreased sharply and significantly with increasing elevation, but foliar concentrations of hemicellulose did not change with elevation. These results are consitent with the hypothesis that as a result of carbon limitation at high elevation, synthesis of the

most expensive fiber constituent (i.e. lignin) is reduced more than that of the least expensive fiber constituent (i.e. hemicellulose). The reduced lignin concentration at high elevation may have implications for nutrient cycling in this ecosystem where cold temperatures limit decomposition rates.

Introduction

From leaf to stand to ecosystem, adequate mineral nutrition is essential for a variety of processes, such as photosynthesis, metabolism, growth and productivity, and decomposition and nutrient cycling (Likens and Bormann 1970, Aber and Melillo 1991, Salisbury and Ross 1992). Understanding the chemical composition of plant foliage is particularly important, as such knowledge can be used to infer nutritional status and identify deficiencies (e.g. van den Driessche 1974) or predict litter decomposition rates (e.g. Melillo et al. 1982).

Understanding the responses of different plant species to environmental gradients is a central focus of physiological ecology. Elevational studies provide us with “natural experiments” from which we can predict possible long-term responses of both populations and individuals to climate change (Körner 1999). Responses to climate change are expected to be largest where ecotones are driven by climate, e.g. at the high-elevation treeline (see Noble 1993 for a review and critique). Although many data have been published on the foliar nutrients of trees growing at low and mid elevation, there are few data available for trees growing at the limits of existence, such as high-elevation *krummholz* (Barrick and Schoettle 1996). The nutritional status of trees at high elevation

In the mountains of the northeastern United States, there is a broad band of coniferous forest, dominated by balsam fir (*Abies balsamea* [L.] Mill.) and red spruce (*Picea rubens* Sarg.), but often featuring a significant *Betula papyrifera* var. *cordifolia* (Regel) Fern. (mountain paper birch) component, between the lowland northern hardwoods (the deciduous beech/sugar maple/yellow birch) and the high-elevation alpine tundra of the summits (Cogbill and White 1991). This spruce-fir zone generally begins around 600-750 m ASL and extends to roughly 1400 m ASL. The primary objective of the present study is to test for elevation-related differences in the foliar chemistry (specifically, macro and micronutrients, trace metals, and fiber content) of red spruce and balsam fir growing below, at, and above the alpine treeline.

Studies of the canopy light gradient provide opportunities for determining an individual’s capacity for phenotypic plasticity (Richardson et al. 2000), and gives insight into the allocation of scarce resources (both nutrients and metabolites) within the canopy (Hollinger 1996). Presumably, individuals that allocate resources more efficiently than others should be more competitive. However, there is little or no consensus in the literature as to whether concentrations should be higher in sun leaves or shade leaves (van den Driessche 1974). The observed patterns are different depending on whether mass-based concentration (g nutrient/g leaf tissue) or area-based content (g nutrient/cm^2 leaf tissue) is considered (e.g. Niinemets 1997). Thus, a second objective of this study is to test whether foliar chemistry differs between sun and shade leaves of red spruce and balsam fir, and to investigate the physiological and ecological implications of these differences. For example, Niinemets (1997) demonstrated that patterns of N partitioning in relation to leaf structure can be directly related to shade tolerance, with shade tolerant species having higher N content in leaves with low LMA (leaf mass to area) ratios, and lower N content in leaves with high LMA, compared to intolerant species.[1]

Methods

The sampling design and sample collection protocol have been described in detail in Chapter 4. Only a brief overview is given here. As in the previous chapters, elevations are abbreviated as L (low), M (mid), and H (high).

Study sites

Study sites were located at different elevations on mountains in three different ranges: Whiteface Mt. (Adirondacks, New York), Mt. Mansfield (Green Mountains, Vermont), and Mt. Moosilauke (White Mountains, New Hampshire). Samples were collected during a single growing season (2000), first from Mt. Moosilauke (early July), then from Whiteface Mt. (late July-early August), and finally from Mt. Mansfield (late

O horizon pH generally ranges from 3.0-4.0, whereas mineral soil pH ranges from 4.0-4.5. The low clay content of these montane soils generally results in CEC being determined by the amount of organic matter present in the solum (Joslin et al. 1992). In

Foliar nitrogen was determined using a Leco CHN 600 combustion analyzer (Leco, St. Joseph, Michigan, USA). For financial reasons, samples were pooled (e.g. red spruce sun needle samples from the three sample trees at each site), reducing the number of analyzed samples to 72 (=216/3). Two replicates were analyzed for each sample. Rye flour standards (Alpha Resources, Stevensville, Michigan, USA) were used to monitor quality control.

Fiber analysis (neutral detergent fiber [NDF], acid detergent fiber [ADF], and 72% sulfuric acid lignin) was conducted using the sequential nylon bag procedure (Goering and Van Soest, 1970) and an Ankom fiber analyzer (Ankom Technology, Fairport, New York, USA) at the USDA's laboratory in Beltsville, MD. Analysis was conducted on the pooled samples, and at a minimum, three replicates were analyzed for each sample. Hemicellulose was calculated as NDF – ADF, and cellulose as ADF – lignin.

Except where explicitly noted, all concentrations are expressed on an oven-dry mass basis. Area-based measures (content) were calculated from concentration data using needle mass and projected area data from Chapter 4. LMA is the leaf (needle) mass to area ratio, expressed in g/m^2.

Statistical analysis

Data were analyzed using a mixed model to properly account for the split-split plot design (further details are given in Chapter 4). Main effects are abbreviated as E (elevation), S (species), and C (crown position). A significance level of $\alpha = 0.05$ was used for all tests. Where necessary, data were log-transformed to improve error term normality and variance homogeneity, and all reported values have been back-transformed.

Results

Element concentrations in relation to elevation, species, and crown position

Averaged across all mountains, concentrations of macronutrients (N, P, K, Ca, Mg) were higher at L than at H. For example, mean K concentrations decreased by 20% between L (3950 mg/kg) and H (3150 mg/kg). However, there were no significant (all P # 0.05) elevation effects in the mixed model analysis (Table 5.1). Concentrations of the micronutrients, Zn and Cu, were similar across all three elevations. There were no significant E×S or E×C interactions for any nutrient (all $P > 0.10$, Table 5.1).

Nutrient content on a needle area basis did not vary significantly among elevations (all $P > 0.05$, data not shown) for any nutrient.

In contrast to this, concentrations of most elements varied significantly with both species and crown position, and, in some cases, the S×C interaction (Table 5.1). For example, N, P, Mg, Ca, Zn, and Cu were significantly higher in balsam fir than red spruce (all P ! 0.05, Figure 5.1). Zn showed the greatest variation between species, with balsam fir concentrations about twice as great as red spruce.

Nutrient investments in sun vs. shade foliage depended on the nutrient in question. Concentrations were higher in sun needles than shade needles for Ca but the reverse was true for N, P, K, and Cu (all P ! 0.05, Table 5.1, Figure 5.1). For Mg and Zn, the crown position effect was not significant (all P # 0.05, Table 5.1).

Table 5.1. *P*

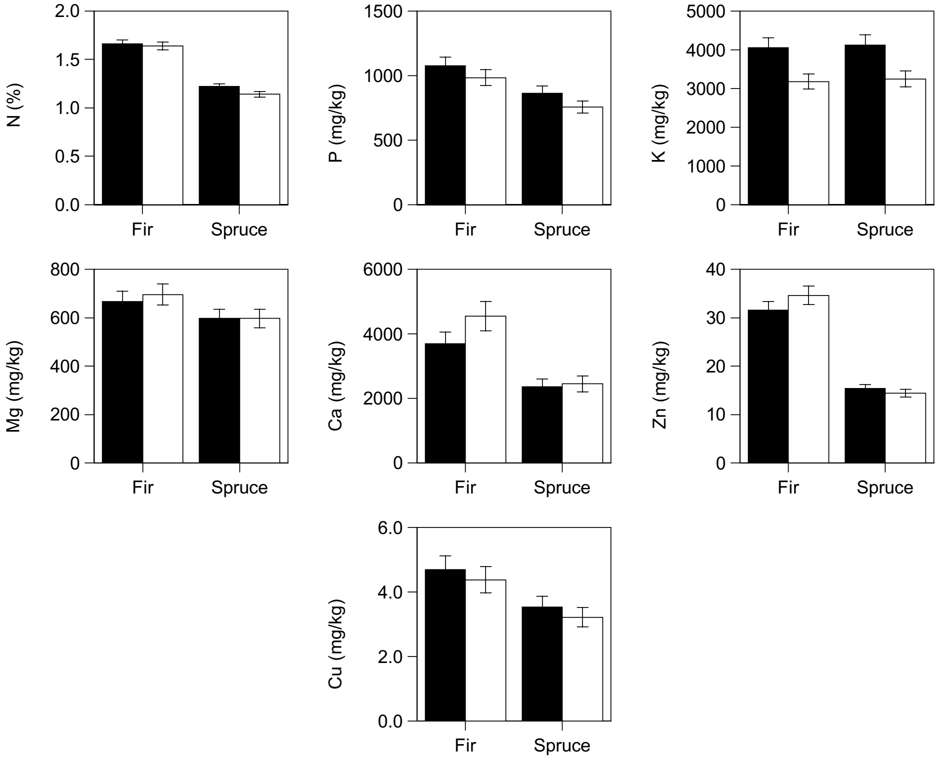


Figure 5.1. Foliar nutrient concentrations (mass basis, mg nutrient/kg needle dry weight, except for N which is % needle dry weight) of red spruce and balsam fir. Black bars are for shade needles, white bars are for sun needles. Error bars indicate ± 1 S.E.

The presence of significant (P ! 0.05) S×C interactions indicates that the sun/shade pattern varied between species for some elements (Table 5.1, Figure 5.1). For N, balsam fir sun and shade concentrations were more or less identical (1.64 and 1.66%, respectively), but they differed by almost 7% in red spruce (1.14 and 1.22%, respectively). For Ca, red spruce sun and shade concentrations were similar (2450 and 2350 mg/kg, respectively), but they differed by 23% in balsam fir (4550 and 3700 mg/kg, respectively). For Zn, balsam fir sun needles had higher concentrations than shade needles but the pattern was reversed in red spruce.

Fiber Content

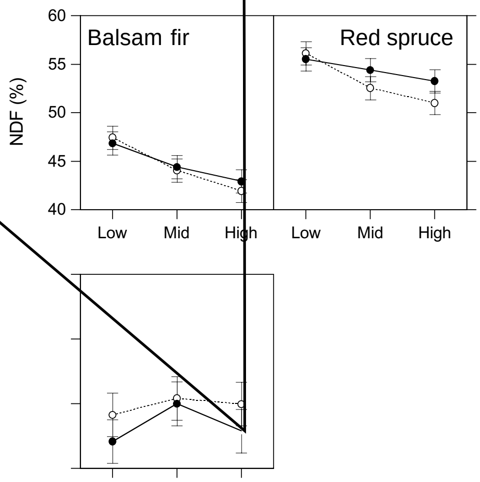
Mean total fiber concentration (= hemicellulose + cellulose + lignin), as measured by NDF, was higher in red spruce (53.8%) than in balsam fir (44.6%, difference significant at P ! 0.001, Table 5.2), and decreased steadily with increasing elevation in both species (Figure 5.2). In both species, shade needle NDF decreased less rapidly with increasing elevation than sun needle NDF, and hence the E×C interaction was significant (Table 5.2).

Hemicellulose did not differ significantly among elevations (P = 0.42, Table 5.2) or crown positions ($P = 0.86$), although the mean hemicellulose concentration of red spruce needles (17.7%, Figure 5.2) was higher (P ! 0.001) than that of balsam fir needles (15.6%).

Both cellulose and lignin decreased steadily from L to H (Figure 5.2). With increasing elevation, the cellulose concentration of shade needles appeared to decrease less slowly than that of sun needles, although the E×C interaction was not significant at

Table 5.2. *P*-values from statistical analysis of needle fiber data for red spruce and balsam fir samples collected along the elevational gradient. Ndf and Ddf indicate numerator and denominator degrees of freedom, respectively, for *F*-tests. *P*-values determined by mixed model analysis of split-split plot design; significant *P*-values (*P* ! 0.05) are shown in bold. NDF and ADF are neutral detergent fiber and acid detergent fiber, respectively; Hemi is hemicellulose.

Factor	**Ndf**	**Ddf**	**NDF**	**ADF**	**Hemi.**	**Cellulose**	**Lignin**
Elevation (E)	2	10	**0.01**	**0.02**	0.42	**0.03**	**0.03**
Species (S)	1	15	!**0.001**	!**0.001**	!**0.001**	!**0.001**	0.39
Crown (C)	1	30	0.87	0.84	0.86	!**0.01**	0.86
E×S	2	15	0.06	0.08	0.92	0.90	0.80
E×C	2	30	**0.04**	0.36	0.29	0.06	0.64
S×C	1	30	0.21	0.83	0.06	0.88	0.74
E×S×C	2	30	0.65	0.92	0.58	0.71	0.77



60
55
50
45
40
NDF (%)
Balsam fir
Red spruce
Low
Mid
High
Low
Mid
High

the $\alpha = 0.05$ level ($P = 0.06$, Table 5.2). In contrast, the elevation pattern for lignin, which decreased from 17.2% at L to 14.3% at H, was virtually identical for sun and shade in both species (Figure 5.2).

Shade needles had higher cellulose concentrations than sun needles (P ! 0.01, Figure 5.2), and cellulose was higher in red spruce (20.4%) than balsam fir (13.8%). Lignin did not differ between species ($P = 0.39$, Table 5.2) or crown positions ($P = 0.86$).

Differences among Mountains

There were some differences in mean element concentrations among the different mountains (e.g. Ca was lowest on Mt. Moosilauke for both species), but in most cases the difference between highest and lowest concentration was not much more than one standard deviation (Table 5.3, data shown for sun needles only). The large variability among samples from the same mountain makes it somewhat difficult to establish clear patterns across mountains. Because samples were collected at different points in the growing season on different mountains, it would be unwise to attribute these differences solely to site effects.

Discussion

Comparison with other nutrient studies

Largely because of the “spruce decline” observed throughout the Appalachians during the 1970s and 1980s, and the possibility that this decline may have been triggered by nutrient deficiencies or toxicities (Friedland et al. 1988, Huntingon et al. 1990, Audley

Table 5.3. Mean (±1 S.D.) concentrations, by species and mountain, for elements and fiber constituents of red spruce and balsam fir foliage collected at three different elevations. NDF is netural detergent fiber, and represents a measure of total fiber content (hemicellulose + cellulose + lignin). Data are for sun foliage only.

a) Balsam fir

	Whiteface Mt.	Mt. Mansfield	Mt. Moosilauke
N (%)	1.56±0.15	1.73±0.13	1.65±0.10
P (mg/kg)	843±97	1136±227	1053±153
K (mg/kg)	2793±660	3196±1006	4112±421
Ca (mg/kg)	5042±983	5599±1381	3878±741

et al. 1998), there has been considerable research into the foliar nutrition of red spruce. Some of these data (along with a number of studies on balsam fir) are listed in Table 5.4.

In agreement with the results from previous studies including both species (Young and Carpenter 1967, Czapowskyj et al. 1980, Siccama and Denny *unpublished data*), concentrations of N, Mg, Ca and Zn were all higher in balsam fir than red spruce. For P and K, inter-specific differences were not consistent among studies.

In this, nutrient concentrations (except N) of both species were generally lower than those reported by others (Table 5.4). This was especially true for P, K, and Zn. While it is acknowledged that differences in digest and analysis procedures makes comparisons between studies somewhat difficult, these results suggest that the trees in the present study lie closer to mineral deficiency than sufficiency. Most nutrient concentrations were below the level at which Swan (1971) found “best growth” to occur in red spruce. Using Swan’s standards for nutrient deficiencies (Table 5.4), the present results suggest moderate N deficiency (red spruce) and acute P deficiency (both species). Concentrations of Zn in red spruce were also deficient (based on Stone 1968, Joslin and Wolfe 1994). Conversely, concentrations of Ca were more than adequate, and concentrations of Mg marginally sufficient (Swan 1971). Nitrogen does not limit primary production in the subalpine environment (Tranquillini 1979), and nitrogen saturation may occur in the northern forests of New England (Aber et al. 1989). Given that N concentrations of both species were equal to, or greater than, those commonly reported (Table 5.4), N deficiency seems unlikely, even though red spruce values are lower than Swan’s (1971) “moderate deficiency” threshold. It should be remembered that Swan’s

Table 5.4. Comparison of results from this study (mean of sun and shade foliage) with

guidelines were developed using seedling experiments, and the requirements of mature trees may be different (MacLean and Robertson 1981, Friedland et al. 1988).

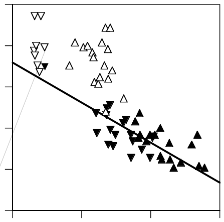
Previous work (Richardson et al. 2001) demonstrated a significant increase in chlorosis (of both red spruce and balsam fir) at and above treeline on Mt. Moosilauke. Since differences in nutrient concentrations with elevation were not significant, then it is hardwood forest at Hubbard Brook, Likens and Bormann (1970) found that foliar chemistry of the major tree species changed very little with elevation, with the exception of Mn, which increased, rather than decreased, with elevation. A more recent analysis from Hubbard Brook (Siccama, *unpublished data*) shows that Ca and Mg decrease with increasing elevation in three northern hardwood species, but P and K do not change much with increasing elevation. Bryant et al. (1997) reported that balsam fir samples collected at 1100 m in the southern Appalachians had lower Ca than samples collected at 980 m. However, in the same study, the opposite pattern was reported for the closely-related *Abies fraseri*. Similarly, neither Huntington et al. (1990) nor Johnson et al. (1994) could detect any clear relationship between elevation and foliar nutrients in the spruce-fir zone.

Table 5.5. Comparison of reported crown position effects on nutrient concentrations (mass basis, g nutrient/g leaf tissue). The present study, Likens and Bormann (1970) and Lichtenthaler (1985) all explicitly compared sun (‘sun’) and shade (‘sh’) leaves, whereas other studies focus on the vertical distribution from the top (‘top’) to the bottom (‘bot’) of the crown, which may or may not correspond to a significant light gradient. Balaguer et al. (2001) data for plants grown in two different light environments, 100% (sun) and 25% (shade) full sunlight. For the review article of van den Driessche (1974), the number of cited examples for each pattern is given in parentheses. Abbreviations: n.s., not significant (at $\alpha = 0.05$); n.r., not reported.

Source	**N**	**P**	**K**	**Ca**	**Mg**
Present study, balsam fir and red spruce	Sh > sun	Sh > sun	Sh > sun	Sh < sun	n.s.
Likens and Bormann 1970,					
Acer saccharum	n.r.	n.r.	Sh " sun	Sh " sun	Sh " sun
Betula alleghaniensis	n.r.	n.r.	Sh < sun	Sh " sun	Sh " sun
Fagus grandifolia	n.r.	n.r.	Sh " sun	Sh " sun	Sh " sun
Lichthenthaler 1985, *Fagus sylvatica*	Sh > sun	Sh " sun	Sh > sun	Sh < sun	Sh > sun
Balaguer et al. 2001, *Quercus coccifera*	Sh > sun	Sh > sun	Sh > sun	n.r.	n.r.
Comerford 1981, *Pinus resinosa*	Bot < top	Bot > top	Bot > top	n.r.	n.r.
MacLean and Robertson 1981, red spruce	Bot < top	Bot < top	n.s.	Bot > top	Bot < top
Myre and Camiré 1996, *Larix decidua*	Bot > top	n.s.	n.s.	n.s.	n.s.
van dan Driessche 1974, Review article	Bot < top (9) Bot > top (1)	Bot < top (7) Bot > top (1)	Bot < top (8) Bot > top (2)	Bot < top (4) Bot > top (4)	Bot < top (6) Bot > top (1)

Driessche (1974) argued for apical control of nutrient distribution within the crown, and suggested that nutrient concentrations in the lower crown would therefore be more sensitive to deficiencies than in the upper crown. For N, optimal allocation theory suggests that the marginal photosynthetic gain per unit of N should be constant throughout the canopy (Field 1983). Simple models predict that N should be allocated in direct proportion to the relative amount of photosynthetically active radiation (PAR) received (Hollinger 1996). This justifies the general result that N concentrations are higher at the top of the canopy (Table 5.5, Hollinger 1996, Stenberg et al. 1998). However, because conifer shade shoots are more efficient at intercepting available PAR than sun shoots (Stenberg et al. 1998, Stenberg et al. 2001), there is potential for this relationship to break down. This may explain the higher N concentrations observed in shade needles compared to sun needles in the present study. Furthermore, additional constraints or costs may limit the flexibility of N allocation, such that the predicted

correlations were found between SLA and area-based measures. Based on these results, mass-based nutrient concentrations should be negatively correlated with LMA, whereas area-based nutrient contents should be positively correlated with LMA. The relationships depend on the degree to which nutrient concentrations become more dilute in higher LMA leaves. In the present study, LMA was negatively correlated with mass-based N (only red spruce significant, *P* ! 0.001), P (both species significant, *P* ! 0.05), and K (both species significant, *P* ! 0.05). LMA was positively correlated with area-based N (both species significant, *P* ! 0.001), P (both species significant *P* ! 0.01) and K (only balsam fir significant, *P* ! 0.01). These relationships are illustrated in Figure 5.3. Note that the 95% confidence intervals for the regression line slopes for the two species overlapped in every case except for area-based N vs. LMA. The steeper slope (*P* ! 0.001) of the area-based N vs. LMA relationship for balsam fir, and the larger intercept (*P* ! 0.01) for red spruce, may have ecological significance. Niinemets (1997) observed that with increasing shade tolerance, the slope of this relationship decreased, and the intercept of this relationship increased. Mass-based N was also generally lower in shade tolerant species. Niinemets (1997) hypothesized that this pattern of nitrogen partitioning both enhances the photosynthetic potential of shade intolerant species in high light (via N allocation to more CO_2-carboxylating enzymes in leaves with high LMA, i.e. sun leaves), and improves light harvesting by shade tolerant species under low light (via N allocation to more chlorophyll in leaves with low LMA, i.e. shade leaves). This then suggests that red spruce and balsam fir differ in shade tolerance. Burns and Honkala (1990) note that balsam fir may be more or less tolerant than red spruce, depending on site conditions and



age. Based on the present results, it is hypothesized that in these montane spruce-fir forests, red spruce is more shade tolerant than balsam fir.

Fiber content

Few data are available for changes in fiber content of leaves along either elevational or light gradients. As a response to a harsh growth environment (e.g. in sun foliage or at high elevation), greater investments might be made in cellulose and lignin. Larsen (1927) observed that shade-enduring conifers, such as *Tsuga heterophylla*, had little or no lignification of the endodermis, whereas light-demanding and drought-tolerant conifers, such as *Pinus contorta*, had a strongly lignified endodermis. Investments in cellulose and lignin should result in stronger, tougher foliage that is more resistant to mechanical damage, and is potentially longer lived (Chabot and Hicks 1982). However, results in the present study showed exactly the opposite—fiber concentrations decrease steadily with increasing elevation. Tranquillini (1979) reported that both the annual bole volume increment and wood quality of *Picea* decrease with increasing elevation: the latter is a consequence of reduced lignification at high elevation sites. Tranquillini hypothesized that, at lower temperatures, photosynthate is preferentially converted to sugars and starch rather than cellulose. This would directly limit the availability of cellulose at high elevation. Alternatively, synthesis of lignin is comparatively expensive in terms of carbon cost (Kozlowski and Pallardy 1997), and thus carbon limitation (due to the shorter growing season and possibly also reduced rates of photosynthesis, Richardson and Berlyn 2002) may further contribute to the reduced concentrations of these compounds at high elevations. It is to be expected that the synthesis of the most expensive fiber constituent, i.e. lignin, would be restricted before that of less expensive fiber constituent, i.e. hemicellulose, as the supply of carbon becomes limiting.

Litter chemistry has important consequences for nutrient cycling, since higher nutrient levels (in particular, N and P) lead to faster rates of decomposition, but decomposition is retarded by lignin and, to a lesser degree, cellulose (Melillo et al. 1982, Taylor et al. 1991, Rutigliano et al. 1996, Kozlowski and Pallardy 1997). The initial lignin:N ratio has been shown to be well correlated with the decomposition rate constant, *k*, in a wide variety of species (Melillo et al. 1982). Since N concentrations were unchanged with elevation, reduced foliar lignin at high elevation leads to smaller lignin:N ratios, which could help to offset the slower decomposition that would naturally accompany cooler temperatures at high elevation.

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References

Aber JD, Melillo JM. 1991. *Terrestrial ecosystems*. Philadelphia: Saunders.

Aber JD, Nadelhoffer KJ, Steudler P, Melillo JM. 1989. Nitrogen saturation in northern forest ecosystems. *BioScience* **39**: 378-386.

Audley DE, Skelly JM, McCormick LH, Jackson WA. 1998. Crown condition and nutrient status of red spruce (*Picea rubens* Sarg.) in West Virginia. *Water, Air and Soil Pollution* **102**: 177-199.

McNulty SG, Aber JD, Newman SD. 1996. Nitrogen saturation in a high elevation New England spruce-fir stand. *Forest Ecology and Management* **84**: 109-121.

Melillo JM, Aber JD, Muratore JF. 1982. Nitrogen and lignin control of hardwood leaf litter decomposition dynamics. *Ecology* **63**: 621-626.

Miller EK, Friedland AJ, Aarons EA, Mohnen VA, Battles JJ, Panek JA, Kadlecek J, Johnson AH. 1993. Atmospheric deposition to forests along an elevational gradient at Whiteface Mountain, NY, U.S.A. *Atmospheric Environment* **27A**: 2121-2136.

Myre R, Camiré C. 1996. The effect of crown position and date of sampling on biomass, nutrient concentrations and contents of needles and shoots in European larch. *Trees* **10**.

Noble IR. 1993. A model of the responses of ecotones to climate change. *Ecological Applications* **3**: 396-403.

Niinemets Ü. 1997. Role of foliar nitrogen in light harvesting and shade tolerance of four temperate deciduous woody species. *Functional Ecology* **11**: 518-531.

Oleksyn J, Reich PB, Zytkowiak R, Karolewski P, Tjoelker MG. 2002. Needle

Salisbury FB, Ross CW. 1992. *Plant Physiology*. Belmont, CA: Wadsworth.

Siccama TG. 1974. Vegetation, soil and climate on the Green Mountains of Vermont. *Ecological Monographs* **44**: 325-349.

Stenberg P, Smolander H, Sprugel D, Smolander S. 1998.

Chapter 6:

Ecophysiological responses

Abstract

Red spruce (*Picea rubens* Sarg.) and balsam fir (*Abies balsamea* [L.] Mill) are the dominant conifer species at treeline in the mountains of the northeastern United States. Physiological methods ranging from “dynamic” (photosynthesis and chlorophyll fluorescence) to “integrated” (spectral reflectance and δ^{13}

are likely due to the greater light-interception efficiency of shade shoots compared to sun shoots.

Chlorophyll fluorescence and several reflectance indices suggested a physiological divergence of sun and shade needles at high elevation. This contrasts with the morphological convergence noted in Chapter 4, and is somewhat surprising given that photosynthetic rates did not show any trends with regard to elevation.

Statistically significant (and, in some cases, previously unreported) correlations were found between $\delta^{13}C$ and other variables. $\delta^{13}C$ was negatively correlated with chl *a* content (as measured by Chl NDI), but positively correlated with area-based N content, epidermis thickness, cuticle thickness, the cross-sectional area of the vascular cylinder, the needle mass-to-area ratio, needle tissue density (significant only for red spruce), and needle thickness. These results confirm the functional significance of variation in leaf structure across different environments.

Introduction

Physiological measurements can be conducted at both different spatial (from molecular to ecosystem) and temporal (from instantaneous or dynamic to highly integrated) scales (cf. Gamon and Qiu 1999). The question of temporal scale is vital, because different factors or processes are importantat different levels of integration. In this chapter, the physiological methods used can be ranked along a temporal continuum, from least integrated and most dynamic to most integrated and least dynamic, as follows: photosynthesis, chlorophyll fluorescence, spectral reflectance, and the stable carbon isotope ratio $\delta^{13}C$.

Along this continuum, there is a tradeoff between the time-scale of integration and the degree to which real physiological processes can be actively quantified. For example, to measure net photosynthesis using an infra-red gas analyzer, the amount of CO estimate of the actual quantum yield. When F_v/F_m is measured on dark-adapted leaves, the result is a more integrated measure of the maximum potential quantum efficiency, which may vary diurnally, seasonally or in response to stress (Bolhàr-Nordenkampf et al. 1989, Krause and Weis 1991, Ball et al. 1994).

Similarly, leaf reflectance has both dynamic and integrated components. Reflectance at visible wavelengths is largely a function of leaf pigmentation, in particular chlorophylls, carotenoids, and anthocyanins (Gamon and Surfus 1999). Under high light conditions, the xanthophyll cycle carotenoids undergo rapid but reversible conversion from violaxanthin to zeaxanthin (Demmig-Adams and Adams 1996). Changes in the epoxidation state of xanthophyll cycle pigments can be detected by subtle reflectance changes at 531 nm (Gamon et al. 1992, Filella et al. 1996). Reflectance can therefore be considered a dynamic method. In comparison, although chlorosis is a common response to stress, foliar chl content is relatively stable over periods of hours or days and reflectance can therefore offer an integrated, rather than dynamic, view of plant function (Carter and Knapp 2001).

This study explores the foliar response of two co-occurring montane conifers, balsam fir (*Abies balsamea* [L] Mill.) and red spruce (*Picea rubens* Sarg.), to environmental gradients. For my model system, I use the canopy light gradient crossed with an elevational gradient centered around treeline. My objective is to use the physiological methods described above to understand how needle function changes along these gradients and to relate observed functional variation to needle and shoot structural properties (see Chapter 4). This direction is motivated by the need to better understand relationships between structure and function, especially at the leaf level (Smith et al. 1997, Gutschick 1999, Press 1999).

Physiological acclimation of a plant's photosynthetic apparatus to prevailing light conditions is one of the best examples of an adaptive phenotypic response to the growth environment (e.g. Boardman 1977, Lichtenthaler et al. 1981, and Lichtenthaler 1985). The functional significance of these responses, many of which occur at the sub-cellular level, is generally enhanced by the accompanying anatomical and morphological responses (Chapter 4 and Smith et al. 1997). Sun foliage is known to be different from shade foliage in terms of photosynthetic properties, such as light compensation and saturation points (Boardman 1977). Sun and shade leaves also differ in terms of photoprotective mechanisms such as chlorophyll fluorescence and the xanthophyll cycle (Lichtenthaler et al. 1981, Demmig-Adams and Adams 1996, Mitchell 1998, Valladares and Pearcy 1999), optical properties such as absorptance (Lee and Graham 1986, St. Jacques et al. 1991) and reflectance (Gausman 1984, Lee and Graham 1986), N allocation to enzymes and pigments (Lichtenthaler et al. 1981), and isotope discrimination (O'Leary 1988).

Plants exhibit a similar range of physiological responses to the elevational gradient. Some evidence suggests that photosynthetic rates of high-elevation plants are impaired (Tranquillini 1979, Wardle 1985, Grace 1989, and Richardson and Berlyn 2002a). However, other have also shown that the photosynthetic efficiency or potential of high-elevation plants may be is intrinsically higher than that of their low-elevation relatives (Körner and Diemer 1994), and there is evidence that this is genetically controlled (Gurevitch 1992, Oleksyn et al. 1998). Elevational trends in F_v/F_m, "green peak" reflectance at 550 nm, and several pigment-based reflectance indices are consistent with a stress response (increased photoprotection and reduced foliar chl content) to high elevation (Richardson et al. 2001, 2003, Richardson and Berlyn 2002a). Although there is considerable variability among studies of isotopic composition in relation to elevation, the mean rate of change in $\delta^{13}C$ appears to be slightly greater than 1.0‰ km^{-1} (Körner et al. 1991, Marshall and Zhang 1994, Hultine and Marshall 2000, Warren et al. 2001). Notably, past studies of physiological responses to elevational gradients have all been conducted exclusively on outer-canopy or sun foliage, which offers little insight to the whole-plant response to environment.

From the above review, it should be clear that the physiological methods I use in this chapter offer a range of different perspectives on the functional response of plants to gradients of light and elevation. In the following chapter, two main questions are investigated:

1) Is the physiological response to the canopy light gradient the same across different elevations?; and
2) Do the different physiological methods offer a consistent interpretation of the physiological responses to these environmental gradients?

Materials and Methods

The study sites, sampling design and sample collection protocol are described in detail in the Chapter 4. In this chapter, only the physiological measurements are described. Reference is made to data presented in the preceding chapters: projected needle area, needle mass, NMA (needle mass to area ratio), needle thickness, and tissue

(1993), Dang et al. (1997), Schaberg et al. (1998), and Nagel and O’Hara (2001) have validated this approach for a number of conifer species, including red spruce. Preliminary investigations confirmed that changes in photosynthesis as a result of branch cutting were minor or nonexistent when the time between cutting and measurement was 10 minutes or less.

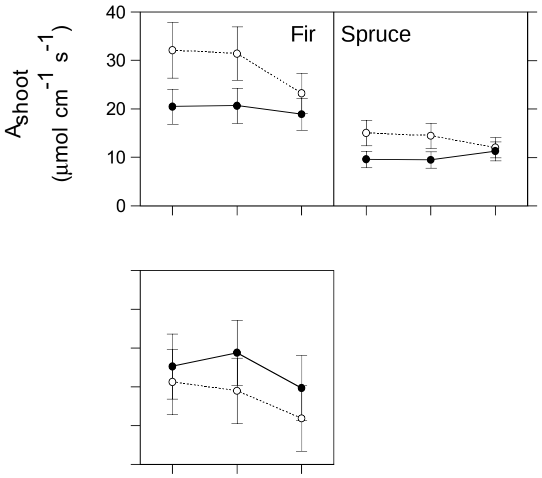
Once in the chamber, shoots were given adequate time to reach equilibrium, which was assessed visually by graphing a strip chart of photosynthesis against time. Measurement times were minimized since a heat-trapping effect within the chamber often

Chlorophyll fluorescence and spectral reflectance

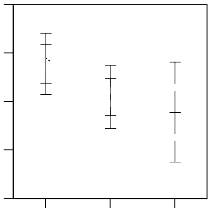
Chlorophyll fluorescence F_v/F_m (variable/maximal fluorescence ratio) is generally measured on dark-adapted leaves because this ensures that photosystem II (PS II) reaction centers are open, and thus the potential efficiency of PS II can be assessed (Bolhàr-Nordenkampf et al. 1989, Krause and Weis 1991, Ball et al. 1994). Additionally, since certain spectral characteristics are known to change rapidly with irradiance (Gamon et al. 1997), reflectance was measured on dark-adapted leaves in order to standardize the measurements across different field sampling days. Both chlorophyll fluorescence and spectral reflectance were measured on dark-adapted foliage in a cool, darkened room at the end of each field day. Samples were analyzed within 12 h of branch cutting. In a standard radiance at wavelength λ). The first-derivative spectrum, D^1_λ, was calculated using difference methods as D^1_λ = (R_n

a flour standard (traceable to IAEA-CH6 reference material) and results are expressed in

Table 6.1


40
30
20
10
0
A$_{shoot}$
(µmol cm^{-1} s^{-1})
Fir
Spruce

construction cost. In neither case was there a significant elevation effect or any elevation-related interactions (all $P > 0.05$, Table 6.1). For both A_{area} and A_{mass}, photosynthetic rates of shade shoots were higher than those of sun shoots (Figure 6.1). Mean A_{area} was virtually identical ("7.2 ± 0.2 μmol m^{-2} s^{-1}, mean ± 1 S.E.) for the two species, but because of differences in NMA (needle mass to area ratio, Chapter 4), mean A_{mass} was

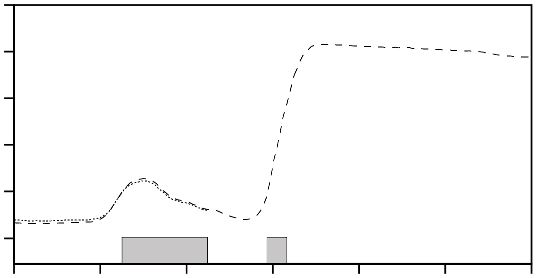


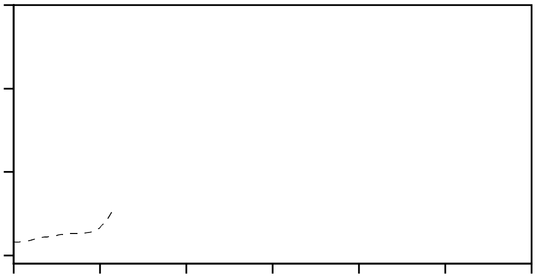
statistically significant, decreases in F_v with increasing elevation that caused the declines in F_v/F_m (Figure 6.2). For both species, the elevation-related decline in F_v was much larger in sun needles than in shade needles (Figure 6.2), which helps to explain the much larger drop in F_v/F_m for sun needles compared to shade needles.

Reflectance spectra

Reflectance spectra differed among elevations, species, and crown positions, but the spectral regions at which these differences occurred depended on the factor in question (Figure 6.3). For each of the main factors E, S, and C, difference spectra (Figure 6.4A) were calculated by subtracting the mean spectra for one grouping under that factor from that for the other grouping, e.g., (mean sun needle reflectance) – (mean shade needle reflectance). Difference spectra were then converted to sensitivity spectra, as suggested by Carter (1991), by dividing through by the mean reflectance across all samples (Figure 6.4B). Difference spectra maxima occur at the wavelengths where the *absolute* difference in reflectance is largest, whereas sensitivity spectra maxima occur where the *relative* difference in reflectance is largest.

Across the entire spectrum from 400–1000 nm, needles from M and H had similar spectra, with slightly higher overall reflectance compared to needles from L (Figure 6.3A). Elevation-related reflectance differences were most pronounced around 550-600 nm, which corresponds to the familiar green-yellow peak of the visible spectrum, and around 700 nm, corresponding to the “red edge” (Figure 6.4A). However, although needles from H and M had higher reflectance above 750 nm than those from L (Figure 6.3A), the elevation effect was not significant (all $P > 0.05$) at these longer wavelengths.





Balsam fir reflectance was consistently higher than red spruce reflectance (Figures 6.3B, 6.4A). Below 700 nm these differences were small (Figure 6.4A), but, with the exception of 568 to 660 nm and 686 to 700 nm, still significant at $P ! 0.05$ (Figure 6.3B). Above 700 nm, the difference between species was much larger: from 750 to 1000 nm, balsam fir needle reflectance was 30% higher than that of red spruce (Figure 6.4B).

Between crown positions, reflectance differences were significant for all but the longest wavelengths (Figure 6.3C). There were two notable reflectance differences between sun and shade needles. First, sun needles had a more pronounced green peak at 550 nm (Figure 6.3C). Second, the difference spectrum reveals an additional feature at 700 nm (Figure 6.4A). Above 738 nm, reflectance differences between sun and shade needles were small and not significant (all $P > 0.05$). Note that although the reflectance difference between sun and shade needles was largest around 550 nm (Figure 6.4A), it was the region around 600 nm where maximum sensitivity to crown position seemed to occur (Figure 6.4B).

The first derivative spectra (Figure 6.5) were somewhat more complex than the untransformed spectra (Figure 6.3), and revealed subtle spectral features that were otherwise masked. As with the untransformed spectra, the first derivative spectra differed among elevations, species, and crown positions. However, the spectral regions at which significant differences occurred were generally narrower, and at different wavelengths, than those for the untransformed spectra. Differences in the first derivative spectra relate to differences in the slopes of the original spectra, but the physiological significance of such shape differences is not yet well understood.

Reflectance indices

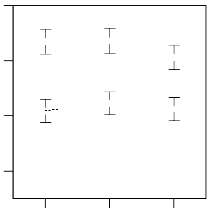
Crown position had the largest effect on Chl NDI (Table 6.1). The chl content of shade needles was significantly higher than that of sun needles. Chl NDI and elevation were negatively correlated, but the significant E×C interaction indicated that the response to elevation was different for the two crown positions. With increasing elevation, sun needle Chl NDI decreased more rapidly than shade needle Chl NDI, and this pattern was clearly expressed in both species (Figure 6.6). The response to elevation was similar for the two species, as indicated by the non-significant E×S interaction.

As with Chl NDI, shade needle PRI was significantly higher than sun needle PRI. However, whereas shade needle PRI did not change with elevation, there was a strong negative correlation between elevation and PRI for sun needles of both species (Figure 6.6), and hence a significant E×C interaction (Table 6.1). Mean PRI of red spruce was significantly lower than that of balsam fir, but, as with Chl NDI, the non-significant E×S and S×C interaction effects (Table 6.1) suggested that the two species responded similarly to these environmental gradients.

Together, these reflectance data indicate only small differences in shade needle pigmentation across elevations. In contrast to this, sun needle pigmentation changed rapidly over the elevational gradient. For both Chl NDI and PRI, the magnitudes of change between M and H and between L and M were similar despite the much smaller elevation difference between M and H ("300 m L–M, "100 m M–H).

Stable carbon isotope ratios

The stable carbon isotope ratio, δ^{13}C, was significantly more negative for shade needles than sun needles, and more negative for balsam fir than that for red spruce (Table 6.1, Figure 6.7). Although the overall elevation effect was not significant ($P = 0.69$, Table 6.1), the significant E×C interaction (P ! 0.01) reflects the fact that the difference between sun and shade needle δ



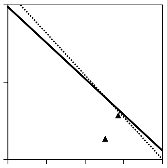


Table 6.2. Pearson correlation coefficients for the linear correlation, *r*, between the stable carbon isotope ratio

Although there were some overall differences between species (i.e. significant S effects in Table 1), the response to environment in these montane forests differed little between species, as demonstrated by the lack of significant S×C or E×S interactions (Table 1). Similarly, in Chapter 4, it was demonstrated that phenotypic plasticity of needle structure in response to crown position was similar for red spruce and balsam fir. This suggests that the species are ecophysiologically similar, despite different ecological and geographical ranges and life histories (Chapter 2). The greater abundance of red spruce and balsam fir (compared to associated species such as mountain paper birch or mountain-ash) in these montane forests suggests that the physiological adaptations of these conifers are truly those that are best suited to, or perhaps required for, the high-elevation environment.

Photosynthesis in relation to crown position

In broadleaf species, light-saturated rates of photosynthesis, when expressed on a leaf area basis, are almost always higher for sun leaves than shade leaves (e.g. Boardman 1977, Lichtenthaler 1981, Lichtenthaler et al. 1985). However, in conifers, there is considerable disagreement as to whether this is true, as different researchers have found contrasting patterns even among trees of the same species. For example, A_{area} of sun needles has been shown to be greater than that of shade needles (e.g. *Pinus contorta* (Schoettle and Smith 1998), *Pinus pondersoa*, *Pseudotsuga menziesii*, and *Tsuga heterophylla* (Bond et al. 1999), *Pseudotsuga menziesii* and *Tsuga heterophylla* (Lewis et al. 2000)). However, other researchers have demonstrated exactly the opposite (e.g. *Taxus brevifolia* (Mitchell 1998), *Abies amabilis* and *Tsuga heterophylla* (Mitchell and Arnott 1995), hybrid *Picea* sp. (Richardson et al. 2000), *Tsuga heterophylla* (Richardson et al. 2001), and *Pinus ponderosa* (Nagel and O'Hara 2001). On a needle mass basis, photosynthetic differences between sun and shade foliage have previously been shown to be negligible (e.g. Bond et al. 1999, Nagel and O'Hara 2001)). In the present study, on both area and mass bases, shade needles photosynthesized more than sun needles.

Conventionally, optimality arguments based on the assumption that natural selection favors plants that can maximize rates of carbon assimilation (e.g. Givnish 1979, Smith et al. 1997) are used to justify the hypothesis that sun leaves should photosynthesize at a greater rate than shade leaves. Given the empirical evidence, one might ask why the pattern for conifers often appears to be different from that for broadleaf species. The observed pattern clearly depends on the basis used to express photosynthetic rates, and more attention needs to be paid to the way in which that basis is selected (Carter and Smith 1985). For broadleaf species, leaf surface area is the most obvious basis, because leaves are flat and leaf area translates directly to the potential for light interception. In contrast, the complex shoot structure of conifers (Stenberg et al. 1998, Stenberg et al. 2001) renders scaling from needle-level to branch-level as challenging as scaling from branch-level to canopy-level (Smith and Knapp 1990). With broadleaf species and modern IRGA technology, it is both sensible and convenient to make leaf-level measurements using individual leaves (with no overlap) arranged perpendicular to the light source. In contrast, for conifers, measurements can be made at either the shoot- or needle-level. Shoot-level measurements preserve the three-dimensional spatial arrangement of needles and may be more realistic than needle-level measurements with needles artificially arranged normal to the light source and with no self-shading (e.g. Lewis et al. 2000). From an ecological perspective, shoot-level measurements are probably more relevant. From a physiological perspective, needle-level measurements could be more telling. However, expressing shoot-level measurements on a projected needle area basis may be deceiving because self-shading and differences in the angle of inclination of different needles results in variation of photosynthesis and conductance among needles on a given shoot (Smith and Knapp 1990). In the present study, results differed when shoot-level measurements were expressed on a shoot basis (sun > shade) or converted to a projected needle area basis (shade > sun). So, although sun shoots photosynthesized more than shade shoots, the difference was smaller than the difference in needle area between sun and shade shoots. The pattern was thus reversed when A_{shoot} was converted to A_{area} (Figure 6.1). An alternative basis may be in terms of shoot silhouette area (SSA), which, compared to projected needle area, is a better measure of the amount of light intercepted.[1] STAR (silhouette to total leaf area ratios) vary between conifer sun and shade shoots (e.g. Carter and Smith 1985). Although I do not have sun and shade shoot STAR data for balsam fir and red spruce, Carter and Smith (1985) provide estimates for the closely related west-coast species, *Abies lasiocarpa* (sun, 0.15 ± 0.04; shade 0.31 ± 0.05) and *Picea engelmanni* (sun, 0.12 ± 0.03; shade, 0.18 ± 0.03). Using these values to get a very rough estimate of photosynthesis on an SSA basis, I calculate that, for both species, sun shoots photosynthesized at a higher rate than shade shoots (80% more in balsam fir; 35% more in red spruce), and red spruce photo-

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[1] Shoot silhouette area is measured on whole shoots, with needles intact in their natural orientation.
Projected needle area is measured with the needles removed from the branch and arranged without oe t7a cted needle f 27 0ed wul(,

synthesized at a higher rate than balsam fir. These results highlight the dilemma, “What is the appropriate basis for expressing photosynthetic rates of conifers?”

Stenberg et al. (2001) used a novel approach to study within-canopy variation in shoot-level photosynthetic efficiency. Efficiency (ε) was defined as (total daily photosynthesis)/(total potential daily light interception). The efficiency term was then itself defined as the product of light-interception efficiency (ε_I, which depends on shoot structure) and photosynthetic conversion efficiency (ε_{PHOT}, which depends on physiological acclimation).[2] Stenberg et al. (2001) found that shade shoots were photosynthetically more efficient (higher ε) than sun shoots due to enhanced ε_I, which was much higher in shade shoots than sun shoots, rather than ε_{PHOT}, which differed little between sun and shade shoots. Although data from the present study do not permit direct calculation of ε_I and ε_{PHOT}, this model illustrates the contribution of shoot-level morphological responses (e.g. needle packing, see Chapter 4) to sun/shade variation in ε_I, whereas needle-level anatomical (e.g. NMA, tissue density, needle thickness, vascular cylinder cross-sectional area, cuticle and epidermis thickness, see Chapter 4) and physiological (e.g. N partitioning and pigmentation, as well as results described in this chapter) responses contribute to variation in ε_{PHOT}. Large enough differences in ε_I between sun foliage and shade foliage can result in shade foliage having a higher overall ε, regardless of differences in ε_{PHOT}.

In some cases, ε_{PHOT} of sun foliage may be lower than that of shade foliage, despite adaptive structural modifications to full sun. In this study, both chlorophyll

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[2] ε_I was defined as (total daily light interception)/(total potential daily light interception). ε_{PHOT} was defined as (total daily photosynthesis)/(total daily light interception).

fluorescence (e.g. F_v/F_m) and reflectance (e.g. PRI) results suggested significantly higher stress in sun foliage compared to shade foliage, which could result in lower ε_{PHOT}. Especially in montane environments, mechanical damage and abiotic stressors are likely to be much more severe in the outer canopy, which (unlike the inner canopy) is exposed directly to the elements (e.g. wind and blowing snow or ice, temperature extremes, and strong vapor pressure deficits). Rather than maximization of carbon gain, it is suggested that for sun needles, stress tolerance is more important than maximizing photosynthetic output. Above all else, leaves need to be "designed" in such a way that water loss and leaf temperature can be efficiently regulated (Parkhurst and Loucks 1972). Needle temperature may be the key variable here. Smith and Knapp (1990) reported that the high density of needles on conifer sun shoots results in a thick boundary layer and elevated leaf temperatures (as much as 8-12°C above ambient temperatures in still air, and 2-4°C above ambient with a wind of 3 m/s). Alexander et al. (1995) found that red spruce photosynthesis is reduced above 20°C. Even small increases in needle temperature could cause this threshold to be exceeded. Elevated needle temperatures result in greater vapor pressure deficits, which can trigger stomatal closure, thereby reducing conductance and leading to diffusion as the most limiting factor for photosynthesis. Note that this scenario is supported by the $\delta^{13}C$ data.

Chlorophyll fluorescence

Following dark adaptation, F_v/F_m of non-stressed leaves is generally in the range photoinhibition and reduced potential quantum yield of photochemistry (Mitchell 1998). A primary cause of photoinhibition is that the D1 reaction center polypeptide, an important component of PS II electron transport, is broken down or destroyed by high light. D1 synthesis is slowed or blocked by a variety of stress factors and therefore replacement of the protein does not always keep up with rate of damage (Long et al. 1994, Taiz and Zeiger 1998). Thus, although both red spruce and balsam fir exhibit some adaptive modifications to shoot morphology (i.e. increased self-shading and vertically-inclined needles, see Chapter 4) along the sun-shade continuum, foliage growing in a full-sun environment may be exposed to more photochemical energy than can be effectively used.

What is especially interesting about the present results is the divergence of sun and shade fluorescence parameters with increasing elevation. Rates of change in F_v and F_v/F_m with increasing elevation were similar for the two species, but much steeper for sun needles than shade needles. While F_v of sun and shade foliage was similar at low elevation, F_v of sun foliage was 10% lower than that of shade foliage at M, and 20% lower at H (Figure 6.2). Photoinhibition of sun needles may be exacerbated at higher elevations by shoot-level changes (i.e. decreasing foliage density of sun shoots at high elevation, which reduces needle self-shading and increases exposure to sun), coupled with increasingly sever abiotic stressors, such as cold temperatures, which inhibit the synthesis of proteins involved in PS II electron transport (e.g. Grace 1989).

In a previous study conducted only on Mt. Moosilauke (Richardson et al. 2001), sun needle F_v/F_m of both red spruce and balsam fir showed a declining trend across a 1000 m elevational gradient from valley floor to treeline, but an increase from treeline to *krummholz*. This may indicate the success of prostrate *krummholz* architecture (which keeps the crown of the "tree" in a far more favorable microclimate) as a stress-avoiding strategy in the harsh alpine environment. However, in the present study, although a similar pattern was once again found on Mt. Moosilauke (this time for both sun and shade needles), it was not seen on either of the other two mountains. Therefore, the treeline inflection point for F_v/F_m does not appear to be a general phenomenon across different mountain ranges.

Reflectance

Characteristically, stress results in increased reflectance at visible, rather than infra-red, wavelengths (Carter 1993). Elevation patterns in overall reflectance correspond to those reported previously for paper birch (Richardson and Berlyn 2002*a*) and two Alaskan spruce species (Richardson et al. 2003). Positive correlations between elevation and both green (around 550-600 nm) and red edge (around 700 nm) reflectance were seen, and are consistent with those noted in response to a variety of stressors, including competition, pathogens, herbicides, and senescence (Carter 1993). The increased reflectance around 700 nm, representing the "blue shift" (i.e. shift to shorter wavelengths) of the red edge transition (clearly visible in the difference spectrum in Figure 6.4A, but also visible to some degree in Figure 6.3) is perhaps the most consistent and characteristic spectral response by plants to stress (Carter and Knapp 2001). This occurs because stress typically results in decreased chl *a* content (Carter 1993). Stress may also have additional spectral signatures, and it is becoming clear that very subtle changes in the *shape* of a number of spectral features may be critical stress indicators. For example,

transmitted or absorbed. Thus, by reflecting less, shade needles have the potential to absorb more radiation. Low reflectance is an appropriate strategy in low light, where it is optimal to maximize light harvesting potential (St. Jacques et al. 1991). In contrast, as a means of photoprotection, sun needles, which experience a light environment where full sun may cause photo-damage, reflect more (and thus likely absorb less) visible radiation; this also helps to minimize the elevation of sun needle temperatures above ambient. Thus it is interesting that in the infra-red (above about 750 nm) little difference in reflectance between sun and shade needles was observed. Presumably this can be attributed to the fact that, at these wavelengths, photons do not have enough energy to power photosynthesis, and consequently high absorptance in this region of the spectrum confers no advantage.

As with the chlorophyll fluorescence data, the reflectance indices Chl NDI and PRI indicate a physiological divergence between sun and shade needles at higher elevations. For shade needles, neither index changed appreciably from L to H, whereas for sun needles there was a clear declining trend in both indices from L to H (Figure 6.6). For both species, index values were higher for shade needles than sun needles. Chl NDI has been shown to be almost linearly related to total chl content (Richardson et al. 2002). Shade needles thus have higher chl contents, and because of the lower NMA relative to sun needles, also have higher chl concentrations than sun needles. Foliar N concentrations (%N) were similar in sun and shade needles for both balsam fir and red spruce, but area-based N content (N_{area}) was higher in sun needles compared to shade needles for both species (Chapter 5). Thus, the ratio of chl to total N was much lower in sun needles than shade needles. In sun needles, more N must therefore be allocated to

different species, functional groups, and seasons (e.g. Gamon et al. 1997, Stylinski et al. 2002). Furthermore, a similarly robust positive correlation has been demonstrated between PRI and the chl:carotenoid ratio (Sims and Gamon 2002). In the present study, PRI was positively correlated with F_v/F_m in both species ($r = 0.59$, P ! 0.001; data not shown), and the elevation- and crown-position patterns of PRI (Figure 6.6) were similar to those for F_v/F_m (Figure 6.2). Taking PRI to be an indicator of the chl:carotenoid ratio, the present results suggest that sun leaves have, on a relative basis, a greater investment in photoprotective carotenoids than shade leaves, which is in agreement with the general pattern (Moran et al. 2000, Poorter et al. 2000). The results also suggest that the sun needle investment in carotenoids increases with increasing elevation, which might be expected as a stress response.

Carbon isotope ratio

In C3 plant tissue, the stable carbon isotope ratio, $\delta^{13}C$ (generally about -28‰), differs from that of the surrounding air (about -8‰ in well-mixed air) because $^{13}CO_2$ diffuses more slowly than $^{14}CO_2$, and because Rubisco further discriminates against $^{13}CO_2$ during carboxylation (Ehleringer 1989). The isotope ratio $\delta^{13}C$ therefore represents the balance between supply (via stomatal conductance) and demand (via photosynthesis) for CO_2 within the leaf (Hultine and Marshall 2000). $\delta^{13}C$ approaches a limit of -36‰ if stomatal diffusion is rapid and carboxylation is limiting (intercellular CO_2 concentration $\Rightarrow$ CO_2 concentration of surrounding air). On the other hand, when diffusion is slow, $\delta^{13}C$ reaches a limit of -12‰ (intercellular CO_2 concentration $\Rightarrow 0$) (O'Leary 1988). Because δ^{13}

indicator of water use efficiency (WUE). Less negative $\delta^{13}C$ implies greater WUE (O'Leary 1988, Ehleringer 1989). On this basis, it is suggested that red spruce has a slightly greater intrinsic WUE than balsam fir.

Past studies have shown that as irradiance increases, $\delta^{13}C$ becomes less negative (O'Leary 1988). Differences in $\delta^{13}C$ between sun and shade leaves can be related to a number of factors, including leaf structure, N partitioning, hydraulic limitation and source composition. For example, results from Chapter 4 indicate that sun needles are thicker and have higher NMA than shade needles, which should result in photosynthesis being limited more by diffusion in sun needles than shade needles. Furthermore, the nutrient and reflectance results described above appear to indicate that shade needles have relatively more N invested in light harvesting pigments, and less in Rubisco, than sun needles. This should result in photosynthesis being limited more by carboxylation in shade needles than sun needles. Additionally, because sun needles are exposed to higher irradiances, they generally have higher leaf-to-air vapor pressure deficits, and their upper canopy position probably results in greater hydraulic limitation (in spite of increased vascular tissue) due to the difficulties inherent in transporting water up a tall trunk (Kozlowski et al. 1991). Finally, Sternberg et al. (1989) have also demonstrated that it is also necessary to consider the isotopic composition of the surrounding air when interpreting foliar $\delta^{13}C$ values. Sternberg et al. (1989) showed that understory or lower canopy leaves generally have more negative $\delta^{13}C$ ratios because air near the ground has more negative $\delta^{13}C$ than the well-mixed air at the top of the canopy. This is a direct result of the fact that CO_{2stive}

decomposing substrate. These factors should all contribute to shade needles having more negative $\delta^{13}C$ values than sun needles, which is what was observed in the present study.

The observation that $\delta^{13}C$ is generally positively correlated with elevation can be attributed not only to changes in the partial pressures of CO_2 and O_2 with elevation (due to decreasing atmospheric pressure), but also to elevation-related changes in foliar anatomy, morphology and physiology that are triggered by temperature (Körner et al. 1991, Hultine and Marshall 2000). For example, high elevation plants generally have thicker leaves with higher mass to area ratios and greater carboxylation efficiency; higher foliar N_{area} at high elevation could also increase photosynthetic capacity and contribute to a lower intercellular/ambient CO_2 ratio (Körner et al. 1991, Körner and Diemer 1994). In the present study, there was no overall elevation trend in $\delta^{13}C$ (Table 6.1, Figure 6.7), and this may be related to the lack of significant trends in leaf thickness or N_{area}.[4] However, the E×C interaction effect (Table 6.1) indicated that the response to elevation differed between sun and shade needles. By taking the difference in $\delta^{13}C$ between sun and shade samples of each species at each site, any confounding variation among sites is effectively controlled for. For both red spruce and balsam fir, the difference in $\delta^{13}C$ between sun and shade foliage converged towards zero with increasing elevation. This may be related to the fact that that tree height is reduced at high elevation (Chapter 4), which could result in less hydraulic limitation of sun needles at H compared to L. The anatomical and

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[4] A contributing factor may be to the small difference in elevation between L and H (" 400 m),: based on results from the literature (e.g. Körner et al. 1991), $\delta^{13}C$ of L and H would be expected to differ by less than 0.5‰. O'Leary (1988) suggested not only that differences of this magnitude can be hard to detect, but also that differences of less than 1.0‰ should be interpreted with caution, as they may not represent truly significant differences. Within each species-crown position class, differences among elevations were consistently less than 0.5‰, although for red spruce sun needles the direction of change in $\delta^{13}C$ from L to H was opposite to that which would be expected ($\delta^{13}C$ became more negative, from -25.5‰ at L to -26.0‰ at H).

morphological convergence of sun and shade needles at H (Chapter 4) may also play a role in the $\delta^{13}C$ E×C pattern.

Correlations of $\delta^{13}C$ with anatomical, morphological, and other physiological variables can be used to infer relationships between structure and function (Figure 6.8). Although the results of Körner et al. (1991) suggest a positive correlation between $\delta^{13}C$ and palisade layer thickness, neither balsam fir nor red spruce have clearly-defined palisade tissue, so it is not possible to make an exact comparison. However, epidermis thickness, cuticle thickness, and vascular cylinder cross-sectional area were all positively correlated with $\delta^{13}C$ in both species. It is difficult to say whether these are functionally significant relationships indicative of cause-and-effect, or arise as an indirect consequence of the simultaneous variation in needle anatomy and $\delta^{13}C$ between sun and shade needles. For example, other things being equal, it would be expected that an increase in vasculature might result in a lower $\delta^{13}C$, since the increased supply of water (enabling greater transpirational losses before desiccation) would permit increased stomatal conductance. Increased stomatal conductance would result in a more negative $\delta^{13}C$ because diffusion would be less limiting to photosynthesis. However, the evaporative demands on the leaf must also be taken into account.

N

Hultine and Marshall (2000) found that N_{area} was correlated significantly with $\delta^{13}C$ for only one (*Pinus contorta*) of the four conifer species they studied in the north-central Rocky Mountains. As in the present study, Hultine and Marshall (2000) reported that %N was not correlated with $\delta^{13}C$ within any species, although across species Körner et al. (1991) found a positive and significant correlation between these variables. In the present study, Chl NDI was negatively correlated with $\delta^{13}C$, and this relationship was similar between red spruce and balsam fir. This pattern is likely related to N partitioning within the needle. Compared to shade needles, sun needles had high N_{area} but low Chl NDI, suggesting a relatively greater investment of N in carboxylating enzymes.

Körner et al. (1991) found that LMA (leaf [needle] mass to area ratio) correlated positively with $\delta^{13}C$ across species. There are a number of reasons that $\delta^{13}C$ should be correlated positively with LMA, and its components, density and thickness (LMA = density × thickness). Thicker leaves can be expected to have more photosynthetic enzymes and thus a greater demand for CO_2, thicker leaves should have longer diffusion pathways for CO_2, and diffusion through leaves with dense mesophyll tissues should be slower than through leaves with greater intercellular space (Valladares and Pearcy 1999, Hultine and Marshall 2000). Results presented by Hultine and Marshall show not only the expected correlation ($\delta^{13}C$ correlates with NMA), but also that this relationship may hold generally across species. In addition, the slope of the relationship reported by Hultine and Marshall was almost identical to that for both red spruce and balsam fir in the present study. The $\delta^{13}C$–NMA relationship illustrates, perhaps more clearly than any of the other correlations, the main thesis of Smith et al. (1997): that variation in leaf structure has functional significance. Although these sorts of relationships have been assumed for decades (e.g. Hanson 1917, Larsen 1927), it has only recently become possible to quantify the physiological effects.

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References

Alexander JD, Donnelly JR, Shane JB. 1995. Photosynthetic and transpirational responses of red spruce understory trees to light and temperature. *Tree Physiology* **15**: 393-398.

Ball MC, Butterworth JA, Roden JS, Christian R, Egerton JJG. 1994. Applications of chlorophyll fluorescence to forest ecology. *Australian Journal of Plant Physiology* **22**: 311-319.

Boardman NK. 1977. Comparative photosynthesis of sun and shade plants. *Annual Review of Plant Physiology* **28**: 355-377.

Bolhàr-Nordenkampf HR, Long SP, Baker NR, Öquist G, Schreiber U, Lechner EG. 1989. Chlorophyll fluorescence as a probe of the photosynthetic competence of leaves in the field: A review of current instrumentation. *Functional Ecology* **3**

Ehleringer JR. 1989. Carbon isotope ratios and physiological processes in aridland plants. In: Rundel PW, Ehleringer JR, and Nagy KA, eds. *Stable isotopes in ecological research*. New York: Springer-Verlag,

Filella I, Amaro T, Araus JL, Peñuelas J. 1996. Relationship between photosynthetic radiation-use efficiency of barley canopies and the photochemical reflectance index (PRI). *Physiologia Plantarum* **96**: 211-216.

Gamon JA, Peñuelas J, Field CB. 1992. A narrow-waveband spectral index that tracks diurnal changes in photosynthetic efficiency. *Remote Sensing of Environment* **41**: 35-44.

Gamon JA, Surfus JS. 1999. Assessing leaf pigment content and activity with a reflectometer. *New Phytologist* **143**: 105-117.

Gamon JA, Qiu H. 1999. Ecological applications of remote sensing at multiple scales. In: Pugnaire FI and Valladares F, eds. *Handbook of functional plant ecology*. New York: Marcel Dekker, 805-846.

Gamon JA, Serrano L, Surfus JS. 1997. The photochemical reflectance index: An optical indicator of photosynthetic radiation use efficiency across species, functional types, and nutrient levels. *Oecologia* **112**: 492-501.

Gausman HW. 1984. Evaluation of factors causing reflectance differences between sun and shade leaves. *Remote Sensing of Environment* **15**: 177-181.

Gitelson A, Merzlyak MN. 1994. Spectral reflectance changes associated with autumn senescence of *Aesculus hippocastanum* L. and *Acer platanoides* L. leaves: Spectral features and relation to chlorophyll estimation. *Journal of Plant Physiology* **143**: 286-292.

Givnish TJ. 1979. On the adaptive significance of leaf form. In: Solbrig OT, Jain S, Johnson GB, and Raven PH, eds. *Topics in plant population biology*. New York: Columbia U.P., 375-407.

Givnish TJ. 1988. Adaptation to sun and shade: a whole plant perspective. *Australian Journal of Plant Physiology* **15**: 63-92.

Grace J. 1989. Tree lines. *Phil. Trans. R. Soc. Lond. B* **324**: 233-245.

Grassi G, Bagnaresi U. 2001. Foliar morphological and physiological plasticity in *Picea abies* and *Abies alba* saplings along a natural light gradient. *Tree Physiology* **21**: 959-967.

Gurevitch J. 1992. Differences in photosynthetic rate in populations of *Achillea lanulosa* from two altitudes. *Functional Ecology* **1992**: 568-574.

Gutschick VP. 1999. Biotic and abiotic consequences of differences in leaf structure. *New Phytologist* **143**: 3-18.

Hanson HC. 1917. Leaf-structure as related to environment. *American Journal of Botany* **4**: 533-560.

Hultine KR, Marshall JD. 2000. Altitude trends in conifer leaf morphology and stable carbon isotope composition. *Oecologia* **123**: 32-40.

Jones HG. 1992. *Plants and microclimate: a quantitative approach to environmental plant physiology*. Cambridge: Cambridge UP.

Körner C. 1999. *Alpine plant life*. Berlin: Springer-Verlag.

Körner C, Diemer M. 1994. Evidence that plants from high altitudes retain their greater photosynthetic efficiency under elevated CO_2. *Functional Ecology* **8**: 58-68.

Körner C, Farquhar GD, Wong SC. 1991. Carbon isotope discrimination by plants follows latitudinal and altitudinal trends. *Oecologia* **88**: 30-40.

Kozlowski TT, Kramer PJ, Pallardy SG. 1991. *The physiological ecology of woody plants*. San Diego: Academic Press.

Krause GH, Weis E. 1991. Chlorophyll fluorescence and photosynthesis: the basics. *Annual Review of Plant Physiology and Plant Molecular Biology* **42**: 313-349.

Larsen JA. 1927. Relation of leaf structure of conifers to light and moisture. *Ecology* **8**: 371-377.

Lee DW, Graham R. 1986. Leaf optical properties of rainforest sun and extreme shade plants. *American Journal of Botany* **73**: 1100-1108.

Lewis JD, McKane RB, Tingey DT, Beedlow PA. 2000. Vertical gradients in

Marshall JD, Zhang J. 1994. Carbon isotope discrimination and water-use efficiency in native plants of the north-central Rockies. *Ecology* **75**: 1887-1895.

Meng F-R, Arp PA. 1993. Net photosynthesis and stomatal conductance of red spruce twigs before and after twig detachment. *Canadian Journal of Forest Research* **23**: 716-721.

Mitchell AK. 1998. Acclimation of Pacific yew (*Taxus brevifolia*) foliage to sun and shade. *Tree Physiology* **18**: 749-757.

Mitchell AK, Arnott JT. 1995. Effects of shade on the morphology and physiology of amabilis fir and western hemlock seedlings. *New Forests* **10**: 79-98.

Moran JA, Mitchell AK, Goodmanson G, Stockburger KA. 2000. Differentiation among effects of nitrogen fertilization treatments on conifer seedlings by foliar

Richardson AD, Berlyn GP. 2002b. Changes in foliar spectral reflectance and chlorophyll fluorescence of four temperate species following branch cutting. *Tree Physiology* **22**: 499-506.

Richardson AD, Berlyn GP, Duigan SP. 2003. Reflectance of Alaskan black spruce and white spruce foliage in relation to elevation and latitude. *Tree Physiology* **In press**.

Richardson AD, Berlyn GP, Gregoire TG. 2001. Spectral reflectance of *Picea rubens* (Pinaceae) and *Abies balsamea* (Pinaceae) needles along an elevational gradient, Mt. Moosilauke, New Hampshire, USA. *American Journal of Botany* **88**: 667-676.

Richardson AD, Duigan SP, Berlyn GP. 2002. An evaluation of noninvasive methods to estimate foliar chlorophyll content. *New Phytologist* **153**: 185-194.

Schaberg PG, Shane JB, Cali PF, Donnelly JR, Strimbeck GR. 1998. Photosynthetic capacity of red spruce during winter. *Tree Physiology* **18**: 271-276.

Schoettle AW, Smith WK. 1998. Interrelationships among light, photosynthesis and nitrogen in the crown of mature *Pinus contorta* ssp. *latifolia*. *Tree Physiology* **19**: 13-22.

Sims DA, Gamon JA. 2002. Relationships between leaf pigment content and spectral reflectance across a wide range of species, leaf structures and developmental stages. *Remote Sensing of Environment* **8**: 337-354.

Smith WK, Knapp AK. 1990. Ecophysiology of high elevation forests. In: Osmond CB, Pitelka LF, and Hidy CM, eds. *Plant biology of the basin and range*. Berlin: Springer-Verlag,

Smith WK, Vogelmann TC, DeLucia EH, Bell DT, Shepherd KA. 1997. Leaf form and photosynthesis. *BioScience* **47**: 785-793.

St. Jacques C, Labrecque M, Bellefleur P. 1991. Plasticity of leaf absorbance in some broadleaf tree seedlings. *Botanical Gazette* **152**: 195-202.

Stenberg P, Smolander H, Sprugel D, Smolander S. 1998. Shoot structure, light interception, and distribution of nitrogen in an *Abies amabilis* canopy. *Tree Physiology* **18**: 759-767.

Stenberg P, Palmroth S, Bond BJ, Sprugel DG, Smolander H. 2001. Shoot structure and photosynthetic efficiency along the light gradient in a Scots pine canopy. *Tree Physiology* **21**: 805-814.

Sternberg LdSL, Mulkey SS, Wright SJ. 1989. Ecological interpretation of leaf carbon isotope ratios: Influence of respired carbon dioxide. *Ecology* **70**: 1317-1324.

Stylinski CD, Gamon JA, Oechel WC. 2002. Seasonal patterns of reflectance indices, carotenoid pigments and photosynthesis of evergreen chaparral species. *Oecologia* **131**: 366-374.

Taiz L, Zeiger E. 1998. *Plant physiology, 2nd ed.* Sunderland, MA: Sinauer.

Tranquillini W. 1979. *Physiological ecology of the alpine timberline*. New York: Springer-Verlag.

Valladares F, Pearcy RW. 1999. The geometry of light interception by shoots of *Heteromeles arbutifolia*: morphological and physiological consequences for individual leaves. *Oecologia* **121**: 171-182.

Wardle P. 1985. New Zealand timberlines. 3. A synthesis. *New Zealand Journal of Botany* **23**: 263-271.

Warren CR, McGrath JF, Adams MA. 2001. Water availability and carbon isotope discrimination in conifers. *Oecologia* **127**: 476-486.

Chapter 7:

Summary and conclusions

This dissertation has focused on the response to growth environment of the foliage of two conifer species, red spruce and balsam fir. As model systems, I have used the canopy light gradient and the elevational gradient. My main question can be phrased in two different ways: either as “Does the response to the canopy light gradient change along the elevation gradient?”, or “Is the response to the elevation gradient the same for sun and shade foliage?” Research was conducted in montane forests where these two species coexist: on Whiteface Mt., in the Adirondacks of New York, Mt. Mansfield, in the Green Mountains of Vermont, and Mt. Moosilauke, in the White Mountains of New Hampshire.

In Chapter 2, I gave an overview of the ecology and biology of these species, which dominate (along with *Betula papyrifera* var. *cordifolia* (Regel) Fern., mountain paper birch) the high-elevation forests of the northeastern United States. Red spruce clearly has a more restricted range—both geographical and ecological—than balsam fir, although both species are of a similar shade tolerance and both species can grow right up to the high-elevation treeline. However, red spruce is much more slow growing, and much more long-lived, than balsam fir. This difference in life history is thought to contribute to the ability of these two species to coexist.

In Chapter 3, I presented results from meteorological studies along the elevational gradient on each mountain. Both mean annual temperatures and air temperature lapse rates showed a modest east-west trend. Mean annual temperatures were strongly correlated with site elevation. Air temperature at treeline on Mt. Mansfield (13.7°C, July-September mean) was slightly warmer than on the other two mountains, but this was largely due to its lower elevation. Lapse rates averaged about –0.6°C/100 m, but there was considerable variation, related mostly to diurnal patterns, in this rate. In contrast to previous studies, soil temperature did not vary as consistently with elevation as air temperature. It has previously been hypothesized that the light environment at high as plasticity at high elevation was significantly lower than that at low elevation, for both species.

In Chapter 5, I compared the foliar chemistry of red spruce and balsam fir. Macronutrient concentrations tended to be lower at high elevation than at low elevation, but the differences among elevations were not significant. This suggests that nutrient limitation does not play a major role as a stress factor at high elevation. However, trends in fiber concentrations suggested that carbon limitation may be a factor at or above treeline.

In Chapter 6, I showed that the physiological response to elevation and crown position was generally quite similar for the two species. This is taken as an indication that in terms of ecophysiology, red spruce and balsam fir are surprisingly alike. The different physiological measurements revealed different patterns with regard to elevation and crown position. For example, although photosynthetic rates did not vary among elevations, A_{shoot} (photosynthesis expressed on a unit shoot length basis) decreased with increasing elevation in sun shoots but not shade shoots. Similarly, the stable carbon isotope ratio, $\delta^{13}C$, of sun and shade needles tended to converge at the highest elevation site. In contrast, fluorescence and reflectance indices suggested a physiological divergence of sun and shade foliage with increasing elevation: sun needles became progressively more stressed, whereas the health of shade needles was unchanged, along the elevational gradient. Correlations between δ^{13}

published studies, and it was suggested that some (in particular, $\delta^{13}C$–needle mass to area ratio) may hold generally across species.

There are four important messages from this research:

1) Niche breadth does not appear to be correlated with the capacity for sun/shade plasticity. This suggests the hypothesis that it is genetic diversity that enables balsam fir to tolerate a wide range of sites, whereas red spruce, which is thought to have very limited genetic diversity, is thereby confined to a much more restricted ecological range.

2) Plasticity is decreased in a harsh growth environment. These results do not give any indication whether this is a phenotypic or genotypic response, but if it is a genotypic response, then this could constrain the ability of high-elevation trees to respond to climate change.

3) Red spruce and balsam fir share similar ecophysiologies. This supports the idea that, for these two species, niche differentiation occurs along a temporal or life-history scale, rather than in terms of resource gradients. At high elevation, balsam fir and red spruce coexist because they can both tolerate the extreme environment, and because of their differing patterns of establishment, growth, senescence and regeneration.

4) When studying the foliar response to environmental gradients, focusing exclusively on sun or shade leaves may give a biased view of the whole-plant response to that gradient. Both the morphology and physiology results clearly demonstrated that the response to elevation is different for these two extreme crown positions.

These results can be put in a more general context by considering that forests in which spruce or fir (either individually or together) are important components are common across virtually the entire temperate zone of the Northern Hemisphere (Liu

References

Archibald OW. 1995. *Ecology of world vegetation*. London: Chapman & Hall.

Arno SF. 1984. *Timberline: Mountain and Arctic forest frontiers*. Seattle: The Mountaineers.

Ellenberg H. 1988. *Vegetation ecology of Central Europe*. Cambridge: Cambridge UP.

Kohyama. 1995. Spatial and temporal patterns of subalpine *Abies* forests in Central Japan. In: Box EO, eds. *Vegetation science in forestry*. Dordrecht: Kluwer, 391-407.

Kojima S. 1995. Boreal forest phytogeocoenoses of Hokkaido Island, Japan. In: Box EO, eds. *Vegetation science in forestry*. Dordrecht: Kluwer, 367-389.

Ling-Zhi C. 1995. The spruce forests of northern China. In: Box EO, eds. *Vegetation science in forestry*. Dordrecht: Kluwer, 352-365.

Liu T-S. 1971. *A monograph of the genus Abies*. Taipei: National Taiwan University.

Reiners WA, Lang GE. 1979. Vegetational patterns and processes in the balsam fir zone, White Mountains, New Hampshire. *Ecology* **60**: 403-417.

Walter H. 1983. *Vegetation of the Earth and ecological systems of the geo-biosphere*. Berlin: Springer-Verlag.

Appendix:

Interpreting main effects and interactions

Interpretation of the results of multi-factor experiments can sometimes be

A) No effect: The overall mean for each species is the same AND the overall mean for each environment is the same AND the species means in each environment are the same.

B) S effect only: The overall mean for species 1 is different from the overall mean for species 2, BUT the overall mean for environment A is the same as the overall mean for environment B AND the difference between species is the same in each environment.

C) E effect only: The overall mean for environment A is different from the overall mean for environment B, BUT the overall mean for species 1 is the same as the overall mean for species 2 AND the species means in each environment are the same.

D) Both S and E effects: The overall mean for environment A is different from the overall mean for environment B, AND the overall mean for species 1 is different from the overall mean for species 2, BUT the difference between species is the same in each environment.

E) E × S interaction effect only. The overall mean for environment A is the same as the overall mean for environment B, AND the overall mean for species 1 is the same as the overall mean for species 2, BUT the species means in each environment are different.

F) S effect and E × S interaction. The overall mean for species 1 is different from the overall mean for species 2, BUT the overall mean for environment A is the same as the overall mean for environment B, AND the species means in each environment are different.

G) E effect and E × S interaction. The overall mean for environment A is different from the overall mean for environment B, BUT the overall mean for species 1 is the same as the overall mean for species 2, AND the species means in each environment are different.

H) S and E effects, and E × S interaction. The overall mean for environment A is different from the overall mean for environment B, AND the overall mean for species 1 is different from the overall mean for species 2, AND the species means in each environment are different (not illustrated).

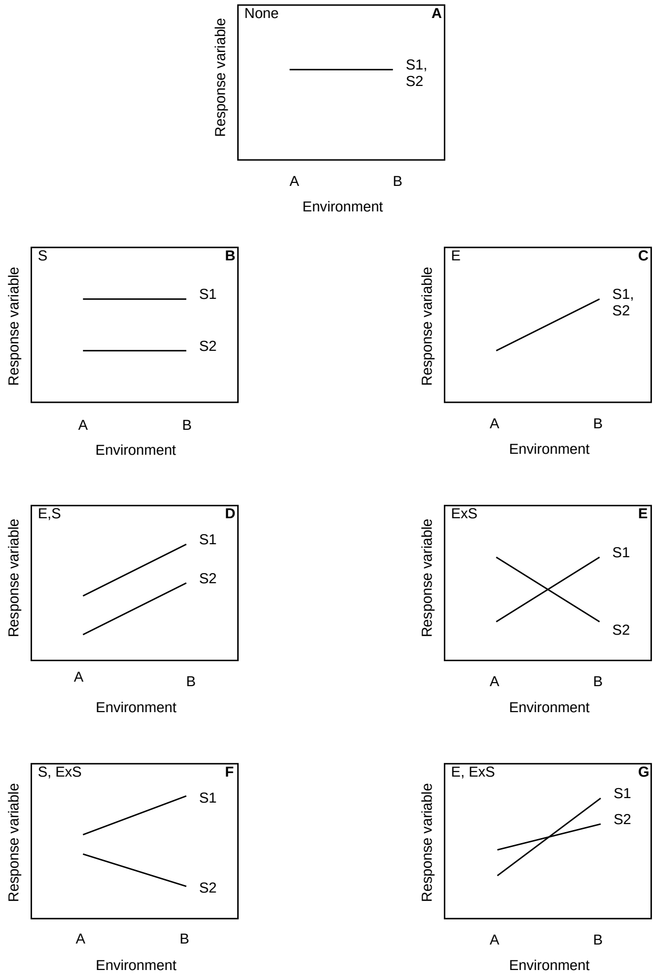

Figure A1. Different outcomes in a two-factor (environment × species) experiment. See text for interpretation.