Optimal partitioning theory revisited: Nonstructural carbohydrates dominate root mass responses to nitrogen

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Abstract. Under optimal partitioning theory (OPT), plants preferentially allocate biomass to acquire the resource that most limits growth. Within this framework, higher root mass under low nutrients is often assumed to reflect an allocation response to build more absorptive surface. However, higher root mass also could result from increased storage of total nonstructural carbohydrates (TNC) without an increase in non-storage mass or root surface area. To test the relative contributions of TNC and non-storage mass as components of root mass responses to resources, we grew seedlings of seven northern hardwood tree species (black, red, and white oak, sugar and red maple, American beech, and black cherry) in a factorial light × nitrogen (N) greenhouse experiment. Because root mass is a coarse metric of absorptive surface, we also examined treatment effects on fine-root surface area (FRSA).

Consistent with OPT, total root mass as a proportion of whole-plant mass generally was greater in low vs. high N. However, changes in root mass were influenced by TNC mass in all seven species and were especially strong in the three oak species. In contrast, non-storage mass contributed to increased total root mass under low N in three of the seven species. Root morphology also responded, with higher fine-root surface area (normalized to root mass) under low vs. high N in four species. Although biomass partitioning responses to resources were consistent with OPT, our results challenge the implicit assumption that increases in root mass under low nutrient levels primarily reflect allocation shifts to build more root surface area. Rather, root responses to low N included increases in: TNC, non-storage mass and fine-root surface area, with increases in TNC being the largest and most consistent of these responses. The greatest TNC accumulation occurred when C was abundant relative to N. Total nonstructural carbohydrates storage could provide seedlings a carbon buffer when respiratory or growth demands are not synchronized with photosynthesis, flexibility in responding to uncertain and fluctuating abiotic and biotic conditions, and increased access to soil resources by providing an energy source for mycorrhizae, decomposers in the rhizosphere, or root uptake of nutrients.

Key words: biomass allocation; biomass partitioning; fine roots; fine root surface area; optimal partitioning theory; soil resources; storage; total nonstructural carbohydrates.

INTRODUCTION

Under optimal partitioning theory (OPT), plants should allocate additional biomass to the organ that takes up the resource that most limits growth (Bloom et al. 1985). For example, under low light, most plant species increase leaf area through some combination of increasing biomass allocation to leaves and producing thinner leaves (e.g., Grubb et al. 1996, Portsmouth and Niinemets 2007). Similarly, increased root biomass under low soil nutrients has been interpreted as evidence of a plastic allocation response to capture limiting soil resources (Fichtner and Schulze 1992, Walters and Reich 2000, Shipley and Meziane 2002, Portsmouth and Niinemets 2007). Optimal biomass partitioning is a critical assumption in theories of plant community dynamics, including the resource ratio hypothesis of plant succession (Tilman 1990) and the competitor–ruderal–stress tolerator life history strategies proposed by Grime (1979).

Increased root mass under low nutrients could reflect increased partitioning to root absorptive surface area, as implicitly assumed in OPT (e.g., Bloom et al. 1985), but empirical evidence is scant and conclusions can depend on approach. For example, apparently counter to OPT, fine-root production can increase with nitrogen (N) availability (Pregitzer et al. 1993, van Vuuren et al. 1996, Espeleta and Donovan 2002); however, because of fine-root mortality, production does not directly translate to standing root surface area. Furthermore, these studies were not explicitly focused on biomass partitioning and whether root mass as a fraction of whole-plant mass (WPM) changed with N. Nonlinear allometric relationships between organ and WPM also have to be taken into account because treatments erroneously could be interpreted as differing in biomass partitioning when in fact all treatments follow a common nonlinear allome-
Biomass partitioning to TNC storage rather than absorptive root surface area could convey a more nimble flexibility for plant interactions with the biotic and abiotic environment during the growing season, which would enhance carbon balance and survivorship under uncertain and fluctuating environments. For example, TNC could continue to be stored or be mobilized to support mycorrhizal or other microbial symbionts, increased uptake kinetics, or rapid construction of fine roots (e.g., Gaudinski et al. 2009) when soil resource levels increased (e.g., Volder et al. 2005). In contrast, biomass partitioning to absorptive root surface area, regardless of whether the proximal carbon source was from storage or current photosynthate, locks in one strategy of nutrient acquisition.

Although this study focuses on within-species variation in TNC in response to resources, interspecific variation in TNC could be an important basis of variation in species life history strategies (Kobe 1997, Kruger and Reich 1997, Canham et al. 1999, Myers and Kitajima 2007). Thus, interspecific variation in partitioning to TNC could have important implications for community processes and the distribution of species across soil resource gradients; we focus more explicitly on species differences in mass partitioning to TNC and community-level implications in another paper (R. K. Kobe et al., unpublished manuscript).

To understand root mass responses to resources, distinguishing TNC from non-storage components is crucial because each is involved in different functions. We define non-storage root mass as: total root mass – TNC root mass (Canham et al. 1999). Partitioning to non-storage implies increased investment in defense (e.g., lignins and phenolics), structure to access soil resources (i.e., fine roots), and/or metabolic compounds (e.g., lipids and proteins) that enhance nutrient uptake (Villar et al. 2006). Although non-storage mass is not a physiological entity, it is a closer approximation to structural mass involved in resource uptake than total root mass.

To test resource effects on biomass partitioning, we adopt a whole-plant perspective that incorporates effects of plant size and the potential for nonlinear allometric trajectories (McConnaughay and Coleman 1999, Weiner 2004). Specifically, we developed species- and resource-specific models of root (total, TNC, and non-storage), leaf, and stem mass, and fine-root surface area (FRSA) based on a greenhouse experiment with seedlings of seven temperate tree species. The models were used to test effects of resources per the following hypotheses: (1) partitioning of biomass to leaves and stems increases under high N and low light; and (2) partitioning of biomass to roots increases under low N and high light. To understand root mass responses, we also tested three alternative but non-mutually exclusive hypotheses. Resource-dependent increases in partitioning to root mass arise from increases in: (3) TNC and/or (4) non-storage root mass. Given that root mass alone is a crude metric of absorptive potential, (5) root surface area per unit non-storage root mass increases under low N availability.

**METHODS**

*Experiment*

To evaluate generality of responses, the study included seven species that vary in shade tolerance and association with site fertility (Table 1). Paper birch also was planted but only eight seedlings survived (all high light, three in low, and five in high N) and limited results are reported. Seed (Sheffield Seed, Locke, New York, USA) was stratified and germinated in perlite. In February 2002, germinants were planted in polyethylene-coated 10.2 × 10.2 × 27 cm³ cardboard containers.
filled with a 10:9:1 by volume silica sand : perlite : field soil mix. For each species–nutrient treatment, we planted 30–36 high-light seedlings and, to compensate for expected shade mortality, 36–45 low-light seedlings. High sand and perlite content facilitated recovery of fine roots during harvests and provided a nutrient-poor medium in which nutrient additions could be controlled. Field soil was obtained from a mesic beech–maple–oak forest at the Michigan State University Tree Research Center, East Lansing, Michigan, USA.

The experiment was designed as a factorial with two levels of light (2% and 22% full sun) and N availability (0.5 mg N/L and 50 mg N/L in a modified Hoagland's solution added every three days). To prevent buildup of salts, containers were flushed weekly with deionized water. Light levels mimicked understory environments beneath closed canopies (2% full sun) and in large tree fall gaps (22% full sun) (Schreeg et al. 2005), and N treatments approximated minimum and maximum KCl-extractable soil N concentrations in northern Michigan (Zak et al. 1986; R. K. Kobe, unpublished data). To achieve light levels, we used an inner layer of black shade cloth and, to minimize heat build-up, an outer layer of reflective knitted poly-aluminum shade cloth. Mean daytime temperatures, monitored with Hobo Dataloggers (Onset Computer, Bourne, Massachusetts, USA), were 23.62°C ± 0.04°C and did not differ between light treatments (t test, P > 0.95).

To examine allometric trajectories, we harvested six seedlings/species–nutrient–light combination at regular intervals to ~100 d (Table 1). Poor survivorship in some low-light treatments precluded some harvests. To minimize diurnal variation in TNC, all harvests were initiated 2.5 h after sunrise and were completed within 3.5 h. Seedlings were washed in deionized water; separated into leaves, stems, and roots; freeze-dried for 2 d; and weighed. Dried roots were pulverized with a ball mill (Kinetic Laboratory Equipment, Visalia, California, USA).

Root and TNC analyses
Washed fresh roots were scanned and the digital images analyzed for surface area with WinRhizo (Regent Instruments, Blain, Quebec, Canada). To measure TNC, we first extracted and analyzed soluble sugars and then analyzed extraction residues for starch. We measured TNC in roots of all harvested seedlings and in stems for a subset of final harvest seedlings to test whether root and stem TNC were correlated. We did not analyze foliar TNC because it is a short-term pool that fluctuates diurnally (Kozlowski 1992).

Soluble carbohydrates in a 20-µg sample were extracted three times in 80% ethanol and centrifuged, and concentrations (as glucose equivalents) in the supernatant were measured with a phenol-sulfuric acid colorimetric assay (Dubois et al. 1956). To measure sugar alcohols (e.g., sorbitol) in black cherry, extracts were derivatized (following Sweeney et al. 1963) and analyzed using gas chromatography (Roper et al. 1988). The pellet remaining after ethanol extraction was analyzed for starch. To gelatinize the starch, we autoclaved and then incubated samples with 10 units of amylglucosidase (Roche Diagnostics, Indianapolis, Indiana, USA) at 55°C for 16 h. Because sample processing can introduce trace amounts of monomers from structural carbohydrates, the extractant was analyzed colorimetrically using glucose-specific trinder reagent (Sigma Chemical, St. Louis, Missouri, USA) (Roper et al. 1988). Total nonstructural carbohydrates concentration was calculated as the sum of glucose equivalents of soluble sugars and starch normalized to total organ mass. Total nonstructural carbohydrates pool sizes were calculated as (concentration × root mass). In black cherry and sugar maple, low-light seedlings had inadequate mass for TNC analysis and were combined into one or more composite samples in a given treatment. See Iyer (2006) for details.

Models and data analysis
Resources could affect component mass and root surface area directly or be mediated through effects on plant mass. Thus, for each species treatment, we modeled the response variable as a function of mass. We modeled mass of leaf, stem, and root total, TNC, and non-storage as functions of WPM and root surface area as a function of non-storage root mass. The tested
models included: (1) simple linear with zero intercept \( (y = ax) \); (2) exponential \( (y = \exp(a \times x) - 1) \); (3) power \( (y = a \times x^b) \); and (4) sigmoid \( (y = a \times \exp(-b/x)) \), where \( a \) and \( b \) are estimated parameters in each model. Linear models with nonzero \( y \) intercepts (a general case of model 1) were tested but are not reported because \( y \) intercepts generally were not different from zero. The model with the strongest empirical support was chosen based on Akaike’s Information Criteria with a correction for small sample size (AICc). For each species–treatment case, we chose the simplest model within two AIC units of minimum AIC (Burnham and Anderson 2002) with significant parameter estimates (i.e., 95% support did not encompass zero).

The nonlinear models are robust for characterizing relationships but a common model is needed to statistically compare across experimental treatments. To test effects of light and N availability, we compared treatment slope estimates for linear zero-intercept models, which provided good fits to most cases. Treatment effects were considered significant \( (P < 0.05) \) when slope estimates had <29% overlap in 95% support (Austin and Hux 2002). When compared treatments had minimal overlap in the independent variable but apparently different slope estimates, we graphically assessed treatment differences in the region closest to overlap to minimize the possibility that treatments followed a common nonlinear allometric trajectory, but occupied different segments of that trajectory. All models (linear and nonlinear) were fit using Gauss-Newton nonlinear estimation in Systat 12 (Chicago, Illinois, USA), based on a loss function with normal error, which was supported by probability plots and \( G \) tests.

**RESULTS**

**Gross mass partitioning to leaves, stems, and roots**

Throughout the results, treatment comparisons are based on differences in mass partitioning to different plant organs (i.e., organ mass as a function of WPM) and are not based on absolute differences in organ mass. Supporting hypothesis 1, mass partitioning to leaves was greater under low irradiance for all species \( \times N \) combinations, except black oak at both N levels and red maple at high N (Fig. 1; also see Appendix A). Even though WPM ranges did not overlap between low- and high-light seedlings, trajectories of leaf mass as a function of WPM clearly separated in all species (Appendix B) for which linear model slopes differed. Linear functions provided the best fits for 17 of 28 cases. High N also led to higher leaf mass under high light but not in the three oak species (Fig. 1). Under low light, N had negligible effects with slight but significantly higher leaf mass per unit WPM for black cherry (0.55 in high vs. 0.48 in low N) and black oak (0.37 in high vs. 0.31 in low N). Stem mass was higher under low vs. high irradiance for all species, within N level. Nitrogen generally did not influence mass partitioning to stems (Fig. 1).

Consistent with OPT and hypothesis 2, root mass as a function of WPM was greater at low vs. high N under at least one light level in all species (Fig. 2; Appendix A). Nitrogen had no effect on biomass partitioning to roots in black and red oak under high light and sugar maple and white oak under low light. Biomass partitioning to roots was greater under high than low irradiance at both N levels for all species (Appendix A), but these results must be interpreted cautiously because of minimal overlap in WPM between low- and high-light seedlings (Fig. 2). We cannot exclude the possibility that seedlings from different light treatments followed the same nonlinear allometric trajectory, with seedlings from low light having low WPM, which was associated with disproportionately lower root mass. Supporting this interpretation, root mass from low- and high-light treatments were similar for each species at overlapping low WPM (Fig. 2) and root mass increased disproportionately with WPM in 13 of 27 cases (best fits to nonlinear models; Appendix C).

Supporting hypothesis 3, root TNC mass was greater under low vs. high N at a given WPM for all species under at least one light level (Fig. 3). Assuming a linear relationship between TNC mass and WPM, TNC mass ranged from 10% to 233% higher in low than high N (Fig. 1) among the 12 pairs of light \( \times \) species treatment combinations that could be tested (Fig. 3). Only black oak under high light and cherry under low light did not differ in TNC between N levels. Small sample sizes precluded comparisons for sugar and red maple under low light.

**Root TNC mass**

Supporting hypothesis 3, root TNC mass was greater under high vs. low irradiance for all tested species (Fig. 1). However, at similar WPM, root TNC mass was comparable in high and low light for most species, except black cherry (Fig. 3). Thus, we cannot distinguish whether seedlings from different light treatments followed different allometric trajectories or occupied different regions on the same
allometric trajectory. In support of the latter, power and sigmoid functions provided the best fit in 12 of 24 cases (Fig. 3, Appendix C), which implies disproportionately less root TNC mass at low WPM, regardless of light treatment.

Starch generally dominated TNC mass and was 15–30 times greater than soluble sugars (including sorbitol and myoinositol) in black cherry (high light) and 1.5–8 times greater in most other species. Paper birch had higher soluble sugar than starch concentrations (Appendix E).
Fig. 2. Root mass as a function of whole-plant mass (WPM; note different scales) for the different light and nutrient treatments. Best-fit equations are graphed; see Appendices A and C, respectively, for linear and nonlinear model parameter estimates and statistics. In some cases, only one line is visible because treatment slope estimates were very similar. For clarity, insets are shown in cases in which it was difficult to distinguish low WPM data points (generally low-light seedlings). See Fig. 1 for an explanation of treatment abbreviations.
Fig. 3. Root total nonstructural carbohydrates (TNC) mass as a function of whole-plant mass (WPM; note different scales). Best-fit equations are graphed; see Appendices A and C, respectively, for linear and nonlinear model parameter estimates and statistics. For clarity, insets are shown in cases in which it was difficult to distinguish low WPM data points (generally low-light seedlings). See Fig. 1 for an explanation of treatment abbreviations.
Root non-storage mass

Contrary to expectation under hypothesis 4, resource effects on non-storage root mass were negligible to modest for most species and in all cases were weaker than for TNC. Based on the linear model results, root non-storage mass responses to lower N ranged from a (nonsignificant) 10% decrease to a 71% increase, with most changes close to zero (Fig. 1). There were no resource effects on non-storage mass for the three oaks or sugar maple. Non-storage mass was a constant proportion of WPM among treatments for the oak species: ~0.4 in red and white oak and 0.38 in black oak (Fig. 4; Appendix A). Similarly for sugar maple, non-storage mass followed a single allometric trajectory for combined treatments (non-storage = 0.39WPM$^{1.18}$, $r^2 = 0.98$; Fig. 4).

Based on slope estimates of the linear models, non-storage mass at a given WPM was higher under low vs. high N in black cherry and American beech (under both low and high light) and red maple (under high light; Figs. 1 and 4). Also for beech and cherry, non-storage root mass was higher under high vs. low irradiance (Fig. 1). However, apparent differences between irradiance levels could have been artifacts of fitting models to different regions of WPM; in overlapping regions at lower WPM, low- and high-light non-storage mass values were similar (Fig. 4).

Surface area of fine roots

Fine-root surface area (<1.0 mm diameter) per unit non-storage root mass responded positively to low N in four of seven species under high light and with similar magnitude as root TNC changes (Fig. 5; Appendix F), providing qualified support for hypothesis 5. Under low light, differences in FRSA between N levels were generally small (~11% to 24%) and only the difference for red oak was significant (17%; Appendix F). We obtained similar results for N effects on FRSA when we analyzed total FRSA and used total root mass as the independent variable. Irradiance level had weaker effects than N on FRSA; under high N for beech and red oak, FRSA per unit non-storage mass was significantly higher under low than high light. In most cases, FRSA increased as a simple linear proportion of root non-storage mass (Fig. 5). For red and sugar maple under high light and high N, FRSA increased less than proportionately with root non-storage mass, as indicated by empirical support for a power function with exponent <1.

Discussion

Root mass responses to N were influenced strongly by TNC and moderately by non-storage mass

Consistent with optimal partitioning theory (OPT) (Bloom et al. 1985, Shipley and Meziane 2002) and hypothesis 2, seedlings of all species had higher root mass under low N. However, counter to hypothesis 4, higher root mass did not simply arise from increased partitioning to non-storage root mass to build more absorptive surface area, as implicitly assumed under OPT. Rather, increased TNC under low N was a large contributor to root mass changes (supporting hypothesis 3), occurred in all species, and increased more on a percentage basis than non-storage mass in all cases (Fig. 1).

These results pose the question: could higher TNC mass under reduced N, documented here and in other woody (Rothstein et al. 2000) and herbaceous species (Mooney et al. 1995, Stitt and Krapp 1999), promote greater access to soil N? Although we did not address this question in our study, we speculate that there are at least three non-mutually exclusive mechanisms through which root TNC would promote greater access to soil N. First, root TNC cues and supports mycorrhizal colonization (Same et al. 1983, Graham et al. 1997). We did not assess mycorrhizae, but decreased mycorrhizal colonization under N fertilization in sugar maple and red oak plantations (Phillips and Fahey 2007) is consistent with this mechanism. Second, root TNC could be the source of rhizosphere carbon exudates (Schwab et al. 1991), which prime populations of microbial decomposers and support mycorrhizae (Dakora and Phillips 2002, Dijkstra and Cheng 2007). The lower levels of root TNC under high N reported here parallels decreased respiration by rhizosphere microbes under N fertilization (Phillips and Fahey 2007). Paper birch had the highest soluble sugar among species here, consistent with its high rates of rhizodeposition (Bradley and Fyles 1995). Third, root TNC could provide energy for mineral N uptake/reduction and carbon skeletons to assimilate N, supported by a positive correlation between N uptake and root TNC in aspen (Rothstein et al. 2000). From an interspecific perspective, species that preferentially take up energetically expensive nitrate over ammonium (Cannell and Thornley 2000) might maintain higher levels of root TNC, but this speculation cannot be evaluated due to the paucity of information on species N form preference. In support of this idea, beech, which prefers NO$_3$- had higher TNC than sugar maple, which has strong preference for NH$_4^+$ (Rothstein et al. 1996, Templer and Dawson 2004). However, red oak has the highest TNC levels and apparently has no N form preference (Templer and Dawson 2004).

The highest levels of root TNC at high light and low nutrients and the lowest at low light and high nutrients are consistent with the carbon-nitrogen balance hypothesis (Lerdau and Coley 2002), which previously has been applied to defenses but also could apply more generally to the storage of non-limiting resources. In high light and low N, relatively abundant C may accumulate because other elements limit construction of new tissue. Conservative allocation strategies in which non-storage mass and FRSA do not respond to low N, as observed for black and white oak here, likely
exacerbate accumulation of carbon and favor forms that later can be mobilized (TNC). Although accumulation of carbon due to carbon–nutrient imbalances may have contributed to observed patterns, our results also support that TNC storage does not arise solely from resource imbalances. Under conditions that presumably would be least favorable for TNC accumulation, low light and high N, TNC concentrations still attained 20–
30% of root mass for all three oak species (Appendices D and E). Foliar nitrate inhibition of gene expression involved in starch synthesis (Scheible et al. 1997) is one process that could regulate TNC.

Fine-root surface area increased under low N for some species

Fine-root surface area is a better functional metric of increased partitioning to soil resource uptake than...
higher absolute FRSA under low N. Thus all species but white and black oak had increased WPM (not observed in any species) or partitioning to root mass (observed in cherry, red maple, and beech). Thus all species but white and black oak had higher absolute FRSA under low N.

Leaf and stem mass

Consistent with OPT and hypothesis 2, partitioning of biomass increased to both carbon-capturing leaves and stems under low light. Greater partitioning to stem mass likely reflects favoring height growth as a plastic response to perceived light competition (Walters et al. 1993, Delagrange et al. 2004). In all species but the oaks, N influenced mass partitioning to leaves under high but not low light, suggesting that leaf production under high light may be N limited. Similarly, woody growth in red oak, American beech, and red maple saplings was limited by foliar N under high but not low irradiance (Kobe 2006). Together, these results suggest that under high N, red oak growth responses are mediated through increased foliar N concentrations, while beech and red maple saplings increase both leaf mass and foliar N.

Caveats

Our snapshot TNC measurements do not estimate total allocation to TNC because these pools, especially soluble sugars, cycle rapidly (Cardon et al. 2002, Phillips and Fahey 2005). Starch, primarily a storage compound, turns over less rapidly than sugars, but can be converted to sugars. Soluble sugar results should be interpreted cautiously.

Differences between light treatments were less ambiguous for leaf and stem than root mass. A single allometric function would not have provided a good fit to leaf or stem mass from both low- and high-light treatments, supporting real differences between light levels. In contrast, mass partitioning to roots in low vs. high light could have arisen from a common nonlinear allometric trajectory with treatments occupying different trajectory segments rather than truly differing (e.g., Weiner 2004). Given that all three biomass components were modeled as functions of WPM, increased partitioning to leaf and stem mass under low light necessitates that root mass decreased. Thus, evidence supports real rather than “allometric” effects of irradiance on mass partitioning to roots.

Yet irradiance results should be interpreted with caution because our light treatments may have been less effective for smaller seedlings. High-light seedlings that had low mass were harvested early in the experiment (Table 1), and there may not have been adequate time for light effects to fully manifest. Previous studies provide conflicting results on irradiance effects on TNC in juvenile trees. In similar species, Canham et al. (1999) found negligible light effects on root TNC in 2-year-old field seedlings. But TNC concentration increased with irradiance in roots of red oak saplings (Naidu and DeLucia 1997) and European beech (Gansert and Sprick 1998).

Seed-derived TNC and nutrients could have carried over to the young seedlings in our experiment and influenced results. However, two lines of evidence suggest that seed influence was negligible and that TNC was largely derived from seedling photosynthate. First, TNC mass increased with WPM in all treatments and species. If measured TNC were largely from seed reserves, then TNC mass would have decreased with WPM as TNC was mobilized for structural growth and respiration. Second, mean seedling mass from harvests 3 and 4 under high light was 714% greater than typical species seed mass (USDA Forest Service 1974) in all species (range = 43% in white oak in high light, low N to 3700% in red maple in high light, high N), except paper birch, which increased mass >5000-fold in high N and >1000-fold in low N. White oak was the only species under high light with decreased mass from seed to seedling.

Some aspects of our methodology may have underestimated the response of FRSA to low N levels. High N mobility in the sand/perlite potting medium may have prevented N depletion in the rhizosphere, precluding benefits of fine-root proliferation. On the other hand, N treatments strongly influenced WPM and mass partitioning and would be expected to affect FRSA as well. Adopting a whole-plant perspective, our goal was to assess how effectively non-storage root mass was deployed as FRSA and thus we did not distinguish between fine- and coarse-root mass, which could underestimate effective FRSA if responses to N treatments were dominated by coarse root mass. This is unlikely because the proportion of root surface area contributed by roots <2.0 mm ranged from 0.70 (white oak in low light and low N) to 1.0 (several species–treatment combinations), with an overall mean of 0.88.

We acknowledge that fine-root rank may be more indicative of function than root diameter (Pregitzer et al. 2002), but we lack rank data. Furthermore, our results superficially contradict observed increases in fine-root mass in response to fertilization, either at the stand level or in finer scale nutrient hot spot studies (e.g., Pregitzer et al. 1993, van Vuuren 1996). However, these studies examine phenomenon at different scales than our study, are not focused on OPT, and thus did not take whole-plant changes into account.

Conclusions

Consistent with OPT, root mass as a proportion of whole-plant mass increased under low N availability. However, the increase in root mass was most strongly influenced by TNC, which could improve plant N status through cuing mycorrhizal colonization, as carbon
exudates that enhance microbial decomposition of organic matter and as energy and assimilation substrate for increased N uptake. For some species, the increase in root mass under low N partly arose from increased biomass partitioning to non-storage root mass, which could increase absorptive surface area. Some species deployed non-storage root mass more efficiently as FRSA under low N. Generally consistent with these results, N fertilizations in the field had weaker effects on fine-root mass and stronger impacts on the TNC-related processes of rhizosphere microbial respiration and mycorrhizal colonization (Phillips and Fahey 2007).

Our results argue for an expanded optimal partitioning theory that goes beyond undifferentiated root mass responses to low fertility and that encompasses how root mass is partitioned to TNC and non-storage and how non-storage mass is deployed as FRSA. Distinguishing among these pools has important implications for plant function. Under low nutrient availability, partitioning mass to build absorptive surface area locks in one major strategy to acquire nutrients necessary for seedling growth and survival. In contrast, partitioning mass to TNC provides flexibility to respond to temporal variation in the local environment; stored TNC could continue to be stored or be mobilized to support mycorrhizae, prime microbial decomposition, or rapidly build fine roots and support metabolic costs of nutrient uptake when soil resources become more available. Storing photosynthate for future deployment when environmental conditions were favorable, rather than immediately constructing fine roots, also would minimize respiratory costs of maintaining living tissue (e.g., Kobe 1997). Furthermore, evaluating OPT by biomass partitioning alone could be misleading since the location of TNC storage is not necessarily the location for the ultimate deployment of TNC. Storage of TNC also could buffer temporary resource shortages (e.g., drought) and loss of tissue to herbivory or physical damage. Through this flexibility and buffering, mass partitioning to TNC may enable a more optimal match between seedling performance (i.e., survivorship and growth) and temporal variation in both the biotic (e.g., mycorrhizae, herbivores) and abiotic environments.

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**Literature Cited**


APPENDIX A

Parameter estimates of biomass partitioning to total root, root total nonstructural carbohydrates (TNC), root non-storage, leaf, and stem mass components (Ecological Archives E091-015-A1).

APPENDIX B

Graphs of leaf mass as a function of whole-plant mass by species and resource treatment (Ecological Archives E091-015-A2).

APPENDIX C

Parameter estimates of biomass partitioning for best-supported models that were nonlinear (Ecological Archives E091-015-A3).

APPENDIX D

Graphs of stem vs. root total nonstructural carbohydrates (TNC) concentration (Ecological Archives E091-015-A4).

APPENDIX E

Graphs of soluble sugar and starch concentrations (Ecological Archives E091-015-A5).

APPENDIX F

Parameter estimates for models relating fine-root surface area to non-storage root mass (Ecological Archives E091-015-A6).