PHYSIOLOGICAL AND MORPHOLOGICAL RESPONSES OF MOUNTAIN MAPLE (*ACER SPICATUM* L.) SEEDLINGS TO LIGHT UNDER DIFFERENT CARBON DIOXIDE CONCENTRATIONS, SOIL TEMPERATURE AND SOIL MOISTURE REGIMES.

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ABSTRACT

Determining the response of mountain maple (*Acer spicatum* L.) to increases in light is confounded by interaction with other environmental factors. Rising atmospheric carbon dioxide concentrations ([CO_2]) and the accompanying global warming and soil moisture depletion are expected to have large impacts on plants, including mountain maple, responses to their biological and physical environments. Yet the effects of global climate change factors on mountain maple response to light is ignored. Mountain maple seedlings were grown in two light regimes (low or shaded and high or unshaded), two [CO_2] (ambient: *c* 380 µmol mol^{-1} and elevated: 760 µmol mol$_{-1}$). Using soil temperature manipulation equipment (17 and 22°C), I tested the effects of [CO_2] and T_{soil} on the physiological and morphological responses of mountain maple to light. Secondly, soil moisture treatment was applied to another set of seedlings to test the effects of [CO_2] and soil moisture on the physiological and morphological responses of mountain maple to light. There were two moisture levels, low and high. Both experiments lasted for two months. The seedlings responded differently to light in terms of physiological and morphological traits; the soil warming tended to reduce the photosynthetic rates of the seedlings in the high light treatment. On the other hand, elevated [CO_2] stimulated the instantaneous water-use efficiency (IWUE) and the ratio between apparent electron transport (J) to maximum rate of carboxylation (V_{cmax}), (J/V_{cmax}) responses to light. The elevated [CO_2] and soil warming constrained the morphological responses to light. The high light had the smallest effect on seedling growth and biomass when exposed to elevated [CO_2] and soil warming. The photosynthetic response of mountain maple to light was unaffected by low soil moisture. The elevated [CO_2] enhanced IWUE response to high light, but decreased the drought tolerance of the seedlings. Increased allocation of biomass to root under elevated [CO_2] that would improve water absorption might have mitigated the susceptibility of mountain maple to drought. The treatment effects on mountain maple suggest that growth response to canopy gaps may be enhanced as [CO_2] continues to increase. However, the accompanying increases in T_{soil} may limit the growth of mountain maple seedling in high light environments.

Key words: mountain maple, light, elevated CO_2, soil warming, soil moisture, photosynthesis, growth, biomass.

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CHAPTER 1: General introduction

Plant response to light

Light is an important environmental factor affecting the growth and survival of plants (Canham 1988a, Canham 1989). Experiments have long established evidence that plant acclimation to changing light environments involves morphological and physiological modifications that enhance plant growth (Zon and Graves 1911, Evans 1988, Lei 1992). Consequently, plant response to light has been researched extensively from a number of perspectives, including growth, biomass allocation patterns, leaf and crown architecture and photosynthetic performance in both natural and controlled environments (Klinka et al. 1992, Lei 1992, Chazdon and Kaufmann 1993, Canham et al. 1996, Kubiske and Pregitzer 1997, Tognetti et al. 1998, Poorter 1999, Parelle et al. 2006, Poorter et al. 2012). The changing light environments that plant need to acclimate to may include high light levels that occur as a result of naturally occurring canopy gaps or the removal of overstory vegetation caused by silvicultural practices (Boucher et al. 1998, Boucher et al. 2007). The ability of plant to acclimate to changes in light conditions plays an important role in its establishment and competitive success (Chazdon 1988, Küppers 1994). Plant species that exhibit relatively high acclimation capacity to relatively high light levels may further be stimulated by changes in other environment factors. For example, significant crown opening in the field that increased soil temperature (T_{soil}) or light and T_{soil} studies in greenhouses show that increasing T_{soil} enhances tree species photosynthetic and growth rates (Boucher et al. 1998, Archibold et al. 2000, Boucher et al. 2007). This may demonstrate that acclimation to high light conditions depend on other environmental factors. The initial phase of seedling establishment after germination appears to pose a considerable bottleneck that influences regeneration success (Clark and Clark 1992, Kobe et al. 1995, Hattenschwiler and Korner 2003). Hence, understanding how seedling morphological and physiological responses to light are influenced by other environmental factors is critical for predicting changes in forest composition or structure in the future.

Plant responses to rising atmospheric carbon dioxide concentration, soil warming and soil moisture

The atmospheric carbon dioxide concentration ([CO_2]) has been increasing in the past 25 million years, and is predicted to reach between 730 and 1200 μmol mol^{-1} by 2100 (Siegenthaler et al. 2005, IPCC 2007b, Meehl et al. 2007, Sitch et al. 2008). An increase in the atmosphere [CO_2] alone has, aside from affecting global climate, instant effect on plants, and thus terrestrial carbon storage (Cox et al. 2000, Körner 2003). CO_2 is a basic substrate for photosynthesis, and in recent years, much attention has been placed on the productivity and alterations of terrestrial ecosystems (forests and grasslands) under future [CO_2]. Such studies have been devoted to tree species because of their sequestration of terrestrial carbon (Huang et al. 2007), although some other studies found that terrestrial ecosystems (e.g. soils) may become a source of CO_2 under future climate (Cox et al. 2000, Hagedorn et al. 2010). Mature trees are less responsive to elevated [CO_2] in terms of growth than seedlings (Nowak et al. 2004, Korner 2006). Nonetheless, elevated [CO_2] has been shown to directly stimulate tree growth in both natural and growth chamber experiments mainly as a result of increased photosynthetic rates (Saxe et al. 1998, Dawes et al. 2011a, Dawes et al. 2011b, Watanabe et al. 2011). Temperate and boreal forest ecosystems are of particular interest, since high elevation and high altitude environments are expected to be more sensitive to global climate change (Saxe et al. 1998, Smith et al. 2009). Studies have shown that elevated [CO_2] can influence plants response to their physical and biological environments (Field et al. 1992, Bazzaz 1996), thus, altering plant distribution and community composition along resource gradient (Bazzaz et al. 1985, Miao et al. 1992, Duff et al. 1994, Catovsky and Bazzaz 1999).

The rising atmospheric [CO_2] and other greenhouse gases will lead to global warming. An increase of 1.4 – 5.8 ºC in the mean global temperature is predicted with the changes in climate (Houghton 1992, IPCC 2001, Wigley 2005, Meehl et al. 2007). T_{soil} is an important ecological factor in temperate and boreal regions that regulates below- and aboveground plant processes such as root growth (Larigauderie et al. 1991, Karlsson and Nordell 1996), nutrient and water absorption (Bowes 1991, DeLucia et al. 1992, Bassirirad 2000), leaf morphology (Stoneman and Dell 1993) and carbon fixation (Day et al. 1991, King et al. 1999, Ambebe et al. 2010). As a consequence, soil warming will inevitably influence plant growth. Evidence of soil warming-induced increases in plant growth and productivity in diverse environments and experimental approaches abounds (Boucher et al. 1998, Archibold et al. 2000, Rustad et al. 2001, Dawes et al. 2011a, De Frenne et al. 2012). Biomass allocation patterns are also found to be influenced by T_{soil} (Peng and Dang 2003, Ambebe et al. 2010, Pumpanen et al. 2012). Decreased root/shoot ratio with increasing T_{soil} has been reported (Larigauderie et al. 1991, Boucher et al. 2007) as a result of increased root functions (Davidson 1969). T_{soil} affects plant response to other factors. For example, significant part of eastern white pine (*Pinus strobus* L.) seedlings to light response is because of changes in T_{soil} (Boucher et al. 2007).

Given the predictions of changes in both the magnitude and degree of variability of precipitation (Houghton 1992, IPCC 2001, Wigley 2005), global climate change will undoubtedly modify soil moisture, thus affecting the growth of temperate and boreal trees**.** Soil warming will also influence soil moisture due to increased rates and depth of evaporation

(Pregitzer and King 2005). Soil moisture has complex effects on plant functioning. It is generally thought that the primary consequence of reduced soil moisture is stomatal closure, thus, reduction of stomatal conductance and inhibition of photosynthesis (Flexas and Medrano 2002, Lawlor and Cornic 2002, Flexas et al. 2004, Lambers et al. 2008, Lawlor and Tezara 2009, Wang et al. 2012). Reduced nutrient availability, due to reduced soil microbial activity (Sardans and Peñuelas 2005, Ruifang et al. 2007) and nutrient absorption by roots (Chapin 1980, Aerts and Chapin Iii 1999) at low soil moisture may also limit plant growth. Shoot growth reduces due to decreased leaf area production and decreased biomass allocation to leaf (Canham et al. 1996, Poorter et al. 2012). Reduced leaf growth at low soil moisture decreases the amount of leaf area displayed for light capture and the photosynthetic capacity of plant (O'Connell et al. 2004, Brisson and Casals 2005). Increased biomass allocation to roots is a key adaptive response of plant at low soil moisture conditions that improves nutrients and water absorption (Canham et al. 1996, Lambers et al. 2008). Reduced soil moisture also limits plant responses to other factors such as CO_2 and light (Samarakoon and Gifford 1995, Muraoka et al. 1997, Volk et al. 2000, Muraoka et al. 2002). Therefore, depletion of soil moisture under future CO_2 and T_{soil} levels may limit plant response to canopy gaps. Experimental manipulations of $[CO_2]$, T_{soil} and soil moisture can help to improve our understanding of plants responses to light and the potential shifts in plant distribution and community composition under environmental change.

The aims of the study

The overall aim of the thesis was to determine the interactive or treatment effects of $[CO_2]$, soil T_{soil} and soil moisture on the physiological and morphological responses of mountain maple (*Acer spicatum* L.) seedlings to light. Mountain maple is an important deciduous shade-tolerant tree/shrub species in North America that persists in shade but responds rapidly to increases in light (Lei 1992, Aubin et al. 2005). Physiological and morphological changes under elevated $[CO_2]$, warmer T_{soil} or low soil moisture that limit water loss and increase carbon gain, enhance water and nutrients absorption might facilitate mountain maple response to increases in light availability. Nonetheless, there is a lack of studies on how global climate change factors might affect mountain maple response to light. The individual chapters below summarises the specific research questions for these topics.

The objective of chapter two was to examine how $[CO_2]$ and T_{soil} may change the pattern of physiological responses of mountain maple (*Acer spicatum* L.) seedling to light. Specifically, I aimed to understand how the interaction between $[CO_2]$ and T_{soil}, or their main effects alone, may influence the physiological performance of mountain maple seedling in high light conditions. I hypothesized that elevated $[CO_2]$ and soil warming would increase A at high light environment due to the direct stimulation of A by elevated $[CO_2]$ and soil warming-related increases in nutrients and water absorption .

The objective of chapter three was to determine how the interaction between $[CO_2]$ and soil warming will change mountain maple (*Acer spicatum* L) seedling growth and biomass responses to light. The specific hypothesis tested was that elevated $[CO_2]$ and soil warming would stimulate growth and biomass responses of mountain maple to light, thus the largest responses to high light would be observed under elevated $[CO_2]$ and warmer T_{soil} treatment combination.

Chapter four examines the effects of $[CO_2]$ and soil moisture on the physiological responses of mountain maple (*Acer spicatum* L.) seedling to light. It focuses on how low soil moisture may limit elevated $[CO_2]$ stimulation of the physiological responses to light. I hypothesize that low soil moisture limits elevated CO_2 stimulation of photosynthesis, and that the effect would be greater in seedlings growing in high light treatment.

Chapter five examines the effects of [CO_2] and soil moisture on growth and biomass responses of mountain maple (*Acer spicatum* L.) seedling to light.It addresses how low soil moisture may limit the growth and biomass responses of the seedlings to elevated [CO2] in high light conditions The hypothesis tested was the low soil moisture that low soil moisture would limit the stimulation of height growth and biomass accumulation by elevated [CO2] in the high light treatment.

References

Aerts, R. and F.S. Chapin Iii. 1999. The mineral nutrition of wild plants revisited: a re-evaluation of processes and patterns. *In* Advances in Ecological Research Eds. A.H. Fitter and D.G. Raffaelli. Academic Press, pp 1-67.

Ambebe, T.F., Q.-L. Dang and J. Li. 2010. Low soil temperature inhibits the effect of high nutrient supply on photosynthetic response to elevated carbon dioxide concentration in white birch seedlings. Tree Physiology. 30:234-243.

Archibold, O.W., C. Acton and E.A. Ripley. 2000. Effect of site preparation on soil properties and vegetation cover, and the growth and survival of white spruce (*Picea glauca*) seedlings, in Saskatchewan. Forest Ecology and Management. 131:127-141.

Aubin, I., C. Messier and D. Kneeshaw. 2005. Population structure and growth acclimation of mountain maple along a successional gradient in the southern boreal forest. Ecoscience. 12:540-548.

Bassirirad, H. 2000. Kinetics of nutrient uptake by roots: responses to global change. New Phytologist. 147:155-169.

Bazzaz, F.A. 1996. Plants in changing environment: Linking physiology, population, and community ecology. Cambridge Univ. Press, Cambridge.

Bazzaz, F.A., K. Garbutt and W.E. William. 1985. Effect of increased atmospjeric carbon dioxide concentration on plant communities. In: *Direct effects of increased carbon dioxide concentration on vegetation*. United States Department of Energy Washington, DC.

Boucher, J.-F., P.Y. Bernier, H.A. Margolis and A.D. Munson. 2007. Growth and physiological response of eastern white pine seedlings to partial cutting and site preparation. Forest Ecology and Management. 240:151-164.

Boucher, J.-F., S. Wetzel and A.D. Munson. 1998. Leaf level response of planted eastern white pine (*Pinus strobus* L.) seven years after intensive silvicultural treatments. Forest Ecology and Management. 107:291-307.

Bowes, G.D. 1991. Soil temperature, root growth, and plant function - In: Plant roots, the hidden half. Marcel Decker Inc.

Brisson, N. and M.-L. Casals. 2005. Leaf dynamics and crop water status throughout the growing cycle of durum wheat crops grown in two contrasted water budget conditions. Agron. Sustain. Dev. 25:151-158.

Canham, C.D. 1988a. Growth and canopy architecture of shade-tolerant trees: response to canopy gaps. Ecology. 69:786-795.

Canham, C.D. 1988b. An index for understory light levels in and around canopy gaps. Ecology. 69:1634-1638.

Canham, C.D. 1989. Different response to gaps among shade-tolerance tree species. Ecology. 70:549 - 550.

Canham, C.D., A.R. Berkowitz, V.R. Kelly, G.M. Lovett, S.V. Ollinger and J. Schnurr. 1996. Biomass allocation and multiple resource limitation in tree seedlings. Canadian Journal of Forest Research. 26:1521-1530.

Catovsky, S. and F.A. Bazzaz. 1999. Elevated CO_2 influences the responses of two birch species to soil moisture: implications for forest community structure. Global Change Biology. 5:507-518.

Chapin, F.S. 1980. The Mineral Nutrition of Wild Plants. Annual review of ecology and systematics. 11:233-260.

Chazdon, R.L. 1988. Sunflecks and their importance to forest understory plants. Adv. Ecol. Research. 18:1–63.

Chazdon, R.L. and S. Kaufmann. 1993. Plasticity of leaf anatomy of two ranin-forest shurbs in relation to photosynthetic light acclimation. Functional Ecology. 7:385-394.

Clark, D.A. and D.B. Clark. 1992. Life history diversity of canopy and emergent trees in a neotropical rain forest. Ecological Monographs. 62:315-344.

Cox, P.M., R.A. Betts, C.D. Jones, S.A. Spall and I.J. Totterdell. 2000. Acceleration of global warming due to carbon-cycle feedbacks in a coupled climate model. Nature. 408:184-187.

Curtis, P.S. and X. Wang. 1998. A meta-analysis of elevated CO_2 effects on woody plant mass, form, and physiology. Oecologia. 113:299-313.

Davidson, R.L. 1969. Effect of root/leaf temperature differentials on root/shoot Ratios in some pasture grasses and clover. Annals of Botany. 33:561-569.

Dawes, M.A., F. Hagedorn, T. Zumbrunn, I.T. Handa, S. Hättenschwiler, S. Wipf and C. Rixen. 2011a. Growth and community responses of alpine dwarf shrubs to in situ CO_2 enrichment and soil warming. New Phytologist. 191:806-818.

Dawes, M.A., S. Hättenschwiler, P. Bebi, F. Hagedorn, I.T. Handa, C. Körner and C. Rixen. 2011b. Species-specific tree growth responses to 9 years of CO_2 enrichment at the alpine treeline. Journal of Ecology. 99:383-394.

Day, T.A., S.A. Heckathorn and E.H. Delucia. 1991. Limitations of photosynthesis in *Pinus taeda* (loblolly Pine) at low soil temperatures. Plant Physiology. 96:1246-1254.

De Frenne, P., B.J. Graae, J. Brunet, A. Shevtsova, A. De Schrijver, O. Chabrerie, S.A.O. Cousins, G. Decocq, M. Diekmann, M. Hermy, T. Heinken, A. Kolb, C. Nilsson, S. Stanton and K. Verheyen. 2012. The response of forest plant regeneration to temperature variation along a latitudinal gradient. Annals of Botany:1 - 10.

DeLucia, E.H., S.A. Heckathorn and T.A. Day. 1992. Effects of soil temperature on growth, biomass allocation and resource acquisition of *Andropogon gerardii* Vitman. New Phytologist. 120:543-549.

Duff, G.A., C.A. Berryman and D. Eamus. 1994. Growth, biomass allocation and foliar nutrient contents of two *Eucalyptus* species of the wet-dry tropics of Australia grown under CO_2 enrichment. Functional Ecology. 8:502-508.

Evans, J.R. 1988. Acclimation by the thylakoid membranes to growth irradiance and the partitioning of nitrogen between soluble and thylakoid proteins Australian Journal of Plant Physiology. 15:93-106.

Field, C.B., F.S.C. Iii, P.A. Matson and H.A. Mooney. 1992. Responses of terrestrial ecosystems to the changing atmosphere: a resource-based approach. Annual Review of Ecology and Systematics. 23:201-235.

Flexas, J., J. Bota, F. Loreto, G. Cornic and T.D. Sharkey. 2004. Diffusive and metabolic limitations to photosynthesis under drought and salinity in C_3 plants. Plant Biology. 6:269-279.

Flexas, J. and H. Medrano. 2002. Drought-inhibition of photosynthesis in C_3 plants: stomatal and non-stomatal limitations revisited. Annals of Botany. 89:183-189.

Hagedorn, F., M. Martin, C. Rixen, S. Rusch, P. Bebi, A. Zürcher, R. Siegwolf, S. Wipf, C. Escape, J. Roy and S. Hättenschwiler. 2010. Short-term responses of ecosystem carbon fluxes to experimental soil warming at the Swiss alpine treeline. Biogeochemistry. 97:7-19.

Hattenschwiler, S. and C. Korner. 2003. Does elevated CO_2 facilitate naturalization of the non-indigenous *Prunus laurocerasus* in Swiss temperate forests? Functional Ecology. 17:778-785.

Houghton, J.T.e.a. 1992. Climate change 1992. The supplementary report to the IPCC scientific assessement.

Huang, J.-G., Y. Bergeron, B. Denneler, F. Berninger and J. Tardif. 2007. Response of forest trees to increased atmospheric CO2. Critical Reviews in Plant Sciences. 26:265-283.

IPCC. 2001. Climate Change 2001: An Assessment of the Intergovernmental Panel on Climate Change.

IPCC. 2007. Summary for policymakers. In: Solomon, S. et al. (eds), Climate Change 2007: the physical science basis. Contribution of Working Group I to the fourth assessment report of the Intergovernmental Panel on Climate Change.

Karlsson, P.S. and K.O. Nordell. 1996. Effects of soil temperature on the nitrogen economy and growth of mountain birch seedlings near its presumed low temperature distribution limit. Ecoscience. 3:183-189.

King, J.S., K.S. Pregitzer and D.R. Zak. 1999. Clonal variation in above- and below-ground growth responses of Populus tremuloides Michaux: Influence of soil warming and nutrient availability. Plant and Soil. 217:119-130.

Klinka, K., Q. Wang, G.J. Kayahara, R.E. Carter and B.A. Blackwell. 1992. Light-growth response relationships in Pacific silver fir (*Abies amabilis*) and subalpine fir (*Abies lasiocarpa*). Canadian Journal of Botany. 70:1919-1930.

Kobe, R.K., S.W. Pacala, J.A. Silander and C.D. Canham. 1995. Juvenile tree survivorship as a component of shade tolerance. Ecological Applications. 5:517-532.

Körner, C. 2003. Ecological impacts of atmospheric CO_2 enrichment on terrestrial ecosystems. Philosophical Transactions of the Royal Society of London Series A. 361: 2023-2041.

Korner, C. 2006. Plant CO_2 responses: an issue of definition, time and resource supply. New Phytologist. 172:393-411.

Kubiske, M.E. and K.S. Pregitzer. 1997. Ecophysiological responses to simulated canopy gaps of two tree species of contrasting shade tolerance in elevated CO_2. Functional Ecology. 11:24-32.

Küppers, M. 1994. Canopy gaps: competitive light interception and economic space filling—a matter of whole plant allocation. In Exploitation of Environmental Heterogeneity by

Plants: Ecophysiological Processes Above- and Belowground. Academic Press, San Diego.

Lambers, H., S.F. Chapin III and T.L. Pons. 2008. Plant physiological eclogy. Springer, New York.

Larigauderie, A., B.A. Ellis, J.N. Mills and J. Kummerow. 1991. The effect of root and shoot temperatures on growth of Ceanothus-Greggii seedlings. Annals of Botany. 67:97-101.

Lawlor, D.W. and G. Cornic. 2002. Photosynthetic carbon assimilation and associated metabolism in relation to water deficits in higher plants. Plant, Cell and Environment. 25:275-294.

Lawlor, D.W. and W. Tezara. 2009. Causes of decreased photosynthetic rate and metabolic capacity in water-deficient leaf cells: a critical evaluation of mechanisms and integration of processes. Annals of Botany. 103:561-579.

Lei, T.T. 1992. Functional design and shade adaptation in *Acer* species. *In* Biology. McGill University.

Meehl, G.A., J.M. Arblaster and C. Tebaldi. 2007. Contributions of natural and anthropogenic forcing to changes in temperature extremes over the United States. Geophys. Res. Lett. 34:L19709.

Miao, S.L., P.M. Wayne and F.A. Bazzaz. 1992. Elevated CO_2 differentially alters the responses of coocurring birch and maple seedlings to a moisture gradient. Oecologia. 90:300-304.

Muraoka, H., Y. Tang, H. Koizumi and I. Washitani. 2002. Effects of light and soil water availability on leaf photosynthesis and growth of *Arisaema heterophyllum* , a riparian forest understorey plant. Journal of Plant Research. 115:419-427.

Muraoka, H., Y. Tang, H. Koizumi, I. Washitani and W. Z. 1997. Combined effects of light and water availability on photosynthesis and growth of *Arisaema Heterophyllum* in the forest understory and an open site. Oecologia. 112:26-34.

Norby, R.J., S.D. Wullschleger, C.A. Gunderson, D.W. Johnson and R. Ceulemans. 1999. Tree responses to rising CO2 in field experiments: implications for the future forest. Plant Cell and Environment. 22:683-714.

Nowak, R.S., D.S. Ellsworth and S.D. Smith. 2004. Functional responses of plants to elevated atmospheric CO_2: Do photosynthetic and productivity data from FACE experiments support early predictions? New Phytologist. 162:253-280.

O'Connell, M.G., G.J. O'Leary, D.M. Whitfield and D.J. Connor. 2004. Interception of photosynthetically active radiation and radiation-use efficiency of wheat, field pea and mustard in a semi-arid environment. Field Crops Research. 85:111-124.

Parelle, J., J.P. Roudaut and M. Ducrey. 2006. Light acclimation and photosynthetic response of beech (*Fagus sylvatica* L.) saplings under artificial shading or natural Mediterranean conditions. Annals of Forest Science. 63:257-266.

Peng, Y.Y. and Q.-L. Dang. 2003. Effects of soil temperature on biomass production and allocation in seedlings of four boreal tree species. Forest Ecology and Management. 180:1-9.

Poorter, H., K.J. Niklas, P.B. Reich, J. Oleksyn, P. Poot and L. Mommer. 2012. Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control. New Phytologist. 193:30-50.

Poorter, L. 1999. Growth responses of 15 rain-forest tree species to a light gradient: the relative importance of morphological and physiological traits. Functional Ecology. 13:396-410.

Pregitzer, K.S. and J.S. King. 2005. Effects of soil temperature on nutrient uptake. - In: Nutrient acquisition by plants: an ecological perspective. Springer.

Pumpanen, J., J. Heinonsalo, T. Rasilo, J. Villemot and H. Ilvesniemi. 2012. The effects of soil and air temperature on CO2 exchange and net biomass accumulation in Norway spruce, Scots pine and silver birch seedlings. Tree Physiology

Ruifang, F., Y. Wanqin, Z. Jian, D. Renju, J. Yi and L. Jin. 2007. Effects of simulated elevated concentration of atmospheric CO_2 and temperature on soil enzyme activity in the subalpine fir forest. Acta Ecologica Sinica. 27:4019-4026.

Rustad, L.E., J.L. Campbell, G.M. Marion, R.J. Norby, M.J. Mitchell, A.E. Hartley, J.H.C. Cornelissen, J. Gurevitch and N. Gcte. 2001. A meta-analysis of the response of soil respiration, net nitrogen mineralization, and aboveground plant growth to experimental ecosystem warming. Oecologia. 126:543-562.

Samarakoon, A.B. and R.M. Gifford. 1995. Soil water content under plants at high CO_2 concentration and interactions with the direct CO_2 effects: a species comparison. Journal of Biogeography. 22:193-202.

Sardans, J. and J. Peñuelas. 2005. Drought decreases soil enzyme activity in a Mediterranean *Quercus ilex* L. forest. Soil Biology and Biochemistry. 37:455-461.

Saxe, H., D.S. Ellsworth and J. Heath. 1998. Tree and forest functioning in an enriched CO_2 atmosphere. New Phytologist. 139:395-436.

Siegenthaler, U., T.F. Stocker, E. Monnin, D. Lüthi, J. Schwander, B. Stauffer, D. Raynaud, J.-M. Barnola, H. Fischer, V. Masson-Delmotte and J. Jouzel. 2005. Stable carbon cycle-climate relationship during the late Pleistocene. Science. 310:1313-1317.

Sitch, S., C. Huntingford, N. Gedney, P.E. Levy, M. Lomas, S.L. Piao, R. Betts, P. Ciais, P. Cox, P. Friedlingstein, C.D. Jones, I.C. Prentice and F.I. Woodward. 2008. Evaluation of the terrestrial carbon cycle, future plant geography and climate-carbon cycle feedbacks using five Dynamic Global Vegetation Models (DGVMs). Global Change Biology. 14:2015-2039.

Smith, W.K., M.J. Germino, D.M. Johnson and K. Reinhardt. 2009. The altitude of alpine treeline: a bellwether of climate change effects. Botanical Review. 75:163-190.

Stoneman, G.L. and B. Dell. 1993. Growth of *Eucalyptus marginata* (Jarah) seedlings in a greenhouse in response to shade and soil temperature Tree Physiology. 13:239-252.

Tissue, D.T., R.B. Thomas and B.R. Strain. 1997. Atmospheric CO_2 enrichment increases growth and photosynthesis of *Pinus taeda*: a 4 year experiment in the field. Plant, Cell & Environment. 20:1123-1134.

Tognetti, R., G. Minotta, S. Pinzauti, M. Michelozzi and M. Borghetti. 1998. Acclimation to changing light conditions of long-term shade-grown beech (*Fagus sylvatica* L.) seedlings of different geographic origins. Trees - Structure and Function. 12:326-333.

Volk, M., P.A. Niklaus and C. Körner. 2000. Soil moisture effects determine CO_2 responses of grassland species. Oecologia. 125:380-388.

Wang, M., S. Shi, F. Lin, Z. Hao, P. Jiang and G. Dai. 2012. Effects of soil water and nitrogen on growth and photosynthetic response of manchurian ash (*Fraxinus mandshurica*) seedlings in northeastern China. PLoS ONE. 7:e30754.

Watanabe, M., Y. Watanabe, S. Kitaoka, H. Utsugi, K. Kita and T. Koike. 2011. Growth and photosynthetic traits of hybrid larch F1 (*Larix gmelinii* var. *japonica* × L. *kaempferi*) under elevated CO_2 concentration with low nutrient availability. Tree Physiology

Wigley, T.M.L. 2005. The Climate Change Commitment. Science. 307:1766-1769.

Zon, R. and H.S. Graves. 1911. Light in relation to tree growth. Forest Service Bulletin 92. U.S. Dept Agriculture, Washington DC, USA.

Chapter 2: Elevated [CO_2] and soil warming change the pattern of physiological responses of mountain maple (*Acer spicatum* L.) seedling to light.

Introduction

Light availability strongly influences plant growth and survival (Pacala et al. 1996, Lin et al. 2002). Optimum carbon gain under different light environments involves the modifications of a suite of physiological and morphological traits (Canham 1988b, Lei and Lechowicz 1990, Kitajima 1994, Chazdon et al. 1996, Kubiske and Pregitzer 1996, Zipperlen and Press 1996, Coomes and Grubb 1998, Dalling et al. 2004). Certain physiological traits at the leaf level are often regarded as being determinants of light acclimation capacity. For example, plant species growing under high light conditions tend to have higher maximum carboxylation rate and electron transport capacity, whereas those growing under low conditions have high light-harvesting capacity (Kubiske and Pregitzer 1996, Niinemets and Tenhunen 1997, Herrick and Thomas 1999, Parelle et al. 2006, Hallik et al. 2012). Directly correlated to light environments and photosynthetic capacity, leaf nitrogen content per unit leaf area and stomatal conductance have been found to influence plant acclimation to light (Harley et al. 1996, Niinemets 2007, Hallik et al. 2012). As such, greater leaf N and photosynthetic rates are observed in plants growing in high than low light conditions (Harley et al. 1996, Herrick and Thomas 1999). While light is often thought to be the major determinant of plant physiological responses to light, other environmental factors such as CO_2 and soil temperature (T_{soil}) may interact to affect plant physiology. Yet, there is little evidence on how CO_2 and T_{soil} might influence plant physiological response to light environment. Hence, a good understanding of plant physiological response to light under different CO_2 and T_{soil} can lead to a better prediction of possible changes in forest composition and distribution in the future.

Increases in atmospheric carbon dioxide concentration ($[CO_2]$) can substantially enhance net photosynthesis (*A*) in C_3 plants (Pearcy 1983, Bunce 1992, Tissue et al. 1997, Curtis and Wang 1998, Saxe et al. 1998, Norby et al. 1999, Sefcik et al. 2006). This is because the present $[CO_2]$ does not saturate photosynthesis in C_3 plant (Long and Drake 1992, Drake et al. 1997, Lambers et al. 2008). Furthermore, elevated $[CO_2]$ suppresses photorespiration, resulting in increased quantum yield and decreased light compensation point of *A* (Pearcy and Björkman 1983, Bowes 1993, Osborne et al. 1997, Saxe et al. 1998). However, photosynthetic responses to elevated $[CO_2]$ vary with experimental protocols. The relative CO_2-stimulation of photosynthesis (*A*) has been reported to be higher in low light conditions (Long and Drake 1991, Gifford 1992, Kubiske and Pregitzer 1996) but others reported no such effect (Herrick and Thomas 1999, Liozon et al. 2000, Takeuchi et al. 2001).

It is predicted that the rise in $[CO_2]$ will cause air and T_{soil} to increase (Mitchell et al. 1990, Houghton et al. 2001, Wigley 2005, IPCC 2007b, Solomon et al. 2007). Soil warming can induce changes in leaf morphology (Stoneman and Dell 1993), root growth and biomass allocation (Boucher et al. 2001), *A* and stomatal conductance (g_s) (Day et al. 1991, DeLucia et al. 1992, Landhäusser et al. 1996, King et al. 1999, Dodd et al. 2000, Ambebe et al. 2010). Previous studies suggest that soil warming influences net *A* through nutrient and water uptake (Pastor et al. 1987, Paré et al. 1993, Fitter et al. 1998, Long and Woodward 1998, Wan et al. 1999) and sink strength (Lyr and Garbe 1995, Boucher et al. 2001). How plants respond to soil warming may affect their distribution in the future (De Frenne et al. 2011). Soil warming may have a profound effect on plants physiological responses to other environmental factors. For example, Ambebe et al. (2010) report that soil warming enhances the stimulatory effects of elevated [CO_2] on photosynthesis in white birch (*Betula papyrifera*) seedlings. Boucher et al. (2001) also found that physiological processes of eastern white pine (*Pinus strobus* L.) seedlings were more responsive to light conditions at warmer soils. The influence of soil warming on photosynthetic response to elevated [CO_2] or light availability has been ascribed to changes in sink strength or nutrient uptake. Increased sink strength (higher root/shoot ratio (R/S) or increased carbohydrates utilization (TPU)) caused by elevated [CO_2] and soil warming should therefore help maintain higher photosynthetic rates in high light conditions. Thus, we hypothesize that elevated [CO_2] and soil warming would increase A at high light environment.

In this study, we investigated the effects of elevated [CO_2] and soil warming on the physiological responses of mountain maple seedlings to light. Mountain maple (*Acer spicatum* Lamb.) is an important understory shrub in boreal mixedwood forests that contributes immensely to the composition, structure and species diversity (Archambault et al. 1998). It colonizes the understory of dense multi-layered forest stands (Post 1967, Post 1970, Sullivan 1993, Rook 2002) although it shows a rapid response to canopy openings (Lei and Lechowicz 1990, Aubin et al. 2005). However, the physiological traits that maximize plants response to increased light availability in gaps differ considerably from traits associated with ability to tolerate low light environments (see responses to light above). The ability of plant species adapted to low light conditions to rapidly respond to canopy gaps may have an important implications on species composition and distribution in the forest ecosystem (Platt and Strong 1989). Elevated [CO_2] and soil warming have the potential to modify physiological responses to light. However, most studies examining the effects of climate change factors on plant physiological responses to light conditions focus on elevated [CO_2] and/or air temperature, but not soil temperature (Kubiske and

Pregitzer 1996, Hättenschwiler 2001, Liang et al. 2001, Takeuchi et al. 2001, Hikosaka 2005). As such, the impact of interactions between [CO_2] and soil warming on physiological responses of plants to different light condition is not well understood.

Materials and methods

Plant material

Mountain maple seeds were collected from Lakehead University Jack Haggerty Forest in the fall of 2010. The forest is located approximately approximately 37km north of Thunder Bay, ON. The seeds were soaked in 1000µmol m^{-1} giberellic acid (GA) solution for 24hrs (Lei 1992). The seeds were placed in germination trays covered with moist paper towels at 4 ºC for two months. After 2 months stratification, seed coats were gently cracked open to facilitate germination. The seeds were germinated in a 2:1(v/v) mixture of peat moss and vermiculite. A total of 160 seedlings (10 seedlings per treatment combination) of relatively uniform height were transplanted into plastic containers (31.5 cm deep, 11 cm top diameter and 9.5 cm bottom diameter). The transplanting was done three weeks after the start of germination.

Experimental design

The treatments consisted of two [CO_2] levels (ambient = 392 µmol mol^{-1} versus elevated = 784 µmol mol^{-1}), two T_{soil} (17 and 22 ºC), and two light levels (high and low). The experiment was a split-split plot design, with [CO_2] as the main plot, T_{soil} as sub-plot and light regime as the sub-sub-plot. The CO_2 elevation was achieved using Argus CO_2 generators (Argus, Vancouver, BC, Canada). Each CO_2 level had two independent replications (greenhouses). The low T_{soil} of 17 ºC represents the mean (July) growing season T_{soil} for mixed forest stand with that included mountain maple (Caners and Kenkel 1997). The warmer soil (22 ºC) in which the average T_{soil} was 5 ºC above the control T_{soil} (17 ºC) represents an expected increase of about 5 ºC by 2100

(IPCC 2007b, Solomon et al. 2007). T_{soil} was regulated by circulating temperature controlled water between pots in a control box. See Cheng et al. (2000) for a detailed description. A metal frame with a layer of neutral density shade cloth was placed over half of the seedlings in each T_{soil} treatment to achieve the low light treatment. The shading reduced the PPFD by 70% relative to the high light treatment. The average PPFD in high and low light treatments were about 600 and 180μmol $m^{-2}s^{-1}$, respectively. High-pressure sodium lamps (Model LR48877, P.L. Systems, Grimspy, ON, Canada) were used to supplement the natural light on cloudy days, and lengthen the photoperiod to 16 hours (maximum summer photoperiod for the Thunder Bay region according to Environment Canada Weather Report, 2010).

Other environmental conditions in each greenhouse were 22/16 °C day/night air temperature and relative humidity of 50%. All the environmental conditions were controlled and monitored by Argus environmental control system (Argus, Vancouver, BC, Canada). Nutrients were added to the irrigation water twice a week at a concentration of 100, 15, 57, 6, 6 and 11 mg/L of N, P, K, Ca, Mg and S, respectively (Ingestad 1981, Canham et al. 1996).

Photosynthetic light response

Five seedlings per treatment combination in each CO_2 replicate were randomly selected for light response measurements. The measurement was done between 10–15 h with an open gas exchange system (LI-6400, LI-COR. Inc., Lincoln. Nebraska. USA). One mature leaf was randomly selected from the top of the canopy (4th - 6th leaf) for the measurement. Light levels were changed in seven steps at 1100, 800, 400, 100, 60, 10 and 0μmol m^{-2} s^{-1}. The light was supplied by an internal LED red light source (LI-6400, LI-COR).The [CO_2] was set at 380 and 760 μmol mol^{-1} for the ambient and elevated CO_2 treatments, respectively. Leaf temperature and relative humidity within the leaf chamber were set at 22 °C and 50%, respectively. Light compensation points (LCP) and apparent quantum efficiency (AQE) of photosynthesis were determined using the Photosyn Assistant software (Dundee Scientific, Scotland, UK).

Photosynthetic CO_2 response

Photosynthetic responses to CO_2 concentration (A/C_i curves) were measured on the same seedlings and leaves used for the light response measurement. The measurements were taken at seven [CO_2]: 50, 100, 200, 400, 800, 1000 and 1500 μmol mol^{-1}. The photosynthetic active radiation (PAR), leaf temperature and relative humidity in the leaf chamber were controlled at 600 μ mol m^{-2} s^{-1}, 22 ºC and 50%, respectively. The A/C_i curves were analyzed using the A/C_i curve fitting utility version 1.1 developed by Sharkey *et al.* (2007). From the analyses, the maximum rate of carboxylation (V_cmax), the rate of photosynthetic electron transport (J), triose phosphate utilization (TPU) and dark respiration (R_d) were obtained. Adjustment of the parameters at the set leaf temperature of 22 °C was done to compensate for fluctuations of temperature among measurements due to differences in leaf transpiration rates (Dreyer et al. 2001, Sharkey et al. 2007).

Statistical analysis

Analysis of variance (ANOVA) was used to test the main and interactive effects of [CO_2], T_soil and light regime. The data were analyzed with Data Desk 6.01 statistical software (Data Description 1996). The normality of distribution and homogeneity of variance were examined graphically using probability plots and histograms, respectively. All the data met those two assumptions of ANOVA. Statistical tests with P-values ≤ 0.05 were considered significant. However, due to pre-existing design of the greenhouses that limited the replication of this study to two, P-values ≤ 0.10 were considered marginally significant. Scheffé's *post-hoc* test was performed on significant means. The linear model for the ANOVA is give below:

$Y_{ijkl}= \mu+ C_i +\varpi(i)_j + T_k + CT_{ik} + \beta_{(ik)\,l} + L_m + CL_{im} + TL_{km} + CTL_{ikm} + \varepsilon_{(ijklm)n}$

$i = 1, 2;$ $j = 1, 2;$ $k = 1, 2;$ $l = 1;$ $m = 1, 2;$ $n = 1;$ Where,

Y_{ijkl} = the measured response of the jth replicate of the lth light regime in the kth T_{soil} and the ith CO_2 concentration.

μ = the overall mean.

C_i = the fixed effect of the ith CO_2 concentration.

$\varpi(i)_j$ = the whole plot error due to restriction on the randomization of the $CO_{2.}$

T_k = the fixed effect of the kth T_{soil}.

CT_{ik} = the interaction effect of the kth T_{soil} in the ith CO_2 level.

$\beta_{(ij)l}$ = the sub-plot error due to the restriction on the randomization of the kth T_{soil} regime in the ith CO_2 level.

L_m= the fixed effect of the mth light regime.

CL_{im} = the interaction effect of the mth light regime in the in the ith CO_2 level.

TL_{km} = the interaction effect of the mth light regime in the kth T_{soil}.

CTL_{ikm} = the interaction effect of the mth light regime in the kth T_{soil} and the ith CO_2 level.

$\varepsilon_{(ijkl)}$ = the sub-sub-plot error.

Appendix 1.1. EMS Table

	df	2 F i	2 F k	2 F m	2 R j	EMS
C_i	1	0	2	2	2	$\sigma^2 + 4\sigma^2_{\varpi} + 6\phi C$
$\varpi(i)_j$	2(2-1)=2	1	2	2	2	$\sigma^2 + 4\sigma^2_{\varpi}$
T_k	1	2	0	2	2	$\sigma^2 + 4\sigma^2_{\beta} + 6\phi_T$
CT_{ik}	1	0	0	2	2	$\sigma^2 + 4\sigma^2_{\beta} + 4\phi_{CT}$
$\beta_{(jk)l}$	2x1=2	1	1	2	2	$\sigma^2 + 4\sigma^2_{\beta}$
L_m	1	2	2	0	2	$\sigma^2 + 8\phi_L$
CL_{im}	1	0	2	0	2	$\sigma^2 + 4\phi_{CL}$
TL_{km}	1	2	0	0	2	$\sigma^2 + 4\phi_{TL}$
CTL_{ikm}	1	0	0	0	2	$\sigma^2 + 2\phi_{CTL}$
$\varepsilon_{(ijklm)n}$	2x1x1=2	1	1	1	1	σ^2
Total	12					

Results

There were significant interactive effects between T_{soil} and light, and $[CO_2]$ and light on *A* (Table 1. 1). The high light treatment increased *A* by 41% at the low T_{soil} but had no significant effect on *A* at the warmer T_{soil} (Fig. 1.1A). In the low light treatment, soil warming increased *A* by 18% (Fig. 1.1A). No significant effect of soil warming on *A* was found in the high light treatment although *A* tended to decrease (Fig. 1.1A). Under the ambient $[CO_2]$, the warmer T_{soil} significantly reduced *A* by 17% (Fig. 1.1B). In contrast, soil warming increased *A* by 13% under the elevated $[CO_2]$ (Fig. 1.1B). Furthermore, the elevated $[CO_2]$ increased *A* by 48% and 100% at the low and warmer T_{soil}, respectively (Fig. 1.1B).

The interaction between T_{soil} and light had a marginal significant effect on g_s (Table 1.1). The high light treatment resulted in higher g_s only under the low but not the warmer T_{soil} where no significant light effect on g_s was observed (Fig. 1.1C). Similarly, soil warming increased g_s at the low light treatment but had no significant effect on g_s at the high light treatment (Fig. 1.1C).

There was a significant interactive effect of $[CO_2]$ and light on IWUE (Table 1.1). The high light treatment had no significant effect on IWUE under the ambient $[CO_2]$ but increased IWUE by 18% under elevated $[CO_2]$ (Fig. 1.1D). The elevated $[CO_2]$ increased IWUE by 79 and 125% in the low and high light, respectively (Fig. 1.1D). No interactions among light, T_{soil} and $[CO_2]$ or between T_{soil} and light or $[CO_2]$ were significant (Table 1.1). The elevated $[CO_2]$ significantly reduced C_i/C_a ratio (0.65 at ambient vs. 0.63 at the elevated $[CO_2]$) but no other factors or their interactions significantly affected C_i/C_a (Table 1.1).

V_{cmax}, *J* and J/V_{cmax} ratio were all significantly affected by the interaction between $[CO_2]$ and light (Table 1.1). The elevated $[CO_2]$ changed the response in V_{cmax}, *J* and J/V_{cmax} to light: the high light treatment significantly reduced V_{cmax} by 19%, had no significant effect on *J* but

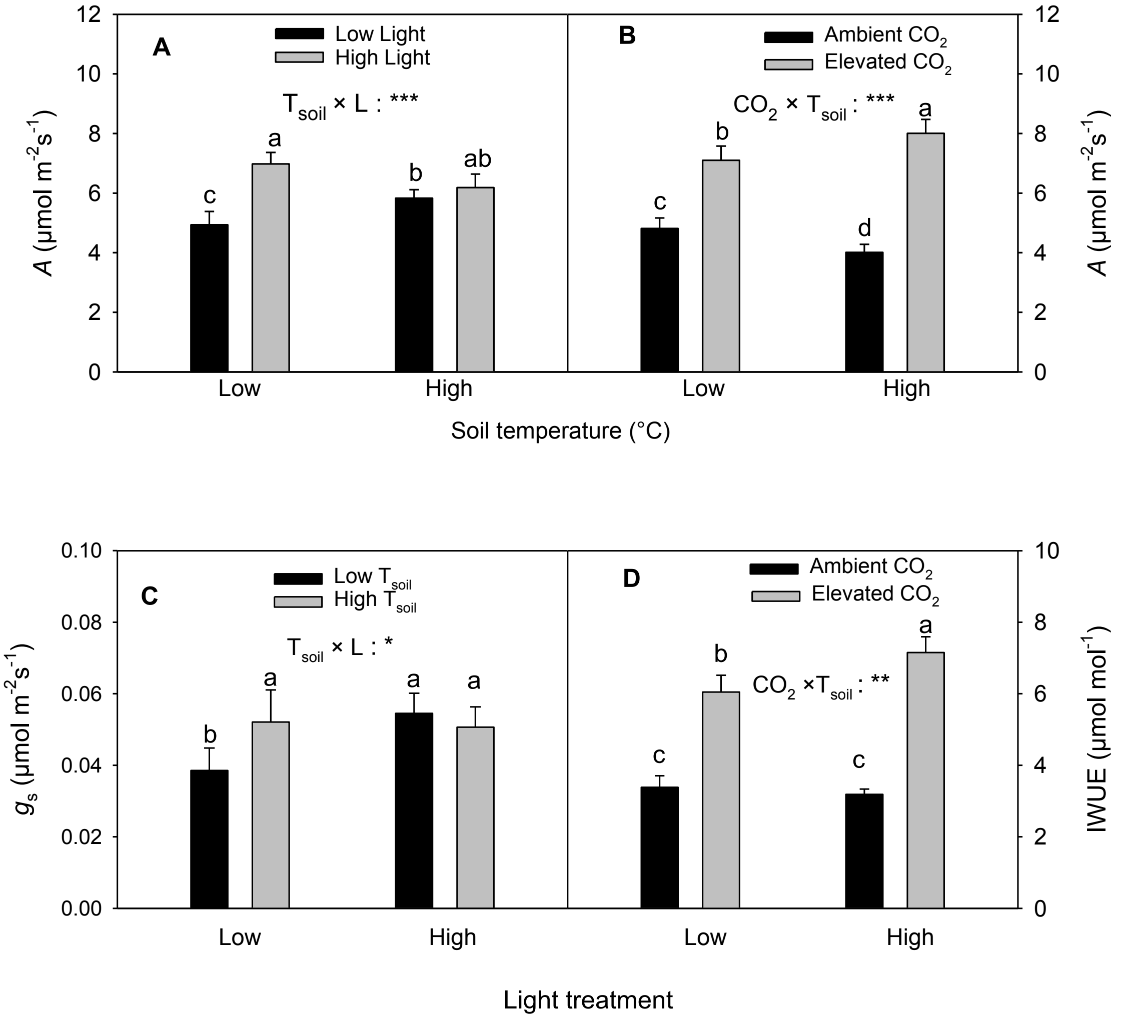

Figure 1.1. Effects of [CO_2], soil temperature (T_{soil}) and light treatment (L) on net photosynthesis (*A*), transpiration rates (*E*) and instantaneous water-use efficiency (IWUE) (mean ± SE, n = 10) of *Acer spicatum*. Seedlings were exposed to two [CO_2] (380 and 760 µmol mol^{-1}), two T_{soil} (17 and 22 °C) and two light treatments (low and high) for two months. The measurements were taken at the corresponding growth [CO_2]. Significant treatment effects are marked as significant: $P \leq 0.01$, ***; $P \leq 0.05$, **; and marginally significant: $P \leq 0.10$, *. Means with same letter(s) are not statistically different ($P > 0.10$).

increased J/V_{cmax} by 20% under elevated [CO_2] (Figs. 1.2A, 1.2B and 1.2C). In contrast, the high light treatment significantly increased V_{cmax} and J by 14 and 15%, but had no significant effect on J/V_{cmax} under the ambient [CO_2] (Figs. 1.2A, 1.2B and 1.2C). The elevated [CO_2] decreased V_{cmax} and J by 29% and 13%, respectively, but increased J/V_{cmax} by 27% in the high but did not have significant effects on any of them in the low light treatment (Figs. 1.2A, 1.2B and 1.2C).

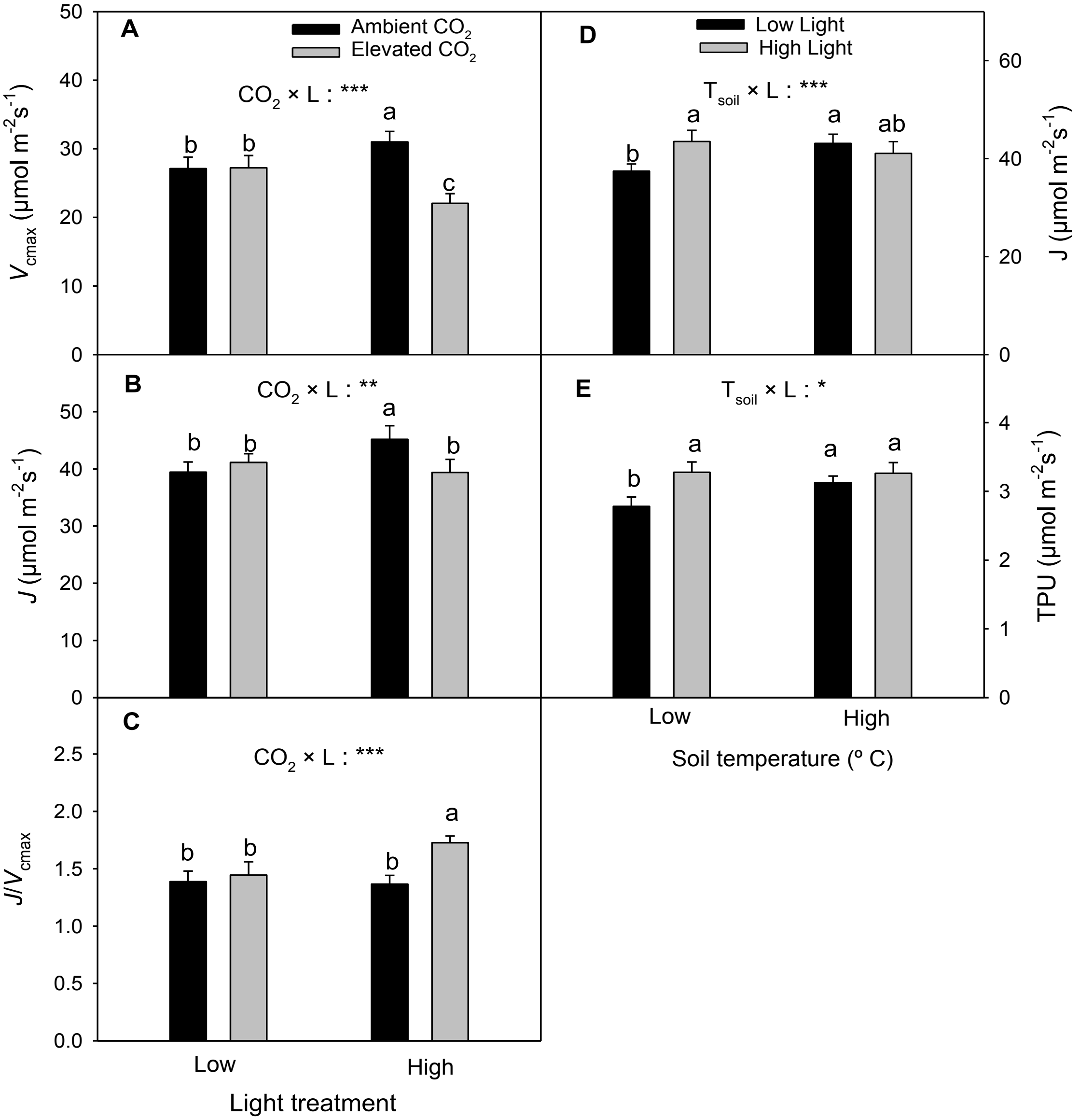

Figure 1.2. Effects of [CO_2], T_{soil} and L on maximum rate of carboxylation (V_{cmax}), rate of photosynthetic electron transport (J), J/V_{cmax} ratio and triose phosphate utilization (TPU) (mean + SE, n = 10) of *Acer spicatum*. Refer to Figure 1.1 for other explanations.

Table 1.1. The *P*-values of ANOVA on the effects of T_{soil} and light (L) on net photosynthesis (A), stomatal conductance (g_s), instantaneous water-use efficiency (IWUE), internal to ambient CO_2 ratio (C_i/C_a), maximum rate of carboxylation (V_{cmax}), rate of photosynthetic electron transport (J), J/V_{cmax} ratio, triose phosphate utilization (TPU), dark respiration (R_d), light compensation point (LCP) and apparent quantum efficiency (AQE) of *Acer spicatum* grown under ambient (380 µmol mol^{-1}) and elevated (760 µmol mol^{-1}) [CO_2]. The seedlings were exposed to 17 and 22° C T_{soil}, and high and low light treatments. Measurements were taken two months after the start of the experiment. Significant differences are highlighted in bold.

Source of variation	CO_2	T_{soil}	CO_2*T_{soil}	L	CO_2*L	$T_{soil}*L$	$CO_2*T_{soil}*L$
A	**0.0065**	0.8071	**0.0114**	**0.0004**	0.6714	**0.0143**	0.9382
g_s	0.7402	0.2021	0.5550	0.2525	0.9746	**0.0694**	0.2424
IWUE	**0.0121**	0.1924	0.4097	**0.0794**	**0.0345**	0.8345	0.9473
C_i/C_a	**0.0403**	0.1465	0.3492	0.1207	0.6408	0.7688	0.3681
V_{cmax}	**0.0088**	**0.0808**	0.9589	0.7091	**0.0078**	0.4507	0.4737
J	0.2312	0.4288	0.4622	0.2529	**0.0331**	**0.0180**	0.4658
J/V_{cmax}	**0.0563**	0.1814	0.4841	**0.0555**	**0.0274**	0.8490	0.6140
TPU	**0.0817**	0.1590	0.7419	**0.0052**	0.3767	**0.0925**	0.4044
R_d	0.1177	0.9318	0.5950	**0.0022**	0.4190	0.9431	0.9800
LCP	**0.0239**	0.9425	0.9227	**≤0.0001**	0.6894	0.7512	0.5888
AQE	**0.0045**	0.9652	0.6799	0.8678	0.2262	0.5753	0.3722

There was a significant interaction between light and T_{soil} on J and a marginal significant interactive effect on TPU (Table 1.1). The high light treatment significantly increased J and TPU by 15% and 13%, respectively, only at the low T_{soil} but had no significant effect on them at the high T_{soil} (Figs. 1.2D and 1.2E). Likewise, soil warming increased *J* and TPU by 16 and 18%, respectively, only in the low but not the high light treatment (Figs. 1.2D and 1.2E). The elevated [CO_2] significantly reduced TPU (3.2 at ambient vs. 3.0 μmol $m^{-2}s^{-1}$ at elevated [CO_2], Table 1.1).

The high light treatment increased R_d by 24% (1.36 at the low light vs. 1.69 μmol $m^{-2}s^{-1}$at the high light, Table 1). However, no other significant effect on R_d was found ($P > 0.10$). The elevated [CO_2] reduced the light compensation point (LCP) of photosynthesis by 21% (14.9 at the ambient vs. 11.7 μmol $m^{-2}s^{-1}$ at the elevated [CO_2], Table 1). Furthermore, the high light treatment increased the LCP by 133% (8.0 at low light vs. 18.6 μmol $m^{-2}s^{-1}$ at high light) (Table 1). The apparent quantum efficiency was significantly increased by elevated [CO_2] (0.045 at the ambient vs. 0.050 mol CO_2 mol^{-1} at the elevated [CO_2]), but no other factors had any significant effect on apparent quantum efficiency (Table 1).

Discussion

Soil warming enhanced the CO_2 stimulation of *A* and reduced the responsiveness of *A*, *J* and TPU to light. Under the elevated [CO_2], *A* increased by 100% at the warmer T_{soil}, but only by 48% at the low T_{soil}. However, there was a significantly reduction of *A* by soil warming under the ambient [CO_2]. Because CO_2 is a substrate for photosynthesis (Lambers et al. 2008), increases in [CO_2] stimulate *A* in C_3 plants (Pearcy 1983, Bunce 1992, Tissue et al. 1997, Curtis and Wang 1998, Saxe et al. 1998, Norby et al. 1999, Sefcik et al. 2006). Elevated [CO_2] also increase the photosynthetic enzyme activity if the increase in [CO_2] is not confounded by low nutrient availability (Long et al. 1996, Zhang and Dang 2006, Ambebe et al. 2010). The higher values of *A* observed under warmer T_{soil} compared to the low T_{soil} could be attributable to enhanced root activity. Water viscosity decreases while roots permeability and growth increase with soil warming (King et al. 1999, Nobel 1999, Boucher et al. 2001). Several authors have reported that soil warming up to a threshold T_{soil} increases water and nutrients absorption (DeLucia et al. 1992, Bassirirad 2000, Dodd et al. 2000, Dong et al. 2001, Weih and Karlsson 2002, Dawes et al. 2011a). More efficient nutrients uptake, coupled with increased substrate (CO_2) availability and enhanced photosynthetic enzyme activity could have led to the greater stimulation of *A* by elevated [CO_2] with soil warming in this study. Ambebe et al. (2010) found similar strong elevated [CO_2] stimulation of *A* with soil warming in white birch (*Betula papyrifera*) seedlings. The reduction of *A* at the warmer soil under ambient [CO_2] is consistent with the findings of (Foster et al. 1991, Ruter and Ingram 1992, Xu et al. 2002). Increased carbon loss through increased roots respiration (Foster et al. 1991, Zogg et al. 1996, Atkin et al. 2000, Pregitzer et al. 2000) might have resulted in the drop under the warmer T_{soil}.

The reduction in *A* stimulation by high light was likely mediated by variations in g_s and morphological acclimation to light at the warmer and low T_{soil}. There was evidence that g_s declined by 3% in response to the high light under the warmer T_{soil}, although the trend was not significant. In contrast, the high light increased g_s by 41% when seedlings were grown at the low T_{soil}. Boucher et al. (2001) also measured lower *A* with decreased g_s in eastern white pine (*Pinus strobus* L.) seedlings under high light and high T_{soil} compared with high light and intermediate T_{soil}. They attributed the decline in *A* and g_s to acclimation to environmental fluctuations of higher amplitude. In this study, the seedlings responded to the high light treatment with thicker leaves (lower SLA, chapter 3 of this thesis) under the warmer than the low T_{soil}. Thicker leaves may have more photosynthetic material per unit leaf area but also more respiratory carbon loss (Boucher et al. 1998, Poorter 1999). The combined effects of high light and warmer T_{soil} on respiratory carbon loss could be partially responsible for the low *A* stimulation by high light under the warmer T_{soil}. This result suggests that soil warming may limit mountain maple response to canopy gaps due to decreased g_s and increased carbon loss.

The elevated [CO_2] enhanced light stimulation of IWUE. The high light treatment increased IWUE by 18% under the elevated [CO_2] but decreased IWUE by 6% under the ambient [CO_2], although the trend was not significant. Decreased g_s, increased *A* or both can improve IWUE (Cowan 1986, Eamus 1991, Drake et al. 1997, Saxe et al. 1998, Lambers et al. 2008, Onoda et al. 2009). However, increases in IWUE may not solely result from photosynthetic responses to experimental treatments, but may also be attributable to leaf morphological acclimations. In this study, g_s did not show significant response to light under either [CO_2], suggesting that the increase in IWUE could be related to increased *A* alone (Samuelson and Seiler 1992, Townend 1993). However, Poorter (1999) found decreased leaf area to root mass ratio (LARMR) in high light environments that balances transpiration with water absorption in seedlings of 15 rain-forest tropical tree species. Consequently, Norby and O´Neil (1991) found that higher WUE under elevated [CO_2] in seedling of yellow poplar (*Liriodendron tulipifera* L.) was caused by decreased LARMR than leaf gas exchange. The leaf area root mass ratio of mountain maple seedlings decreased in response to high light and elevated [CO_2] (chapter 3 of thesis), and thus, might partly be responsible for the increased IWUE observed in this study. The elevated CO_2-mediated increase IWUE may be beneficial in improving mountain maple drought tolerance in canopy gaps, as it is has thinner leaves and is less drought of drought (Paula 2004).

The elevated [CO_2] modified mountain maple photosynthetic acclimation to light. The high light resulted in increased V_{cmax} and *J* under the ambient [CO_2] but the trend for V_{cmax} was reversed under the elevated [CO_2]. Consequently, the J/V_{cmax} ratio was increased by the high light condition under elevated [CO_2]. At a saturating light level, either V_{cmax} or J can limit photosynthetic rates in C_3 plants (Farquhar et al. 1980). However, elevated [CO_2] influences N allocation between V_{cmax} and J such that maximum photosynthesis is achieved. In many cases, elevated [CO_2] decreases N allocation to V_{cmax} because J but not Rubisco limits photosynthesis at high [CO_2] (Stitt 1991, Mitchell et al. 2000, Takeuchi et al. 2001, Lambers et al. 2008, Onoda et al. 2009). The decreased allocation of N to V_{cmax} may be a compensatory response that balances N between non-limiting and limiting processes for maximum carbon gain. The differential responses of V_{cmax} and J to high light under different [CO_2] may reflect a change in N allocation between biochemical and photochemical components of photosynthesis (Hikosaka 2005, Onoda et al. 2005, Onoda et al. 2009). The results may suggest that mountain maple seedlings growing in canopy gaps may improve photosynthetic nitrogen use efficiency by allocating N to rate-limiting component of photosynthetic under future [CO_2].

Elevated [CO_2] increased stomatal factors limitation to A but positively affected light response parameters of A. The C_i/C_a ratio and LCP decreased while apparent quantum yield increased in response to elevated [CO_2]. Reduction in C_i/C_a ratio is generally observed when A is more limited by stomatal than by non-stomatal factors (Cornic 2000, Flexas and Medrano 2002, Dang and Cheng 2004, Flexas et al. 2004, Ambebe and Dang 2009). The accumulation of carbohydrates in leaves due to higher photosynthetic rate under elevated [CO_2] has been found to restrict CO_2 diffusion (Stitt 1991). Although there was no significant reduction of g_s in response to the elevated [CO_2], this result indicates that stomatal limitation to A still reduced the potential stimulation of A by elevated [CO_2]. Increased quantum yield and lower LCP resulting from higher RuBP carboxylation (inhibition of photosynthetic carbon oxidation, PCO) and lower respiration rates have been reported in C_3 plants under elevated [CO_2] (Long and Drake 1991, Kubiske and Pregitzer 1996, Osborne et al. 1997). More efficient photosynthetic light-use and decreased respiratory carbon loss increase *A* in elevated [CO_2] (Kubiske and Pregitzer 1996, Drake et al. 1997). The elevated CO_2-induced decreases in LCP and increases in quantum yield may compensate for the increased stomatal limitation of *A*, and improve plants carbon gain (Jones et al. 1995, Kubiske and Pregitzer 1996, Hättenschwiler et al. 1997, Liang et al. 2001).

In conclusion, soil warming greatly increased the responses of *A* to elevated [CO_2] but did not stimulate the responses of *A* to high light. Soil warming may alleviate low light effect on *A* by influencing RuBP regeneration and TPU of mountain maple. The results suggest that mountain maple may show higher growth rates at resource-rich environments. However, elevated CO_2-induced photosynthetic acclimation may limit *A* responses to high light environments. Increased light use efficiency and reduced carbon loss under elevated [CO_2] could have important implications on the duration of photosynthetic carbon gain in mountain maple seedlings at resource-limited environments, such as under low soil moisture and low light conditions. This study was restricted to short-term responses of mountain maple seedlings and may not exactly reflect the responses of mature trees in the field (Yokota and Hagihara 1996, Pritchard et al. 1999, Bond 2000, Cavender-Bares and Bazzaz 2000, Wieser et al. 2002, Zotz et al. 2005). Therefore, considerable care must be taken in extrapolating the physiological responses of mountain maple seedlings to climate change scenarios. Further long-term experiments with mature mountain maple trees are needed to determine any future response patterns

References

Ambebe, T.F. and Q.-L. Dang. 2009. Low moisture availability inhibits the enhancing effect of increased soil temperature on net photosynthesis of white birch (*Betula papyrifera*) seedlings grown under ambient and elevated carbon dioxide concentrations. Tree Physiology. 29:1341-1348.

Ambebe, T.F., Q.-L. Dang and J. Li. 2010. Low soil temperature inhibits the effect of high nutrient supply on photosynthetic response to elevated carbon dioxide concentration in white birch seedlings. Tree Physiology. 30:234-243.

Archambault, L., J. Morissette and M. Bernier-Cardou. 1998. Forest succession over a 20-year period following clearcutting in balsam fir yellow birch ecosystems of eastern Quebec, Canada. Forest Ecology and Management. 102:61-74.

Atkin, O.K., J.R. Evans, M.C. Ball, H. Lambers and T.L. Pons. 2000. Leaf respiration of snow gum in the light and dark. interactions between temperature and irradiance. Plant Physiology. 122:915-923.

Aubin, I., C. Messier and D. Kneeshaw. 2005. Population structure and growth acclimation of mountain maple along a successional gradient in the southern boreal forest. Ecoscience. 12:540-548.

Bassirirad, H. 2000. Kinetics of nutrient uptake by roots: responses to global change. New Phytologist. 147:155-169.

Bond, B.J. 2000. Age-related changes in photosynthesis of woody plants. Trends in Plant Science. 5:349-353.

Boucher, J.-F., S. Wetzel and A.D. Munson. 1998. Leaf level response of planted eastern white pine (*Pinus strobus* L.) seven years after intensive silvicultural treatments. Forest Ecology and Management. 107:291-307.

Boucher, J.F., P.Y. Bernier and A.D. Munson. 2001. Radiation and soil temperature interactions on the growth and physiology of eastern white pine (*Pinus strobus* L.) seedlings. Plant and Soil. 236:165-174.

Bowes, G. 1993. Facing the inevitable - Plants and increasing atmospheric CO_2 Annual Review of Plant Physiology and Plant Molecular Biology. 44:309-332.

Bunce, J.A. 1992. Stomatal conductance, photosynthesis and respiration of temperate deciduous tree seedlings grown outdoors at an elevated concentration of carbon dioxide. Plant Cell and Environment. 15:541-549.

Caners, R. and N.C. Kenkel. 1997. Uncommon plant species of eastern deciduous floristic affinity, Rising Mountain National Park. UFS (Delta Marsh) Annual Report. Department of Botany, University of Manitoba, pp 80 – 82. .

Canham, C.D. 1988. An index for understory light levels in and around canopy gaps. Ecology. 69:1634-1638.

Canham, C.D., A.R. Berkowitz, V.R. Kelly, G.M. Lovett, S.V. Ollinger and J. Schnurr. 1996. Biomass allocation and multiple resource limitation in tree seedlings. Canadian Journal of Forest Research. 26:1521-1530.

Cavender-Bares, J. and F.A. Bazzaz. 2000. Changes in drought response strategies with ontogeny in *Quercus rubra* : implications for scaling from seedlings to mature trees. Oecologia. 124:8-18.

Chazdon, R.L., R. Pearcy, D. Lee and N. Fetcher. 1996. Photosynthetic responses of tropical forest plants to contrasting light environments. In S. Mulkey, R. L. Chazdon, and A. P Smith [eds.], Tropical forest plant ecophysiology. Chapman and Hall, New York.

Cheng, S., Q.-L. Dang and T.-B. Cai. 2000. A soil temperature control system for ecological research in greenhouses. Journal of Forest Research. 5:205-208.

Coomes, D.A. and P.J. Grubb. 1998. Responses of juvenile trees to above- and belowground competition in nutrient-starved Amazonian rain forest. Ecology. 79:768-782.

Cornic, G. 2000. Drought stress inhibits photosynthesis by decreasing stomatal aperture - not by affecting ATP synthesis. Trends in Plant Science. 5:187-188.

Cowan, I.R. 1986. Economics of carbon fixation in higher plants. In: On the economy of plant form and function. Cambridge University Press, Cambridge, UK.

Curtis, P.S. and X. Wang. 1998. A meta-analysis of elevated CO_2 effects on woody plant mass, form, and physiology. Oecologia. 113:299-313.

Dalling, J.W., K. Winter and S.P. Hubbell. 2004. Variation in growth responses of neotropical pioneers to simulated forest gaps. Functional Ecology. 18:725-736.

Dang, Q.-L. and S. Cheng. 2004. Effects of soil temperature on ecophysiological traits in seedlings of four boreal tree species. Forest Ecology and Management. 194:379-387.

Dawes, M.A., F. Hagedorn, T. Zumbrunn, I.T. Handa, S. Hättenschwiler, S. Wipf and C. Rixen. 2011. Growth and community responses of alpine dwarf shrubs to in situ CO_2 enrichment and soil warming. New Phytologist. 191:806-818.

Day, T.A., S.A. Heckathorn and E.H. Delucia. 1991. Limitations of photosynthesis in *Pinus taeda* (loblolly Pine) at low soil temperatures. Plant Physiology. 96:1246-1254.

De Frenne, P., J. Brunet, A. Shevtsova, A. Kolb, B.J. Graae, O. Chabrerie, S.A. Cousins, G. Decocq, A.N. De Schrijver, M. Diekmann, R. Gruwez, T. Heinken, M. Hermy, C. Nilsson, S. Stanton, W. Tack, J. Willaert and K. Verheyen. 2011. Temperature effects on forest herbs assessed by warming and transplant experiments along a latitudinal gradient. Global Change Biology. 17:3240-3253.

DeLucia, E.H., S.A. Heckathorn and T.A. Day. 1992. Effects of soil temperature on growth, biomass allocation and resource acquisition of *Andropogon gerardii* Vitman. New Phytologist. 120:543-549.

Dodd, I.C., J. He, C.G.N. Turnbull, S.K. Lee and C. Critchley. 2000. The influence of supra-optimal root-zone temperatures on growth and stomatal conductance in Capsicum annuum L. Journal of Experimental Botany. 51:239-248.

Dong, S., C.F. Scagel, L. Cheng, L.H. Fuchigami and P.T. Rygiewicz. 2001. Soil temperature and plant growth stage influence nitrogen uptake and amino acid concentration of apple during early spring growth. Tree Physiology. 21:541-547.

Drake, B.G., M.A. Gonzàlez-Meler and S.P. Long. 1997. More efficient plants: A consequence of rising atmospheric CO_2? Annual Review of Plant Physiology and Plant Molecular Biology. 48:609-639.

Dreyer, E., X. Le Roux, P. Montpied, F.A. Daudet and F. Masson. 2001. Temperature response of leaf photosynthetic capacity in seedlings from seven temperate tree species. Tree Physiology. 21:223-232.

Eamus, D. 1991. The interaction of rising CO_2 and temperatures with water use efficiency. Plant, Cell and Environment. 14:843-852.

Farquhar, G.D., S. Caemmerer and J.A. Berry. 1980. A biochemical model of photosynthetic CO_2 assimilation in leaves of C_3 species. Planta. 149:78-90.

Fitter, A.H., J.D. Graves, G.K. Self, T.K. Brown, D.S. Bogie and K. Taylor. 1998. Root production, turnover and respiration under two grassland types along an altitudinal gradient: influence of temperature and solar radiation. Oecologia. 114:20-30.

Flexas, J., J. Bota, F. Loreto, G. Cornic and T.D. Sharkey. 2004. Diffusive and metabolic limitations to photosynthesis under drought and salinity in C_3 plants. Plant Biology. 6:269-279.

Flexas, J. and H. Medrano. 2002. Drought-inhibition of photosynthesis in C_3 plants: stomatal and non-stomatal limitations revisited. Annals of Botany. 89:183-189.

Foster, W.J., D.L. Ingram and T.A. Nell. 1991. Photosynthesis and root respiration in *Ilex crenata* `Rotundifolia' at supraoptimal root-zone temperatures. HortScience. 26:535-537.

Gifford, R.M. 1992. Interaction of carbon dioxide with growth-limiting environmental factors in vegetation productivity: implications for the global carbon cycle. In Advances in Bioclimatology. Ed. G. Stanhill. Springer-Verlag, New York.

Hallik, L., Ü. Niinemets and O. Kull. 2012. Photosynthetic acclimation to light in woody and herbaceous species: a comparison of leaf structure, pigment content and chlorophyll fluorescence characteristics measured in the field. Plant Biology. 14:88-99.

Harley, P., A. Guenther and P. Zimmerman. 1996. Effects of light, temperature and canopy position on net photosynthesis and isoprene emission from sweetgum (*Liquidambar styraciflua*) leaves. Tree Physiology. 16:25-32.

Hättenschwiler, S. 2001. Tree seedling growth in natural deep shade: functional traits related to interspecific variation in response to elevated CO_2. Oecologia. 129:31-42.

Hättenschwiler, S., F. Miglietta, A. Raschi and C. KöRner. 1997. Thirty years of in situ tree growth under elevated CO_2: a model for future forest responses? Global Change Biology. 3:463-471.

Herrick, J.D. and R.B. Thomas. 1999. Effects of CO_2 enrichment on the photosynthetic light response of sun and shade leaves of canopy sweetgum trees (*Liquidambar styraciflua*) in a forest ecosystem. Tree Physiology. 19:779-786.

Hikosaka, K. 2005. Nitrogen partitioning in the photosynthetic apparatus of *Plantago asiatica* leaves grown under different temperature and light conditions: similarities and differences between temperature and light acclimation. Plant and Cell Physiology. 46:1283-1290.

Houghton, J.T., Y. Ding, J.J. J. J. Griggs and e. al. 2001. Climate Change 2001: The Scientific Basis. . Cambridge University Press, Cambridge.

Ingestad, T. 1981. Nutrition and growth of birch and grey alder seedlings in low conductivity solutions and at varied relative rates of nutrient addition. Physiologia Plantarum. 52:454-466.

IPCC. 2007. Summary for policymakers. In: Solomon, S. et al. (eds), Climate Change 2007: the physical science basis. Contribution of Working Group I to the fourth assessment report of the Intergovernmental Panel on Climate Change.

Jones, M.B., J.C. Brown, A. Raschi and F. Miglietta. 1995. The effects on Arbutus unedo L. of long-term exposure to elevated CO2. Global Change Biology. 1:295-302.

King, J.S., K.S. Pregitzer and D.R. Zak. 1999. Clonal variation in above- and below-ground growth responses of Populus tremuloides Michaux: Influence of soil warming and nutrient availability. Plant and Soil. 217:119-130.

Kitajima, K. 1994. Relative importance of photosynthetic traits and allocation patterns as correlates of seedling shade tolerance of 13 tropical trees. Oecologia. 98:419-428.

Kubiske, M.E. and K.S. Pregitzer. 1996. Effects of elevated CO_2 and light availability on the photosynthetic light response of trees of contrasting shade tolerance. Tree Physiology. 16:351-358.

Lambers, H., S.F. Chapin III and T.L. Pons. 2008. Plant physiological eclogy. Springer, New York.

Landhäusser, S.M., R.W. Wein and P. Lange. 1996. Gas exchange and growth of three arctic tree-line tree species under different soil temperature and drought preconditioning regimes. Canadian Journal of Botany. 74:686-693.

Lei, T.T. and M.J. Lechowicz. 1990. Shade adaptation and shade tolerance in saplings of three *Acer* species from eastern north America. Oecologia. 84:224-228.

Liang, N.H., Y. Tang and T. Okuda. 2001. Is elevation of carbon dioxide concentration beneficial to seedling photosynthesis in the understory of tropical rain forests? Tree Physiology. 21:1047-1055.

Lin, J., P.A. Harcombe, M.R. Fulton and R.W. Hall. 2002. Sapling growth and survivorship as a function of light in a mesic forest of southeast Texas, USA. Oecologia. 132:428-435.

Liozon, R., F.W. Badeck, B. Genty, S. Meyer and B. Saugier. 2000. Leaf photosynthetic characteristics of beech (*Fagus sylvatica*) saplings during three years of exposure to elevated CO_2 concentration. Tree Physiology. 20:239-247.

Long, S.P. and B.G. Drake. 1991. Efeect of the long-term elevation of CO_2 concentration in the field on the quantum yield of photosynthesis of the C_3 sedge, *Scirpus olneyi*. Plant Physiology. 96:221-226.

Long, S.P. and B.G. Drake. 1992. Photosynthetic CO_2 assimilation and rising atmospheric CO_2 concentrations. In Crop Photosynthesis: spatial and temporal determinants. Elsevier Science Publishers B.V., Amsterdam.

Long, S.P., C.P. Osborne and S.W. Humphries. 1996. Photosynthesis, rising atmospheric carbon dioxide concentration and climate change. In SCOPE 56 - Global change: effects on coniferous forests and grasslands. John Wiley and Sons, New York.

Long, S.P. and F.I. Woodward. 1998. Plant and Temperature. The Company of Biologists Limited, Department of Zoology. University of Cambridge, Cambridge.

Lyr, H. and V. Garbe. 1995. Influence of root temperature on growth of <i>Pinus sylvestris , <i>Fagus sylvatica, Tilia cordata and <i>Quercus robur</i>. Trees - Structure and Function. 9:220-223.

Mitchell, J.F., B.S. Manabe, V. Meleshko and T. Tokioka. 1990. Equilibrium climate change - and its implications for the future. In: Climate Change: The IPCC scientific assessment. Cambridge Univ. Press, Cambridge.

Mitchell, R.A.C., J.C. Theobald, M.A.J. Parry and D.W. Lawlor. 2000. Is there scope for improving balance between RuBP-regeneration and carboxylation capacities in wheat at elevated CO_2? Journal of Experimental Botany. 51:391-397.

Niinemets, Ü. 2007. Photosynthesis and resource distribution through plant canopies. Plant, Cell and Environment. 30:1052-1071.

Niinemets, Ü. and J.D. Tenhunen. 1997. A model separating leaf structural and physiological effects on carbon gain along light gradients for the shade-tolerant species *Acer saccharum*. Plant, Cell and Environment. 20:845-866.

Nobel, P.S. 1999. Physicochemical and environmental plant physiology. Academic Press, San Diego, CA, USA.

Norby, R.J. and E.G. O'Neill. 1991. Leaf area compensation and nutrient interactions in CO_2-enriched seedlings of yellow-poplar (*Liriodendron tulipifera* L.). New Phytologist. 117:515-528.

Norby, R.J., S.D. Wullschleger, C.A. Gunderson, D.W. Johnson and R. Ceulemans. 1999. Tree responses to rising CO2 in field experiments: implications for the future forest. Plant Cell and Environment. 22:683-714.

Onoda, Y., K. Hikosaka and T. Hirose. 2005. Seasonal change in the balance between capacities of RuBP carboxylation and RuBP regeneration affects CO_2 response of photosynthesis in *Polygonum cuspidatum*. Journal of Experimental Botany. 56:755-763.

Onoda, Y., T. Hirose and K. Hikosaka. 2009. Does leaf photosynthesis adapt to CO_2-enriched environments? An experiment on plants originating from three natural CO_2 springs. New Phytologist. 182:698-709.

Osborne, C.P., B.G. Drake, J. LaRoche and S.P. Long. 1997. Does long-term elevation of CO_2 concentration increase photosynthesis in forest floor vegetation? Indiana strawberry in a Maryland forest. Plant Physiology. 114:337-344.

Pacala, S.W., C.D. Canham, J. Saponara, J.A.S. Jr, R.K. Kobe and E. Ribbens. 1996. Forest models defined by field measurements: estimation, error analysis and dynamics. Ecological Monographs. 66:1-43.

Paré, D., Y. Bergeron and C. Camiré. 1993. Changes in the forest floor of Canadian southern boreal forest after disturbance. Journal of Vegetation Science. 4:811-818.

Parelle, J., J.P. Roudaut and M. Ducrey. 2006. Light acclimation and photosynthetic response of beech (*Fagus sylvatica* L.) saplings under artificial shading or natural Mediterranean conditions. Annals of Forest Science. 63:257-266.

Pastor, J., R.H. Gardner, V.H. Dale and W.M. Post. 1987. Successional changes in nitrogen availability as a potential factor contributing to spruce declines in boreal North America. Canadian Journal of Forest Research. 17:1394-1400.

Paula, M.P. 2004. Plant Physiologist. U.S. Department of Agriculture, Forest Service, North Central Research Station, Hardwood Tree Improvement and Regeneration Center, 195 Marsteller Street, West Lafayette, IN 47907.

Pearcy, R.W. 1983. The light environment and growth of C_3 and C_4 tree species in the understory of a Hawaiian forest. Oecologia. 58:19-25.

Pearcy, R.W. and O. Björkman. 1983. Physiological effects. In *CO_2 and plants, the response of plants to rising levels of atmospheric carbon dioxide*. AAAS selected symposium 84. Westview Press, Boulder, Colorado.

Platt, W.J. and D.R. Strong. 1989. Special feature - treefall gaps and forest dynamic. Ecology 70:535-576.

Poorter, L. 1999. Growth responses of 15 rain-forest tree species to a light gradient: the relative importance of morphological and physiological traits. Functional Ecology. 13:396-410.

Post, L.J. 1967. Effects of intensity of radiation on height growth, weight increment, and maximum rate of photosynthesis, in mountain maple seedlings. M.Sc. Thesis, University of New Brunswick, Fredericton, New Brunswick.

Post, L.J. 1970. Dry-matter production of mountain maple and balsam fir in northwestern New Brunswick. Ecology. 51:548-550.

Pregitzer, K.S., J.S. King, A.J. Burton and S.E. Brown. 2000. Responses of tree fine roots to temperature. New Phytologist. 147:105-115.

Pritchard, S.G., H.H. Rogers, S.A. Prior and C.M. Peterson. 1999. Elevated CO_2 and plant structure: a review. Global Change Biology. 5:807-837.

Rook, E.J.S. 2002. Shrubs of the northwoods. http://www.rook. org/earl / bwca /nature / shrubs / index.html.

Ruter, J.M. and D.L. Ingram. 1992. High root-zone temperatures influence Rubisco activity and pigment accumulation in leaves of `Rotundifolia' Holly. Journal of the American Society for Horticultural Science. 117:154-157.

Samuelson, L.J. and J.R. Seiler. 1992. Fraser fir seedling gas exchange and growth in response to elevated CO_2. Environmental and Experimental Botany. 32:351-356.

Saxe, H., D.S. Ellsworth and J. Heath. 1998. Tree and forest functioning in an enriched CO_2 atmosphere. New Phytologist. 139:395-436.

Sefcik, L.T., D.R. Zak and D.S. Ellsworth. 2006. Photosynthetic responses to understory shade and elevated carbon dioxide concentration in four northern hardwood tree species. Tree Physiology. 26:1589-1599.

Sharkey, T.D., C.J. Bernacchi, G.D. Farquhar and E.L. Singsaas. 2007. Fitting photosynthetic carbon dioxide response curves for C3 leaves. Plant, Cell & Environment. 30:1035-1040.

Solomon, S., D. Qin, M. Manning, Z. Chen, M. Marquis, K.B. Averyt, M. Tignor and H.L. Miller. 2007. *Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change*. . Cambridge University Press, Cambridge.

Stitt, M. 1991. Rising CO_2 levels and their potential significance for carbon flow in photosynthetic cells. Plant, Cell and Environment. 14:741-762.

Stoneman, G.L. and B. Dell. 1993. Growth of *Eucalyptus marginata* (Jarah) seedlings in a greenhouse in response to shade and soil temperature Tree Physiology. 13:239-252.

Sullivan, J. 1993. *Acer spicatum*. In: U.S. Department of agriculture, Forest Service, Rocky Mountain Research Station. Fire Sciences Laboratory. Fire effects information system.

Takeuchi, Y., M.E. Kubiske, J.G. Isebrands, K.S. Pregtizer, G. Hendrey and D.F. Karnosky. 2001. Photosynthesis, light and nitrogen relationships in a young deciduous forest canopy under open-air CO_2 enrichment. Plant, Cell & Environment. 24:1257-1268.

Tissue, D.T., R.B. Thomas and B.R. Strain. 1997. Atmospheric CO_2 enrichment increases growth and photosynthesis of *Pinus taeda*: a 4 year experiment in the field. Plant, Cell & Environment. 20:1123-1134.

Townend, J. 1993. Effects of elevated carbon dioxide and drought on the growth and physiology of clonal Sitka spruce plants (*Picea stichensis* (Bong) Carr). Tree Physiology. 13:389-399.

Wan, X.C., S.M. Landhausser, J.J. Zwiazek and V.J. Lieffers. 1999. Root water flow and growth of aspen (*Populus tremuloides*) at low root temperatures. Tree Physiology. 19:879-884.

Weih, M. and P.S. Karlsson. 2002. Low winter soil temperature affects summertime nutrient uptake capacity and growth rate of mountain birch seedlings in the subarctic, Swedish lapland. Arctic, Antarctic, and Alpine Research. 34:434-439.

Wieser, G., K. Tegischer, M. Tausz, K.-H. Häberle, T.E.E. Grams and R. Matyssek. 2002. Age effects on Norway spruce (*Picea abies*) susceptibility to ozone uptake: a novel approach relating stress avoidance to defense. Tree Physiology. 22:583-590.

Wigley, T.M.L. 2005. The Climate Change Commitment. Science. 307:1766-1769.

Xu, Q., B. Huang and Z. Wang. 2002. Photosynthetic responses of creeping bentgrass to reduced root-zone temperatures at supraoptimal air temperature. Journal of the American Society for Horticultural Science. 127:754-758.

Yokota, T. and A. Hagihara. 1996. Dependence of the aboveground CO_2 exchange rate on tree size in field-grown hinoki cypress (*Chamaecyparis obtusa*). Journal of Plant Research. 109:177-184.

Zhang, S. and Q.-L. Dang. 2006. Effects of carbon dioxide concentration and nutrition on photosynthetic functions of white birch seedlings. Tree Physiology. 26:1457-1467.

Zipperlen, S.W. and M.C. Press. 1996. Photosynthesis in relation to growth and seedling ecology of two dipterocarp rain forest tree species. Journal of Ecology. 84:863-876.

Zogg, G.P., D.R. Zak, A.J. Burton and K.S. Pregitzer. 1996. Fine root respiration in northern hardwood forests in relation to temperature and nitrogen availability. Tree Physiology. 16:719-725.

Zotz, G., S. Pepin and C. Körner. 2005. No Down-Regulation of Leaf Photosynthesis in Mature Forest Trees after Three Years of Exposure to Elevated CO2. Plant Biology. 7:369-374.

Chapter 3: Effects of elevated [CO_2] and soil warming on the morphological responses of mountain maple (*Acer spicatum* L) seedlings to light.

Introduction

Plant acclimated to low light conditions shows adaptive morphological and physiological traits that maximize light interception for improved carbon gain. The fraction of biomass allocated to leaf (LMR) is higher in low light environment. The leaf area per unit leaf mass (specific leaf area, SLA) increases under shaded environment. As a result, the product of the two traits, leaf area ratio (LAR), is generally greater in low compared with high light environment (Anten and Hirose 1998, Boucher et al. 2001, Poorter et al. 2012, Semchenko et al. 2012). Plants in low light environments often have slow growth rates but can increase their growth as light increases with the removal of the overstory vegetation (Clark and Clark 1992, Poorter 1999). Some plants growing in low light environment exhibit growth habit that enable them to discover and exploit high light environments (Rincon and Grime 1989, Lei and Lechowicz 1990). In natural habitats, the light acclimation differs between low and high light environments. Alternatively, plants are growing in high light environments should show opposite acclimation relative to those growing in low light environments. For example, acclimation to high light environments would be predicted to shift more resources into roots that are associated with water and nutrients absorption (Grime 1994, Pearcy and Sims 1994, Poorter 1999, Boucher et al. 2001). Morphological and structural attributes such as smaller-sized and thicker leaves, with low SLA and LAR, and high root-shoot ratio are among the acclimatory responses to high light environments (Poorter 1999, Sultan 2000, Ray et al. 2004, Lambers et al. 2008). Thus, plants are expected to acclimate to changing light environments by partitioning biomass among various plants parts to optimize below and above resource capture for maximum growth (Hibbs et al. 1980, Dewar 1993, McConnaughay and Coleman 1999). However, other environmental factors may influence plants ability to acclimate to changes in light environment and thus the dynamics and composition of forests.

The atmospheric carbon dioxide concentration ([CO_2]) is predicted to continue to increase, reaching between 730 – 1200 μmol mol^{-1} by the year 2100 (IPCC 2007a, Meehl et al. 2007). As a result of the increases in [CO_2], both air and soil temperatures are expected to rise (Adams et al. 1990, Houghton 1992, IPCC 2001, Wigley 2005, IPCC 2007b). Increases in [CO_2] enhance plant growth and biomass accumulation through its direct stimulation of photosynthesis under adequate nutrients supplies (Bazzaz and Miao 1993, Drake et al. 1997, Curtis and Wang 1998, Wand et al. 1999). Elevated [CO_2] enhances plants establishment and growth in their physical and biological environments through improved carbon balance because of decrease light compensation point of photosynthesis (Osborne et al. 1997, Liang et al. 2001), increases light use efficiency (Naumburg and Ellsworth 2000). Hättenschwiler and Körner (2000) have shown that growth and biomass responses of *Acer pseudoplatanus*, *Quercus robur* and *Abies alba* to high light increases more under elevated [CO_2]. The enhanced growth responses to light, under different [CO_2] could alter species distribution and composition in the ecosystem in the future.

Few studies have addressed growth and biomass responses to light under soil warming (Boucher et al. 2001). Soil temperature is a key factor in the boreal forest that limits the growth of plants (Tamm 1991). Soil warming has been shown to increase plant growth and biomass production (Rustad et al. 2001, Peng and Dang 2003, Walker et al. 2006, Dawes et al. 2011a, Ambebe et al. *in press*). Plants growth and survival responses to light generally depend on other resources such as soil nutrients (Canham et al. 1996, Coomes and Grubb 2000, Catovsky and Bazzaz 2002). Positive correlations between warming and biochemical and ecological processes such as increased rate of organic matter decomposition (Jansson and Berg 1985, Hobbie 1996, Melillo et al. 2002), nitrogen (N) mineralization (MacDonald et al. 1995), and water and nutrient uptake (Bowes 1991, DeLucia et al. 1992, Karlsson and Nordell 1996, Bassirirad 2000, Rennenberg et al. 2006) indicate the significance of warming on plants growth. There is paucity of information in this area despite the importance of soil warming on plants growth and survival.

This study examined the interactive effects of [CO_2] and T_{soil} on the growth and biomass responses of mountain maple to light. Mountain maple (*Acer spicatum* L.) is an important shrub or tree species in the boreal forest understory of North America (Sullivan 1993, Aubin et al. 2005). It survives and grows slowly under the forest canopy for a long period of time, but can rapidly dominate canopy gaps caused by the removal of overstory vegetation (Lei and Lechowicz 1997a, Archambault et al. 1998, Bergeron 2000, Rook 2002, Aubin et al. 2005). Increased light availability in canopy gaps results in increases in T_{soil} (Breshears et al. 1998), which may make it difficult to separate the effects of light and T_{soil} on plant growth particularly under field conditions (Stoneman 1992, Boucher et al. 1998). Boucher et al. (2001) hypothesized that a significant part of the growth responses of eastern white pine seedlings to light was actually due to a change in T_{soil}. Despite the predictions of simultaneous increases in [CO_2] and T_{soil}, we still lack a good understanding of how increasing [CO_2] and soil warming may together affect mountain maple's growth responses to light. The effects of elevated [CO_2] and soil warming on the growth response of mountain maple to canopy gaps may have important ecological implications. We hypothesize that elevated [CO_2] and soil warming would cause a greater enhancement of growth and biomass responses of mountain maple to light.

Materials and Methods

Plant material

Seeds of mountain maple (*Acer spicatum*) were collected from Jack Haggerty Forest in Thunder Bay, Ontario, Canada (48°22′56″ N, 89°14′46″ W). Seeds were soaked in a 1000µmol m^{-1} giberellic acid (GA) for 24hr and stratified at 4°C for 2 months. The hard seed coats were gently cracked opened after 2 months to facilitate germination. Germination was carried out on a 2:1 mixture of vermiculite and peat moss in horticultural trays at Lakehead University greenhouse. Average environmental conditions during germination were as follows; temperature 22/16 °C (day/night), relative humidity (RH) of 50% and 16hr photoperiod (summertime day length for Thunder Bay, ON. based on Environment Canada Weather Report, 2010). 160 similar-sized seedlings (about 8cm tall) were transplanted into plastic pots (31.5 cm deep, 11 cm top diameter and 9.5 cm bottom diameter) three weeks after germination. The pots were filled with the same composition of growing medium used in the germination process.

Experimental design

The experiment followed a split-split- plot design. The main-plot treatment comprised of two levels of CO_2 (392 and 784 µmol mol^{-1}) with two independent replications for each level. The elevated [CO_2] (760 µmol mol^{-1}) was achieved using Argus CO_2 generators (Argus, Vancouver, BC, Canada). The [CO_2] in each greenhouse was monitored using Argus environmental control system (Argus, Vancouver, BC, Canada). The sub-plot treatment consisted of two T_{soil}, low (17 ºC) and high (22 ºC), in each greenhouse. The low T_{soil} represented the mean (July) growing season T_{soil} for mountain maple (Caners and Kenkel 1997). The high T_{soil} represented an assumed future T_{soil} if an increase of 5 ºC in T_{soil} occurs by end of 2100 (IPCC 2007b, Solomon et al. 2007). T_{soil} was achieved by circulating temperature-controlled water between pots in a T_{soil}

control box, (See Cheng et al. (2000) for a detailed description). The sub-sub-plot treatment consisted of two light levels (low and high) at each T_{soil}. Each T_{soil} control box was divided into two halves with one half unshaded and the other shaded. The shading was achieved suing neutral density shading cloth mounted on metal frames. The shading reduced the PAR by 70% relative to the unshaded light treatment. High-pressure sodium lamps (Model LR48877, P.L. Systems, Grimspy, ON, Canada) were mounted above the T_{soil} control boxes to provide supplemental light on cloudy days and to extend the photoperiod to 16hr.

The environmental conditions were controlled and monitored by an Argus environmental control system (Argus, Vancouver, BC, Canada). Nutrients were added to irrigation water twice a week at a concentration of 100, 15, 57, 6, 6 and 11 mg/L of N, P, K, Ca, Mg and S, respectively. The nutrient concentrations were determined based on the studies of (Canham et al. 1996) and (Ingestad 1981) on *Acer* species and other deciduous tree species. However, supplemental watering was done depending on the moisture content readings on an HH2 moisture meter at the different treatment combinations.

Growth and biomass measurements

After two months of the treatments, five seedlings per treatment combination were measured for height (*H*) and root collar diameter (RCD). The seedlings were harvested and fractioned into leaves, stems, and roots. The total leaf area per seedling was measured using WinFolia (Regent Instrument Inc., Quebec, Canada). The samples were dried to a constant weight at 70 ºC for 48hrs. Specific leaf area (SLA) and root-shoot ratio (RSR) were calculated by dividing the total leaf area by dry leaf mass and by dividing dry root mass by dry shoot mass, respectively. Other indices of biomass allocation calculated were leaf mass ratio (LMR) and root mass ratio (RMR). Leaf area ratio (LAR) was calculated as a product of SLA and LMR.

Statistical analysis

The data were analyzed with Data desk 6.01 Statistical Package k (Data Description 1996). The assumptions of normality of distribution and homogeneity of variance were examined graphically using probability plots and histograms of the residuals, respectively, before the Analysis of Variance (ANOVA) was done. The data met both assumptions. Three-way split-split-plot ANOVA was used to test the effects of CO_2, T_{soil} and light treatments and their interactions. *P*-values ≤ 0.05 were considered significant and *P*-values ≤ 0.10 were considered marginally significant due to pre-existing design of the greenhouses that limited the replication of this study. Scheffé's *post-hoc* test was performed when an interaction was significant.

Results

The interaction between CO_2 and soil warming had a significant effect on seedling growth responses to light treatments (Table 2.1). While the high light treatment resulted in significantly greater *H* and RCD in both [CO_2] and at both T_{soil}, the magnitude of increase was smallest in the elevated [CO_2] and high T_{soil} and in the ambient [CO_2] and high T_{soil} treatment combinations for *H*, and in the ambient [CO_2] and low T_{soil} for RCD (Figs. 2.1A and 2.1B). The response of *H* to soil warming was modified by [CO_2] and light interaction: the warmer T_{soil} generally resulted in a greater *H*, but *H* appeared to show an opposite trend (statistically no-significance) in the elevated [CO_2] and high light treatment combination (Fig. 2.1A). The elevated [CO_2] significantly increased *H* in all T_{soil}-light treatment combinations except in the high T_{soil} and high light treatment combination where no significant [CO_2] effect on *H* was observed (Fig. 2.1A).

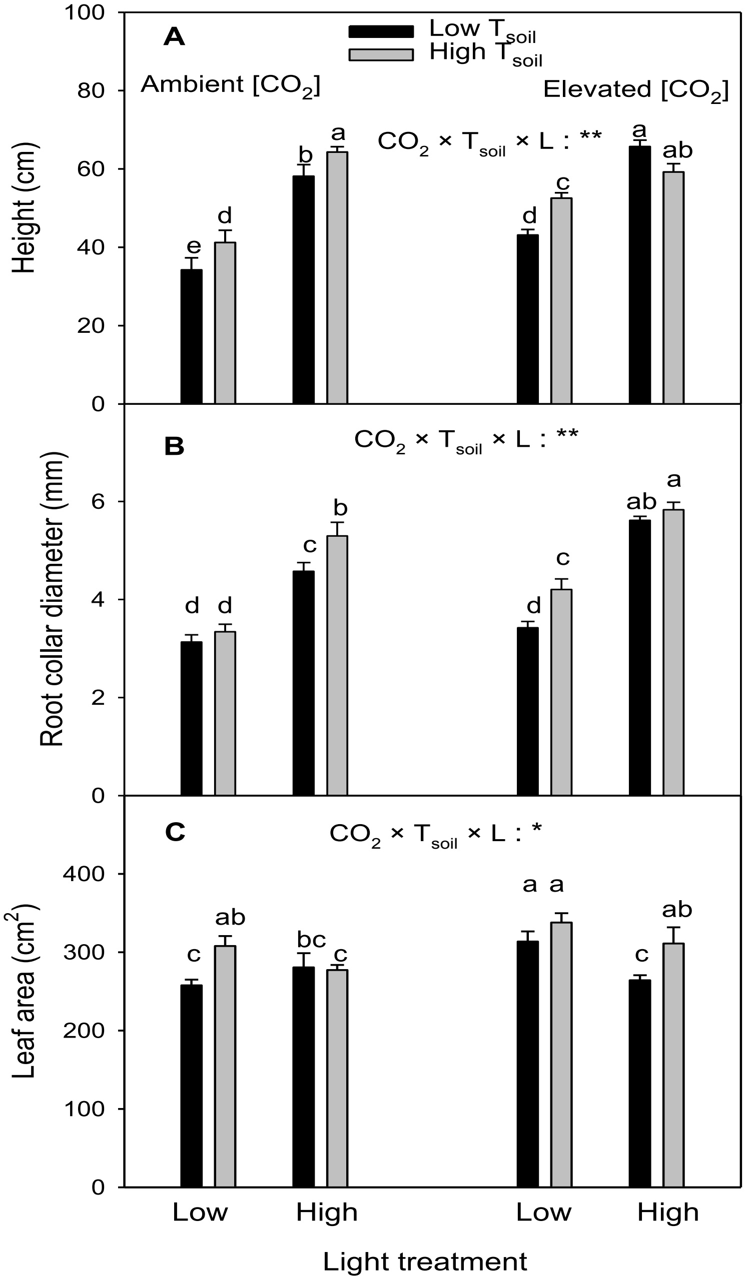

Figure 2.1. Effects of [CO_2], soil temperature (T_{soil}) and light treatment (L) on seedling height (*H*), root collar diameter (RCD) and leaf area (LA) (mean ± SE, n = 10) of *Acer spicatum*. Seedlings were exposed to two [CO_2] (380 and 760 μmol mol^{-1}), two T_{soil} (17 and 22 °C) and two light treatments (high and low) for two months. Significant treatment effects are marked as significant: $P \leq 0.01$, ***; $P \leq 0.05$, **; and marginally significant: $P \leq 0.10$, *. Bars with same letter(s) are not significant different ($p > 0.10$) from each other or one another.

RCD significantly increased with soil warming in the high light and ambient [CO_2] and in the low light and elevated [CO_2] treatment combinations. However, RCD showed no significant response to soil warming in the low light and ambient [CO_2] or in high light and elevated [CO_2] treatment combinations (Fig. 2.1B). The elevated [CO_2] significantly increased RCD in all T_{soil}-light combinations except in the low light and low T_{soil} where no significant [CO_2] effect on RCD was observed (Fig. 2.1B).

There was a marginally significant interactive effect among [CO_2], Tsoil and light on leaf area (LA) (Table 2.1). LA decreased in response to the high light treatment at the warmer T_{soil} under the ambient [CO_2] but the trend was the opposite under the elevated [CO_2] (Fig. 2.1C). There was no significant of high light on LA at low T_{soil} under the ambient [CO_2] or at the warmer T_{soil} under the elevated [CO_2] (Fig. 2.1C). Soil warming resulted in significantly greater LA in the low light under ambient [CO_2] but in the high light treatment under the elevated [CO_2] (Fig. 2.1C). Furthermore, the elevated [CO_2] increased LA in the low light treatment under the low T_{soil} and in the high light treatment under the warmer T_{soil} (Fig. 2.1C).

The interaction among [CO_2], T_{soil} and light significantly affected LAR and SLA and they showed similar patterns of responses (Table 2.1). LAR and SLA generally decreased in response to high light under both [CO_2] and at both T_{soil} (Figs. 2.2A and 2.2B). The magnitude of reduction was greatest under the elevated [CO_2] and low T_{soil} and under the ambient [CO_2] and warmer T_{soil} but lowest under the ambient [CO_2] and low T_{soil} and under the elevated [CO_2] and warmer T_{soil} for both LAR and SLA. Soil warming significantly increased LAR only under the elevated [CO_2] and high light but decreased LAR under the ambient [CO_2] and high light (Fig.

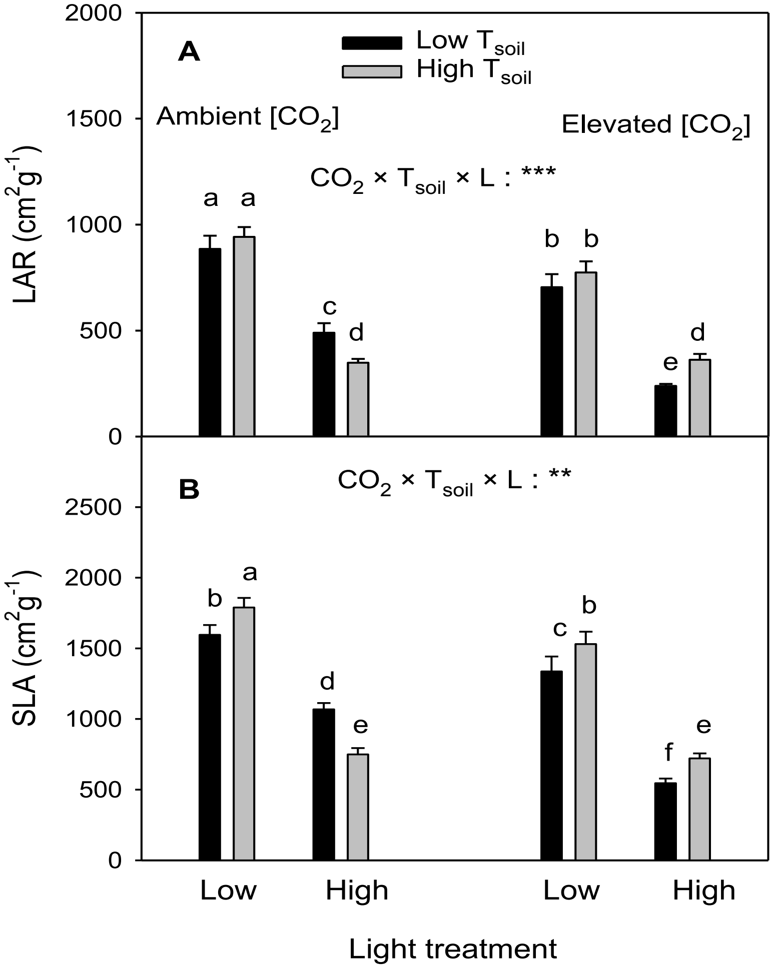

Figure 2.2. Effects of [CO_2], T_{soil} and L on specific leaf area (SLA) and leaf area ratio (LAR) (mean ± SE, n = 10) of *Acer spicatum*. Refer to Figure 2.1 for other explanations.

2.2B). No significant T_{soil} effect on LAR was observed under either [CO_2] and at the low light (Fig. 2.2B). Soil warming significantly increased SLA at all [CO_2]-light treatment combinations except under the ambient [CO_2] and high light (Fig. 2.2B). The elevated [CO_2] significantly decreased LAR and SLA in all T_{soil} – light treatment combinations except under the warmer T_{soil} and low light where no significant [CO_2] effect on LAR and SLA was observed (Figs. 2.2A and 2.2B).

Table 2.1. Analysis of variance (ANOVA) of main treatments effects and treatment interactions on height (*H*), root collar diameter (RCD), leaf area (LA), specific leaf area (SLA), leaf area ratio (LAR), total seedling biomass (TB), root: shoot ratio (RSR), leaf mass ratio (LMR) and root mass ratio (RMR) in mountain maple (*Acer spicatum*) seedlings grown under ambient (380 µmol mol^{-1}) or elevated (760 µmol mol^{-1}) [CO_2], 17 or 22° C soil temperature (T_{soil}), and high (100%) or low (30%) light (L) environment. Measurements were taken two months after the start of the treatment. Significant effects ($P \leq 0.10$) are in bold.

Source of variation	CO_2	T_{soil}	CO_2*T_{soil}	L	CO_2*L	T_{soil}*L	CO_2*T_{soil}*L
H	0.1159	**0.0211**	0.1366	**≤ 0.0001**	**0.0123**	**0.0173**	**0.0353**
RCD	**0.0571**	**0.0004**	0.8124	**≤0.0001**	0.3895	0.9751	**0.0453**
LA	0.2907	**0.0148**	0.5831	**0.0288**	0.1176	0.5634	**0.0606**
SLA	**0.0201**	0.1897	**0.0110**	**≤0.0001**	0.8685	**0.0061**	**0.0099**
LAR	**0.0771**	0.3946	**0.0304**	**≤0.0001**	0.3761	0.2576	**0.0493**
TB	**0.0891**	**0.0021**	0.3196	**≤ 0.0001**	0.1428	0.6795	**0.0046**
RSR	0.1629	0.3632	**0.0289**	**≤ 0.0001**	0.9494	0.2567	**0.0176**
LMR	0.2865	0.1582	0.9103	**≤0.0001**	**≤ 0.0001**	**0.0006**	0.9951
RMR	**0.0058**	**0.0288**	0.1066	**≤0.0001**	0.9408	0.2235	**0.0831**

The interaction between CO_2 and T_{soil} affected seedling biomass response to light (Table 2.1). While the high light generally increased seedling biomass at all [CO_2]-T_{soil} treatment combinations, the magnitude of increase was greatest under the elevated [CO_2] and low T_{soil} and under the ambient [CO_2] and warmer T_{soil} but lowest under the elevated [CO_2] and warmer T_{soil} and under the ambient [CO_2] and low T_{soil} (Fig. 2.3A). Soil warming significantly increased seedling biomass under the ambient [CO_2] and high light and under the elevated [CO_2] and low light treatment combinations (Fig. 2.3A). No significant T_{soil} effect on seedling biomass was observed under the ambient [CO_2] and low light or under the elevated [CO_2] and high light (Fig. 2.3A). The elevated [CO_2] increased seedling biomass at the warmer T_{soil} and low light and at the low T_{soil} and high light treatment combinations (Fig. 2.3A). However, the [CO_2] did not affect biomass at the low T_{soil} and low light or at the warmer T_{soil} and high light (Fig. 2.3A).

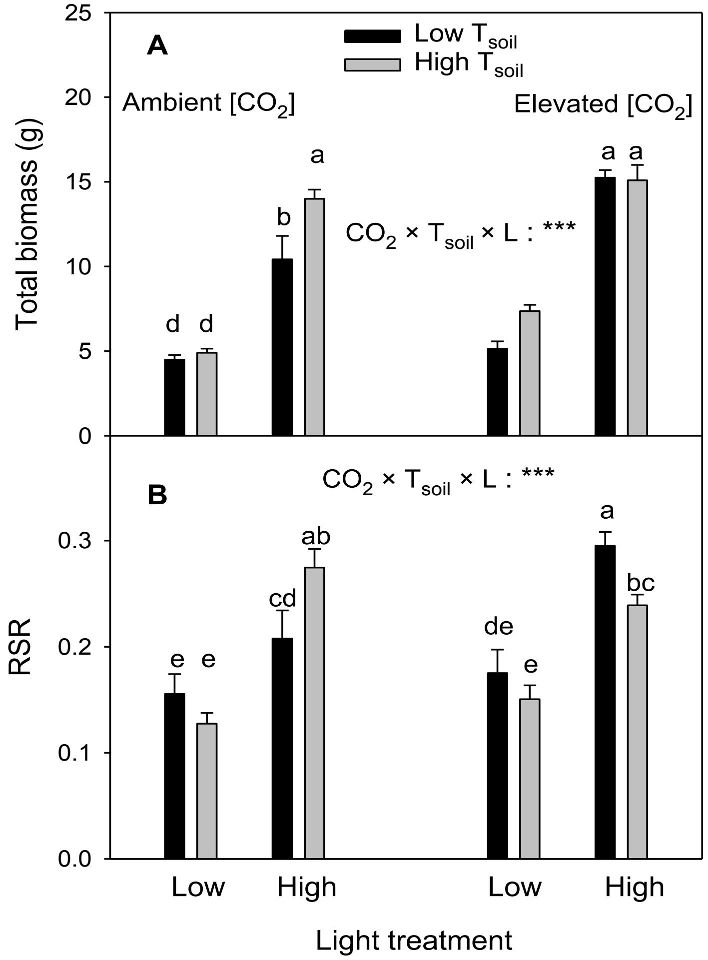

Figure 2.3. Effects of [CO_2], T_{soil} and L on total seedling biomass and root-shoot ratio (RSR) (mean ± SE, n = 10) of *Acer spicatum*. Refer to Figure 2.1 for other explanations.

The three-way interaction among [CO_2], T_{soil} and light had a significant effect on root-shoot ratio (RSR) (Table 2.1). The high light generally resulted in a higher RSR at all [CO_2]-T_{soil} treatment combinations (Fig. 2.3B). RSR increased in response to high light in the order of ambient [CO_2]-low T_{soil} (34%) < elevated [CO_2]-warmer T_{soil} (59%) < elevated [CO_2]-low T_{soil} (68%) < ambient [CO_2]-warmer T_{soil} (116%). The soil warming significantly RSR at the high light under ambient [CO_2] but decreased it at the corresponding light under elevated [CO_2] (Fig.

2.3B). No significant T_{soil} effect on RSR was observed at the low light under either [CO_2] (Fig. 2.3B). The elevated [CO_2] significantly increased RSR only at the low T_{soil} under high light but had no significant effect on RSR at the other treatment combinations (Fig. 2.3B).

The LMR was significantly affected by the interactions between T_{soil} and light, and [CO_2] and light (Table 2.1). The high light treatment generally decreased LMR at both T_{soil} but the magnitude of decrease was greater at the low than the high T_{soil} (-21% vs. -9%, Fig. 2.4A). Soil warming significantly increased LMR by 11% in the high but not the low light treatment where no significant warming effect on LMR was found (Fig. 2.4A). The high light treatment generally resulted in a significantly lower LMR under both [CO_2] but the reduction was greater under the ambient than under the elevated [CO_2] (-23% vs. -7%, Fig. 2.4B). The elevated [CO_2] significantly reduced LMR in the low light but increased it at the high light treatment (Fig. 2.4B).

There was a marginal significant interactive effect between [CO_2] and T_{soil} on the response of RMR to light (Table 2.1). The high light generally increased RMR under both [CO_2] and T_{soil} treatments but the magnitude of increase was greatest under the ambient [CO_2] and warmer T_{soil} (Fig. 2.4C) and lowest under the ambient [CO_2] and low T_{soil} treatment combinations. Soil warming significantly decreased RMR under all [CO_2]-light treatment combinations except under ambient [CO_2] and high light where RMR showed no significant response to T_{soil} (Fig. 2.4C). The elevated [CO_2] significantly increased RMR only at the low T_{soil} in the high light treatment (Fig. 2.4C).

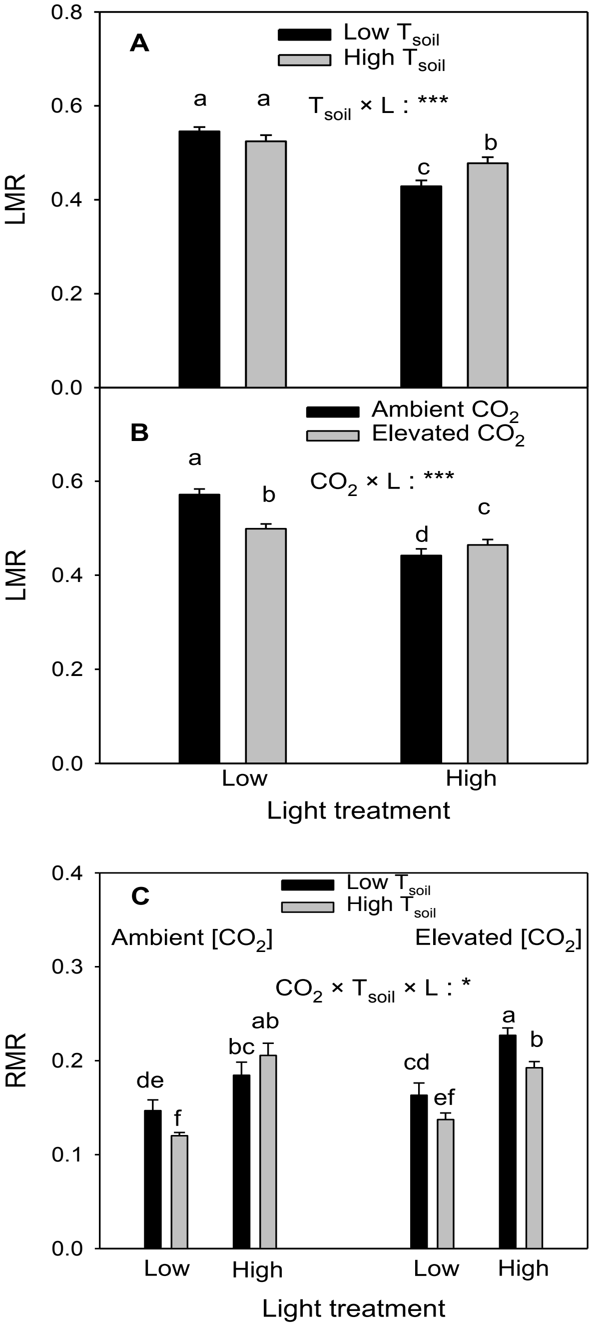

Figure 2.4. Effects of $[CO_2]$, T_{soil} and L on leaf mass ratio (LMR), stem mass ratio (SMR) and root mass ratio (RMR) (mean ± SE, n = 10) of *Acer spicatum*. Figs. 4A and 4B represent $T_{soil} \times L$ interaction. Fig. 4C represents $CO_2 \times L$ interaction while Fig. 4D represents $CO_2 \times T_{soil} \times L$ interaction. Figs. 4A and 4B are means of pooled $[CO_2]$ treatments and Fig. 4C is means of pooled T_{soil} treatments. Refer to Figure 2.1 for other explanations.

Discussion

Elevated [CO_2] and soil warming influenced growth responses of mountain maple seedlings to light. The results show that soil warming under ambient [CO_2], and the control T_{soil} under elevated [CO_2] resulted in maximum height growth stimulation by light, and that, the minimum growth stimulation by light occurred at the elevated [CO_2] and warmer T_{soil}. Specifically, height growth stimulation by the high light treatment decreased in the order of ambient [CO_2]-low T_{soil} (70%) > ambient [CO_2]-warmer T_{soil} (56%) > elevated [CO_2]-low T_{soil} (52%) > elevated [CO_2]-warmer T_{soil} (13%). The minimum RCD growth stimulation by high light also occurred under elevated [CO_2] and soil warming. In contrast to the maximum height growth stimulation by light, the greatest RCD stimulation by high light occurred at the elevated [CO_2]-low T_{soil} treatment combination. The results contrast with the hypothesis that mountain maple would show larger growth responses to high light under elevated [CO_2] and warmer T_{soil}. The results suggest that the interaction between elevated [CO_2] and soil warming did not improve seedling growth and may create physiological constraints that limit growth responses to high light environments. The results could mean that mountain maple seedlings establishment in canopy gaps may be limited under future climate change scenarios of higher [CO_2] and warmer T_{soil}. The low growth stimulation by light under elevated [CO_2] and soil warming may influence forest composition and dynamics in the future since mountain maple is known to rapid canopy gaps after the removal of overstory vegetation (Archambault et al. 1998, Bergeron 2000, Rook 2002, Aubin et al. 2005). This may have important ecological implications given that mountain maple competitively suppresses the growth and establishment of other tree or shrub species (Post 1970, Aubin et al. 2005). However, elevated [CO_2] and soil warming can ameliorate the high light-induced reduction of mountain maple seedling leaf growth from 9% to 7%, which can potentially facilitate continued growth under high light environments.

The experimental treatments led to changes in leaf traits, with reductions in LAR and SLA. LAR and SLA are known to correlate with growth rate, being low in slow growing species and high in fast growing species (Rice and Bazzaz 1989a, Poorter and Remkes 1990, Lambers and Poorter 1992, Ray et al. 2004, Lambers et al. 2008). These leaf traits decrease with increasing light availability (Anten and Hirose 1998, Boucher et al. 2001, Poorter et al. 2012) and elevated [CO_2] (Norby and O'Neill 1991, Roumet and Roy 1996, Temperton et al. 2003), but increase with soil warming (Weih and Karlsson 2001). In this study, the greatest reductions in these leaf traits in response to light occurred at the elevated [CO_2] and low T_{soil} treatment combination where the greatest RCD growth also occurred. This may indicate that reductions in LAR and SLA are not strictly linked the treatments alone. Thus, reasons for the minimal growth stimulation by light under the elevated [CO_2] and soil warming treatment combination remain unclear, but the inability of the seedlings to adjust leaf characteristics in response to the treatments could be implicated. Both LAR and SLA did not differ in response to light under the elevated [CO_2] and soil warming.

The interaction between [CO_2] and T_{soil} changed the pattern of biomass response to light. Mountain maple seedling showed particularly large increases in biomass in response to light when seedlings were exposed to low T_{soil} under the elevated [CO_2], and lowest increases in biomass response to light when both [CO_2] and T_{soil} were high. The total seedling biomass decreased in response to light in the order of elevated [CO_2] - low T_{soil} (197%) > ambient [CO_2] - warmer T_{soil} (186%) > ambient [CO_2] – low T_{soil} (132%) > elevated [CO_2] –warmer T_{soil} (105%) treatment combinations, respectively. This follows the same pattern of response to light under all the [CO_2]-T_{soil} treatment combinations as that of RCD. The results contrast with the hypothesis that elevated [CO_2] and soil warming would stimulate biomass responses to high light, due to elevated CO_2-induced aboveground growth and soil warming-related increases in water and nutrient absorption. In a previous study, increases in growth and biomass in *Acer pseudoplatanus*, *Quercus robur* and *Abies alba* by 3.4% light compared to 1.3% light level were attributed to elevated CO_2 stimulation of leaf photosynthesis (Hättenschwiler and Körner 2000). However, nutrients limitation and physiological controls within the leaf can have negative feedback on plant growth and biomass responses to high light (Körner 2000). For example, accumulation of non-structural carbohydrates due to decreases in leaf nitrogen concentrations in high light conditions (Kubiske and Pregitzer 1996, Zotz et al. 2006) causes down-regulation of photosynthesis and reduces growth and biomass production in plants. We found that the photosynthetic capacity reduced in response to high light under elevated [CO_2], although the response was independent of T_{soil} (chapter 2 of this thesis). Lack of increases in photosynthesis in response high light, coupled with warming-induced increases in C loss through roots respiration (Lambers et al. 1996, Zogg et al. 1996, Atkin et al. 2000, Pregitzer et al. 2000) might have resulted in the smaller increases in biomass in response to the high light treatment. We observed that root biomass decreased significantly in response to high light conditions under the elevated [CO_2] and soil warming (data not shown), in agreement with the findings of Ambebe and Dang (2010). These results demonstrate that increasing [CO_2] and soil warming may have minimal positive effect on mountain maple seedlings carbon status in canopy gaps.

The proportion of biomass allocated to leaf was lower at the low T_{soil} and ambient [CO_2] in response to light treatment. Plants in high light environments are faced with increased heat load, thus they allocate less biomass to leaves in order to reduce transpiration (Givnish 1988, Pearcy and Sims 1994). Soil warming is shown to improve water and nutrient uptake (Bowes 1991, DeLucia et al. 1992, Karlsson and Nordell 1996, Bassirirad 2000, Rennenberg et al. 2006) which may result in increased leaf production (Boucher et al. 2001). Similar increase in LMR under elevated as a result of higher leaf production has been reported (Barrett and Gifford 1995). There was an increased allocation of biomass to roots in response to high light. Relatively higher allocation to roots in response to high light increase water absorption and compensate for increased transpiration demands (Rice and Bazzaz 1989a, Rice and Bazzaz 1989b, Poorter 1999, Boucher et al. 2001). However, the relative proportion of biomass allocated to roots in response to light was greatest under the ambient [CO_2]-warmer T_{soil}, and lowest under the ambient [CO_2]-low T_{soil} treatment combinations. Even though soil warming increases roots activities, and thus decreases allocation to roots (Davidson 1969), increased evaporation rate that reduces soil moisture content under soil warmer soils (Pregitzer and King 2005, Hagedorn et al. 2010) could counteract such response. This could be implicated in the pattern of biomass allocation to roots observed in this study.

In conclusion, elevated [CO_2] and soil warming reduced the stimulatory effect of high light on mountain maple seedling growth. Elevated [CO_2] caused mountain maple to improve biomass accumulation at the low T_{soil} in the high light treatment, but soil warming greatly reduced biomass accumulation under elevated [CO_2]. This demonstrates that efficient light capture and assimilation into biomass that enhances maple establish in canopy gaps will partly be dependent on T_{soil}.

References

References

Abrams, M.D., M.E. Kubiske and K.C. Steiner. 1990. Drought adaptations and responses in five genotypes of *Fraxinus pennsylvanica* Marsh.: photosynthesis, water relations and leaf morphology. Tree Physiology. 6:305-315.

Abrams, M.D. and S.A. Mostoller. 1995. Gas exchange, leaf structure and nitrogen in contrasting successional tree species growing in open and understory sites during a drought. Tree Physiology. 15:361-370.

Adams, R.M., C. Rosenzweig, R.M. Peart, J.T. Ritchie, B.A. McCarl, J.D. Glyer, R.B. Curry, J.W. Jones, K.J. Boote and L.H. Allen. 1990. Global climate change and US agriculture. Nature. 345:219-224.

Aerts, R. and F.S. Chapin Iii. 1999. The mineral nutrition of wild plants revisited: a re-evaluation of processes and patterns. *In* Advances in Ecological Research Eds. A.H. Fitter and D.G. Raffaelli. Academic Press, pp 1-67.

Ainsworth, E.A. and S.P. Long. 2005. What have we learned from 15 years of free-air CO_2 enrichment (FACE)? A meta-analytic review of the responses of photosynthesis, canopy properties and plant production to rising CO_2. New Phytologist. 165:351-372.

Allen, L.H., R.R. Valle, J.W. Jones and P.H. Jones. 1998. Soybean leaf water potential responses to carbon dioxide and drought. Agron. J. 90:375-383.

Ambebe, T.F. and Q.-L. Dang. 2009. Low moisture availability inhibits the enhancing effect of increased soil temperature on net photosynthesis of white birch (*Betula papyrifera*) seedlings grown under ambient and elevated carbon dioxide concentrations. Tree Physiology. 29:1341-1348.

Ambebe, T.F. and Q.-L. Dang. 2010. Low moisture availability reduces the positive effect of increased soil temperature on biomass production of white birch (*Betula papyrifera*) seedlings in ambient and elevated carbon dioxide concentration. Nordic Journal of Botany. 28:104-111.

Ambebe, T.F., Q.-L. Dang and J. Li. 2010. Low soil temperature inhibits the effect of high nutrient supply on photosynthetic response to elevated carbon dioxide concentration in white birch seedlings. Tree Physiology. 30:234-243.

Ambebe, T.F., G. Danyagri and Q.-L. Dang. *in press*. Low soil temperature inhibits the stimulatory effect of elevated [CO_2] on height and biomass accumulation of white birch seedlings grown under three non-limiting phosphorus conditions. Nordic Journal of Botany

Anten, N.P.R. and T. Hirose. 1998. Biomass allocation and light partitioning among dominant and subordinate individuals in Xanthium canadense stands. Annals of Botany. 82:665-673.

Archambault, L., J. Morissette and M. Bernier-Cardou. 1998. Forest succession over a 20-year period following clearcutting in balsam fir yellow birch ecosystems of eastern Quebec, Canada. Forest Ecology and Management. 102:61-74.

Archibold, O.W., C. Acton and E.A. Ripley. 2000. Effect of site preparation on soil properties and vegetation cover, and the growth and survival of white spruce (*Picea glauca*) seedlings, in Saskatchewan. Forest Ecology and Management. 131:127-141.

Atkin, O.K., J.R. Evans, M.C. Ball, H. Lambers and T.L. Pons. 2000. Leaf respiration of snow gum in the light and dark. interactions between temperature and irradiance. Plant Physiology. 122:915-923.

Aubin, I., M. Beaudet and C. Messier. 2000. Light extinction coefficients specific to the understory vegetation of the southern boreal forest, Quebec. Canadian Journal of Forest Research-Revue Canadienne De Recherche Forestiere. 30:168-177.

Aubin, I., C. Messier and D. Kneeshaw. 2005. Population structure and growth acclimation of mountain maple along a successional gradient in the southern boreal forest. Ecoscience. 12:540-548.

Barrett, D.J. and R.M. Gifford. 1995. Photosynthetic acclimation to elevated CO_2 in relation to biomass allocation in cotton. Journal of Biogeography. 22:331-339.

Bassirirad, H. 2000. Kinetics of nutrient uptake by roots: responses to global change. New Phytologist. 147:155-169.

Bazzaz, F.A. 1996. Plants in changing environment: Linking physiology, population, and community ecology. Cambridge Univ. Press, Cambridge.

Bazzaz, F.A., J.S. Coleman and S.R. Morse. 1990. Growth responses of seven major co-occurring tree species of the northeastern United States to elevated CO_2. Canadian Journal of Forest Research. 20:1479-1484.

Bazzaz, F.A., K. Garbutt and W.E. William. 1985. Effect of increased atmospjeric carbon dioxide concentration on plant communities. In: *Direct effects of increased carbon dioxide concentration on vegetation*. United States Department of Energy Washington, DC.

Bazzaz, F.A. and S.L. Miao. 1993. Successional status, seed size, and responses of tree seedlings to CO_2, light, and nutrients. Ecology. 74:104-112.

Beaudet, M., C. Messier, D.W. Hilbert, E. Lo, Z.M. Wang and M.J. Lechowicz. 2000. Leaf- and plant-level carbon gain in yellow birch, sugar maple, and beech seedlings from contrasting forest light environments. Canadian Journal of Forest Research. 30:390-404.

Beier, C.M., J.L. Horton, J.F. Walker, B.D. Clinton and E.T. Nilsen. 2005. Carbon limitation leads to suppression of first year oak seedlings beneath evergreen understory shrubs in Southern Appalachian hardwood forests. Plant Ecology. 176:131-142.

Belote, R.T., J.F. Weltzin and R.J. Norby. 2004. Response of an understory plant community to elevated [CO2] depends on differential responses of dominant invasive species and is mediated by soil water availability. New Phytologist. 161:827-835.

Bergeron, Y. 2000. Species and stand dynamics in the mixed woods of Quebec's southern boreal forest. Ecology. 81:1500-1516.

Bjôrkman, O. 1981. Responses to different quantum flux densities. In: Encyclopaedia of plant physiology. Plant physiological ecology I. Springer, Berlin, Germany.

Bond, B.J. 2000. Age-related changes in photosynthesis of woody plants. Trends in Plant Science. 5:349-353.

Boucher, J.-F., P.Y. Bernier, H.A. Margolis and A.D. Munson. 2007. Growth and physiological response of eastern white pine seedlings to partial cutting and site preparation. Forest Ecology and Management. 240:151-164.

Boucher, J.-F., S. Wetzel and A.D. Munson. 1998. Leaf level response of planted eastern white pine (*Pinus strobus* L.) seven years after intensive silvicultural treatments. Forest Ecology and Management. 107:291-307.

Boucher, J.F., P.Y. Bernier and A.D. Munson. 2001. Radiation and soil temperature interactions on the growth and physiology of eastern white pine (*Pinus strobus* L.) seedlings. Plant and Soil. 236:165-174.

Bowes, G. 1993. Facing the inevitable - Plants and increasing atmospheric CO_2 Annual Review of Plant Physiology and Plant Molecular Biology. 44:309-332.

Bowes, G.D. 1991. Soil temperature, root growth, and plant function - In: Plant roots, the hidden half. Marcel Decker Inc.

Breshears, David D., John W. Nyhan, Christopher E. Heil and Bradford P. Wilcox. 1998. Effects of woody plants on microclimate in a semiarid woodland: soil temperature and evaporation in canopy and intercanopy patches. International Journal of Plant Sciences. 159:1010-1017.

Brisson, N. and M.-L. Casals. 2005. Leaf dynamics and crop water status throughout the growing cycle of durum wheat crops grown in two contrasted water budget conditions. Agron. Sustain. Dev. 25:151-158.

Bunce, J.A. 1992. Stomatal conductance, photosynthesis and respiration of temperate deciduous tree seedlings grown outdoors at an elevated concentration of carbon dioxide. Plant Cell and Environment. 15:541-549.

Burdett, A.N. 1990. Physiological processes in plantation establishment and the development of specifications for forest planting stock. Canadian Journal of Forest Research. 20:415-427.

Campbell, B.D., D.M. Stafford Smith and G.M. McKeon. 1997. Elevated CO_2 and water supply interactions in grasslands: A pastures and rangelands management perspective. Global Change Biology. 3:177-187.

Caners, R. and N.C. Kenkel. 1997. Uncommon plant species of eastern deciduous floristic affinity, Rising Mountain National Park. UFS (Delta Marsh) Annual Report. Department of Botany, University of Manitoba, pp 80 – 82. .

Canham, C.D. 1988a. Growth and canopy architecture of shade-tolerant trees: response to canopy gaps. Ecology. 69:786-795.

Canham, C.D. 1988b. An index for understory light levels in and around canopy gaps. Ecology. 69:1634-1638.

Canham, C.D. 1989. Different response to gaps among shade-tolerance tree species. Ecology. 70:549 - 550.

Canham, C.D., A.R. Berkowitz, V.R. Kelly, G.M. Lovett, S.V. Ollinger and J. Schnurr. 1996. Biomass allocation and multiple resource limitation in tree seedlings. Canadian Journal of Forest Research. 26:1521-1530.

Catovsky, S. and F.A. Bazzaz. 1999. Elevated CO_2 influences the responses of two birch species to soil moisture: implications for forest community structure. Global Change Biology. 5:507-518.

Catovsky, S. and F.A. Bazzaz. 2002. Nitrogen availability influences regeneration of temperate tree species in the understory seedling bank. Ecological Applications. 12:1056-1070.

Cavender-Bares, J. and F.A. Bazzaz. 2000. Changes in drought response strategies with ontogeny in *Quercus rubra* : implications for scaling from seedlings to mature trees. Oecologia. 124:8-18.

Chapin, F.S. 1980. The Mineral Nutrition of Wild Plants. Annual review of ecology and systematics. 11:233-260.

Chapin III, S.F., A.J. Bloom, C.B. Field and R.H. Waring. 1987. Plant responses to multiple environmental factors. BioScience. 37:49-57.

Chazdon, R.L. 1988. Sunflecks and their importance to forest understory plants. Adv. Ecol. Research. 18:1–63.

Chazdon, R.L. and C.B. Field. 1987. Determinants of photosynthetic capacity in six rainforest *Piper* species. Oecologia. 73:222-230.

Chazdon, R.L. and S. Kaufmann. 1993. Plasticity of leaf anatomy of two ranin-forest shurbs in relation to photosynthetic light acclimation. Functional Ecology. 7:385-394.

Chazdon, R.L., R. Pearcy, D. Lee and N. Fetcher. 1996. Photosynthetic responses of tropical forest plants to contrasting light environments. In S. Mulkey, R. L. Chazdon, and A. P Smith [eds.], Tropical forest plant ecophysiology. Chapman and Hall, New York.

Cheng, S., Q.-L. Dang and T.-B. Cai. 2000. A soil temperature control system for ecological research in greenhouses. Journal of Forest Research. 5:205-208.

Clark, D.A. and D.B. Clark. 1992. Life history diversity of canopy and emergent trees in a neotropical rain forest. Ecological Monographs. 62:315-344.

Coomes, D.A. and P.J. Grubb. 1998. Responses of juvenile trees to above- and belowground competition in nutrient-starved Amazonian rain forest. Ecology. 79:768-782.

Coomes, D.A. and P.J. Grubb. 2000. Impacts of root competition in forests and woodlands: a theoretical framework and review of experiments. Ecological Monographs. 70:171-207.

Cornic, G. 2000. Drought stress inhibits photosynthesis by decreasing stomatal aperture - not by affecting ATP synthesis. Trends in Plant Science. 5:187-188.

Cowan, I.R. 1986. Economics of carbon fixation in higher plants. In: On the economy of plant form and function. Cambridge University Press, Cambridge, UK.

Cox, P.M., R.A. Betts, C.D. Jones, S.A. Spall and I.J. Totterdell. 2000. Acceleration of global warming due to carbon-cycle feedbacks in a coupled climate model. Nature. 408:184-187.

Craine, J.M. and P.B. Reich. 2005. Leaf-level light compensation points in shade-tolerant woody seedlings. New Phytologist. 166:710-713.

Curtis, P.S. and X. Wang. 1998. A meta-analysis of elevated CO_2 effects on woody plant mass, form, and physiology. Oecologia. 113:299-313.

Dalling, J.W., K. Winter and S.P. Hubbell. 2004. Variation in growth responses of neotropical pioneers to simulated forest gaps. Functional Ecology. 18:725-736.

Dang, Q.-L. and S. Cheng. 2004. Effects of soil temperature on ecophysiological traits in seedlings of four boreal tree species. Forest Ecology and Management. 194:379-387.

Davidson, R.L. 1969. Effect of root/leaf temperature differentials on root/shoot Ratios in some pasture grasses and clover. Annals of Botany. 33:561-569.

Dawes, M.A., F. Hagedorn, T. Zumbrunn, I.T. Handa, S. Hättenschwiler, S. Wipf and C. Rixen. 2011a. Growth and community responses of alpine dwarf shrubs to in situ CO_2 enrichment and soil warming. New Phytologist. 191:806-818.

Dawes, M.A., S. Hättenschwiler, P. Bebi, F. Hagedorn, I.T. Handa, C. Körner and C. Rixen. 2011b. Species-specific tree growth responses to 9 years of CO_2 enrichment at the alpine treeline. Journal of Ecology. 99:383-394.

Day, T.A., S.A. Heckathorn and E.H. Delucia. 1991. Limitations of photosynthesis in *Pinus taeda* (loblolly Pine) at low soil temperatures. Plant Physiology. 96:1246-1254.

De Frenne, P., J. Brunet, A. Shevtsova, A. Kolb, B.J. Graae, O. Chabrerie, S.A. Cousins, G. Decocq, A.N. De Schrijver, M. Diekmann, R. Gruwez, T. Heinken, M. Hermy, C. Nilsson, S. Stanton, W. Tack, J. Willaert and K. Verheyen. 2011. Temperature effects on forest herbs assessed by warming and transplant experiments along a latitudinal gradient. Global Change Biology. 17:3240-3253.

De Frenne, P., B.J. Graae, J. Brunet, A. Shevtsova, A. De Schrijver, O. Chabrerie, S.A.O. Cousins, G. Decocq, M. Diekmann, M. Hermy, T. Heinken, A. Kolb, C. Nilsson, S. Stanton and K. Verheyen. 2012. The response of forest plant regeneration to temperature variation along a latitudinal gradient. Annals of Botany:1 - 10.

De Grandpré, L.D., D. Gagnon and Y. Bergeron. 1993. Changes in the understory of Canadian southern boreal forest after fire. Journal of Vegetation Science. 4:803-810.

DeLucia, E.H., S.A. Heckathorn and T.A. Day. 1992. Effects of soil temperature on growth, biomass allocation and resource acquisition of *Andropogon gerardii* Vitman. New Phytologist. 120:543-549.

Derner, J.D., H.B. Johnson, B.A. Kimball, P.J. Pinter, H.W. Polley, C.R. Tischler, T.W. Boutton, R.L. Lamorte, G.W. Wall, N.R. Adam, S.W. Leavitt, M.J. Ottman, A.D. Matthias and T.J. Brooks. 2003. Above- and below-ground responses of C_3–C_4 species mixtures to elevated CO_2 and soil water availability. Global Change Biology. 9:452-460.

Dewar, R.C. 1993. A root-shoot partitioning model based on carbon-nitrogen-water interactions and munch phloem flow. Functional Ecology. 7:356-368.

Dodd, I.C., J. He, C.G.N. Turnbull, S.K. Lee and C. Critchley. 2000. The influence of supra-optimal root-zone temperatures on growth and stomatal conductance in Capsicum annuum L. Journal of Experimental Botany. 51:239-248.

Dong, S., C.F. Scagel, L. Cheng, L.H. Fuchigami and P.T. Rygiewicz. 2001. Soil temperature and plant growth stage influence nitrogen uptake and amino acid concentration of apple during early spring growth. Tree Physiology. 21:541-547.

Drake, B.G., M.A. Gonzàlez-Meler and S.P. Long. 1997. More efficient plants: A consequence of rising atmospheric CO_2? Annual Review of Plant Physiology and Plant Molecular Biology. 48:609-639.

Dreyer, E., X. Le Roux, P. Montpied, F.A. Daudet and F. Masson. 2001. Temperature response of leaf photosynthetic capacity in seedlings from seven temperate tree species. Tree Physiology. 21:223-232.

Duff, G.A., C.A. Berryman and D. Eamus. 1994. Growth, biomass allocation and foliar nutrient contents of two *Eucalyptus* species of the wet-dry tropics of Australia grown under CO_2 enrichment. Functional Ecology. 8:502-508.

Eamus, D. 1991. The interaction of rising CO_2 and temperatures with water use efficiency. Plant, Cell and Environment. 14:843-852.

Èermák, J. and J. Kuèera. 1993. Extremely fast changes in xylem flow rate in tall trees caused by atmospheric, soil and mechanical factors. In Water Transport in Plants Under Climatic Stress. Cambridge University Press, Cambridge, UK.

Èermák, J., R. Matyssek and J. Kuèera. 1993. Rapid responses of large, drought-stressed beech trees to irrigation. Tree Physiology. 12:281-290.

Epron, D. and E. Dreyer. 1993. Long-term effects of drought on photosynthesis of adult oak trees [*Quercus petraea* (Matt.) Liebl. and *Quercus robur* L.] in a natural stand. New Phytologist. 125:381-389.

Evans, J.R. 1988. Acclimation by the thylakoid membranes to growth irradiance and the partitioning of nitrogen between soluble and thylakoid proteins Australian Journal of Plant Physiology. 15:93-106.

Farquhar, G.D., S. Caemmerer and J.A. Berry. 1980. A biochemical model of photosynthetic CO_2 assimilation in leaves of C_3 species. Planta. 149:78-90.

Farquhar, G.D. and S. von Caemmerer. 1982. Modelling of photosynthetic response to environmental conditions. In encyclopaedia of plant physiology, physiological plant ecology II: water relations and carbon assimilation Springer Verlag, Berlin.

Field, C.B., F.S.C. Iii, P.A. Matson and H.A. Mooney. 1992. Responses of terrestrial ecosystems to the changing atmosphere: a resource-based approach. Annual Review of Ecology and Systematics. 23:201-235.

Fitter, A.H., J.D. Graves, G.K. Self, T.K. Brown, D.S. Bogie and K. Taylor. 1998. Root production, turnover and respiration under two grassland types along an altitudinal gradient: influence of temperature and solar radiation. Oecologia. 114:20-30.

Flexas, J., J. Bota, F. Loreto, G. Cornic and T.D. Sharkey. 2004. Diffusive and metabolic limitations to photosynthesis under drought and salinity in C_3 plants. Plant Biology. 6:269-279.

Flexas, J. and H. Medrano. 2002. Drought-inhibition of photosynthesis in C_3 plants: stomatal and non-stomatal limitations revisited. Annals of Botany. 89:183-189.

Foster, W.J., D.L. Ingram and T.A. Nell. 1991. Photosynthesis and root respiration in *Ilex crenata* `Rotundifolia' at supraoptimal root-zone temperatures. HortScience. 26:535-537.

Franklin, O., J. Johansson, R.C. Dewar, U. Dieckmann, R.E. McMurtrie, Å. Brännström and R. Dybzinski. 2012. Modeling carbon allocation in trees: a search for principles. Tree Physiology

Gauthier, S., J. Gagnon and Y. Bergeron. 1993. Population age structure of *Pinus banksiana* at the southern edge of the Canadian boreal forest. Journal of Vegetation Science. 4:783-790.

Gifford, R.M. 1992. Interaction of carbon dioxide with growth-limiting environmental factors in vegetation productivity: implications for the global carbon cycle. In Advances in Bioclimatology. Ed. G. Stanhill. Springer-Verlag, New York.

Givnish, T. 1988. Adaptation to sun and shade: a whole-plant perspective. Functional Plant Biology. 15:63-92.

Givnish, T.J. 1995. Plant stems: biomechanical adaptation for energy capture and influence on species distributions. In Plant stems: shysiology and functional forphology. Academic Press, San Diego, Califonia.

Grime, J.P. 1994. The role of plasticity in exploiting environmental heterogeneity. *In* Exploitation of Environmental Heterogeneity by Plants. Ecophysiological Processes Above- and Belowground. Academic Press, Inc., USA.

Hagedorn, F., M. Martin, C. Rixen, S. Rusch, P. Bebi, A. Zürcher, R. Siegwolf, S. Wipf, C. Escape, J. Roy and S. Hättenschwiler. 2010. Short-term responses of ecosystem carbon fluxes to experimental soil warming at the Swiss alpine treeline. Biogeochemistry. 97:7-19.

Hallik, L., Ü. Niinemets and O. Kull. 2012. Photosynthetic acclimation to light in woody and herbaceous species: a comparison of leaf structure, pigment content and chlorophyll fluorescence characteristics measured in the field. Plant Biology. 14:88-99.

Harley, P., A. Guenther and P. Zimmerman. 1996. Effects of light, temperature and canopy position on net photosynthesis and isoprene emission from sweetgum (*Liquidambar styraciflua*) leaves. Tree Physiology. 16:25-32.

Hättenschwiler, S. 2001. Tree seedling growth in natural deep shade: functional traits related to interspecific variation in response to elevated CO_2. Oecologia. 129:31-42.

Hattenschwiler, S. and C. Korner. 2003. Does elevated CO_2 facilitate naturalization of the non-indigenous *Prunus laurocerasus* in Swiss temperate forests? Functional Ecology. 17:778-785.

Hättenschwiler, S. and C. Körner. 2000. Tree seedling responses to in situ CO_2-enrichment differ among species and depend on understorey light availability. Global Change Biology. 6:213-226.

Hättenschwiler, S., F. Miglietta, A. Raschi and C. KöRner. 1997. Thirty years of in situ tree growth under elevated CO_2: a model for future forest responses? Global Change Biology. 3:463-471.

Herrick, J.D. and R.B. Thomas. 1999. Effects of CO_2 enrichment on the photosynthetic light response of sun and shade leaves of canopy sweetgum trees (*Liquidambar styraciflua*) in a forest ecosystem. Tree Physiology. 19:779-786.

Hibbs, D.E., F.W. Brayton and B.C. Fischer. 1980. Habitat requirements and growth of striped maple (*Acer Pensylvanicum* L.). Ecology. 61:490-496.

Hikosaka, K. 2005. Nitrogen partitioning in the photosynthetic apparatus of *Plantago asiatica* leaves grown under different temperature and light conditions: similarities and differences between temperature and light acclimation. Plant and Cell Physiology. 46:1283-1290.

Hobbie, S.E. 1996. Temperature and Plant Species Control Over Litter Decomposition in Alaskan Tundra. Ecological Monographs. 66:503-522.

Houghton, J.T., Y. Ding, J.J. J. J. Griggs and e. al. 2001. Climate Change 2001: The Scientific Basis. . Cambridge University Press, Cambridge.

Houghton, J.T.e.a. 1992. Climate change 1992. The supplementary report to the IPCC scientific assessement.

Huang, J.-G., Y. Bergeron, B. Denneler, F. Berninger and J. Tardif. 2007. Response of forest trees to increased atmospheric CO2. Critical Reviews in Plant Sciences. 26:265-283.

Ingestad, T. 1981. Nutrition and growth of birch and grey alder seedlings in low conductivity solutions and at varied relative rates of nutrient addition. Physiologia Plantarum. 52:454-466.

IPCC. 2001. Climate Change 2001: An Assessment of the Intergovernmental Panel on Climate Change.

IPCC. 2007a. Climate Change 2007: Synthesis Report. Contribution of Working Groups I, II and III to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change, IPCC, Geneva, Switzerland.

IPCC. 2007b. Summary for policymakers. In: Solomon, S. et al. (eds), Climate Change 2007: the physical science basis. Contribution of Working Group I to the fourth assessment report of the Intergovernmental Panel on Climate Change.

James, J.J., R.L. Tiller and J.H. Richards. 2005. Multiple resources limit plant growth and function in a saline-alkaline desert community. Journal of Ecology. 93:113-126.

Jansson, P.E. and B. Berg. 1985. Temporal variation of litter decomposition in relation to simulated soil climate. Long-term decomposition in a Scots pine forest. V. Canadian Journal of Botany. 63:1008-1016.

Jones, M.B., J.C. Brown, A. Raschi and F. Miglietta. 1995. The effects on Arbutus unedo L. of long-term exposure to elevated CO2. Global Change Biology. 1:295-302.

Karlsson, P.S. and K.O. Nordell. 1996. Effects of soil temperature on the nitrogen economy and growth of mountain birch seedlings near its presumed low temperature distribution limit. Ecoscience. 3:183-189.

King, J.S., K.S. Pregitzer and D.R. Zak. 1999. Clonal variation in above- and below-ground growth responses of Populus tremuloides Michaux: Influence of soil warming and nutrient availability. Plant and Soil. 217:119-130.

Kitajima, K. 1994. Relative importance of photosynthetic traits and allocation patterns as correlates of seedling shade tolerance of 13 tropical trees. Oecologia. 98:419-428.

Klinka, K., Q. Wang, G.J. Kayahara, R.E. Carter and B.A. Blackwell. 1992. Light-growth response relationships in Pacific silver fir (*Abies amabilis*) and subalpine fir (*Abies lasiocarpa*). Canadian Journal of Botany. 70:1919-1930.

Knapp, A.K., E.P. Hamerlynck and E.O. Clenton. 1993. Photosynthetic and water relations responses to elevated CO_2 in the C_4 grass *Andropogon gerardii*. International Journal of Plant Sciences. 154:459-466.

Kobe, R.K., S.W. Pacala, J.A. Silander and C.D. Canham. 1995. Juvenile tree survivorship as a component of shade tolerance. Ecological Applications. 5:517-532.

Koerner, C. 2006. Plant CO_2 responses: an issue of definition, time and resource supply. New Phytologist. 172:393-411.

Korner, C. 2006. Plant CO_2 responses: an issue of definition, time and resource supply. New Phytologist. 172:393-411.

Körner, C. 2000. Biosphere responses to CO_2 enrichment. Ecological Applications. 10:1590-1619.

Körner, C. 2003. Ecological impacts of atmospheric CO_2 enrichment on terrestrial ecosystems. Philosophical Transactions of the Royal Society of London Series A. 361:2023-2041.

Kubiske, M.E. and M.D. Abrams. 1994. Ecophysiological analysis of woody species in contrasting temperate communities during wet and dry years. Oecologia. 98:303-312.

Kubiske, M.E. and K.S. Pregitzer. 1996. Effects of elevated CO_2 and light availability on the photosynthetic light response of trees of contrasting shade tolerance. Tree Physiology. 16:351-358.

Kubiske, M.E. and K.S. Pregitzer. 1997. Ecophysiological responses to simulated canopy gaps of two tree species of contrasting shade tolerance in elevated CO_2. Functional Ecology. 11:24-32.

Küppers, M. 1994. Canopy gaps: competitive light interception and economic space filling—a matter of whole plant allocation. In Exploitation of Environmental Heterogeneity by Plants: Ecophysiological Processes Above- and Belowground. Academic Press, San Diego.

Lambers, H., O.K. Atkin and F.K. Millenaar. 1996. Respiratory patterns in roots in relation to their functioning. In: Waisel Y, Eshel A, Kafkaki U, eds. *Plant roots. The hidden half*, 2nd Edn. Marcel Dekker, New York.

Lambers, H., S.F. Chapin III and T.L. Pons. 2008. Plant physiological eclogy. Springer, New York.

Lambers, H. and H. Poorter. 1992. Inherent variation in growth rate between higher plants: A search for physiological causes and eological consequences. *In* Advances in Ecological Research Eds. M. Begon and A.H. Fitter. Academic Press, pp 187-261.

Landhäusser, S.M., R.W. Wein and P. Lange. 1996. Gas exchange and growth of three arctic tree-line tree species under different soil temperature and drought preconditioning regimes. Canadian Journal of Botany. 74:686-693.

Larigauderie, A., B.A. Ellis, J.N. Mills and J. Kummerow. 1991. The effect of root and shoot temperatures on growth of Ceanothus-Greggii seedlings. Annals of Botany. 67:97-101.

Lawlor, D.W. 2002. Limitation to photosynthesis in water-stressed leaves: stomata vs. metabolism and the role of ATP. Annals of Botany. 89:871-885.

Lawlor, D.W. and G. Cornic. 2002. Photosynthetic carbon assimilation and associated metabolism in relation to water deficits in higher plants. Plant, Cell and Environment. 25:275-294.

Lawlor, D.W. and W. Tezara. 2009. Causes of decreased photosynthetic rate and metabolic capacity in water-deficient leaf cells: a critical evaluation of mechanisms and integration of processes. Annals of Botany. 103:561-579.

LeCain, D.R., J.A. Morgan, G.L. Hutchinson, J.D. Reeder and F.A. Dijkstra. 2012. Interactions between elevated atmospheric CO_2 and defoliation on North American rangeland plant species at low and high N availability. Grass and Forage Science:no-no.

Lei, T.T. 1992. Functional design and shade adaptation in *Acer* species. *In* Biology. McGill University.

Lei, T.T. and M.J. Lechowicz. 1990. Shade adaptation and shade tolerance in saplings of three *Acer* species from eastern north America. Oecologia. 84:224-228.

Lei, T.T. and M.J. Lechowicz. 1997a. Functional responses of *Acer* species to two simulated forest gap environments: leaf-level properties and photosynthesis. Photosynthetica. 33:277-289.

Lei, T.T. and M.J. Lechowicz. 1997b. The photosynthetic response of eight species of *Acer* to simulated light regimes from the centre and edges of gaps. Functional Ecology. 11 16 - 23.

Li, S., S.R. Pezeshki and S. Goodwin. 2004. Effects of soil moisture regimes on photosynthesis and growth in cattail (*Typha latifolia*). Acta Oecologica. 25:17-22.

Liang, N.H., Y. Tang and T. Okuda. 2001. Is elevation of carbon dioxide concentration beneficial to seedling photosynthesis in the understory of tropical rain forests? Tree Physiology. 21:1047-1055.

Lin, J., P.A. Harcombe, M.R. Fulton and R.W. Hall. 2002. Sapling growth and survivorship as a function of light in a mesic forest of southeast Texas, USA. Oecologia. 132:428-435.

Liozon, R., F.W. Badeck, B. Genty, S. Meyer and B. Saugier. 2000. Leaf photosynthetic characteristics of beech (*Fagus sylvatica*) saplings during three years of exposure to elevated CO_2 concentration. Tree Physiology. 20:239-247.

Liu, F. and H. Stützel. 2004. Biomass partitioning, specific leaf area, and water use efficiency of vegetable amaranth (*Amaranthus* spp.) in response to drought stress. Scientia Horticulturae. 102:15-27.

Long, S.P. and B.G. Drake. 1991. Effect of the long-term elevation of CO_2 concentration in the field on the quantum yield of photosynthesis of the C_3 sedge, *Scirpus olneyi*. Plant Physiology. 96:221-226.

Long, S.P. and B.G. Drake. 1992. Photosynthetic CO_2 assimilation and rising atmospheric CO_2 concentrations. In Crop Photosynthesis: spatial and temporal determinants. Elsevier Science Publishers B.V., Amsterdam.

Long, S.P., C.P. Osborne and S.W. Humphries. 1996. Photosynthesis, rising atmospheric carbon dioxide concentration and climate change. In SCOPE 56 - Global change: effects on coniferous forests and grasslands. John Wiley and Sons, New York.

Long, S.P. and F.I. Woodward. 1998. Plant and temperature. The Company of Biologists Limited, Department of Zoology. University of Cambridge, Cambridge.

Lyr, H. and V. Garbe. 1995. Influence of root temperature on growth of <i>Pinus sylvestris , <i>Fagus sylvatica, Tilia cordata and <i>Quercus robur</i>. Trees - Structure and Function. 9:220-223.

MacDonald, N.W., D.R. Zak and K.S. Pregitzer. 1995. Temperature effects on kinetics of microbial respiration and net nitrogen and sulfur mineralization. Soil Sci. Soc. Am. J. 59:233-240.

McConnaughay, K.D.M. and J.S. Coleman. 1999. Biomass allocation in plants: Ontogeny or optimum? A Test along three resource gradients Ecology. 80:2581-2593.

Medlyn, B.E., F.W. Badeck, D.G.G. De Pury, C.V.M. Barton, M. Broadmeadow, R. Ceulemans, P. De Angelis, M. Forstreuter, M.E. Jach, S. Kellomäki, E. Laitat, M. Marek, S. Philippot, A. Rey, J. Strassemeyer, K. Laitinen, R. Liozon, B. Portier, P. Roberntz, K. Wang and P.G. Jstbid. 1999. Effects of elevated [CO_2] on photosynthesis in European forest species: a meta-analysis of model parameters. Plant, Cell & Environment. 22:1475-1495.

Meehl, G.A., J.M. Arblaster and C. Tebaldi. 2007. Contributions of natural and anthropogenic forcing to changes in temperature extremes over the United States. Geophys. Res. Lett. 34:L19709.

Melillo, J.M., P.A. Steudler, J.D. Aber, K. Newkirk, H. Lux, F.P. Bowles, C. Catricala, A. Magill, T. Ahrens and S. Morrisseau. 2002. Soil warming and carbon-cycle feedbacks to the climate system. Science. 298:2173-2176.

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

Miao, S.L., P.M. Wayne and F.A. Bazzaz. 1992. Elevated CO_2 differentially alters the responses of coocurring birch and maple seedlings to a moisture gradient. Oecologia. 90:300-304.

Mitchell, J.F., B.S. Manabe, V. Meleshko and T. Tokioka. 1990. Equilibrium climate change - and its implications for the future. In: Climate Change: The IPCC scientific assessment. Cambridge Univ. Press, Cambridge.

Mitchell, R.A.C., J.C. Theobald, M.A.J. Parry and D.W. Lawlor. 2000. Is there scope for improving balance between RuBP-regeneration and carboxylation capacities in wheat at elevated CO_2? Journal of Experimental Botany. 51:391-397.

Moore, B.D., S.H. Cheng, D. Sims and J.R. Seemann. 1999. The biochemical and molecular basis for photosynthetic acclimation to elevated atmospheric CO_2. Plant, Cell and Environment. 22:567-582.

Morison, J.I.L. 1993. Response of plants to CO_2 under water limited conditions. Plant Ecology. 104-105:193-209.

Muraoka, H., Y. Tang, H. Koizumi and I. Washitani. 2002. Effects of light and soil water availability on leaf photosynthesis and growth of *Arisaema heterophyllum* , a riparian forest understorey plant. Journal of Plant Research. 115:419-427.

Muraoka, H., Y. Tang, H. Koizumi, I. Washitani and W. Z. 1997. Combined effects of light and water availability on photosynthesis and growth of *Arisaema Heterophyllum* in the forest understory and an open site. Oecologia. 112:26-34.

Muraoko, H., Y. Tang, H. Koizumi and I. Washitan. 2002. Effects of light and soil water availability on leaf photosynthesis and growth of *Arisaema heterophyllum*, a riparian forest understorey plant. Journal of Plant Research. 115:419 - 427.

Naumburg, E. and D.S. Ellsworth. 2000. Photosynthetic sunfleck utilization potential of understory saplings growing under elevated CO_2 in FACE. Oecologia. 122:163-174.

Niinemets, Ü. 2007. Photosynthesis and resource distribution through plant canopies. Plant, Cell and Environment. 30:1052-1071.

Niinemets, Ü. and J.D. Tenhunen. 1997. A model separating leaf structural and physiological effects on carbon gain along light gradients for the shade-tolerant species *Acer saccharum*. Plant, Cell and Environment. 20:845-866.

Nobel, P.S. 1999. Physicochemical and environmental plant physiology. Academic Press, San Diego, CA, USA.

Norby, R.J. and E.G. O'Neill. 1991. Leaf area compensation and nutrient interactions in CO_2-enriched seedlings of yellow-poplar (*Liriodendron tulipifera* L.). New Phytologist. 117:515-528.

Norby, R.J., S.D. Wullschleger, C.A. Gunderson, D.W. Johnson and R. Ceulemans. 1999. Tree responses to rising CO2 in field experiments: implications for the future forest. Plant Cell and Environment. 22:683-714.

Nowak, R.S., D.S. Ellsworth and S.D. Smith. 2004. Functional responses of plants to elevated atmospheric CO_2: Do photosynthetic and productivity data from FACE experiments support early predictions? New Phytologist. 162:253-280.

O'Connell, M.G., G.J. O'Leary, D.M. Whitfield and D.J. Connor. 2004. Interception of photosynthetically active radiation and radiation-use efficiency of wheat, field pea and mustard in a semi-arid environment. Field Crops Research. 85:111-124.

Ohashi, Y., N. Nakayama, H. Saneoka and K. Fujita. 2006. Effects of drought stress on photosynthetic gas exchange, chlorophyll fluorescence and stem diameter of soybean plants. Biologia Plantarum. 50:138-141.

Onoda, Y., K. Hikosaka and T. Hirose. 2005. Seasonal change in the balance between capacities of RuBP carboxylation and RuBP regeneration affects CO_2 response of photosynthesis in *Polygonum cuspidatum*. Journal of Experimental Botany. 56:755-763.

Onoda, Y., T. Hirose and K. Hikosaka. 2009. Does leaf photosynthesis adapt to CO_2-enriched environments? An experiment on plants originating from three natural CO_2 springs. New Phytologist. 182:698-709.

Örlander, G., G. Hallsby, P. Gemmel and C. Wilhelmsson. 1998. Inverting improves establishment of Pinus contorta and *Picea abies*— 10-year results from a site preparation trial in Northern Sweden. Scandinavian Journal of Forest Research. 13:160-168.

Osborne, C.P., B.G. Drake, J. LaRoche and S.P. Long. 1997. Does long-term elevation of CO_2 concentration increase photosynthesis in forest floor vegetation? Indiana strawberry in a Maryland forest. Plant Physiology. 114:337-344.

Owensby, C.E., P.I. Coyne, J.M. Ham, L.M. Auen and A.K. Knapp. 1993. Biomass production in a tallgrass prairie ecosystem exposed to ambient and elevated CO_2. Ecological Applications. 3:644-653.

Pacala, S.W., C.D. Canham, J. Saponara, J.A.S. Jr, R.K. Kobe and E. Ribbens. 1996. Forest models defined by field measurements: estimation, error analysis and dynamics. Ecological Monographs. 66:1-43.

Palanisamy, K. 1999. Interactions of elevated CO_2 concentration and drought stress on photosynthesis in *Eucalyptus Cladocalyx* F. Muell. Photosynthetica. 36:635-638.

Paré, D., Y. Bergeron and C. Camiré. 1993. Changes in the forest floor of Canadian southern boreal forest after disturbance. Journal of Vegetation Science. 4:811-818.

Parelle, J., J.P. Roudaut and M. Ducrey. 2006. Light acclimation and photosynthetic response of beech (*Fagus sylvatica* L.) saplings under artificial shading or natural Mediterranean conditions. Annals of Forest Science. 63:257-266.

Pastor, J., R.H. Gardner, V.H. Dale and W.M. Post. 1987. Successional changes in nitrogen availability as a potential factor contributing to spruce declines in boreal North America. Canadian Journal of Forest Research. 17:1394-1400.

Paula, M.P. 2004. Plant Physiologist. U.S. Department of Agriculture, Forest Service, North Central Research Station, Hardwood Tree Improvement and Regeneration Center, 195 Marsteller Street, West Lafayette, IN 47907.

Pearcy, R.W. 1983. The light environment and growth of C_3 and C_4 tree species in the understory of a Hawaiian forest. Oecologia. 58:19-25.

Pearcy, R.W. 1990. Sunflecks and photosynthesis in plant canopies. Annual Review of Plant Physiology and Plant Molecular Biology. 41:421-453.

Pearcy, R.W. and O. Björkman. 1983. Physiological effects. In *CO_2 and plants, the response of plants to rising levels of atmospheric carbon dioxide*. AAAS selected symposium 84. Westview Press, Boulder, Colorado.

Pearcy, R.W. and D.A. Sims. 1994. Photosynthetic acclimation to changing light environments: Scaling from the leaf to the whole plant. *In* Exploitation of Environmental Heterogeneity by Plants. Ecophysiological Processes Above- and Belowground. Academic Press, Inc., USA.

Peng, Y.Y. and Q.-L. Dang. 2003. Effects of soil temperature on biomass production and allocation in seedlings of four boreal tree species. Forest Ecology and Management. 180:1-9.

Platt, W.J. and D.R. Strong. 1989. Special feature - treefall gaps and forest dynamic. Ecology 70:535-576.

Polley, H.W., C.R. Tischler, H.B. Johnson and R.E. Pennington. 1999. Growth, water relations, and survival of drought-exposed seedlings from six maternal families of honey mesquite (*Prosopis glandulosa*): responses to CO_2 enrichment. Tree Physiology. 19:359-366.

Poorter, H., K.J. Niklas, P.B. Reich, J. Oleksyn, P. Poot and L. Mommer. 2012. Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control. New Phytologist. 193:30-50.

Poorter, H. and C. Remkes. 1990. Leaf area ratio and net assimilation rate of 24 wild species differing in relative growth rate. Oecologia. 83:553-559.

Poorter, L. 1999. Growth responses of 15 rain-forest tree species to a light gradient: the relative importance of morphological and physiological traits. Functional Ecology. 13:396-410.

Poorter, L. and S.F. Oberbauer. 1993. Photosynthetic induction responses of two rainforest tree species in relation to light environment. Oecologia. 96:193-199.

Post, L.J. 1967. Effects of intensity of radiation on height growth, weight increment, and maximum rate of photosynthesis, in mountain maple seedlings. M.Sc. Thesis, University of New Brunswick, Fredericton, New Brunswick.

Post, L.J. 1970. Dry-matter production of mountain maple and balsam fir in northwestern New Brunswick. Ecology. 51:548-550.

Pothier, D. and M. Prévost. 2002. Photosynthetic light response and growth analysis of competitive regeneration after partial cutting in a boreal mixed stand. Trees - Structure and Function. 16:365-373.

Pregitzer, K.S. and J.S. King. 2005. Effects of soil temperature on nutrient uptake. - In: Nutrient acquisition by plants: an ecological perspective. Springer.

Pregitzer, K.S., J.S. King, A.J. Burton and S.E. Brown. 2000. Responses of tree fine roots to temperature. New Phytologist. 147:105-115.

Pritchard, S.G., H.H. Rogers, S.A. Prior and C.M. Peterson. 1999. Elevated CO_2 and plant structure: a review. Global Change Biology. 5:807-837.

Pumpanen, J., J. Heinonsalo, T. Rasilo, J. Villemot and H. Ilvesniemi. 2012. The effects of soil and air temperature on CO2 exchange and net biomass accumulation in Norway spruce, Scots pine and silver birch seedlings. Tree Physiology

Ray, D., S.K. Dey and G. Das. 2004. Significance of the leaf area ratio in *Hevea brasiliensis* under high irradiance and low temperature stress. Photosynthetica. 42:93-97.

Rennenberg, H., F. Loreto, A. Polle, F. Brilli, S. Fares, R.S. Beniwal and A. Gessler. 2006. Physiological responses of forest trees to heat and drought. Plant Biology. 8:556-571.

Rice, S.A. and F.A. Bazzaz. 1989a. Growth consequences of plasticity of plant traits in response to light conditions. Oecologia. 78:508-512.

Rice, S.A. and F.A. Bazzaz. 1989b. Quantification of plasticity of plant traits in response to light intensity: comparing phenotypes at a common weight. Oecologia. 78:502-507.

Ridolfi, M. and E. Dreyer. 1997. Responses to water stress in an ABA-unresponsive hybrid poplar (*Populus koreana*×*trichocarpa* cv. Peace) III. Consequences for photosynthetic carbon assimilation. New Phytologist. 135:31-40.

Rincon, E. and J.P. Grime. 1989. Plasticity and light interception by six bryophytes of contrasted ecology. Journal of Ecology. 77:439-446.

Rind, D., R. Goldberg, J. Hansen, C. Rosenzweig and R. Ruedy. 1990. Potential evapotranspiration and the likelihood of future drought. J. Geophys. Res. 95:9983-10004.

Rook, E.J.S. 2002. Shrubs of the northwoods. http://www.rook. org/earl / bwca /nature / shrubs / index.html.

Roumet, C. and J. Roy. 1996. Prediction of the growth response to elevated CO_2: A search for physiological criteria in closely related grass species. New Phytologist. 134:615-621.

Ruifang, F., Y. Wanqin, Z. Jian, D. Renju, J. Yi and L. Jin. 2007. Effects of simulated elevated concentration of atmospheric CO_2 and temperature on soil enzyme activity in the subalpine fir forest. Acta Ecologica Sinica. 27:4019-4026.

Rustad, L.E., J.L. Campbell, G.M. Marion, R.J. Norby, M.J. Mitchell, A.E. Hartley, J.H.C. Cornelissen, J. Gurevitch and N. Gcte. 2001. A meta-analysis of the response of soil respiration, net nitrogen mineralization, and aboveground plant growth to experimental ecosystem warming. Oecologia. 126:543-562.

Ruter, J.M. and D.L. Ingram. 1992. High root-zone temperatures influence Rubisco activity and pigment accumulation in leaves of `Rotundifolia' Holly. Journal of the American Society for Horticultural Science. 117:154-157.

Samarakoon, A.B. and R.M. Gifford. 1995. Soil water content under plants at high CO_2 concentration and interactions with the direct CO_2 effects: a species comparison. Journal of Biogeography. 22:193-202.

Samuelson, L.J. and J.R. Seiler. 1992. Fraser fir seedling gas exchange and growth in response to elevated CO_2. Environmental and Experimental Botany. 32:351-356.

Sardans, J. and J. Peñuelas. 2005. Drought decreases soil enzyme activity in a Mediterranean *Quercus ilex* L. forest. Soil Biology and Biochemistry. 37:455-461.

Saxe, H., D.S. Ellsworth and J. Heath. 1998. Tree and forest functioning in an enriched CO_2 atmosphere. New Phytologist. 139:395-436.

Sefcik, L.T., D.R. Zak and D.S. Ellsworth. 2006. Photosynthetic responses to understory shade and elevated carbon dioxide concentration in four northern hardwood tree species. Tree Physiology. 26:1589-1599.

Semchenko, M., M. Lepik, L. Götzenberger and K. Zobel. 2012. Positive effect of shade on plant growth: amelioration of stress or active regulation of growth rate? Journal of Ecology. 100:459-466.

Sharkey, T.D., C.J. Bernacchi, G.D. Farquhar and E.L. Singsaas. 2007. Fitting photosynthetic carbon dioxide response curves for C3 leaves. Plant, Cell & Environment. 30:1035-1040.

Siegenthaler, U., T.F. Stocker, E. Monnin, D. Lüthi, J. Schwander, B. Stauffer, D. Raynaud, J.-M. Barnola, H. Fischer, V. Masson-Delmotte and J. Jouzel. 2005. Stable carbon cycle-climate relationship during the late Pleistocene. Science. 310:1313-1317.

Sitch, S., C. Huntingford, N. Gedney, P.E. Levy, M. Lomas, S.L. Piao, R. Betts, P. Ciais, P. Cox, P. Friedlingstein, C.D. Jones, I.C. Prentice and F.I. Woodward. 2008. Evaluation of the terrestrial carbon cycle, future plant geography and climate-carbon cycle feedbacks using five Dynamic Global Vegetation Models (DGVMs). Global Change Biology. 14:2015-2039.

Smith, S., T. Huxman, S. Zitzer, T. Charlet, D. Housman, J. Coleman, L. Fenstermaker, J. Seemann and R. Nowak. 2000. Elevated CO_2 increases productivity and invasive species success in an arid ecosystem. Nature. 408:79-82.

Smith, W.K., M.J. Germino, D.M. Johnson and K. Reinhardt. 2009. The altitude of alpine treeline: a bellwether of climate change effects. Botanical Review. 75:163-190.

Solomon, S., D. Qin, M. Manning, Z. Chen, M. Marquis, K.B. Averyt, M. Tignor and H.L. Miller. 2007. *Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change*. . Cambridge University Press, Cambridge.

Stitt, M. 1991. Rising CO_2 levels and their potential significance for carbon flow in photosynthetic cells. Plant, Cell and Environment. 14:741-762.

Stoneman, G.L. 1992. Factors affecting the establishment of jarrah (*Eucalyptus marginatu)* from seed in the northern jarrah forest of Western Australia. Murdoch Univ., Australia.

Stoneman, G.L. and B. Dell. 1993. Growth of *Eucalyptus marginata* (Jarah) seedlings in a greenhouse in response to shade and soil temperature Tree Physiology. 13:239-252.

Sullivan, J. 1993. *Acer spicatum*. In: U.S. Department of agriculture, Forest Service, Rocky Mountain Research Station. Fire Sciences Laboratory. Fire effects information system.

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

Takeuchi, Y., M.E. Kubiske, J.G. Isebrands, K.S. Pregtizer, G. Hendrey and D.F. Karnosky. 2001. Photosynthesis, light and nitrogen relationships in a young deciduous forest canopy under open-air CO_2 enrichment. Plant, Cell & Environment. 24:1257-1268.

Tamm, C.O. 1991. Nitrogen and terrestrial ecosystems. . Springer, Verlag, Berlin-Heidelberg. 81, 115 p.

Temperton, V.M., S.J. Grayston, G. Jackson, C.V.M. Barton, P. Millard and P.G. Jarvis. 2003. Effects of elevated carbon dioxide concentration on growth and nitrogen fixation in *Alnus glutinosa* in a long-term field experiment. Tree Physiology. 23:1051-1059.

Tezara, W., V.J. Mitchell, S.D. Driscoll and D.W. Lawlor. 1999. Water stress inhibits plant photosynthesis by decreasing coupling factor and ATP. Nature. 401:914-917.

Tissue, D.T., R.B. Thomas and B.R. Strain. 1997. Atmospheric CO_2 enrichment increases growth and photosynthesis of *Pinus taeda*: a 4 year experiment in the field. Plant, Cell & Environment. 20:1123-1134.

Tognetti, R., G. Minotta, S. Pinzauti, M. Michelozzi and M. Borghetti. 1998. Acclimation to changing light conditions of long-term shade-grown beech (*Fagus sylvatica* L.) seedlings of different geographic origins. Trees - Structure and Function. 12:326-333.

Tognetti, R., A. Raschi and M.B. Jones. 2000. Seasonal patterns of tissue water relations in three Mediterranean shrubs co-occurring at a natural CO_2 spring. Plant, Cell & Environment. 23:1341-1351.

Townend, J. 1993. Effects of elevated carbon dioxide and drought on the growth and physiology of clonal Sitka spruce plants (*Picea stichensis* (Bong) Carr). Tree Physiology. 13:389-399.

Tschaplinski, T.J., R.J. Norby and S.D. Wullschleger. 1993. Responses of loblolly pine seedlings to elevated CO_2 and fluctuating water supply. Tree Physiology. 13:283-296.

Van Den Boogaard, R., E.J. Veneklaas and H. Lambers. 1996. The association of biomass allocation with growth and water use efficiency of two *Triticum aestivum* cultivars. Functional Plant Biology. 23:751-761.

Volk, M., P.A. Niklaus and C. Körner. 2000. Soil moisture effects determine CO_2 responses of grassland species. Oecologia. 125:380-388.

Volkova, L., L. Bennett, A. Merchant and M. Tausz. 2010. Shade does not ameliorate drought effects on the tree fern species *Dicksonia antarctica* and *Cyathea australis*. Trees - Structure and Function. 24:351-362.

Walker, M.D., C.H. Wahren, R.D. Hollister, G.H.R. Henry, L.E. Ahlquist, J.M. Alatalo, M.S. Bret-Harte, M.P. Calef, T.V. Callaghan, A.B. Carroll, H.E. Epstein, I.S. Jónsdóttir, J.A. Klein, B.ó. Magnússon, U. Molau, S.F. Oberbauer, S.P. Rewa, C.H. Robinson, G.R. Shaver, K.N. Suding, C.C. Thompson, A. Tolvanen, Ø. Totland, P.L. Turner, C.E. Tweedie, P.J. Webber and P.A. Wookey. 2006. Plant community responses to experimental warming across the tundra biome. Proceedings of the National Academy of Sciences of the United States of America. 103:1342-1346.

Wan, X.C., S.M. Landhausser, J.J. Zwiazek and V.J. Lieffers. 1999. Root water flow and growth of aspen (*Populus tremuloides*) at low root temperatures. Tree Physiology. 19:879-884.

Wand, S.J.E., G.F. Midgley, M.H. Jones and P.S. Curtis. 1999. Responses of wild C_4 and C_3 grass (Poaceae) species to elevated atmospheric CO_2 concentration: a meta-analytic test of current theories and perceptions. Global Change Biology. 5:723-741.

Wang, M., S. Shi, F. Lin, Z. Hao, P. Jiang and G. Dai. 2012. Effects of soil water and nitrogen on growth and photosynthetic response of manchurian ash (*Fraxinus mandshurica*) seedlings in northeastern China. PLoS ONE. 7:e30754.

Watanabe, M., Y. Watanabe, S. Kitaoka, H. Utsugi, K. Kita and T. Koike. 2011. Growth and photosynthetic traits of hybrid larch F1 (*Larix gmelinii* var. *japonica* × L. *kaempferi*) under elevated CO_2 concentration with low nutrient availability. Tree Physiology

Wayne, P.M. and F.A. Bazzaz. 1993. Morning vs afternoon sun patches in experimental forest gaps: consequences of temporal incongruency of resources to birch regeneration. Oecologia. 94:235-243.

Weih, M. and P.S. Karlsson. 2001. Growth response of Mountain birch to air and soil temperature: is increasing leaf-nitrogen content an acclimation to lower air temperature? New Phytologist. 150:147-155.

Weih, M. and P.S. Karlsson. 2002. Low winter soil temperature affects summertime nutrient uptake capacity and growth rate of mountain birch seedlings in the subarctic, Swedish lapland. Arctic, Antarctic, and Alpine Research. 34:434-439.

Wieser, G., K. Tegischer, M. Tausz, K.-H. Häberle, T.E.E. Grams and R. Matyssek. 2002. Age effects on Norway spruce (*Picea abies*) susceptibility to ozone uptake: a novel approach relating stress avoidance to defense. Tree Physiology. 22:583-590.

Wigley, T.M.L. 2005. The Climate Change Commitment. Science. 307:1766-1769.

Wilson, B.F. and B.C. Fischer. 1977. Striped maple: shoot growth and bud formation related to light intensity. Canadian Journal of Forest Research. 7:1-7.

Wullschleger, S.D., P.J. Hanson and T.J. Tschaplinski. 1998a. Whole-plant water flux in understory red maple exposed to altered precipitation regimes. Tree Physiology. 18:71-79.

Wullschleger, S.D., F.C. Meinzer and R.A. Vertessy. 1998b. A review of whole-plant water use studies in tree. Tree Physiology. 18:499-512.

Wullschleger, S.D., T.J. Tschaplinski and R.J. Norby. 2002. Plant water relations at elevated CO_2– implications for water-limited environments. Plant, Cell & Environment. 25:319-331.

Xu, Q., B. Huang and Z. Wang. 2002. Photosynthetic responses of creeping bentgrass to reduced root-zone temperatures at supraoptimal air temperature. Journal of the American Society for Horticultural Science. 127:754-758.

Yokota, T. and A. Hagihara. 1996. Dependence of the aboveground CO_2 exchange rate on tree size in field-grown hinoki cypress (*Chamaecyparis obtusa*). Journal of Plant Research. 109:177-184.

Zhang, S. and Q.-L. Dang. 2006. Effects of carbon dioxide concentration and nutrition on photosynthetic functions of white birch seedlings. Tree Physiology. 26:1457-1467.

Zipperlen, S.W. and M.C. Press. 1996. Photosynthesis in relation to growth and seedling ecology of two dipterocarp rain forest tree species. Journal of Ecology. 84:863-876.

Ziska, L.H., K.P. Hogan, A.P. Smith, B.G. Drake and K.P. Hogen. 1991. Growth and photosynthetic response of nine tropical species with long-term exposure to elevated carbon dioxide. Oecologia. 86:383-389.

Zogg, G.P., D.R. Zak, A.J. Burton and K.S. Pregitzer. 1996. Fine root respiration in northern hardwood forests in relation to temperature and nitrogen availability. Tree Physiology. 16:719-725.

Zon, R. and H.S. Graves. 1911. Light in relation to tree growth. Forest Service Bulletin 92. U.S. Dept Agriculture, Washington DC, USA.

Zotz, G., N. Cueni and C. Koerner. 2006. In situ growth stimulation of a temperate zone liana (*Hedera helix*) in elevated CO_2. Functional Ecology. 20:763-769.

Zotz, G., S. Pepin and C. Körner. 2005. No Down-Regulation of Leaf Photosynthesis in Mature Forest Trees after Three Years of Exposure to Elevated CO2. Plant Biology. 7:369-374.

Chapter 4: Effects of elevated [CO_2] and low soil moisture on the physiological responses of mountain maple (*Acer spicatum* L.) seedlings to light.

Introduction

The responses of shade tolerant plant species to light availability are at two endpoints. Some shade tolerant plant species grow slowly and consistently in the understory and do not respond considerably to canopy gaps. On the other hand, some persist in the understory and only show considerable increases in growth once canopy gaps are created (Canham 1989, Clark and Clark 1992). These shade tolerant plant species exhibit light foraging growth habits that enable them to discover and take advantage of the canopy gaps (Rincon and Grime 1989, Lei and Lechowicz 1990). The response of shade tolerant plant species to the creation of canopy gaps has been associated with the plasticity in suite of physiological traits (Chazdon 1988, Pearcy 1990, Kubiske and Pregitzer 1997). Plant acclimation to canopy gaps may involve higher photosynthesis per leaf area and higher stomatal conductance compared with shade plant (Canham 1988a). While light availability in canopy gaps is considered a primary determinant of photosynthetic capacity (Chazdon and Field 1987, Poorter and Oberbauer 1993, Wayne and Bazzaz 1993), other factors may influence photosynthesis. Any factors that enhance the photosynthetic rates of shade tolerant plant species in canopy gaps may play an important role in forest dynamics.

The atmospheric carbon dioxide concentration ([CO_2]) has increased since the industrial revolution, and carbon-climate models predict the increase to continue (IPCC 2007b, Meehl et al. 2007, Sitch et al. 2008). An increase in the atmosphere [CO_2] alone has, aside from affecting global climate, instant effect on plants, and thus terrestrial carbon storage (Cox et al. 2000, Körner 2003). Because CO_2 is the primary substrate for photosynthesis, and the photosynthetic machinery of C_3 plants is able to handle higher than the current [CO_2], the increase in [CO_2] is predicted to have a 'fertilization' effect on C_3 plants (Farquhar and von Caemmerer 1982, Koerner 2006, Lambers et al. 2008, Watanabe et al. 2011). The positive effect elevated [CO_2] on the photosynthetic rate is related to increased activity of the photosynthetic enzyme, ribulose-1, 5-bisphosphate carboxylase/oxygenase (Rubisco) and inhibition of photorespiration due to the shift in CO_2 : O ratio (Long and Drake 1992, Drake et al. 1997, Moore et al. 1999, Liang et al. 2001). Elevated [CO_2] also has been shown to increase apparent quantum efficiency (AQE) and lower the light compensation point (LCP) of photosynthesis, and thus increasing photosynthetic carbon gain (Kubiske and Pregitzer 1996, Osborne et al. 1997, Saxe et al. 1998). Numerous studies have reported increases in photosynthesis under elevated [CO_2], at least in the short term (Medlyn et al. 1999, Nowak et al. 2004, Ainsworth and Long 2005). Higher absolute photosynthetic rates in high light environment (Poorter and Oberbauer 1993, Lei and Lechowicz 1997b), suggests that elevated [CO_2] can be expected to further increase photosynthetic carbon gain in high light environment. However, environmental stress can influence CO_2 responsiveness. For example, low soil moisture reduces the stimulatory effect elevated [CO_2] (Owensby et al. 1993, Samarakoon and Gifford 1995, Campbell et al. 1997, Smith et al. 2000, Volk et al. 2000), and may limit the photosynthetic carbon gain in high light environment . However, the effect of [CO_2] and soil moisture on the physiological responses of plants to high light regime is less understood. A good understanding of the effects of climate change on shade tolerant species in simulated canopy gaps may be critical for a reliable prediction of forest dynamics in the future.

The changes in the global climate are predicted to be accompanied by 1.4 – 5.8 °C increase in global mean temperatures by the end of this century (Mitchell et al. 1990, IPCC 2007b, Meehl et al. 2007, Solomon et al. 2007). Increases in temperature will likely cause a decrease in soil moisture due to increased rate and depth of evapotranspiration (Rind et al. 1990, Pregitzer and King 2005). The negative effect of low soil moisture on mature trees (Èermák et al. 1993, Wullschleger et al. 1998b, Èermák and Kuèera 1993), seedlings and saplings (Abrams et al. 1990, Kubiske and Abrams 1994, Wullschleger et al. 1998a) has been demonstrated. Plants that are growing at low soil moisture conditions have lower photosynthetic rates because of decreased stomatal conductance (Ridolfi and Dreyer 1997, Cornic 2000, Flexas and Medrano 2002, Lawlor 2002, Lawlor and Cornic 2002, Ohashi et al. 2006). Therefore, we hypothesize that low soil moisture limits elevated CO_2 stimulation of photosynthesis, and that low soil moisture causes a greater reduction of photosynthesis in mountain maple seedlings in grown in high light.

In the present study, the physiological responses of mountain maple (*Acer spicatum* L.) to light under different [CO_2] and soil moisture were studied. Mountain maple is an important understory shrub or tree species in the boreal mixedwood that contributes immensely to the composition, structure and diversity of such forests (Sullivan 1993, Archambault et al. 1998). It influences the amount of light reaching the forest floor, thus affecting the growth of other plant species (Aubin et al. 2000). Mountain maple grows on a wide range of habitats and persists through all the stages of forest development (Gauthier et al. 1993). Its phenotypic plasticity in response to light allows it to acclimate to a large range of light conditions and respond rapidly to canopy gaps after the removal of overstory vegetation (Lei and Lechowicz 1990, De Grandpré et al. 1993, Aubin et al. 2005). Previous studies have also indicated that mountain maple is sensitive to low soil moisture conditions (Paula 2004). Despite the evidence that mountain maple respond rapidly to canopy gaps, we still lack insights into how low soil moisture may affect mountain maple's physiological responses to light under future climate.

Materials and Methods

Plant materials and germination procedure

Mountain maple seeds were collected from the Jack Haggerty Forest located approximately 37km north of Thunder Bay, ON. Seeds were soaked in a 1000μmol mol^{-1} giberellic acid (GA) for 24hrs. The seeds were kept at 4 °C temperature for 2 months on moistened paper towels. The seed coats were cracked to facilitate germination after the stratification. Germination occurred in a 2:1 mixture of vermiculite and peat moss in horticultural trays at Lakehead University greenhouse. The average environmental conditions in the greenhouse during the germination were: 22/16 °C day/night temperature and a photoperiod of 16hr (maximum summertime photoperiod for Thunder Bay, ON, according to Environment Canada Weather Report, 2010). The growing medium was maintained moist by sprinkling them with water daily. Three weeks after germination, 160 relatively similar-sized seedlings were transplanted into plastic containers (31.5 cm deep, 11 cm top diameter and 9.5 cm bottom diameter) with the same growing medium composition as described above.

Experimental design

The experiment followed a split-split- plot factorial design. The main-plot treatment comprised of two levels of CO_2 (392 and 784 μmol mol^{-1}) with two independent replications for each level. The elevated [CO_2] (760 μmol mol^{-1}) was achieved using Argus CO_2 generators (Argus, Vancouver, BC, Canada). The [CO_2] in each greenhouse was monitored using Argus environmental control system (Argus, Vancouver, BC, Canada). The sub-plot treatment consisted of two levels of light within each CO_2 treatment. The light levels were high light (HL) and low light (LL). Metal frames with their tops and sides covered with neutral density shade cloth were constructed to provide the LL level. The shading reduced the amount of light by 70%. High pressure sodium lamps were mounted above the benches to provide supplemental light on cloudy days and to extend the photoperiod to 16hr. The sub-sub-plot treatment comprised of two soil moisture treatments within each sub-plot. In the high soil moisture treatment, the seedlings were watered daily. The seedlings were watered to the dripping point during each watering event. Seedlings in the low soil moisture treatment were watered every 2 - 4 days. The moisture content was about 40 – 50% of that in the high soil moisture. The moisture level in the growing medium was measured daily using HH2 moisture meter. The low soil moisture treatment started one week after the seedlings were transplanted to allow for establishment.

The environmental conditions of the greenhouses were set at 22/16 °C day/night air temperature and relative humidity of 50%. These were controlled and monitored by Argus environmental control system (Argus, Vancouver, BC, Canada). The seedlings were fertilized once or twice a week with 100, 15, 57, 6, 6 and 11 mg/L of N, P, K, Ca, Mg and S, respectively. The nutrient concentrations were determined based on the studies of Canham *et al*. (1996) and Ingestad (1981) on *Acer* species and other deciduous tree species.

Photosynthetic light and CO_2 responses

The light response curves of photosynthesis at the corresponding growth [CO_2] were measured at seven PAR levels: 1100, 800, 400, 100, 60, 10 and 0μmol m^{-2} $s^{-1.}$ The measurement was carried out on three randomly selected seedlings in each treatment condition. The measurement was done between 10:00–15:00 h with a Li-Cor 6400 open gas exchange system (LI-6200. LI-COR. Inc., Lincoln. Nebraska. USA). The relative humidity (RH) and temperature within the leaf chamber were 50% and 22 ºC, respectively. The light compensation points (LCP) and apparent quantum efficiency (AQE) of the seedlings were obtained using the Photosyn Assistant software (Dundee Scientific, Scotland, UK). Values were expressed on one-sided leaf area basis.

The A/C_i curves were measured on the same seedlings and leaves used in the light response measurements. The measurements were taken at 50, 100, 200, 400, 800, 1000 and 1500 μmol mol^{-1} CO_2. PAR of 600μmol m^{-2} s^{-1}, RH of 50% and leaf temperature of 22 ºC were used. The gas exchange variables (net photosynthesis, A and stomatal conductance, g_s) of the seedlings were expressed on one-sided leaf area basis. The A/C_i curves were analyzed using the Curve Fitting Utility version 1.1 developed by Sharkey *et al.* (2007) to estimate the maximum rate of Rubisco carboxylation (V_{cmax}), photosynthetic electron transport rate (J), triose phosphate utilization (TPU) and dark respiration (R_d). They were adjusted to the measurement leaf temperature of 22 °C to obtain the appropriate estimates (Sharkey et al. 2007) because actual leaf temperature differed as a result of different transpiration rates.

Xylem water potential measurements

The seedlings used in the above measurements were also used to measure midday xylem water potential (Ψ_x). The measurement was done with a Scholander pressure chamber (Soil Moisture Stress, Inc., Santa Barbara, CA, USA) between 12: 00 and 15: 00 hours. The terminal shoot from each seedling was placed in the chamber with the cut end protruding outside the chamber. The pressure inside the chamber was gradually increased until sap appeared at the cut end. The reading on the pressure gauge was then immediately recorded.

Statistical analysis

The data were analyzed with Data desk 6.01 Statistical Package (Data Description 1996). The normality and homogeneity of variance were examined graphically using probability plots and histograms, respectively, before analyses of variance (ANOVA) was done. All the data met the two assumptions. The effects of [CO_2], light regimes, soil moisture regimes, and their interactions were tested by three-factor, split-split plot ANOVA. The significant level was set at $P \leq 0.05$ but P –values ≤ 0. 10 were considered marginally significant due to restriction on the replication of this study. Scheffe's *post hoc* test was done when an interaction was significant.

The linear model for the ANOVA is give below:

$Y_{ijkl} = \mu + C_i + \varpi(i)_j + L_k + CL_{ik} + \beta_{(ik)\,l} + M_m + CM_{im} + LM_{km} + CLM_{ikm} + \varepsilon_{(ijklm)n}$

i = 1, 2; j = 1, 2; k = 1, 2; l = 1; m = 1, 2; n = 1; Where,

Y_{ijkl} = the measured response of the j^{th} replicate of the l^{th} light regime in the k^{th} T_{soil} and the i^{th} CO_2 concentration.

μ = the overall mean.

C_i = the fixed effect of the i^{th} CO_2 concentration.

$\varpi(i)_j$ = the whole plot error due to restriction on the randomization of the $CO_{2.}$

L_k = the fixed effect of the k^{th} light régime.

CL_{ik} = the interaction effect of the k^{th} light regime in the i^{th} CO_2 level.

$\beta_{(ij)l}$ = the sub-plot error due to the restriction on the randomization of the k^{th} light regime in the i^{th} CO_2 level.

M_m= the fixed effect of the m^{th} soil moisture regime.

CM_{im} = the interaction effect of the m^{th} soil moisture regime in the in the i^{th} CO_2 level.

LM_{km} = the interaction effect of the m^{th} soil moisture in the k^{th} light regime.

CLM_{ikm} = the interaction effect of the m^{th} soil moisture regime in the k^{th} light regime and the i^{th} CO_2 level.

$\varepsilon_{(ijkl)}$ = the sub-sub-plot error.

Apendix 2.1. EMS Table

	df	2 F i	2 F k	2 F m	2 R j	EMS
C_i	1	0	2	2	2	$\sigma^2 + 4\sigma^2_{\varpi} + 6\phi C$
$\varpi(i)_j$	2(2-1)=2	1	2	2	2	$\sigma^2 + 4\sigma^2_{\varpi}$
L_k	1	2	0	2	2	$\sigma^2 + 4\sigma^2_{\beta} + 6\phi_L$
CL_{ik}	1	0	0	2	2	$\sigma^2 + 4\sigma^2_{\beta} + 4\phi_{CL}$
$\beta_{(jk)l}$	2x1=2	1	1	2	2	$\sigma^2 + 4\sigma^2_{\beta}$
M_m	1	2	2	0	2	$\sigma^2 + 8\phi_M$
CM_{im}	1	0	2	0	2	$\sigma^2 + 4\phi_{CM}$
LM_{km}	1	2	0	0	2	$\sigma^2 + 4\phi_{LM}$
CLM_{ikm}	1	0	0	0	2	$\sigma^2 + 2\phi_{CLM}$
$\varepsilon_{(ijklm)n}$	2x1x1=2	1	1	1	1	σ^2
Total	12					

Results

The interaction between light and soil moisture significantly affected A and C_i/C_a (Table 3.1). Additionally, there was a marginal significant interactive effect between light and soil moisture on g_s (Table 3.1). The high light treatment increased A, g_s and C_i/C_a ratio only at the high soil moisture treatment. However, the high light treatment had no significant effect on A, but decreased g_s and C_i/C_a by 22% and 14%, respectively, at the low soil moisture treatment (Fig. 3.1A, 3.1B and 3.1C). In the low light treatment, no significant soil moisture effect on A, g_s or C_i/C_a was found. However, the low soil moisture treatment significantly decreased A, g_s and C_i/C_a by 29%, 43% and 17%, respectively, in the high light treatment (Figs. 1A, 1B, and 1C). Furthermore, the elevated [CO$_2$] significantly (Table 3.1) increased A by 72% but its interaction with light or soil moisture did not have any significant effect on A.

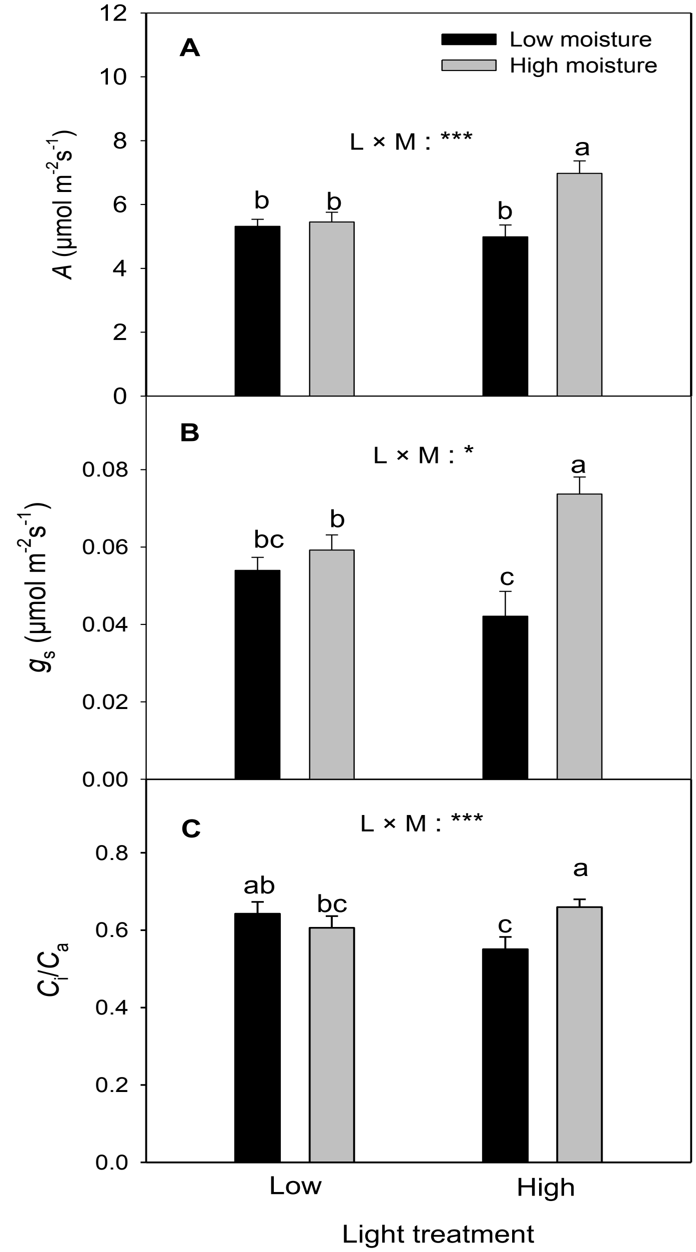

Figure 3.1. Effects of [CO_2], light (L) and soil moisture (M) on net photosynthesis (*A*), stomatal conductance to CO_2 (g_s) and internal to ambient [CO_2] ratio (C_i/C_a) (mean ± SE, n = 10) of mountain maple. Seedlings were exposed to two [CO_2] (380 and 760 µmol mol^{-1}), two light regimes (high and low) and two soil moisture regimes (high and low) for two months. The letters on the bars represent L × M interaction. Significant treatment effects are marked as significant: $P \leq 0.01$, ***; $P \leq 0.05$, **; and marginally significant: $P \leq 0.10$, *. Means with same letter(s) are not statistically significant ($P > 0.10$) from each other or one another.

The interaction between CO_2 and soil moisture had a significant effect on C_i/C_a (Table 3.1). C_i/C_a was significantly lower in the low soil moisture under ambient [CO_2] but no significant soil moisture effect on C_i/C_a was found under the elevated [CO_2] (Fig. 3.2A). The elevated [CO_2], however, significantly increased C_i/C_a in the low soil moisture treatment (Fig. 3.2A).

The interaction between CO_2 and light had a significant effect on IWUE (Table 3.1). The high light increased IWUE by 51% under elevated [CO_2] but had no significant effect on IWUE under the ambient [CO_2] (Fig. 3.2B). The elevated [CO_2] increased IWUE at both light treatments; the magnitude of increase was however, higher in the high compared with the low light treatment (117% vs. 55%, Fig. 3.2B).

The interaction between CO_2 and light had a marginal significant effect on V_{cmax} but a significant effect on J (Table 3.1). The high light treatment resulted in higher V_{cmax} (20%) and J (19%) under elevated [CO_2] but had no significant effect on V_{cmax} or J under the ambient [CO_2] (Figs. 3.3A and 3.3B). There was no significant [CO_2] affect on V_{cmax} in either light treatments or on J in the low light treatment (Figs. 3.3A and 3.3B). However, the elevated [CO_2] significantly increased J by 44% in the high light treatment (Fig. 3.3B).

The interaction between CO_2 and light had a significant effect on V_{cmax} and J (Table 3.1). Elevated [CO_2] did not significantly affect V_{cmax} in either light treatments or J in the low light treatment (Figs. 3.3A and 3.3B). However, the elevated [CO_2] significantly increased J by 44% in the high light treatment (Fig. 3B). Furthermore, the low light treatment resulted in lower V_{cmax} (20%) and J (19%) compared with the high light under elevated [CO_2] but not under ambient [CO_2] (Figs. 3.3A and 3.3B).

TPU was significantly and marginally affected by light × soil moisture and CO_2 × soil moisture interactions, respectively (Table 3.1). The high light treatment significantly increased TPU by 20% at the high but had no significant effect on TPU at the low soil moisture treatment (Fig. 3.3C). Furthermore, the low soil moisture decreased TPU by 9% at the high light but no significant soil moisture effect on TPU was found at the low light treatment (Fig. 3.3C). Soil

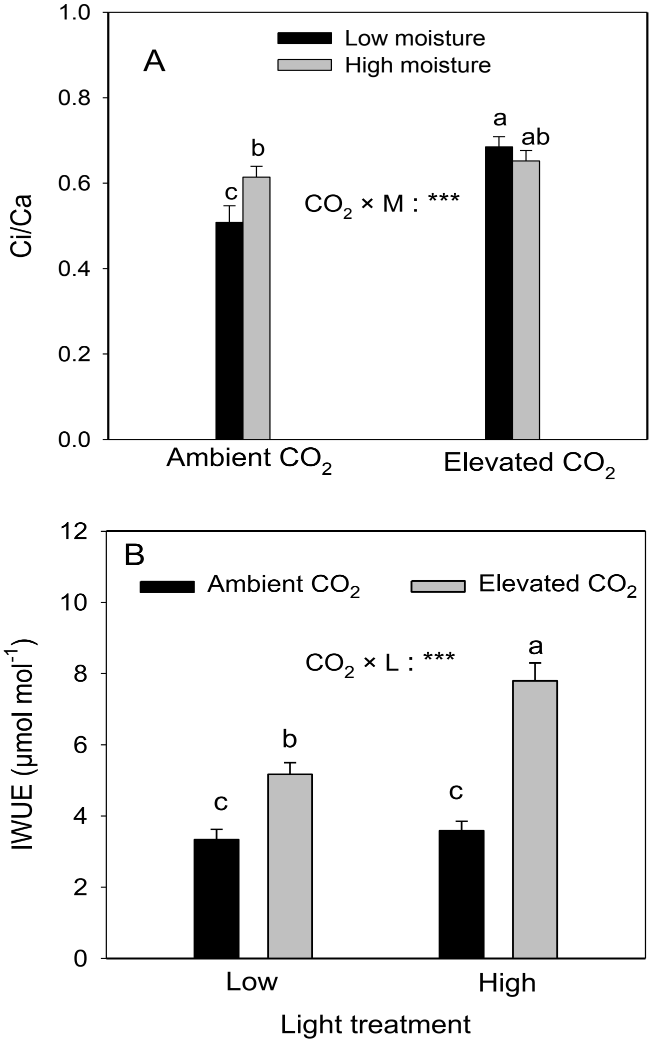

Figure 3.2. Effects of $[CO_2]$, L and M on C_i/C_a ratio and instantaneous water-use efficiency (IWUE) (mean ± SE, n = 10) of *Acer spicatum*. Refer to Figure 3.1 for other explanations.

moisture did not significantly affect TPU under either [CO_2] (Fig. 3D). However, the elevated [CO_2] significantly increased TPU by 19% at the low but had no significant effect on TPU at the high soil moisture (Fig. 3.3D).

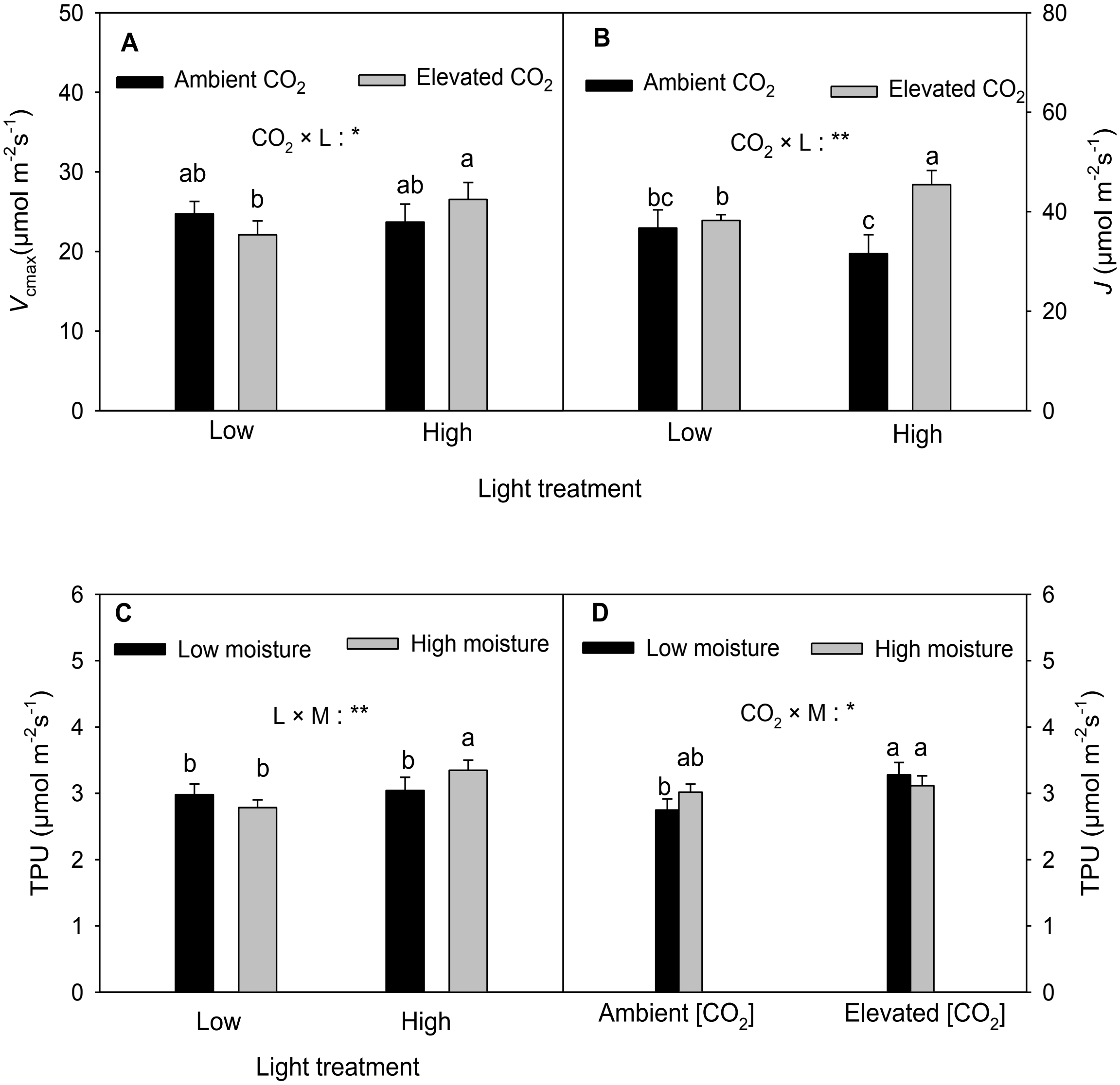

Figure 3.3. Effects of [CO_2], and L and M on the maximum rate of carboxylation (V_{cmax}), photosynthetic electron transport rate (*J*) and triose phosphate utilization (TPU) (mean ± SE, n = 10) of *Acer spicatum* seedlings. Refer to Figure 3.1 for other explanations.

There was a marginal significant interactive effect among CO_2, light and soil moisture on LCP (Table 3.1). The high light treatment increased LCP under all the CO_2 and soil moisture combinations (Fig. 3.4A). LCP increased in response to low soil moisture under the ambient $[CO_2]$ and low light treatment while it showed an opposite trend under the elevated $[CO_2]$ and low light treatment (Fig. 3.4A).

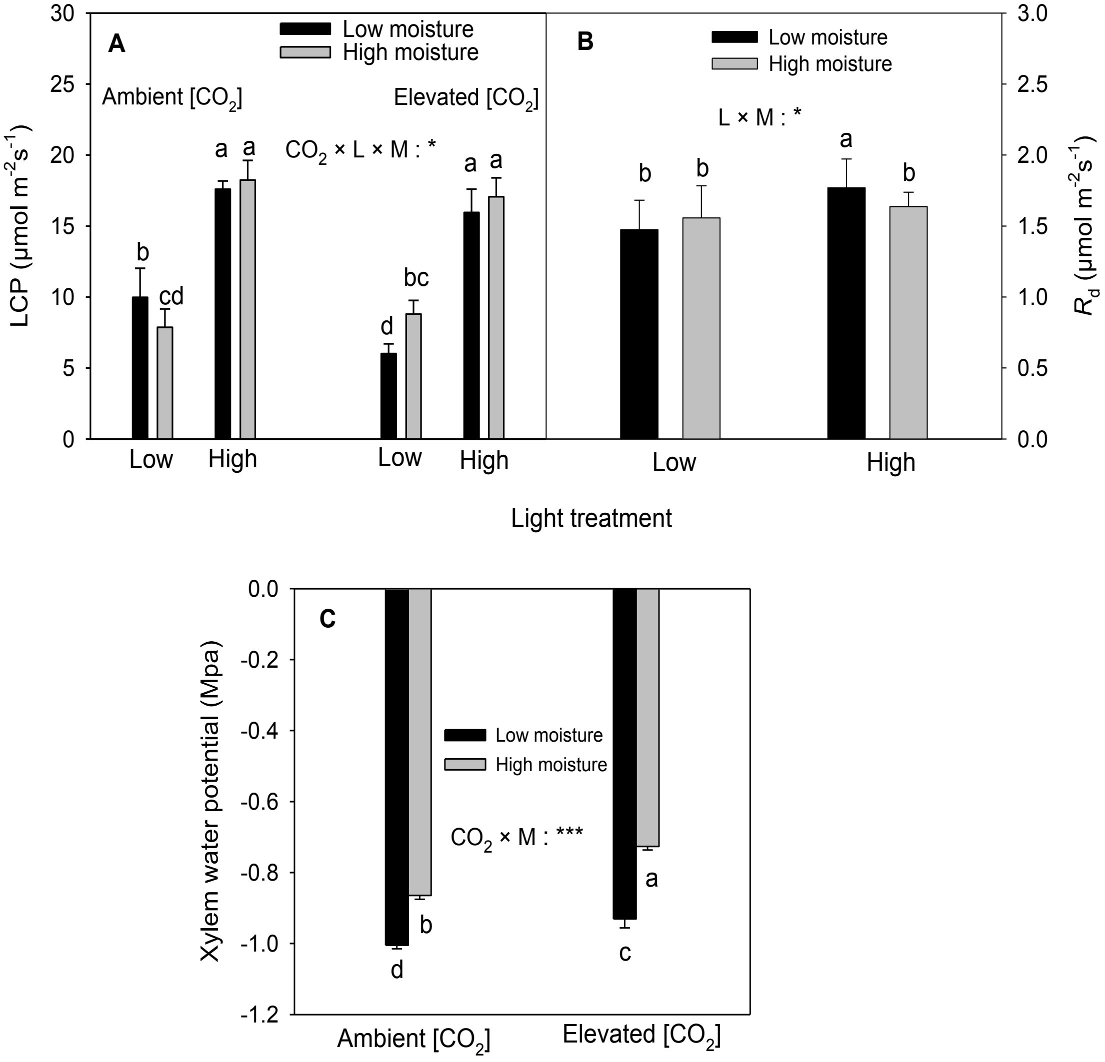

Figure 3.4. Effects of $[CO_2]$, L and M on photosynthetic light compensation point (LCP), dark respiration rate (Rd) and midday xylem water potential (mean ± SE, n = 10) of *Acer spicatum* seedlings. Refer to Figure 3.1 for other explanations.

No significant soil moisture effect on LCP was found at the high light under either [CO_2]. The elevated [CO_2] significantly reduced LCP only in the low soil moisture and low light treatment combination but had no significant effect on LCP at the other treatment combinations (Fig. 3.4A).

The elevated [CO_2] significantly increased the AQE of photosynthesis by 24% but no other significant effects on AQE were found (Table 3.1). The interaction between light and soil moisture had a marginal significant effect on R_d (Table 3.1). The high light treatment increased R_d at the low soil moisture but had no significant effect on R_d at the high soil moisture treatment (Fig. 3.4B). At the low light treatment, the low soil moisture did not significantly affect R_d (Fig. 3.4B). However, the low soil moisture significantly increased R_d at the high light treatment.

The interaction between CO_2 and soil moisture significantly affected midday Ψ_x (Table 3.1). Ψ_x significantly decreased in response to low soil moisture under both ambient and elevated [CO_2] (Fig. 3.4C). Furthermore, the elevated [CO_2] significantly increased Ψ_x (less negative) at both soil moisture treatments, but the magnitude was greater in the high (19%) than low (8%) soil moisture (Fig. 3.4C). The low light treatment significantly increased Ψ_x compared with high light treatment (Table 3.1).

Discussion

This study shows that the net photosynthesis of mountain maple seedling was less sensitive to high light under the low soil moisture. It is a little surprising that the seedlings at the low soil moisture showed a relatively stable *A* in response to the high light. This contrasts partially with the hypothesis that the low soil moisture would limit *A* response to the high light treatment.

Table 3.1. The p-values of ANOVA on the effects of CO_2 (C), soil moisture (M) and light (L) on net photosynthesis (A), stomatal conductance to CO_2 (g_s), water-use efficiency (IWUE), C_i/C_a ratio, maximum rate of carboxylation (V_{cmax}), photosynthetic electron transport rate (J), triose phosphate utilization (TPU), light compensation point (LCP), apparent quantum efficiency (AQE), dark respiration rates (R_d) and midday xylem water potential (Ψ_x) of mountain maple seedlings. The seedlings were grown under ambient (380 μmol mol^{-1}) and elevated (760 μmol mol^{-1}) [CO_2]. They were exposed to well-watered and water –stressed treatments in high and low light environments. Measurements were taken two months after the start of the treatment.

Source of variation	CO_2	L	CO_2*L	M	CO_2*M	L*M	CO_2*L*M
A	**0.0208**	0.1220	0.1630	**0.0015**	0.4976	**0.0049**	0.3142
g_s	0.1585	0.8028	0.8833	**0.0025**	0.2629	**0.0985**	0.7501
IWUE	**0.0327**	**≤ 0.0001**	**≤ 0.0001**	0.4910	0.1338	0.4724	0.7446
C_i/C_a	**0.0378**	0.4321	0.5166	0.1210	**0.0039**	**0.0029**	0.4671
V_{cmax}	0.8930	0.3022	**0.0688**	0.3443	0.1489	0.3211	0.3606
J	0.2986	0.5772	**0.0192**	**0.0231**	0.1926	0.3105	0.4820
TPU	0.4220	**0.0150**	0.5392	0.6351	**0.0897**	**0.0478**	0.5511
LCP	0.6357	**≤ 0.0001**	0.8361	0.5474	**0.0874**	0.3028	**0.0481**
AQE	**0.0415**	0.9659	0.6591	0.3753	0.5789	0.4096	0.9901
R_d	0.1332	**0.0567**	0.5171	0.3160	0.3386	**0.0931**	0.1079
Ψ_x	**0.0064**	**≤0.0001**	0.8820	**≤0.0001**	**0.0056**	0.6838	0.5897

The results are also in contrast with the findings of other studies that report that low soil moisture decreases *A* response to high light conditions because of reductions in g_s and leaf area (Muraoka et al. 1997, Muraoka et al. 2002). I expected decreased g_s or leaf area in the high light under low soil moisture to have a negative effect on *A*. Although g_s showed a trend towards a decrease, there was no significant difference between the low and high light under the low soil moisture. Similarly, the high light treatment did not decrease leaf area production when seedlings were exposed to low soil moisture (no significant L ×M, Chapter 5 of this thesis). The results mean that the high light response was not constrained by g_s or the size photosynthetic machinery under the low soil moisture. The results show that reduction in soil moisture supply alone may have little effect on *A* response of mountain maple seedling to high light conditions. However, multiple resource limitations may act to affect mountain maple physiological performances in high light conditions (Chapin III et al. 1987, Canham et al. 1996, James et al. 2005).

Both stomatal and non-stomatal limitation to *A* in response to high light under the low soil moisture was found in this study, but one of them appeared to be the primary limiting factor. Stomatal and non-stomatal limitations to *A* appear vary among plants species or the conditions of the study (Epron and Dreyer 1993, Palanisamy 1999, Tezara et al. 1999, Cornic 2000, Ambebe and Dang 2009). Although the high light did not decrease g_s under the low soil moisture, I observed an increased stomatal limitation to *A*. C_i/C_a ratio which decreases when stomatal limitation to *A* is high (Dang and Cheng 2004, Ambebe and Dang 2009), was observed in the high light treatments under low soil moisture. However, this did not result in a concomitant decline in *A*, suggesting that stomatal limitation to *A* was not the primary limiting factor. The high light had parallel effect on *A* and TPU under the low soil moisture, indicating that potential biochemical limitations to *A* were similar between the low and high light under low soil moisture. Thus, non-stomatal factors might be the primarily regulators of A response to high light under low soil moisture in this study. Although light availability is considered the primary determinant of A (Chazdon and Field 1987, Poorter and Oberbauer 1993, Wayne and Bazzaz 1993, Muraoka et al. 1997), the results reveal that the response of A to light high conditions might be considerably limited under low soil moisture due to its strong effect on g_s and TPU.

The photosynthetic light response parameters suggest that mountain maple seedlings in low light condition maintain positive carbon balance by reducing R_d and LCP. Lower values of R_d and LCP and higher AQE are considered to be shade-tolerance mechanisms which allow plants to maintain a positive carbon balance and extend the duration of photosynthetic carbon gain (Liang et al. 2001, Pothier and Prévost 2002, Craine and Reich 2005). Our results show that seedlings in low light seem to adopt a strategy that extend the net photosynthetic gain time during the day when grown under elevated [CO_2] and low soil moisture. Carbon conversation strategy (lower R_d) in seedling in low light treatment was adopted when soil moisture was low, but the response was independent of [CO_2] treatment. Reasons for such response are unclear, but varied treatments effect on the physiological mechanisms that ensure the maintenance of positive carbon balance might have played a role. The significance of lower LCP in low light under elevated [CO_2] and low soil moisture remains to be determined, as LCP alone does not necessarily indicate positive carbon balance (Messier et al. 1999, Pothier and Prévost 2002). The finding that low light treatment had no significant effect on AQE is consistent with those of (Sefcik et al. 2006) but inconsistent with the results of (Liang et al. 2001). However, our results demonstrate that the physiological mechanisms controlling positive carbon balance in low light may be species-specific and is mediated by other environmental factors.

The elevated [CO_2] decreased drought tolerance of mountain maple seedlings. The elevated [CO_2] increased seedlings xylem water potential under both soil moisture but the magnitude was greater under the high compared with the low soil moisture. Elevated CO_2-induced increases in seedlings growth can deplete soluble concentration in leaves, thus increasing osmotic potential and decreasing drought tolerance (Wullschleger et al. 2002). As a consequence, elevated [CO_2] has been shown to increase xylem water potential in six families of *Prosopis glandulosa* and *Andropogon gerardii* (Knapp et al. 1993, Polley et al. 1999). Tschaplinski *et al*. (1993) used a pressure chamber to determine the effect of elevated [CO_2] and low soil moisture on osmotic adjustment in shoots of loblolly pine seedlings. They found that elevated [CO_2] increased osmotic potential at turgor loss point in the seedlings at low and high soil moisture by 17.42% and 17.02%, respectively. However, they found that the elevated [CO_2] increased biomass allocation to roots, which could potentially mitigate the drought effect and enhance continued growth. I found that the elevated [CO_2] increased RMR at both soil moisture treatments but the magnitude of increase was greater under the low compared with the high soil moisture (chapter 5 of this thesis, Fig. 4.4B). This is in agreement with previous studies that found increased biomass allocation to roots as a drought tolerant mechanism under elevated [CO_2] (Morison 1993, Townend 1993, Allen et al. 1998, Tognetti et al. 2000). Therefore, the higher xylem water potential observed at both soil moisture treatments under elevated [CO_2] should be mitigated by increased biomass allocation to roots for improved absorption and increased drought tolerance in mountain maple seedlings.

In conclusion, elevated [CO_2] and low soil moisture resulted in different physiological responses of mountain maple seedlings to light. I found no significant CO_2 × M interactive effect on the observed physiological traits, except LCP, responses to light, indicating that the stimulation of physiological responses of mountain maple in high light might not be limited by low soil moisture. The photosynthetic response of mountain maple to light was associated with sink strength under low soil moisture but showed no significant difference between low and high light treatments. The elevated [CO_2] decreased mountain maple drought tolerance by increasing xylem water potential but appear to offset that by increasing biomass allocation to roots. Although the physiological responses of mountain maple seedling to light in this short-term study cannot be extrapolated to mature trees in the field, the study highlights the potential responses of mountain maple seedling to different light conditions in future climate change scenarios.

References

Abrams, M.D., M.E. Kubiske and K.C. Steiner. 1990. Drought adaptations and responses in five genotypes of *Fraxinus pennsylvanica* Marsh.: photosynthesis, water relations and leaf morphology. Tree Physiology. 6:305-315.

Ainsworth, E.A. and S.P. Long. 2005. What have we learned from 15 years of free-air CO_2 enrichment (FACE)? A meta-analytic review of the responses of photosynthesis, canopy properties and plant production to rising CO_2. New Phytologist. 165:351-372.

Allen, L.H., R.R. Valle, J.W. Jones and P.H. Jones. 1998. Soybean leaf water potential responses to carbon dioxide and drought. Agron. J. 90:375-383.

Ambebe, T.F. and Q.-L. Dang. 2009. Low moisture availability inhibits the enhancing effect of increased soil temperature on net photosynthesis of white birch (*Betula papyrifera*) seedlings grown under ambient and elevated carbon dioxide concentrations. Tree Physiology. 29:1341-1348.

Archambault, L., J. Morissette and M. Bernier-Cardou. 1998. Forest succession over a 20-year period following clearcutting in balsam fir yellow birch ecosystems of eastern Quebec, Canada. Forest Ecology and Management. 102:61-74.

Aubin, I., M. Beaudet and C. Messier. 2000. Light extinction coefficients specific to the understory vegetation of the southern boreal forest, Quebec. Canadian Journal of Forest Research-Revue Canadienne De Recherche Forestiere. 30:168-177.

Aubin, I., C. Messier and D. Kneeshaw. 2005. Population structure and growth acclimation of mountain maple along a successional gradient in the southern boreal forest. Ecoscience. 12:540-548.

Campbell, B.D., D.M. Stafford Smith and G.M. McKeon. 1997. Elevated CO_2 and water supply interactions in grasslands: A pastures and rangelands management perspective. Global Change Biology. 3:177-187.

Canham, C.D. 1988. Growth and canopy architecture of shade-tolerant trees: response to canopy gaps. Ecology. 69:786-795.

Canham, C.D. 1989. Different response to gaps among shade-tolerance tree species. Ecology. 70:549 - 550.

Canham, C.D., A.R. Berkowitz, V.R. Kelly, G.M. Lovett, S.V. Ollinger and J. Schnurr. 1996. Biomass allocation and multiple resource limitation in tree seedlings. Canadian Journal of Forest Research. 26:1521-1530.

Chapin III, S.F., A.J. Bloom, C.B. Field and R.H. Waring. 1987. Plant responses to multiple environmental factors. BioScience. 37:49-57.

Chazdon, R.L. 1988. Sunflecks and their importance to forest understory plants. Adv. Ecol. Research. 18:1–63.

Chazdon, R.L. and C.B. Field. 1987. Determinants of photosynthetic capacity in six rainforest *Piper* species. Oecologia. 73:222-230.

Clark, D.A. and D.B. Clark. 1992. Life history diversity of canopy and emergent trees in a neotropical rain forest. Ecological Monographs. 62:315-344.

Cornic, G. 2000. Drought stress inhibits photosynthesis by decreasing stomatal aperture - not by affecting ATP synthesis. Trends in Plant Science. 5:187-188.

Cox, P.M., R.A. Betts, C.D. Jones, S.A. Spall and I.J. Totterdell. 2000. Acceleration of global warming due to carbon-cycle feedbacks in a coupled climate model. Nature. 408:184-187.

Dang, Q.-L. and S. Cheng. 2004. Effects of soil temperature on ecophysiological traits in seedlings of four boreal tree species. Forest Ecology and Management. 194:379-387.

De Grandpré, L.D., D. Gagnon and Y. Bergeron. 1993. Changes in the understory of Canadian southern boreal forest after fire. Journal of Vegetation Science. 4:803-810.

Drake, B.G., M.A. Gonzàlez-Meler and S.P. Long. 1997. More efficient plants: A consequence of rising atmospheric CO_2? Annual Review of Plant Physiology and Plant Molecular Biology. 48:609-639.

Èermák, J. and J. Kuèera. 1993. Extremely fast changes in xylem flow rate in tall trees caused by atmospheric, soil and mechanical factors. In Water Transport in Plants Under Climatic Stress. Cambridge University Press, Cambridge, UK.

Èermák, J., R. Matyssek and J. Kuèera. 1993. Rapid responses of large, drought-stressed beech trees to irrigation. Tree Physiology. 12:281-290.

Epron, D. and E. Dreyer. 1993. Long-term effects of drought on photosynthesis of adult oak trees [*Quercus petraea* (Matt.) Liebl. and *Quercus robur* L.] in a natural stand. New Phytologist. 125:381-389.

Farquhar, G.D. and S. von Caemmerer. 1982. Modelling of photosynthetic response to environmental conditions. In encyclopaedia of plant physiology, physiological plant ecology II: water relations and carbon assimilation Springer Verlag, Berlin.

Flexas, J. and H. Medrano. 2002. Drought-inhibition of photosynthesis in C_3 plants: stomatal and non-stomatal limitations revisited. Annals of Botany. 89:183-189.

Gauthier, S., J. Gagnon and Y. Bergeron. 1993. Population age structure of *Pinus banksiana* at the southern edge of the Canadian boreal forest. Journal of Vegetation Science. 4:783-790.

Ingestad, T. 1981. Nutrition and growth of birch and grey alder seedlings in low conductivity solutions and at varied relative rates of nutrient addition. Physiologia Plantarum. 52:454-466.

IPCC. 2007. Summary for policymakers. In: Solomon, S. et al. (eds), Climate Change 2007: the physical science basis. Contribution of Working Group I to the fourth assessment report of the Intergovernmental Panel on Climate Change.

James, J.J., R.L. Tiller and J.H. Richards. 2005. Multiple resources limit plant growth and function in a saline-alkaline desert community. Journal of Ecology. 93:113-126.

Knapp, A.K., E.P. Hamerlynck and E.O. Clenton. 1993. Photosynthetic and water relations responses to elevated CO_2 in the C_4 grass *Andropogon gerardii*. International Journal of Plant Sciences. 154:459-466.

Koerner, C. 2006. Plant CO_2 responses: an issue of definition, time and resource supply. New Phytologist. 172:393-411.

Körner, C. 2003. Ecological impacts of atmospheric CO_2 enrichment on terrestrial ecosystems. Philosophical Transactions of the Royal Society of London Series A. 361:2023-2041.

Kubiske, M.E. and M.D. Abrams. 1994. Ecophysiological analysis of woody species in contrasting temperate communities during wet and dry years. Oecologia. 98:303-312.

Kubiske, M.E. and K.S. Pregitzer. 1996. Effects of elevated CO_2 and light availability on the photosynthetic light response of trees of contrasting shade tolerance. Tree Physiology. 16:351-358.

Kubiske, M.E. and K.S. Pregitzer. 1997. Ecophysiological responses to simulated canopy gaps of two tree species of contrasting shade tolerance in elevated CO_2. Functional Ecology. 11:24-32.

Lambers, H., S.F. Chapin III and T.L. Pons. 2008. Plant physiological eclogy. Springer, New York.

Lawlor, D.W. 2002. Limitation to photosynthesis in water-stressed leaves: stomata vs. metabolism and the role of ATP. Annals of Botany. 89:871-885.

Lawlor, D.W. and G. Cornic. 2002. Photosynthetic carbon assimilation and associated metabolism in relation to water deficits in higher plants. Plant, Cell and Environment. 25:275-294.

Lei, T.T. 1992. Functional design and shade adaptation in *Acer* species. *In* Biology. McGill University.

Lei, T.T. and M.J. Lechowicz. 1990. Shade adaptation and shade tolerance in saplings of three *Acer* species from eastern north America. Oecologia. 84:224-228.

Lei, T.T. and M.J. Lechowicz. 1997. The photosynthetic response of eight species of *Acer* to simulated light regimes from the centre and edges of gaps. Functional Ecology. 11 16 - 23.

Liang, N.H., Y. Tang and T. Okuda. 2001. Is elevation of carbon dioxide concentration beneficial to seedling photosynthesis in the understory of tropical rain forests? Tree Physiology. 21:1047-1055.

Long, S.P. and B.G. Drake. 1992. Photosynthetic CO_2 assimilation and rising atmospheric CO_2 concentrations. In Crop Photosynthesis: spatial and temporal determinants. Elsevier Science Publishers B.V., Amsterdam.

Medlyn, B.E., F.W. Badeck, D.G.G. De Pury, C.V.M. Barton, M. Broadmeadow, R. Ceulemans, P. De Angelis, M. Forstreuter, M.E. Jach, S. Kellomäki, E. Laitat, M. Marek, S. Philippot, A. Rey, J. Strassemeyer, K. Laitinen, R. Liozon, B. Portier, P. Roberntz, K.

Wang and P.G. Jstbid. 1999. Effects of elevated [CO_2] on photosynthesis in European forest species: a meta-analysis of model parameters. Plant, Cell & Environment. 22:1475-1495.

Meehl, G.A., J.M. Arblaster and C. Tebaldi. 2007. Contributions of natural and anthropogenic forcing to changes in temperature extremes over the United States. Geophys. Res. Lett. 34:L19709.

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

Mitchell, J.F., B.S. Manabe, V. Meleshko and T. Tokioka. 1990. Equilibrium climate change - and its implications for the future. In: Climate Change: The IPCC scientific assessment. Cambridge Univ. Press, Cambridge.

Moore, B.D., S.H. Cheng, D. Sims and J.R. Seemann. 1999. The biochemical and molecular basis for photosynthetic acclimation to elevated atmospheric CO_2. Plant, Cell and Environment. 22:567-582.

Morison, J.I.L. 1993. Response of plants to CO_2 under water limited conditions. Plant Ecology. 104-105:193-209.

Muraoka, H., Y. Tang, H. Koizumi and I. Washitani. 2002. Effects of light and soil water availability on leaf photosynthesis and growth of *Arisaema heterophyllum* , a riparian forest understorey plant. Journal of Plant Research. 115:419-427.

Muraoka, H., Y. Tang, H. Koizumi, I. Washitani and W. Z. 1997. Combined effects of light and water availability on photosynthesis and growth of *Arisaema Heterophyllum* in the forest understory and an open site. Oecologia. 112:26-34.

Nowak, R.S., D.S. Ellsworth and S.D. Smith. 2004. Functional responses of plants to elevated atmospheric CO_2: Do photosynthetic and productivity data from FACE experiments support early predictions? New Phytologist. 162:253-280.

Ohashi, Y., N. Nakayama, H. Saneoka and K. Fujita. 2006. Effects of drought stress on photosynthetic gas exchange, chlorophyll fluorescence and stem diameter of soybean plants. Biologia Plantarum. 50:138-141.

Osborne, C.P., B.G. Drake, J. LaRoche and S.P. Long. 1997. Does long-term elevation of CO_2 concentration increase photosynthesis in forest floor vegetation? Indiana strawberry in a Maryland forest. Plant Physiology. 114:337-344.

Owensby, C.E., P.I. Coyne, J.M. Ham, L.M. Auen and A.K. Knapp. 1993. Biomass production in a tallgrass prairie ecosystem exposed to ambient and elevated CO_2. Ecological Applications. 3:644-653.

Palanisamy, K. 1999. Interactions of elevated CO_2 concentration and drought stress on photosynthesis in *Eucalyptus Cladocalyx* F. Muell. Photosynthetica. 36:635-638.

Paula, M.P. 2004. Plant Physiologist. U.S. Department of Agriculture, Forest Service, North Central Research Station, Hardwood Tree Improvement and Regeneration Center, 195 Marsteller Street, West Lafayette, IN 47907.

Pearcy, R.W. 1990. Sunflecks and photosynthesis in plant canopies. Annual Review of Plant Physiology and Plant Molecular Biology. 41:421-453.

Polley, H.W., C.R. Tischler, H.B. Johnson and R.E. Pennington. 1999. Growth, water relations, and survival of drought-exposed seedlings from six maternal families of honey mesquite (*Prosopis glandulosa*): responses to CO_2 enrichment. Tree Physiology. 19:359-366.

Poorter, L. and S.F. Oberbauer. 1993. Photosynthetic induction responses of two rainforest tree species in relation to light environment. Oecologia. 96:193-199.

Pothier, D. and M. Prévost. 2002. Photosynthetic light response and growth analysis of competitive regeneration after partial cutting in a boreal mixed stand. Trees - Structure and Function. 16:365-373.

Pregitzer, K.S. and J.S. King. 2005. Effects of soil temperature on nutrient uptake. - In: Nutrient acquisition by plants: an ecological perspective. Springer.

Ridolfi, M. and E. Dreyer. 1997. Responses to water stress in an ABA-unresponsive hybrid poplar (*Populus koreana*×*trichocarpa* cv. Peace) III. Consequences for photosynthetic carbon assimilation. New Phytologist. 135:31-40.

Rincon, E. and J.P. Grime. 1989. Plasticity and light interception by six bryophytes of contrasted ecology. Journal of Ecology. 77:439-446.

Rind, D., R. Goldberg, J. Hansen, C. Rosenzweig and R. Ruedy. 1990. Potential evapotranspiration and the likelihood of future drought. J. Geophys. Res. 95:9983-10004.

Samarakoon, A.B. and R.M. Gifford. 1995. Soil water content under plants at high CO_2 concentration and interactions with the direct CO_2 effects: a species comparison. Journal of Biogeography. 22:193-202.

Saxe, H., D.S. Ellsworth and J. Heath. 1998. Tree and forest functioning in an enriched CO_2 atmosphere. New Phytologist. 139:395-436.

Sharkey, T.D., C.J. Bernacchi, G.D. Farquhar and E.L. Singsaas. 2007. Fitting photosynthetic carbon dioxide response curves for C3 leaves. Plant, Cell & Environment. 30:1035-1040.

Sitch, S., C. Huntingford, N. Gedney, P.E. Levy, M. Lomas, S.L. Piao, R. Betts, P. Ciais, P. Cox, P. Friedlingstein, C.D. Jones, I.C. Prentice and F.I. Woodward. 2008. Evaluation of the terrestrial carbon cycle, future plant geography and climate-carbon cycle feedbacks using five Dynamic Global Vegetation Models (DGVMs). Global Change Biology. 14:2015-2039.

Smith, S., T. Huxman, S. Zitzer, T. Charlet, D. Housman, J. Coleman, L. Fenstermaker, J. Seemann and R. Nowak. 2000. Elevated CO_2 increases productivity and invasive species success in an arid ecosystem. Nature. 408:79-82.

Solomon, S., D. Qin, M. Manning, Z. Chen, M. Marquis, K.B. Averyt, M. Tignor and H.L. Miller. 2007. *Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change*. . Cambridge University Press, Cambridge.

Sullivan, J. 1993. *Acer spicatum*. In: U.S. Department of agriculture, Forest Service, Rocky Mountain Research Station. Fire Sciences Laboratory. Fire effects information system.

Tezara, W., V.J. Mitchell, S.D. Driscoll and D.W. Lawlor. 1999. Water stress inhibits plant photosynthesis by decreasing coupling factor and ATP. Nature. 401:914-917.

Tognetti, R., A. Raschi and M.B. Jones. 2000. Seasonal patterns of tissue water relations in three Mediterranean shrubs co-occurring at a natural CO_2 spring. Plant, Cell & Environment. 23:1341-1351.

Townend, J. 1993. Effects of elevated carbon dioxide and drought on the growth and physiology of clonal Sitka spruce plants (*Picea stichensis* (Bong) Carr). Tree Physiology. 13:389-399.

Tschaplinski, T.J., R.J. Norby and S.D. Wullschleger. 1993. Responses of loblolly pine seedlings to elevated CO_2 and fluctuating water supply. Tree Physiology. 13:283-296.

Volk, M., P.A. Niklaus and C. Körner. 2000. Soil moisture effects determine CO_2 responses of grassland species. Oecologia. 125:380-388.

Watanabe, M., Y. Watanabe, S. Kitaoka, H. Utsugi, K. Kita and T. Koike. 2011. Growth and photosynthetic traits of hybrid larch F1 (*Larix gmelinii* var. *japonica* × L. *kaempferi*) under elevated CO_2 concentration with low nutrient availability. Tree Physiology

Wayne, P.M. and F.A. Bazzaz. 1993. Morning vs afternoon sun patches in experimental forest gaps: consequences of temporal incongruency of resources to birch regeneration. Oecologia. 94:235-243.

Wullschleger, S.D., P.J. Hanson and T.J. Tschaplinski. 1998a. Whole-plant water flux in understory red maple exposed to altered precipitation regimes. Tree Physiology. 18:71-79.

Wullschleger, S.D., F.C. Meinzer and R.A. Vertessy. 1998b. A review of whole-plant water use studies in tree. Tree Physiology. 18:499-512.

Wullschleger, S.D., T.J. Tschaplinski and R.J. Norby. 2002. Plant water relations at elevated CO_2– implications for water-limited environments. Plant, Cell & Environment. 25:319-331.

Chapter 5: Effects of CO_2, light and soil moisture on growth and biomass allocation in mountain maple (*Acer spicatum* L.) seedlings.

Introduction

Light is one of the most important ecological factors that strongly affect plant growth (Kobe et al. 1995, Canham et al. 1996, Hättenschwiler 2001). However, the occurrence of canopy gaps from natural causes (e.g. tree-falls, fire) or silvicultural practices (e.g., selection cutting) results in increased availability of light to understory plants. Some understory plant species have foraging growth habits that allow them to discover and exploit these canopy gaps (Rincon and Grime 1989, Lei and Lechowicz 1990). Their survival and regenerative success in such high light conditions must depend on physiological and morphological acclimations at the leaf and whole plant level (Canham 1988a, Givnish 1988, Canham et al. 1996, Messier et al. 1999, Beaudet et al. 2000, Lambers et al. 2008, Bjôrkman 1981). Plant traits and environmental factors that enhance the exploitation of high light environment may influence the structure of forest stands and species. Carbon dioxide concentration ([CO_2]) (Bazzaz et al. 1990, Bazzaz and Miao 1993, Hättenschwiler 2001, Beier et al. 2005) and soil moisture (Abrams and Mostoller 1995, Givnish 1995, Muraoka et al. 1997, Muraoka et al. 2002, Volkova et al. 2010) are known to affect plant response to light environment. Most studies have however, ignored the interactive effects of [CO_2] and soil moisture on plants growth response to light, particularly shade-tolerant species.

The atmospheric carbon dioxide concentration ([CO_2]) is predicted to reach between 730 - 1200 μmol mol^{-1} by the year 2100 (IPCC 2007a, Meehl et al. 2007, Sitch et al. 2008). Plants responses to the rising [CO_2] have been of interest to many researchers because they play an important role in carbon sequestration (Huang et al. 2007, Dawes et al. 2011a, Dawes et al. 2011b). Studies have shown that plant growth and biomass production increase under elevated [CO_2] at least in the short-term (Bazzaz et al. 1990, Körner 2000, Hättenschwiler 2001, LeCain et al. 2012). However, others reported no increase or even decrease in growth response to elevated [CO_2] (Norby and O'Neill 1991, Ziska et al. 1991, Dawes et al. 2011b). In addition to its effect on growth and biomass, elevated [CO_2] also affect the functional relationship between plant parts through biomass allocation (Norby and O'Neill 1991, Duff et al. 1994). Elevated [CO_2] increases plants vigor, allowing them to explore their microhabitats and expand their ecological niches (Hattenschwiler and Korner 2003, Zotz et al. 2006). Therefore, elevated [CO_2] might facilitate plant response to light environment. This may have far-ranging effect on forest composition and dynamics.

The increase in [CO_2] is predicted to be accompanied by increasing global mean temperatures (IPCC 2007b, Meehl et al. 2007, Sitch et al. 2008). Increases in temperature may lead to decrease in soil moisture because of increased evaporation (Pregitzer and King 2005). Low soil moisture affects many plants activities including stomatal conductance and photosynthesis (Flexas and Medrano 2002, Flexas et al. 2004, Lambers et al. 2008, Wang et al. 2012) and nutrient absorption by roots (Chapin 1980, Aerts and Chapin Iii 1999). Soil moisture availability affects plants responses to elevated [CO_2]. Studies have shown that low soil moisture attenuates plants responsiveness to elevated [CO_2] (Owensby et al. 1993, Samarakoon and Gifford 1995, Campbell et al. 1997, Catovsky and Bazzaz 1999, Smith et al. 2000, Volk et al.

2000, Derner et al. 2003, Belote et al. 2004). The adverse effect of low soil moisture on the stimulation of plants growth by elevated [CO_2] is often related to increased stomatal limitation to photosynthesis (Li et al. 2004, Lawlor and Tezara 2009, Ambebe and Dang 2010). Further, increased biomass allocation to roots for improved water absorption under low soil moisture (Lambers et al. 2008, Ambebe and Dang 2010) may limit the stimulation of plant growth by elevated [CO_2]. Muroako et al. (1997) also report that low soil moisture reduces the responses of plant growth and biomass to high light environment. Thus, we hypothesize that low soil moisture would limit the stimulation of height growth and biomass accumulation by elevated [CO_2] in high light.

In the present study, the interactive effects of CO_2 and soil moisture on the growth and biomass responses of mountain maple (*Acer spicatum* L.) seedlings to increase in light was investigated. Mountain maple regenerates in dense forest understory but respond rapidly to canopy openings (Lei and Lechowicz 1997a, Archambault et al. 1998, Bergeron 2000, Rook 2002, Aubin et al. 2005). High growth response to increase light conditions has been observed in other maple species (Wilson and Fischer 1977, Canham 1988a). However, growth and biomass responses of mountain maple to increases in light may be hindered by its low drought tolerance ability (Paula 2004). This is particularly a concern because mountain maple has high soil moisture demands (Paula 2004). This may limit the response of mountain maple to high light conditions. To our knowledge, no one has examined how the interaction between [CO_2] and soil moisture might affect the response of mountain maple seedling to increases in light conditions. Therefore, it is important to examine how mountain maple may respond to increases in light conditions in the future.

Materials and Method

Plant materials

Seeds of mountain maple (*Acer spicatum* L.) were collected from Jack Haggerty Forest in Thunder Bay, Ontario, Canada (48°22′56″ N, 89°14′46″ W). Seeds were soaked in a 1000µmol m^{-1} giberellic acid (GA) for 24hr and stratified at 4°C for 2 months (Lei 1992). The hard seed coats were gently cracked open after 2 months to facilitate germination. Germination was carried out on a 2:1 mixture of vermiculite and peat moss in horticultural trays at Lakehead University greenhouse. Average environmental conditions during germination were as follows; temperature 22/16 °C (day/night) and 16hr photoperiod (maximum summertime day length for Thunder Bay, ON. based on Environment Canada Weather Report, 2010). One hundred and sixty similar-sized seedlings were transplanted into plastic pots (31.5 cm deep, 11 cm top diameter, and 9.5 cm bottom diameter) three weeks after germination. The pots were filled with the same composition of growing medium used in the germination process.

Experimental design

The experiment had two [CO_2], two light and two soil moisture treatments in a split-split-plot design. The [CO_2] were ambient (*c*. 392µmol mol^{-1}) and elevated (target concentration 784 µmol mol^{-1}). The elevated [CO_2] was generated with CO_2 generators and monitored by an Argus environmental control system (Argus systems Ltd, Vancouver, BC, Canada). Each CO_2 level had two replicates (greenhouses). The sub-plot treatment consisted of two levels of light within each CO_2 treatment. Half of the seedlings in each CO_2 treatment were shaded using neutral density shade cloths at the top and sides. The shading reduced the PAR by 70% relative to the unshaded treatment. High-pressure sodium lamps (Model LR48877, P.L. Systems, Grimspy, ON, Canada) were used to provide supplemental light on cloudy days and to extend the photoperiod to 16hr. The sub-sub-plot treatment comprised of two soil moisture treatments within each light treatment. In the high soil moisture treatment, the seedlings were watered to the dripping point daily. Growing medium moisture content in the low soil moisture treatment was about 40% – 50% relative to high soil moisture. The moisture content was measured using HH2 moisture meter. The low soil moisture treatment started one week after the seedlings were transplanted to allow for establishment.

Other environmental conditions in each greenhouse were 22/16 °C day/night air temperature and relative humidity of 50%. The environmental conditions were controlled and monitored by an Argus environmental control system (Argus systems Ltd, Vancouver, BC, Canada). Nutrients were added to irrigation water twice a week at a concentration of 100, 15, 57, 6, 6 and 11 mg/L of N, P, K, Ca, Mg and S, respectively (Ingestad 1981, Canham et al. 1996) The fertilization was done during the days when seedlings in the low soil moisture were watered.

Growth and biomass measurements

After two months of the treatment, five seedlings from each treatment combination were measured for height (*H*) and root collar diameter (RCD). The total leaf area per seedling was measured using WinFolia (Regent Instrument Inc., Quebec, Canada). The seedlings were harvested, separated into leaves, stems, and roots, and oven-dried to a constant weight at 70 º C. Specific leaf area (SLA) and shoot: root ratio (SRR) were calculated by dividing the total leaf area by the corresponding dry leaf mass and by dividing total shoot (stem + leaf) by the root mass, respectively. Other indices of biomass allocation calculated were root mass ratio (RMR) and leaf mass ratio (LMR).

Statistical analysis

The data were analyzed with Data desk 6.01 statistical package k (Data Description 1996). The assumptions of normality of distribution and homogeneity of variance were examined using graphically using probability plots and histograms, respectively, before Analyses of Variance (ANOVA) was done. Both assumptions were met. The effects of [CO_2], light and soil moisture regimes, and their interactions were tested using split-split plot ANOVA. The significant level was set at $P \leq 0.05$ but P-values ≤ 0.10 were considered marginally significant. Scheffé's *post hoc* test was done when an interaction was significant.

Results

There was a marginally significant interactive effect among CO_2, light and soil moisture on seedling height (Table 4.1).The high light increased H greatest in the elevated [CO_2]-low moisture combination (103%) and smallest in the elevated [CO_2]-high moisture combination (55%). The low soil moisture generally decreased H but the effect was statistically significant only under the high light and ambient [CO_2] combination and the low light and elevated [CO_2] combination (Fig. 4.1A). The elevated [CO_2] significantly increased H in both soil moisture treatments under the high light and the in high soil moisture only in the low light treatment (Fig. 4.1A).

The effect of light on the leaf area (LA) per seedling also varied with soil moisture and [CO_2] (Table 4.1). The high light treatment significantly affected LA only in the high soil moisture treatment but the effect was the opposite in the two [CO_2]: it decreased it by 16% under the ambient but increased by 16% under the elevated [CO_2] (Fig. 4.1B). The low soil moisture significantly decreased LA in both light treatments under the ambient [CO_2] but only in the low light under the elevated [CO_2] (Fig. 4.1B). The elevated [CO_2] significantly increased LA at both soil moisture treatments under the low light but only at the low soil moisture under high light treatment (Fig. 4.1B).

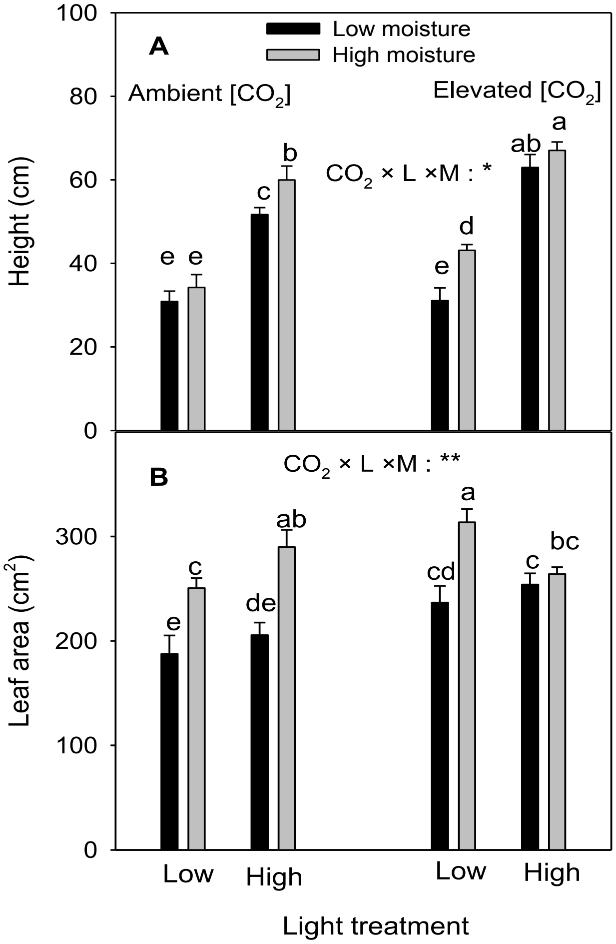

Figure 4.1. Effects of [CO_2], light treatment (L) and soil moisture (M) on seedling height (*H*) and leaf area (LA) (mean ± SE, n = 10) of *Acer spicatum*. Seedlings were exposed to two [CO_2] (380 and 760 μmol mol^{-1}), two light treatments (high , 100% and low, 30%) and two soil moisture (high and low) regimes for two months. Significant treatment effects are marked as significant: $P \leq 0.01$, ***; $P \leq 0.05$, **; and marginally significant: $P \leq 0.10$, *. Bars with same letter(s) are not statistically significant ($P >$ 0.10) from each other or one another.

The interaction between [CO_2] and light had a significant effect on root collar diameter (RCD) (Table 4.1). The high light significantly increased RCD under both [CO_2] but the magnitude was higher under elevated than ambient [CO_2] (65 vs. 42%, Fig. 4.2A). The elevated [CO_2] significantly increased RCD (by 19%) only in the high light treatment (Fig.4. 2A). Soil moisture or its interactions with [CO_2] or light did not have significant effect on RCD (Table 4.1).

The interaction between [CO_2] and light had a significant effect on specific leaf area (SLA) (Table 4.1). Additionally, there was a marginally significant interactive between [CO_2] and soil moisture SLA (Table 4.1). The high light significantly decreased SLA under both [CO_2] but the magnitude of reduction was greater under the elevated than ambient [CO_2] (50 vs. 27%, Fig. 4.2B). Furthermore, the elevated [CO_2] significantly decreased SLA in both light treatments but the effect was greater in the high light treatment (Fig. 4.2B). The low soil moisture significantly increased SLA (by 21%) only under the elevated but not the ambient [CO_2] (Fig. 4.2C). The elevated [CO_2] significantly decreased SLA (by 15 and 28%) at the low and high soil moisture treatments, respectively, (Fig. 4.2C).

Total seedling biomass was significantly affected by CO_2 × L and L ×M interactions (Table 4.1). Total seedling biomass significantly increased in response to the high light treatment under both [CO_2] but the increase was greater under the elevated than ambient [CO_2] (215vs. 131%, Fig 4.3A). The elevated [CO_2] significantly increased seedling biomass by 53% only in the high but not low light treatment (Fig. 4.3A). Seedling biomass significantly increased in response to the high light at both soil moisture treatments but the degree of increase was greater when the soil moisture was high (156% vs. 127%, Fig. 4.3B). The low soil moisture significantly reduced seedling biomass in both light treatments and the magnitude of reduction was greater at the high compared with the low light treatment (57 vs. 51%, Fig. 4.3B).

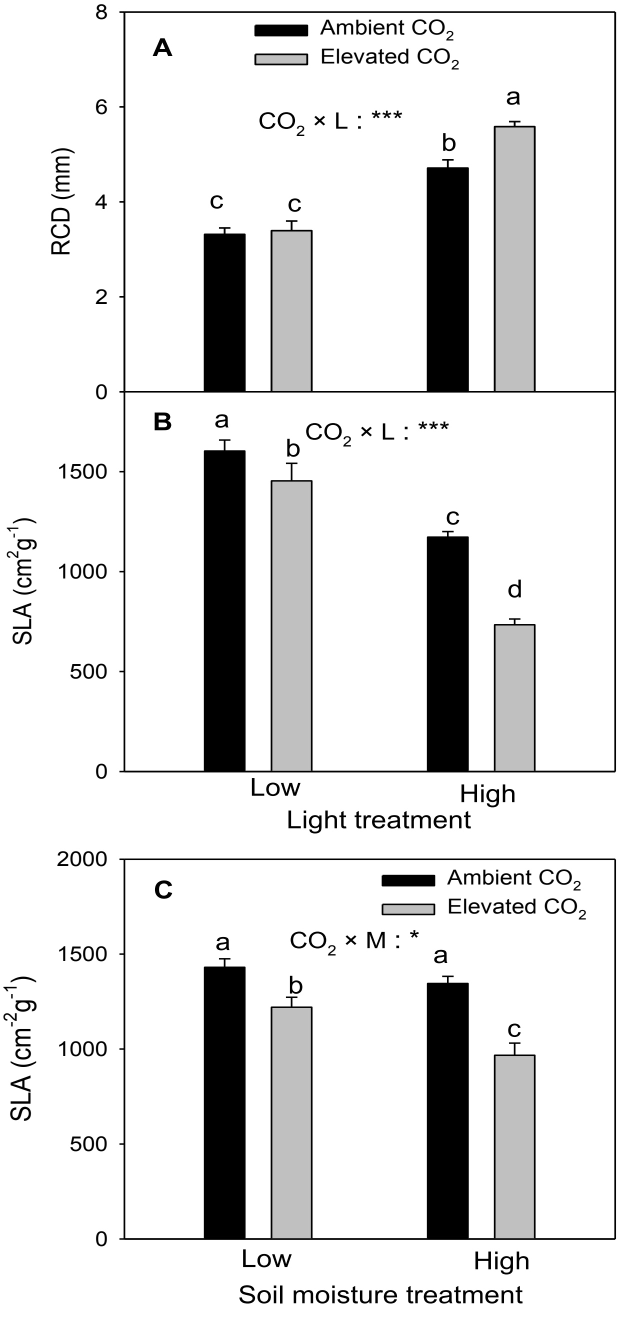

Figure 4.2. Effects of [CO_2], L and M on root collar diameter (RCD) and specific leaf area (SLA) (mean ± SE, n = 10) of *Acer spicatum*. Refer to Fig. 4.1 for other descriptions.

There were significant L × M and CO_2 × M interactive effects on SRR (Table 4.1). SRR significantly decreased in response to the high light treatment when soil moisture was low but no significant high light effect on SRR was found at the high soil moisture (Fig. 4.3C). The low soil moisture significantly decreased SRR only in the high light but not significantly affect SRR in the low light treatment (Fig. 4.3C). Furthermore, the low soil moisture significantly decreased SRR only under the ambient but not under the elevated [CO_2] (Fig. 4.3D). The elevated [CO_2] resulted in a significantly lower SRR at the high soil moisture treatment but did not significantly affect SRR when seedlings were grown at the low soil moisture regime (Fig. 4.3D).

The root mass ratio (RMR) significantly varied with L × M and CO_2 × M combinations (Table 4.1). The high light treatment significantly increased RMR only at the low but not at the high soil moisture (Fig. 4.4A). The low soil moisture significantly increased RMR only at the high treatment (Fig. 4A). Furthermore, the low soil moisture significantly increased RMR (by 43%) under the ambient but not under the elevated [CO_2] (Fig. 4.4B). The elevated [CO_2] significantly increased RMR in both soil moisture treatments but the magnitude of increase was greater at the high compared with the low soil moisture (58 vs. 26%, Fig**.** 4.4B). The LMR significantly decreased in response to the elevated [CO_2], high light and the low soil moisture treatments but no significant interactive effect on LMR was found (Table 1).

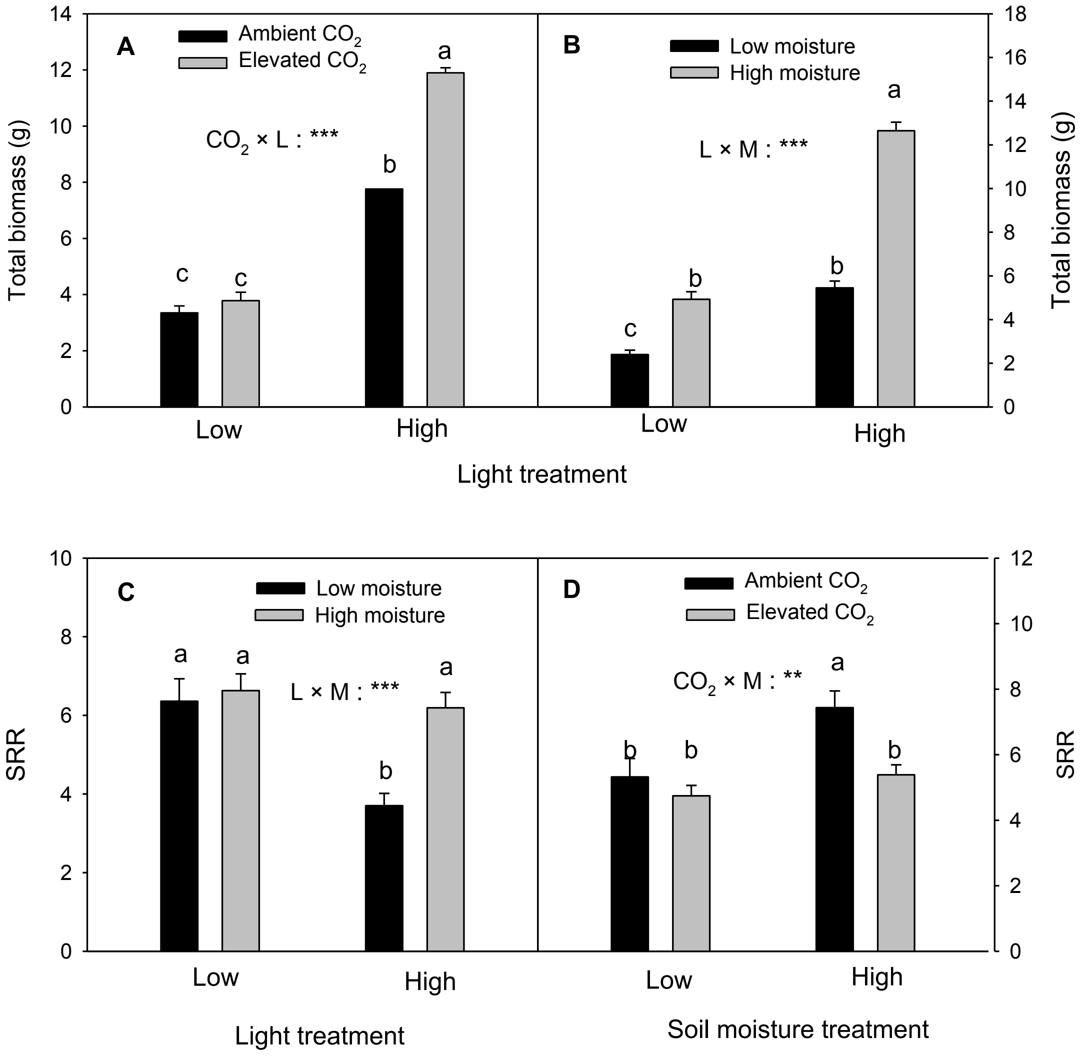

Figure 4.3. Effects of [CO_2], L and M on total seedling biomass and shoot: root ratio (SRR) (mean ± SE, n = 10) of *Acer spicatum*. Refer to Fig. 4.1 for other descriptions.

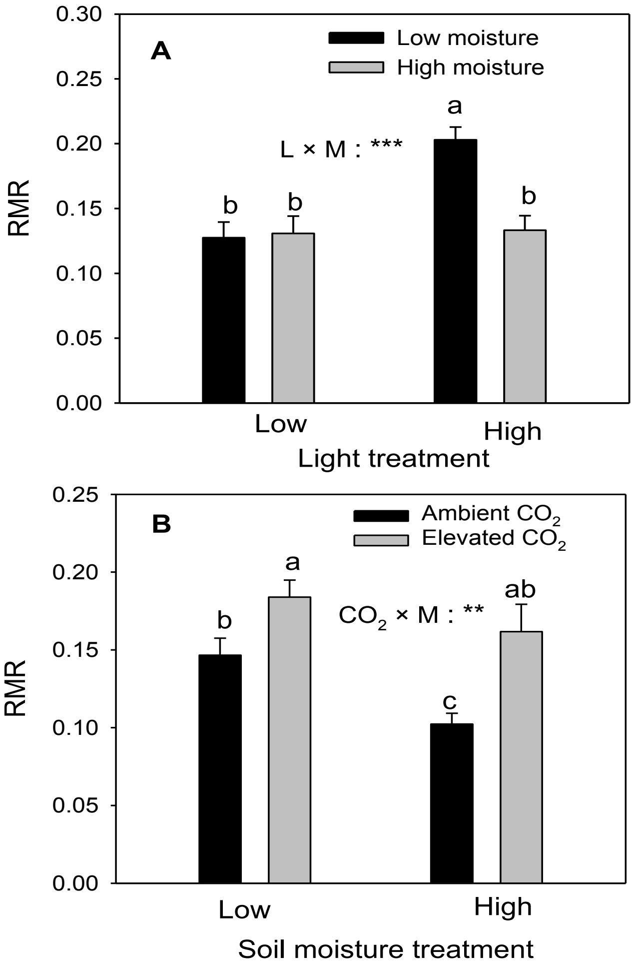

Figure 4.4. Effects of [CO_2], L and M on root mass ratio (RMR) (mean ± SE, n = 10) of *Acer spicatum*. Refer to Fig. 4.1 for other explanations.

Discussion

The hypothesis that low soil moisture would limit mountain maple growth stimulation by elevated [CO_2] in the high light condition was not supported. The seedling height growth response to high light was greater at elevated [CO_2], but the relative enhancement was much greater at the low soil moisture treatment. It was expected that low soil moisture would reduce leaf area growth (Muraoka et al. 1997, Muraoka et al. 2002) and thus, reduce the stimulatory effect of elevated [CO_2] on seedling growth. The low soil moisture did not decrease leaf area production when seedlings were exposed to elevated [CO_2] in the high light treatment. Leaf area plays a significant role in plants growth (Curtis and Wang 1998, Pritchard et al. 1999, Muraoko et al. 2002). In other studies, low soil moisture reduces the growth response of *Arisaema heterophyllum* to high light conditions due reduction in leaf growth (Muraoka et al. 1997). Norby and O'Neil (1991) attributed a lack of growth responses of yellow poplar (*Liriodendron tulipifera* L)-seedlings to elevated [CO_2] to decline in LAR. In this study, the interaction among CO_2, light and soil moisture did not significantly affect LAR. This could be implicated in the lack low soil moisture effect on the growth response of mountain maple seedling to light under elevated [CO_2]. It is also possible that the low soil moisture used in this study might not be stressful enough to limit mountain maple growth response to high light under elevated [CO_2].

Our results showed distinctly different effects of [CO_2] and soil moisture on total seedling biomass response to light. The high light increased biomass by 215% under elevated [CO_2] compared with 131% under the ambient [CO_2]. The high light increased seedling biomass by 157% and 127% at the high and low soil moisture treatments, respectively. The elevated [CO_2] increased biomass at the high but not at the low light treatment, indicating that high light and elevated [CO_2] have synergistic effects on biomass of mountain maple seedlings. The relatively strong response of seedlings biomass to high light conditions, mediated by the elevated [CO_2] might be related to leaf traits and physiological activity. For example, the high light reduced SLA more under the elevated [CO_2] than when seedlings were grown under ambient [CO_2]. At high light conditions, thicker leaf (lower SLA) maximizes carbon gain through reduced water loss (Poorter 1999, Aubin et al. 2005). In cases where leaf thickness is caused by increases in photosynthetically active parenchyma layers, the leaf photosynthetic rate is improved (Poorter 1999).

Table 4.1. Analysis of variance (ANOVA) on the effects of CO2 concentration, soil moisture and light regimes on seedling height (*H*), leaf area (LA), root collar diameter (RCD), specific leaf area (SLA), leaf area ratio (LAR), total seedling biomass (TB), shoot: root ratio (SRR), root mass ratio (RMR) and leaf mass ratio (LMR) in mountain maple (*Acer spicatum* L.) seedlings. The seedlings grown under ambient (380 µmol mol^{-1}) or elevated (760 µmol mol^{-1}) [CO_2], high (100%) or low (30%) light (L) and high or low soil moisture (M) regimes. Measurements were taken two months after the start of the treatment. Significant ($P \leq 0.10$) effects are in bold.

Source of variation	CO_2	L	$CO_2 \times L$	M	$CO_2 \times M$	$L \times M$	$CO_2 \times L \times M$
H	0.2878	**≤0.0001**	0.1700	**0.0001**	0.5068	0.6539	**0.0583**
LA	0.1815	0.5280	**0.0285**	**≤0.0001**	0.1796	0.1845	**0.0478**
RCD	**0.0190**	**≤0.0001**	**0.0119**	0.3278	0.7781	0.9564	0.3349
SLA	**0.0240**	**≤ 0.0001**	**0.0028**	**0.0006**	**0.0856**	0.8923	0.8322
LAR	**0.0379**	**≤ 0.0001**	0.8294	**≤0.0001**	0.305	0.9648	0.8639
TB	**0.0696**	**≤ 0.0001**	**0.0059**	**≤ 0.0001**	0.2802	**≤ 0.0001**	0.5923
SRR	**0.0438**	**0.0003**	0.7336	**0.0009**	**0.0517**	**0.0047**	0.6226
RMR	0.1112	**0.0005**	0.8447	**≤ 0.0001**	**0.0528**	**0.0047**	0.2736

Additionally, the high light treatment increased the instantaneous water-use efficiency of the seedlings (IWUE) more under elevated than ambient [CO_2] (chapter 4 of this thesis, Fig 2B). Increased IWUE under elevated [CO_2] has been found to be a mechanism by which plants increase biomass production without a concomitant increase in leaf area (Norby and O'Neill 1991). In this study, there was no significant effect of high light on leaf area under the elevated

[CO_2] (no significant CO_2 × light interaction), suggesting that the increase in biomass in high light might be attributable to increased carbon assimilation.

The proportion of biomass allocated to roots increased in response to the low soil moisture more under the ambient than the under the elevated [CO_2]. Increased biomass allocation to roots at low soil moisture is a functional response associated with decrease in water and nutrients absorption (Davidson 1969, Poorter 1999, Franklin et al. 2012). The ability of mountain maple to allocate more biomass to roots (increased root size) might confer drought avoidance and survival (Burdett 1990, Örlander et al. 1998, Boucher et al. 2001) at sites where soil moisture is insufficient. However, the low soil moisture did not change the pattern of biomass allocation to roots under the elevated [CO_2], indicating that elevated [CO_2] might have ameliorated the stressful effects of low soil moisture on mountain maple. There was a decreased in biomass allocated to leaf at the low soil moisture treatment but the response pattern did not differ between [CO_2]. The differential patterns of biomass allocation to roots or leaf is consistent with other studies (Van Den Boogaard et al. 1996, Liu and Stützel 2004, Ambebe and Dang 2010).

The soil moisture modified light effects on biomass allocation root. At the high light treatment the low soil moisture increased biomass allocation to root. Similar pattern of biomass allocation in a shade-tolerant but drought-intolerant sugar maple (*Acer saccharum* Marsh) has been reported (Canham et al. 1996). However, the high light did not affect biomass allocation at the high soil moisture, suggesting that mountain maple biomass allocation response to light is governed by soil resource availability. Despite the higher biomass allocated to root when moisture was low in the high light treatment, low soil moisture might still limit mountain maple growth compared with moist environments (Canham et al. 1996). This is because mountain maple is known to be drought-intolerant (Paula 2004). Therefore, the response of mountain maple to high light conditions where other environmental factors such as soil moisture co-vary might hinder its competitive ability and establishment.

In conclusion, the low soil moisture did not reduce the stimulatory effect of elevated [CO_2] on seedling height growth response to light. The high light treatment had much greater effect on leaf area production than the low soil moisture under elevated [CO_2]. Hence, the results suggest that high light rather than low soil moisture limited the positive effect of elevated [CO_2] on seedling height growth. Improved IWUE at the high light conditions, mediated by elevated [CO_2] increased biomass production without a corresponding increase in leaf growth. Further, the combined effect of high light and elevated [CO_2] had a greater effect on biomass than both factors alone. Despite the higher biomass allocated to root at high light conditions when soil moisture was low, mountain maple competitive success in canopy gaps might be limited in future climates of higher [CO_2].

References

Abrams, M.D. and S.A. Mostoller. 1995. Gas exchange, leaf structure and nitrogen in contrasting successional tree species growing in open and understory sites during a drought. Tree Physiology. 15:361-370.

Ambebe, T.F. and Q.-L. Dang. 2010. Low moisture availability reduces the positive effect of increased soil temperature on biomass production of white birch (*Betula papyrifera*) seedlings in ambient and elevated carbon dioxide concentration. Nordic Journal of Botany. 28:104-111.

Archambault, L., J. Morissette and M. Bernier-Cardou. 1998. Forest succession over a 20-year period following clearcutting in balsam fir yellow birch ecosystems of eastern Quebec, Canada. Forest Ecology and Management. 102:61-74.

Aubin, I., C. Messier and D. Kneeshaw. 2005. Population structure and growth acclimation of mountain maple along a successional gradient in the southern boreal forest. Ecoscience. 12:540-548.

Bazzaz, F.A., J.S. Coleman and S.R. Morse. 1990. Growth responses of seven major co-occurring tree species of the northeastern United States to elevated CO_2. Canadian Journal of Forest Research. 20:1479-1484.

Bazzaz, F.A. and S.L. Miao. 1993. Successional status, seed size, and responses of tree seedlings to CO_2, light, and nutrients. Ecology. 74:104-112.

Beaudet, M., C. Messier, D.W. Hilbert, E. Lo, Z.M. Wang and M.J. Lechowicz. 2000. Leaf- and plant-level carbon gain in yellow birch, sugar maple, and beech seedlings from contrasting forest light environments. Canadian Journal of Forest Research. 30:390-404.

Beier, C.M., J.L. Horton, J.F. Walker, B.D. Clinton and E.T. Nilsen. 2005. Carbon limitation leads to suppression of first year oak seedlings beneath evergreen understory shrubs in Southern Appalachian hardwood forests. Plant Ecology. 176:131-142.

Belote, R.T., J.F. Weltzin and R.J. Norby. 2004. Response of an understory plant community to elevated [CO2] depends on differential responses of dominant invasive species and is mediated by soil water availability. New Phytologist. 161:827-835.

Bergeron, Y. 2000. Species and stand dynamics in the mixed woods of Quebec's southern boreal forest. Ecology. 81:1500-1516.

Bjôrkman, O. 1981. Responses to different quantum flux densities. In: Encyclopaedia of plant physiology. Plant physiological ecology I. Springer, Berlin, Germany.

Boucher, J.-F., P.Y. Bernier, H.A. Margolis and A.D. Munson. 2007. Growth and physiological response of eastern white pine seedlings to partial cutting and site preparation. Forest Ecology and Management. 240:151-164.

Boucher, J.F., P.Y. Bernier and A.D. Munson. 2001. Radiation and soil temperature interactions on the growth and physiology of eastern white pine (*Pinus strobus* L.) seedlings. Plant and Soil. 236:165-174.

Burdett, A.N. 1990. Physiological processes in plantation establishment and the development of specifications for forest planting stock. Canadian Journal of Forest Research. 20:415-427.

Campbell, B.D., D.M. Stafford Smith and G.M. McKeon. 1997. Elevated CO_2 and water supply interactions in grasslands: A pastures and rangelands management perspective. Global Change Biology. 3:177-187.

Canham, C.D. 1988. Growth and canopy architecture of shade-tolerant trees: response to canopy gaps. Ecology. 69:786-795.

Canham, C.D., A.R. Berkowitz, V.R. Kelly, G.M. Lovett, S.V. Ollinger and J. Schnurr. 1996. Biomass allocation and multiple resource limitation in tree seedlings. Canadian Journal of Forest Research. 26:1521-1530.

Catovsky, S. and F.A. Bazzaz. 1999. Elevated CO_2 influences the responses of two birch species to soil moisture: implications for forest community structure. Global Change Biology. 5:507-518.

Curtis, P.S. and X. Wang. 1998. A meta-analysis of elevated CO_2 effects on woody plant mass, form, and physiology. Oecologia. 113:299-313.

Davidson, R.L. 1969. Effect of root/leaf temperature differentials on root/shoot Ratios in some pasture grasses and clover. Annals of Botany. 33:561-569.

Dawes, M.A., F. Hagedorn, T. Zumbrunn, I.T. Handa, S. Hättenschwiler, S. Wipf and C. Rixen. 2011a. Growth and community responses of alpine dwarf shrubs to in situ CO_2 enrichment and soil warming. New Phytologist. 191:806-818.

Dawes, M.A., S. Hättenschwiler, P. Bebi, F. Hagedorn, I.T. Handa, C. Körner and C. Rixen. 2011b. Species-specific tree growth responses to 9 years of CO_2 enrichment at the alpine treeline. Journal of Ecology. 99:383-394.

Derner, J.D., H.B. Johnson, B.A. Kimball, P.J. Pinter, H.W. Polley, C.R. Tischler, T.W. Boutton, R.L. Lamorte, G.W. Wall, N.R. Adam, S.W. Leavitt, M.J. Ottman, A.D. Matthias and T.J. Brooks. 2003. Above- and below-ground responses of C_3–C_4 species mixtures to elevated CO_2 and soil water availability. Global Change Biology. 9:452-460.

Duff, G.A., C.A. Berryman and D. Eamus. 1994. Growth, biomass allocation and foliar nutrient contents of two *Eucalyptus* species of the wet-dry tropics of Australia grown under CO_2 enrichment. Functional Ecology. 8:502-508.

Franklin, O., J. Johansson, R.C. Dewar, U. Dieckmann, R.E. McMurtrie, Å. Brännström and R. Dybzinski. 2012. Modeling carbon allocation in trees: a search for principles. Tree Physiology

Givnish, T. 1988. Adaptation to sun and shade: a whole-plant perspective. Functional Plant Biology. 15:63-92.

Givnish, T.J. 1995. Plant stems: biomechanical adaptation for energy capture and influence on species distributions. In Plant stems: shysiology and functional forphology. Academic Press, San Diego, Califonia.

Hättenschwiler, S. 2001. Tree seedling growth in natural deep shade: functional traits related to interspecific variation in response to elevated CO_2. Oecologia. 129:31-42.

Hattenschwiler, S. and C. Korner. 2003. Does elevated CO_2 facilitate naturalization of the non-indigenous *Prunus laurocerasus* in Swiss temperate forests? Functional Ecology. 17:778-785.

Huang, J.-G., Y. Bergeron, B. Denneler, F. Berninger and J. Tardif. 2007. Response of forest trees to increased atmospheric CO2. Critical Reviews in Plant Sciences. 26:265-283.

Ingestad, T. 1981. Nutrition and growth of birch and grey alder seedlings in low conductivity solutions and at varied relative rates of nutrient addition. Physiologia Plantarum. 52:454-466.

IPCC. 2007a. Climate Change 2007: Synthesis Report. Contribution of Working Groups I, II and III to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change, IPCC, Geneva, Switzerland.

IPCC. 2007b. Summary for policymakers. In: Solomon, S. et al. (eds), Climate Change 2007: the physical science basis. Contribution of Working Group I to the fourth assessment report of the Intergovernmental Panel on Climate Change.

Kobe, R.K., S.W. Pacala, J.A. Silander and C.D. Canham. 1995. Juvenile tree survivorship as a component of shade tolerance. Ecological Applications. 5:517-532.

Körner, C. 2000. Biosphere responses to CO_2 enrichment. Ecological Applications. 10:1590-1619.

Lambers, H., S.F. Chapin III and T.L. Pons. 2008. Plant physiological eclogy. Springer, New York.

Lawlor, D.W. and W. Tezara. 2009. Causes of decreased photosynthetic rate and metabolic capacity in water-deficient leaf cells: a critical evaluation of mechanisms and integration of processes. Annals of Botany. 103:561-579.

LeCain, D.R., J.A. Morgan, G.L. Hutchinson, J.D. Reeder and F.A. Dijkstra. 2012. Interactions between elevated atmospheric CO_2 and defoliation on North American rangeland plant species at low and high N availability. Grass and Forage Science:no-no.

Lei, T.T. 1992. Functional design and shade adaptation in *Acer* species. *In* Biology. McGill University.

Lei, T.T. and M.J. Lechowicz. 1990. Shade adaptation and shade tolerance in saplings of three *Acer* species from eastern north America. Oecologia. 84:224-228.

Lei, T.T. and M.J. Lechowicz. 1997. Functional responses of *Acer* species to two simulated forest gap environments: leaf-level properties and photosynthesis. Photosynthetica. 33:277-289.

Li, S., S.R. Pezeshki and S. Goodwin. 2004. Effects of soil moisture regimes on photosynthesis and growth in cattail (*Typha latifolia*). Acta Oecologica. 25:17-22.

Liu, F. and H. Stützel. 2004. Biomass partitioning, specific leaf area, and water use efficiency of vegetable amaranth (*Amaranthus* spp.) in response to drought stress. Scientia Horticulturae. 102:15-27.

Meehl, G.A., J.M. Arblaster and C. Tebaldi. 2007. Contributions of natural and anthropogenic forcing to changes in temperature extremes over the United States. Geophys. Res. Lett. 34:L19709.

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

Muraoka, H., Y. Tang, H. Koizumi and I. Washitani. 2002. Effects of light and soil water availability on leaf photosynthesis and growth of *Arisaema heterophyllum* , a riparian forest understorey plant. Journal of Plant Research. 115:419-427.

Muraoka, H., Y. Tang, H. Koizumi, I. Washitani and W. Z. 1997. Combined effects of light and water availability on photosynthesis and growth of *Arisaema Heterophyllum* in the forest understory and an open site. Oecologia. 112:26-34.

Muraoko, H., Y. Tang, H. Koizumi and I. Washitan. 2002. Effects of light and soil water availability on leaf photosynthesis and growth of *Arisaema heterophyllum*, a riparian forest understorey plant. Journal of Plant Research. 115:419 - 427.

Norby, R.J. and E.G. O'Neill. 1991. Leaf area compensation and nutrient interactions in CO_2-enriched seedlings of yellow-poplar (*Liriodendron tulipifera* L.). New Phytologist. 117:515-528.

Örlander, G., G. Hallsby, P. Gemmel and C. Wilhelmsson. 1998. Inverting improves establishment of Pinus contorta and *Picea abies*— 10-year results from a site preparation trial in Northern Sweden. Scandinavian Journal of Forest Research. 13:160-168.

Owensby, C.E., P.I. Coyne, J.M. Ham, L.M. Auen and A.K. Knapp. 1993. Biomass production in a tallgrass prairie ecosystem exposed to ambient and elevated CO_2. Ecological Applications. 3:644-653.

Paula, M.P. 2004. Plant Physiologist. U.S. Department of Agriculture, Forest Service, North Central Research Station, Hardwood Tree Improvement and Regeneration Center, 195 Marsteller Street, West Lafayette, IN 47907.

Pearcy, R.W. and D.A. Sims. 1994. Photosynthetic acclimation to changing light environments: Scaling from the leaf to the whole plant. *In* Exploitation of Environmental Heterogeneity by Plants. Ecophysiological Processes Above- and Belowground. Academic Press, Inc., USA.

Poorter, L. 1999. Growth responses of 15 rain-forest tree species to a light gradient: the relative importance of morphological and physiological traits. Functional Ecology. 13:396-410.

Pregitzer, K.S. and J.S. King. 2005. Effects of soil temperature on nutrient uptake. - In: Nutrient acquisition by plants: an ecological perspective. Springer.

Pritchard, S.G., H.H. Rogers, S.A. Prior and C.M. Peterson. 1999. Elevated CO_2 and plant structure: a review. Global Change Biology. 5:807-837.

Reich, P.B., M.G. Tjoelker, M.B. Walters, D.W. Vanderklein and C. Buschena. 1998. Close association of RGR, leaf and root morphology, seed mass and shade tolerance in seedlings of nine boreal tree species grown in high and low light. Functional Ecology. 12:327-338.

Rincon, E. and J.P. Grime. 1989. Plasticity and light interception by six bryophytes of contrasted ecology. Journal of Ecology. 77:439-446.

Rook, E.J.S. 2002. Shrubs of the northwoods. http://www.rook. org/earl / bwca /nature / shrubs / index.html.

Samarakoon, A.B. and R.M. Gifford. 1995. Soil water content under plants at high CO_2 concentration and interactions with the direct CO_2 effects: a species comparison. Journal of Biogeography. 22:193-202.

Smith, S., T. Huxman, S. Zitzer, T. Charlet, D. Housman, J. Coleman, L. Fenstermaker, J. Seemann and R. Nowak. 2000. Elevated CO_2 increases productivity and invasive species success in an arid ecosystem. Nature. 408:79-82.

Van Den Boogaard, R., E.J. Veneklaas and H. Lambers. 1996. The association of biomass allocation with growth and water use efficiency of two *Triticum aestivum* cultivars. Functional Plant Biology. 23:751-761.

Volk, M., P.A. Niklaus and C. Körner. 2000. Soil moisture effects determine CO_2 responses of grassland species. Oecologia. 125:380-388.

Volkova, L., L. Bennett, A. Merchant and M. Tausz. 2010. Shade does not ameliorate drought effects on the tree fern species *Dicksonia antarctica* and *Cyathea australis*. Trees - Structure and Function. 24:351-362.

Wilson, B.F. and B.C. Fischer. 1977. Striped maple: shoot growth and bud formation related to light intensity. Canadian Journal of Forest Research. 7:1-7.

Ziska, L.H., K.P. Hogan, A.P. Smith, B.G. Drake and K.P. Hogen. 1991. Growth and photosynthetic response of nine tropical species with long-term exposure to elevated carbon dioxide. Oecologia. 86:383-389.

Zotz, G., N. Cueni and C. Koerner. 2006. In situ growth stimulation of a temperate zone liana (*Hedera helix*) in elevated CO_2. Functional Ecology. 20:763-769.

Chapter 6: Synthesis, Summary and conclusions

Synthesis

This dissertation investigates the responses of the shade-tolerant species, mountain maple (*Acer spicatum* L), seedlings to light under climate change scenarios of increasing [CO_2], soil warming and low soil moisture. The main objective of the dissertation has been to determine how the global climate changes scenarios such as rising atmospheric [CO_2], soil warming and soil moisture depletion may change the physiological and morphological responses of mountain maple seedling responses to light. While the effect of light on photosynthesis and growth are well known, there is a lack of empirical data upon which to predict pattern of shade-tolerant plant species response to light in the future. This has led to the design of two experiments that addressed two key themes: CO_2 and T_{soil}; and CO_2 and soil moisture interactive effects on mountain maple seedling response to light.

The T_{soil} interacted with [CO_2] and light to affect photosynthesis and growth of mountain maple seedling in different ways (Chapters 2 and 3). While photosynthesis increased in response to elevated [CO_2] when soil was warmer, no stimulating effect of soil warming on the photosynthetic response of mountain maple to light conditions was observed. The prediction that growth response to light would increase under elevated [CO_2] and warmer soils was rejected. It was expected that growth response to high light conditions would result from increases in carbon assimilation mediated by elevated [CO_2] and soil warming. The elevated [CO_2] and soil warming increased leaf area but the effect did not differ between light treatments. In fact, leaf area tended to decrease with increasing light conditions. Leaf traits such as LAR and SLA did not differ between CO_2 treatments at warmer soil and high light conditions. Also, the seedlings responded to high light conditions by producing thinker leaves (low SLA) which may have higher photosynthetic rates but also carbon losses. The lack of plasticity in leaf traits and high carbon losses might have limited the enhancement of growth and biomass by the high light treatment. The study shows that, elevated [CO_2] and soil warming do not promote mountain maple seedling photosynthetic and growth responses to light, and might present physiological constraints that hinder mountain maple growth in canopy gaps.

Reduction in soil moisture was not found to limit photosynthetic and growth responses of mountain maple seedling to light (chapters 4 and 5). While it is established that low soil moisture reduces physiological performances and growth of plants in high light conditions (Muraoka et al. 1997, Muraoka et al. 2002), this study shows that low moisture may not be a limiting factor on mountain maple photosynthetic and growth rates in high light environments. However, it is worth noting that the low soil moisture used in this study might not be stressful enough to have any detrimental effects on mountain maple. Hence, some caution is necessary in interpreting soil moisture effect on mountain maple seedling response to light. The data indicate that low soil moisture did not reduce leaf growth response to light when seedlings were exposed to elevated [CO_2]. However, the seedlings maintained high growth and biomass responses to high light by increasing IWUE but not morphological adjustment in terms of leaf traits under elevated [CO_2].

Studies on mountain maple and other shade-tolerant plant species physiological and morphological responses to increases in light availability and how these responses are affected by other environmental factors such as [CO_2], T_{soil} and soil moisture are not-existent. Although mountain maple respond to increases in light availability (Wilson and Fischer 1977, Canham 1988a, Lei 1992), interactions with T_{soil}, soil moisture, along with several other environmental factors may confound their effects. This study is quite unique in separating the effect of light on mountain maple physiology and morphology, and indicate overall lower responsiveness to high light in biomass production under elevated [CO_2] and warmer treatment combination than when either [CO_2] or T_{soil} alone was increased. The experiment lasted only four months and may not reflect the long-term physiological and morphological responses of mountain maple seedling to light under [CO_2], T_{soil} and soil moisture. Nonetheless, the study shows that the respond of mountain maple seedling to the creation of canopy gaps should be evaluated taken into account, the interactive effect of light with other environmental factors rather than the main effect of light alone.

References

Canham, C.D. 1988. Growth and canopy architecture of shade-tolerant trees: response to canopy gaps. Ecology. 69:786-795.

Lei, T.T. 1992. Functional design and shade adaptation in *Acer* species. *In* Biology. McGill University.

Muraoka, H., Y. Tang, H. Koizumi and I. Washitani. 2002. Effects of light and soil water availability on leaf photosynthesis and growth of *Arisaema heterophyllum* , a riparian forest understorey plant. Journal of Plant Research. 115:419-427.

Muraoka, H., Y. Tang, H. Koizumi, I. Washitani and W. Z. 1997. Combined effects of light and water availability on photosynthesis and growth of *Arisaema Heterophyllum* in the forest understory and an open site. Oecologia. 112:26-34.

Wilson, B.F. and B.C. Fischer. 1977. Striped maple: shoot growth and bud formation related to light intensity. Canadian Journal of Forest Research. 7:1-7.

Summary

Physiological responses to light under elevated [CO_2] and warmer T_{soil}

Soil warming has opposing effects on *A* responses to [CO_2] and light. While *A* responded strongly to elevated [CO_2] under warmer T_{soil}, *A* of mountain maple seedlings in the high light was slightly reduced by soil warming. The effect of soil warming on *A* response to light could be possible if soil warming-related increases in carbohydrates consumption were not met with sufficient replenishment of carbon. However, physiological (somewhat decreased g_s) and morphological (thicker leaves) acclimations to high light could also be implicated.

The elevated [CO_2] improves resource use efficiency in mountain maple seedling growing in high light treatment. The [CO_2] enrichment resulted in significant positive IWUE response to light. Under the elevated [CO_2], IWUE was 18% higher in high compared with the low light treatment. However, the high light decreased IWUE by 6% under the ambient [CO_2]. It appeared that leaf gas exchange parameters alone were not solely responsible for the increased IWUE response to light under elevated [CO_2], but also leaf morphological acclimation to light and CO_2 enrichment. Under elevated [CO_2], the allocation of N to *J* at the expense of V_{cmax} increased by 20% in response to high light treatment. However, there was no significant light effect on J/V_{cmax} when seedlings were grown under ambient [CO_2]. The results show that N was allocated to rate-limiting process of photosynthesis as a compensatory response that maximizes photosynthetic carbon gain. Mountain maple seedlings growing in canopy gaps may improve water conservation and photosynthetic nitrogen use efficiency (PNUE) through increases in IWUE and J/V_{cmax}, respectively, in the future.

Morphological responses to light under elevated [CO_2] and warmer T_{soil}

Results from the elevated [CO_2] and soil warming effects on mountain maple morphological responses to light indicate that plant size (height, RCD and TB), biomass allocation (LMR, RMR and SRR) and morphology (leaf area, SLA, LAR) responded in a similar pattern to the high light treatment but showed different relative maximum growth at different [CO_2]-T_{soil} treatment combinations. While seedlings height responded more positively to high light treatment under the ambient [CO_2]-low T_{soil}, RCD and TB showed maximum growth responses to high light under the elevated [CO_2] and low T_{soil} treatment combinations. However, increasing [CO_2] and soil warming reduced the relative effect of high light treatment on all plant size variables. Although biomass allocation to roots (RSR and RMR) increased in response to high light across [CO_2]-T_{soil} treatment combinations, the maximum response to high light was observed when seedlings were exposed to warmer T_{soil} under the ambient [CO_2]. Both variables were lowest at the ambient [CO_2]-low T_{soil} treatment combination, suggesting that the higher values at the warmer T_{soil} might be related to soil warming induced moisture depletion. In contrast, the [CO_2] and T_{soil} independently modified the response of biomass allocation to leaf, although the high light reduced LMR under both treatments. Morphological responses to light were different leaf area, SLA and LAR. Leaf area showed no positive response to high light when [CO_2] and T_{soil} were high. The elevated [CO_2] and soil warming caused a similar magnitude of decrease in LAR and SLA in response to high light treatment. The smallest reductions in these leaf traits in response to high light occurred under the ambient [CO_2] and low T_{soil} where the maximum relative effect of light on height growth was observed. Smaller reductions in these leaf traits may mean that there is a larger photosynthetic surface displayed per unit biomass invested and relatively higher growth potential in seedlings under ambient [CO_2] and low T_{soil}. However, the results demonstrates that mountain maple growth responses to the creation of canopy gaps in the future might be limited as [CO_2] rises and soils get warmer.

Physiological responses to light under elevated [CO_2] and low soil moisture

Light stimulated *A* differently under the low and high soil moisture treatments. The effect of light on *A* of mountain maple was about 28% higher under the high soil moisture. In contrast, the high light treatment reduced *A* by 6% when soil moisture was low, although the effect was not statistically significant. The results showed that both physiological (g_s) and morphological (leaf area) responses to light contributed to the lack of light on *A* when the soil moisture was low. Both g_s and leaf area did not did not differ between light treatments when seedlings were exposed to low soil moisture. However, the response of *A* to light depended on which one of the two major factors of *A* limitation was driving the response. If *A* had responded in the same pattern as C_i/C_a, then the response would primarily be associated with a strong stomatal limitation to *A*. The pattern of C_i/C_a ratio observed in this study suggests that stomatal limitation to *A* was not an important limiting factor. V_{cmax}, *J* or TPU responses to the treatments can reveal non-stomatal limitation to *A*. The pattern of TPU response to light under low soil moisture provided strong evidence for non-stomatal factors limiting *A*. The high light treatment reduced TPU by 2%, but like *A*, there was no significant difference between the high and low light treatments when soil moisture was low. Therefore, the results show that the lack decline in TPU in response to light when soil moisture was low particularly regulated *A* response to light.

Light compensation of point of photosynthesis was the only parameter that showed a significant response to the interaction among [CO_2], soil moisture and light. Relative to the low light treatment, the relative effect of high light on LCP was in the order of ambient [CO_2]-low soil moisture (10%) < elevated [CO_2]-high soil moisture (70%) < ambient [CO_2]-high soil moisture (168%) < elevated [CO_2]-low soil moisture (178%). The results show that the low soil moisture had a large negative effect on LCP (increased LCP) response to high light when seedlings were grown under the elevated [CO_2]. This indicates that, low soil moisture could shorten the time during which carbon assimilation can occur in mountain maple in future [CO_2]. However, this might not necessarily regulate mountain maple carbon balance because LCP alone does not determine plants positive carbon balance.

The CO_2 enrichment led to a reduction in drought tolerance mechanism in mountain maple seedlings. While the xylem water potential was 0.14MPa lower in response to the low soil moisture under ambient [CO_2], it decreased to 0.2MPa under the elevated [CO_2]. These adjustments resulted in higher xylem water potentials for seedlings grown under the elevated compared to the ambient [CO_2]. Theoretically, elevated [CO_2] could decrease drought tolerance of plants if increased growth rates result in decreased availability of substrates for osmotic adjustment (Wullschleger et al. 2002). However, the elevated [CO_2] increased the proportion of biomass allocated to roots at both soil moisture treatments, thus, potentially increasing the water absorption ability of the seedlings.

Morphological responses to light under elevated [CO_2] and low soil moisture

Results from this chapter indicate that the low soil moisture increased the magnitude of height growth response to high light under the elevated [CO_2], contrary to the expectation of this study. The lack growth reduction in response to light under the low soil and elevated [CO_2] appeared to be related to the response of plant morphological characteristics (leaf area). This is because there no reduction in leaf area in response to light under the same treatment combination, suggesting that low plasticity of leaf area in response to the treatment might have played a significant role. The lowest height growth response to high light was observed under the elevated [CO_2] and high soil moisture. Similarly, leaf area was reduced by the high light treatment under the elevated [CO_2] and high soil moisture. The lower values of height and leaf area could be the consequence of an acclimation of the seedlings to environmental fluctuations of greater amplitude, but a greater relative enhancement of seedlings with decreasing soil moisture by elevated [CO_2] cannot be ruled out.

The high light treatment showed striking increases in seedling biomass, but the degree of influence differed with [CO_2] or soil moisture treatment. Increase in biomass production due to increased light availability went from 131% under ambient [CO_2], to 215% under elevated [CO_2]. Similarly, biomass production as a result of increased high light availability went from 127% under low soil moisture, to 157% under the high soil moisture. The large increases in biomass in response to the high light conditions under the elevated [CO_2] was likely mediated by SLA and IWUE. High light-induced reduction in SLA and increase in IWUE under elevated [CO_2] were observed. Lower SLA in high light environments can maximize carbon gain while decreasing water loss, thus, increasing IWUE and consequently seedling biomass.

Observation of biomass allocation response to high light indicates that allocation to roots increased considerably under low soil moisture. However, light had no significant effect on biomass allocation to roots when soil moisture was high. The low soil moisture increased biomass allocation to roots under the ambient but not the elevated [CO_2]. This contrasts with the theory that low soil moisture and elevated [CO_2] increase biomass to below-ground parts to enhance water and nutrients absorption. The pattern of biomass allocation in response to light under low soil moisture is a common response in plant species, and may reflect higher transpiration demands or drought tolerance mechanism.

Main conclusions

Contrasting effects of soil warming on A response to light and [CO_2]. The decreased in *A* under ambient [CO_2] and the lack of stimulation of *A* by high light might support the theory that increased carbon consumption by roots depressed under soil warming. In contrast, elevated CO_2-induced stimulation of *A* was more when seedlings were exposed to warmer T_{soil} than low T_{soil}. The results show that seedlings in elevated [CO_2] might have benefited from increased carbon availability and possibly root activity.

Lower responsiveness of total biomass to light under elevated [CO_2] and warmer T_{soil}. Significant increase in light availability favors the growth of mountain maple seedlings. However, the results show that increases in [CO_2] and T_{soil} may not create ideal conditions for growth stimulation by light in mountain maple seedlings. Reductions in leaf traits such as SLA and LAR in high light environment and elevated [CO_2] could limit the growth potential of mountain maple seedlings. Mountain maple seedlings growth in canopy gaps after the removal of the overstory vegetation may be limited in future climate change scenarios of higher [CO_2] and warmer T_{soil}. This could have serious ecological implications on forest composition and dynamics.

Higher growth response to light under low soil moisture and elevated [CO_2]. Maximum growth response of mountain maple seedling to light was when soil moisture was low and [CO_2] was high. This is contrary to the prediction that low soil moisture would limit growth responses to light under elevated [CO_2]. Low soil moisture did not reduce leaf growth response to light under the elevated [CO_2], which could mean that low plasticity of photosynthetic surface played a role. It's deduced that, growth responses to light under elevated [CO_2] can only occur when low soil moisture significantly reduce leaf area production.