

EFFECTS OF MOISTURE AND NITROGEN STRESS ON GAS EXCHANGE AND NUTRIENT RESORPTION IN *QUERCUS RUBRA* SEEDLINGS

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Abstract.—The effects of simulated soil fertility at three levels (poor, medium, and rich soils) and moisture stress at two levels (well watered versus moisture stressed) on gas exchange and foliar nutrient resorption in 1+0 bareroot northern red oak (*Quercus rubra*) seedlings were evaluated. Current nitrogen (N) uptake was labeled with the stable isotope ¹⁵N to enable discrimination and quantification of the different N pools in new growth of plants, and the proportion remobilized and/or resorbed following senescence. Predawn leaf xylem water potential (Ψ_L) was -0.92 MPa in stressed plants and -0.76 MPa in well watered seedlings. Photosynthetic assimilation (A), stomatal conductance (gs), and transpiration (E) decreased with moisture and nutrient stress. Gas exchange rates were higher in well watered plants than in stressed plants. Simulated soil fertility increased A by 38 percent, E by 40 percent, and g_s by 50 percent in seedlings grown on the rich soil relative to those established on the poor soil. Seventy percent of N was resorbed in leaves of well watered plants compared to 47 percent in leaves of stressed plants. Resorption efficiency was 70 percent on the poor soil, but decreased to 40 percent on the rich soil. Increased foliar N resorption at low fertility suggests that the resorption process may have evolved as an important nutrient conservation strategy on infertile soils.

INTRODUCTION

Foliar nutrient resorption is an important nutrient conservation mechanism in deciduous forest ecosystems because the process may withdraw about 50 to 90 percent of the nutrients from senesced leaves for storage in plant tissues (Aerts 1996, Tagliavini and others 1998). However, the importance and underlying mechanisms of nutrient resorption and retention in plant tissues, and remobilization for new growth, are not well elucidated for northern red oak (*Quercus rubra*) seedlings. Additionally, the significance of the interactive effects of moisture and nutrient stress on red oak physiology, and on nutrient resorption efficiency, which is defined as percent N reduction between green and senesced leaves (Killingbeck 1996, Teklay 2004, Singh and others 2005, Yuan and others 2005), is not well studied. Moreover, how soil fertility might regulate resorption efficiency in red oak has yet to be elucidated. A better understanding of how moisture and nutrients might interact to influence physiological processes and resorption efficiency in red oak may be used to help manipulate plants at the nursery stage to increase nutrient retention in tissues for later utilization to benefit early establishment success of out-planted seedlings (Birge and others 2006, Salifu and Jacobs 2006).

OBJECTIVE

The objective of this study was to evaluate the effects of moisture stress and simulated soil fertility on gas exchange and N resorption in red oak seedlings. We hypothesized that resorption efficiency will increase on poor soils, but diminish on rich soils, following a nutrient conservation strategy on infertile soils. Northern

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red oak is selected for this study because of its economic importance and increased use in environmental plantings in the Central Hardwood Forest Region of the United States (Jacobs and others 2004).

MATERIALS

Materials and Growth Conditions

Bareroot northern red oak seedlings were grown from acorns for 1 year under operational conditions at the Vallonia State Nursery (38°85'N, 86°10'W) south of Indianapolis, IN, based on procedures described in Jacobs (2003). For the current study, 1-year-old bareroot red oak seedlings were obtained from Vallonia State Nursery and transplanted in sand culture using 6.2-l Treepots™ (Stuewe and Sons, Corvallis, OR). The potting soil consisted of 4:1:1 (sand:vermiculite:perlite). Plants were grown in the Department of Horticulture and Landscape Architecture Plant Growth Facility at Purdue University, West Lafayette, IN (40°25'N, 86°55'W). Mean day and night temperatures in the greenhouse were of 24 °C and 20 °C, respectively, under ambient light conditions.

Transplanted seedlings received one of three fertilizer rates: 0 [control], 500, or 1,000 mg N plant⁻¹, simulating poor, medium, and rich soils, respectively. The 500 and 1000 mg N plant⁻¹ rates approximated 125 and 250 kg N ha⁻¹, respectively, under field conditions based on the weight of soil in pots (8 kg) in relation to silvicultural prescriptions under field conditions (Brady and Weil 2002, Salifu and Timmer 2003). Nitrogen was supplied with the irrigation as [¹⁵NH₄]₂SO₄ enriched to five atoms percent ¹⁵N (34-0-0, ISOTECH, Inc.). Current N uptake was labeled with the stable isotope ¹⁵N, which allowed discrimination and quantification of the different N pools (labeled vs. unlabeled N) in new growth. Chelated micronutrients were applied at the rate of 0.08 g l⁻¹ and phosphorus (P) supplemented by KH₂P₂O₅ (0-52-34, Plant Products Co. Ltd., Brampton Ont.) at the rate of 30 kg P ha⁻¹ to avert deficiency of other nutrients. Plants were exposed to water stress treatments at two levels (well watered versus moisture stressed). Well watered plants were those that were irrigated back to container capacity (White and Mastalerz 1966) three times a week. Moisture-stressed plants were supplied irrigation only once per week, which approximates one-third of the total irrigation received by well watered seedlings. The third irrigation schedule for well watered plants coincided with that for moisture-stressed seedlings. Thus, moisture-stressed seedlings were watered using container capacity determinations from the third watering regime for well watered plants. Pots were periodically weight to determine the amount of water to be added (150 to 200 ml pot⁻¹) to bring pots back to container capacity.

Seedlings flushed one week after planting at which time fertilization commenced. Five equal split applications were conducted at weekly intervals for the first 5 weeks following flushing to improve uptake efficiency. Fertilizer was dissolved in the desired amount of irrigation and supplied to plants once a week.

Experimental Design, Plant Sampling and Chemical Analyses

The experimental design was a 2 x 3 factorial design testing moisture stress at two levels and simulated soil fertility at three levels, which was replicated three times. Foliar samples were collected from plants at two time periods: in mid-August (3 months after planting), when leaves were green and fully mature; and in November, when leaves senesced. Individual plants were enclosed in a plastic wire mesh to collect senesced leaves. Foliar data were processed for total N and ¹⁵N analyses following protocols detailed in Rundel and others (1989). Total N and ¹⁵N were determined using a Stable Isotope Mass Spectrometer coupled to a Micro-Dumas Elemental analyzer at the Stable Isotope Laboratory located at the University of Georgia,

Athens, GA. Nitrogen resorption efficiency was calculated based on procedures described by Teklay (2004) and Yuan and others (2005) as shown in the following equation:

$$\text{Resorption Efficiency (\%)} = \left[\frac{N_g - N_s}{N_g} \right] \times 100$$

Where N_g is total leaf N (mg plant^{-1}) in green and mature leaves collected prior to senescence and N_s is total leaf N (mg plant^{-1}) in senesced leaves.

Mid-day leaf gas exchange was measured on three plants per treatment in mid-August. Net photosynthesis, stomatal conductance, and transpiration were measured with LI-6400 portable infrared gas analyzer equipped with a red LED light source and a CO_2 mixer control unit (LI-COR Inc., NE). Measurements were taken on the second fully formed leaf moving basipetally from the terminal point of the second flush. External light, provided by an LED red light source (LI6400-02) built into the top of the leaf chamber, was set to ambient greenhouse light intensity during measurements. CO_2 was controlled with the LI-6400 CO_2 injection system. Relative humidity was maintained at 55 to 65 percent. Leaf temperature was maintained at ambient conditions (28 to 30 °C). Pre-dawn Ψ_L was measured a day following gas exchange measurements. Leaf xylem water potential was measured with a pressure chamber (Model 600, PMS Instruments, Inc., Corvallis, OR).

Gas exchange, Ψ_L , and resorption efficiency data were evaluated using analysis of variance and effects tested at $P < 0.05$. Significant treatment means were ranked according to Tukey's HSD test at $\alpha = 0.05$. No significant treatment interaction effect was observed between simulated soil fertility x moisture stress treatments.

RESULTS AND DISCUSSION

Moisture stress significantly affected gas exchange parameters, but not plant water potential (Table 1). For example, relative to water-stressed plants, increases in net photosynthesis (85 percent), transpiration (126 percent) and stomatal conductance (160 percent) were observed in well watered seedlings. Similarly, simulated soil fertility treatments increased net photosynthesis (38 percent), transpiration (40 percent) and stomatal conductance (50 percent) in seedlings grown on the rich soil relative to those established on the poor soil although the differences were not statistically significant (Table 1). By contrast, simulated soil fertility treatments significantly influenced plant water potential. Foliar N resorption efficiency was significantly higher ($P < 0.0016$) in well watered plants (70 percent in comparison with moisture stressed seedlings (47 percent) (Fig. 1). Simulated soil fertility significantly affected foliar N resorption ($P < 0.0027$), which was greater at lower soil fertility, but declined with increasing nutrient input (Fig. 1). For instance, resorption efficiency was 70, 65, and 40 percent on respective poor, medium, and rich soils. Diminished foliar N resorption with soil fertility suggests the resorption process may have evolved as a mechanism for N conservation at low soil fertility.

The higher foliar N resorption efficiency noted for red oak in the current study compares well with about 50 to 90 percent suggested for hardwoods (Aerts 1996, Tagliavini and others 1998, Duchesne and others 2001). This high efficiency has important implications for nutrient loading of hardwoods (Birge and others 2006, Salifu and Jacobs 2006) to build nutrient reserves for subsequent utilization. Higher resorption suggests significant quantities of nutrients may be withdrawn from leaves of loaded seedlings into storage

Table 1.—Mean (SE) of physiological responses of red oak seedlings to moisture and nitrogen stresses grown in greenhouse environments for 6 months.

Treatments	A ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	E ($\text{mmol m}^{-2} \text{s}^{-1}$)	g_s ($\text{mol m}^{-2} \text{s}^{-1}$)	Ψ (MPa)
Well watered	9.56 (0.72)a	4.03 (0.44)a	0.13 (0.02)a	-0.48 (0.05)a
Moisture-stressed	5.16 (0.50)b	1.78 (0.19)b	0.05 (0.01)b	-0.54 (0.05)a
Simulated soil fertility				
Poor	6.03 (1.15)a	2.21 (0.52)a	0.06 (0.02)a	-0.44 (0.03)b
Medium	7.72 (1.31)a	3.43 (0.83)a	0.11 (0.03)a	-0.64 (0.04)a
Rich	8.34 (1.06)a	3.09 (0.43)a	0.09 (0.02)a	-0.45 (0.05)b
P < F				
Moisture	0.0002	0.0002	0.0005	0.2911
Simulated fertility	0.0963	0.0925	0.1440	0.0109

Column means followed by different letters within treatment differ significantly according to Tukey's HSD test at $\alpha = 0.05$.

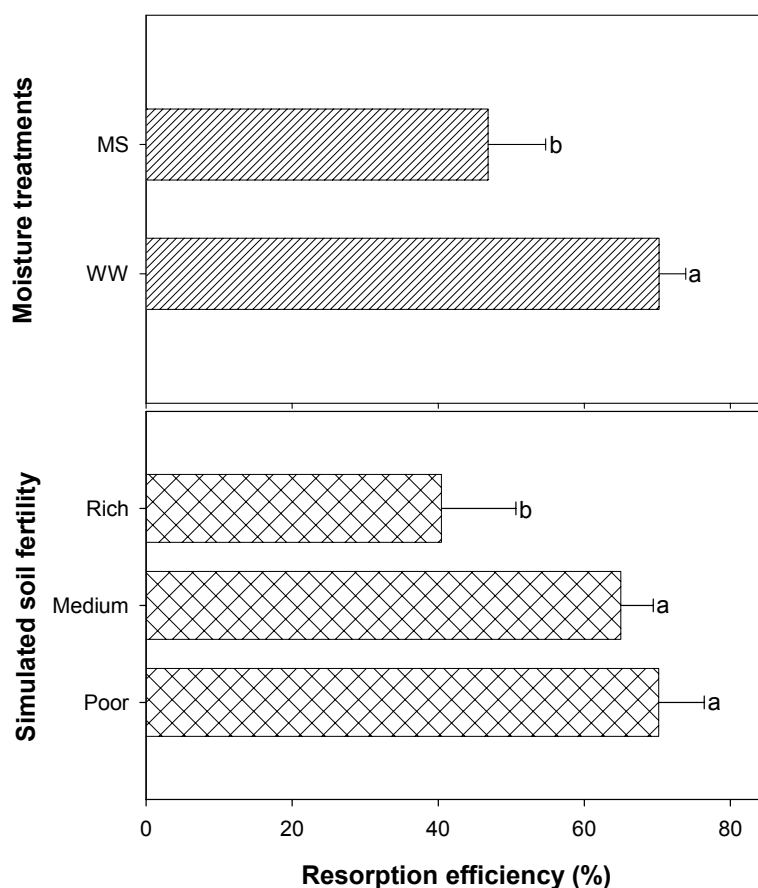


Figure 1.—Mean foliar resorption efficiency in red oak seedlings in response to moisture and nitrogen stress treatments under controlled greenhouse conditions. Plants were well water (WW), or exposed to moisture stress (MS), and unfertilized (poor soil), or received 500 mg N plant⁻¹ (medium soil), or 1,000 mg N plant⁻¹ (rich soil). Error bars indicate standard error of the mean estimate. Bars marked with same letters within treatment are not statistically different according to Tukey's highly significant difference test at $\alpha = 0.05$.

tissues prior to senescence to benefit future growth. It is suggested that roots and shoots serve as important sinks for N storage during senescence (Dickson 1989, Lacoite and others 1994, Duchesne and others 2001), and sources for N in new growth in spring (Dickson 1989). Thus, conserved nutrients may be drawn upon immediately in spring to meet increased sink demand, especially for red oak during episodic growth events (Reich and others 1980, Crow 1988, Dickson and others 2000). Data (unpublished) to be generated by quantifying the various N pools in the plant-soil system will be used to further explore morphological and physiological mechanisms in relation to how moisture and simulated soil fertility might interact to influence N remobilization, resorption and utilization in new growth of red oak seedlings. Understanding of the proportional contribution of the various N pools in new growth will enable us to identify the most important N pool, which can then be manipulated to enhance plant response. In summary, results suggest red oak may withdraw between 40 and 70 percent of nutrients from foliage into storage tissues prior to senescence. Conserved N will likely serve as a critical N source for remobilization in spring (Cheng and Fuchigami 2002, Salifu and Timmer 2003) to benefit survival, growth, and early establishment success following field transplant.

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