Chapter 7

THE RESPONSE OF BELOWGROUND CARBON ALLOCATION IN FORESTS TO GLOBAL CHANGE

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ABSTRACT

Belowground carbon allocation (BCA) in forests regulates soil organic matter formation and influences biotic and abiotic properties of soil such as bulk density, cation exchange capacity, and water holding capacity. On a global scale, the total quantity of carbon allocated belowground by terrestrial plants is enormous, exceeding by an order of magnitude the quantity of carbon emitted to the atmosphere through combustion of fossil fuels. Despite the importance of BCA to the functioning of plant and soil communities, as well as the global carbon budget, controls on BCA are relatively poorly understood. Consequently, our ability to predict how BCA will respond to changes in atmospheric greenhouse gases, climate, nutrient deposition, and plant community composition remains rudimentary. In this synthesis, we examine BCA from three perspectives: coarse-root standing stock, belowground net primary production (BNPP), and total belowground carbon allocation (TBCA). For each, we examine methodologies and methodological constraints, as well as constraints of terminology. We then examine available

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data for any predictable variation in BCA due to changes in species composition, mean annual temperature, or elevated CO_2 in existing Free Air CO_2 Exposure (FACE) experiments. Finally, we discuss what we feel are important future directions for belowground carbon allocation research, with a focus on global change issues.

INTRODUCTION

Belowground carbon allocation (BCA) links the soil ecosystem and foodweb with the forest canopy, providing a flow of organic carbon (C) to the soil from the CO_2 fixed by photosynthesis from the air. From an evolutionary perspective, BCA represents the currency with which photosynthetic cyanobacterial endosymbionts in leaves (chloroplasts) acquire nutrients, water and structural support from their symbiotic partners belowground (plant roots, mycorrhizal fungi, and in some cases nitrogen-fixing bacteria). This flow of organic C between aboveground endosymbiont and belowground symbionts has a substantial impact on the global carbon cycle. BCA is the Earth's third largest biologically mediated C flux, after terrestrial photosynthesis (from which BCA is derived) and oceanic photosynthesis. Terrestrial plants allocate belowground some 60 Pg C out of the 120 Pg C fixed annually by terrestrial vegetation through photosynthesis, with most this gross carbon flux occurring in ecosystems with trees (Schimel 1995, Grace and Rayment 2000). By comparison, the annual flux of combusted fossil fuel C into the atmosphere is about 6 Pg C (Schimel 1995). At the stand scale, plants allocate large quantities of carbon belowground for the construction and maintenance of roots and mycorrhizae, such that BCA may represent the largest sink for gross primary production (Ryan et al. 1996, Janssens et al. 2001). In resource-limited environments typical of terrestrial ecosystems, high plant investment in BCA is necessary to secure the water and nutrients that drive terrestrial primary production.

Despite the magnitude of BCA, both globally and locally, BCA remains the least understood C flux in plant communities (Ryan et al. 1996, Clark et al. 2001a, Giardina and Ryan 2002, Giardina et al. 2004). In contrast to aboveground plant physiology, which is precisely captured in leaf-based physiological process models (Landsberg and Gower 1997), controls on belowground processes are poorly captured in process models. Efforts to validate belowground models are hindered by the complexity of above and belowground interactions with local and global changes in environmental factors (Figure 1). Further, the soil matrix complicates nearly all aspects of BCA. There are a wide range of approaches to characterizing belowground carbon cycling (Figure 2), but robust validation of these approaches remains problematic. As a result, conceptual and theoretical models describing BCA response to global change variables are highly uncertain (Giardina and Ryan 2000, Holland et al. 2000, Pendall et al. 2004), with ecosystem models often

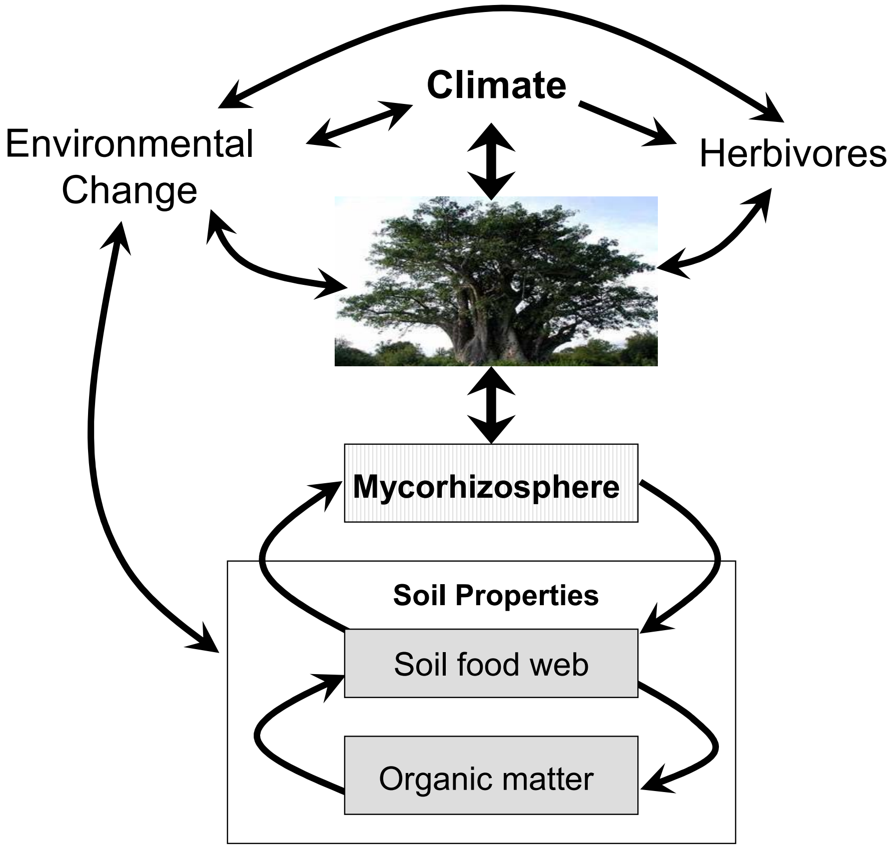

Figure 1. Diagram of the direct and indirect effects of environmental change (e.g., increased CO_2, O_3 and other greenhouse gases, change in fire regimes, elevated nutrient deposition rates, altered species succession, species invasion, etc.) on belowground carbon allocation through changes in canopy function, aboveground herbivore communities and soil properties (soil carbon and nutrient quality, soil food web including belowground herbivores).

relying on the assumption that the functioning and dynamics of aboveground tissues adequately describe those of belowground tissues (e.g., VEMAP et al 1994).

Forests are dynamic, with belowground process rates depending on factors such as tree species composition, nutrient and water supplies, and temperature. These factors influence BCA, and determine BCA response to global change. Species change is a dominant feature of global change (Figure 3, and other chapters in this volume), with composition varying over long and short periods. Dramatic species change can occur in response to climatic change in just centuries (Figure 3). Species change in response to exploitation (e.g., loss of white pine in the Great Lakes forests) or disease (e.g., loss of chestnut to blight) can occur in decades or less. Change can be even faster when short-term droughts are coupled with severe fires. Human management of ecosystems has altered species composition across plant life forms – annual

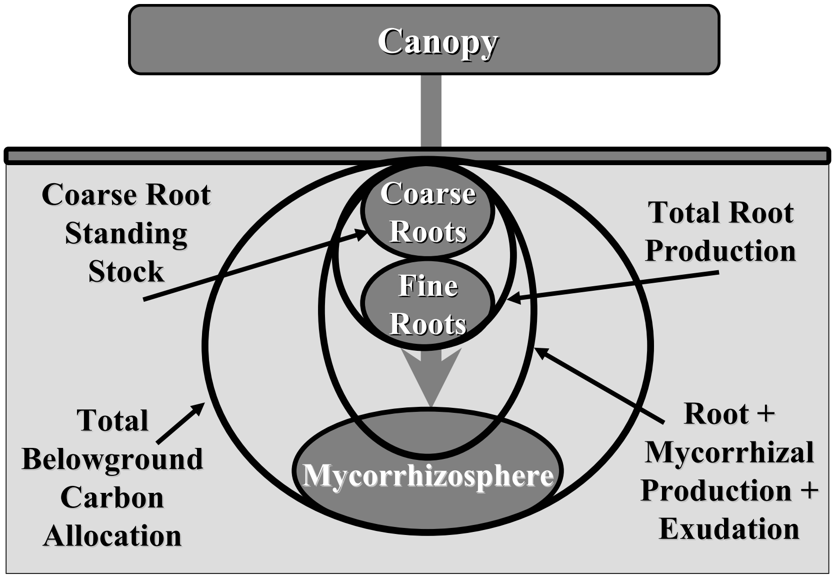

Figure 2. Various approaches to examining BCA in forests. Coarse root standing stock is a pool of carbon in soil measured by excavation and weighing at a single point in time. Total root production is comprised of coarse and fine root NPP, but typically excludes exudation or mycorrhizal production. Root + mycorrhizal production + exudation is all the C allocated belowground except for root respiration. Total belowground carbon allocation is all the C allocated belowground.

grasses in agricultural systems, perennial grasses and forbs in managed pastures, and long-lived trees in forest plantations.

Agricultural land use impacts soils, and most of these impacts are negative with regards to organic matter content (Paul and Clark 1996, Davidson et al. 2002). Forest management alters species composition (by planting, and use of fire and herbicides), nutrient supply (through fertilization or indirectly with harvesting and other silvicultural operations), and even water supply. These modifications typically increase aboveground process rates, but the response of BCA is less clear and probably variable. For example, intensive forest management usually increases aboveground net primary production (ANPP), but BCA may be reduced as a result of species (and genotype) change and improved tree nutrition. Afforestation in the 20th Century may have increased soil quality through increased organic matter content and reduced bulk density in many regions (see Six et al. 2002 for agricultural lands; Minkinnen et al. 1999 for a peatland case study), but we have little idea of the magnitude of changes in BCA that account for soil C changes (Bashkin and Binkley 1998, Binkley and Resh 1999, Paul et al. 2002, Giardina et al. 2004). Rising concentrations of gases in the atmosphere affect plants directly and indirectly. Increased concentrations of CO_2 in the atmosphere may stimulate productivity, including BCA (Pregitzer et al. 2000b, Zak et al. 2000a, King et al. 2001, Norby et al. 2002). Other gases

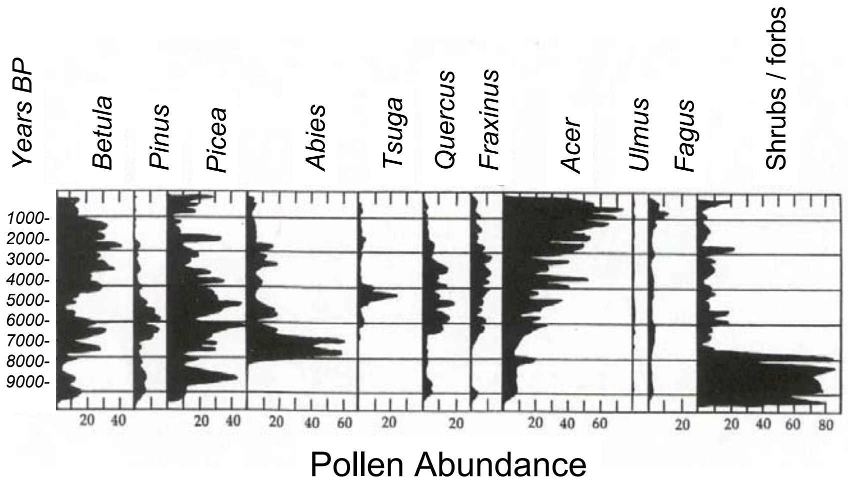

Figure 3. 9000 year record of change in vegetation (years before present) in Nova Scotia, as captured by a change in the quantity of pollen from different genera of common North American trees (Adapted from Livingston, 1968).

such as ozone (O_3) inhibit productivity (Reich 1983, Reich and Amundson 1985, King et al. 2001, Karnosky et al. 2003). Increases in atmospheric CO_2 and other greenhouse gases may indirectly affect plants by changing global climate (Norby and Luo 2004, Pendall et al. 2004). Taken together, these direct and indirect effects on plants will likely impact BCA but above to belowground links (Figure 1) remain poorly quantified.

Concern exists that rising greenhouse gases may warm the biosphere (Norby and Luo 2004) with micro to meso-scale studies often show strong temperature and moisture effects on plants and microbes (Uselman et al. 1999, Pregitzer et al. 2000a, Zak et al. 2000b, Pendall et al. 2004). The effects of increased greenhouse gases on BCA in forests remain uncertain, including direct alteration of canopy processes and indirect influences through warming and changing hydrology. Experiments on these individual processes have documented impacts on aboveground plant productivity (Reich 1983, Townsend et al. 1996, Holland et al. 1996, Karnosky et al. 2003), plant and mycorrhizal community composition (Karnosky et al. 2003, Lilleskov et al. 2001, Lilleskov et al. 2002), and soil heterotrophic organisms (Zak et al. 2000a, Zak et al. 2000b). However, the overall response of BCA to global change has been difficult to quantify King et al. (2001) because BCA integrates above and belowground changes, and the direct and indirect effects of global change factors on ecosystems can offset one another (Figure 1).

Few generalizations about controls on BCA have been made because methods and resulting estimates of BCA range widely (Ovington 1957, Raich and Nadelhoffer 1989, Albaugh et al. 1998, Reich and Bolstad 2001, Shaver and Jonasson 2001, Gower et al. 2001a, King et al. 2001, Davidson et al. 2002, Giardina and Ryan 2002), and responses to environmental variables are diverse (King et al. 1999, Pregitzer et al. 2000a, King et al. 2001, Giardina and Ryan 2002, Litton et al. *in review*). However, advances in belowground carbon science are occurring rapidly, particularly where stable isotopes permit investigators to track the flow of carbon through soil (Loya et al. 2003, Giardina et al. 2004, Matamala et al. 2004).

Two sets of findings point to an important change in our understanding of how global change factors control BCA. First, belowground processes in forests may be less responsive to temperature perturbations than previously believed, with root acclimation and substrate limitation on soil surface CO_2 efflux (Fitter et al. 1999, Giardina and Ryan 2000, Hogberg et al. 2001, Janssens et al. 2001, Melillo et al. 2002) potentially reducing the sensitivity of "soil respiration" to global warming (but see Burton and Pregitzer 2003, Burton et al. 2003). Secondly, plant canopies are tightly coupled to soil surface CO_2 efflux, with efflux being derived largely from recent photosynthesis (Horwath et al. 1994, Fitter et al. 1999, Janssens et al. 2001, Hogberg et al. 2001, Giardina et al. 2004). The degree of coupling was highlighted in a boreal forest by Hogberg et al. (2001), who reported up to a 40% reduction in soil CO_2 efflux within days of eliminating phloem transport of carbon to roots and mycorrhizae through girdling. Giardina et al. (2004) used ^{13}C isotopic methods to calculate that 90% of soil surface CO_2 efflux in a humid tropical forest was derived from current-year photosynthesis.

Further advances in belowground science have become possible with experiments exposing whole stands of trees to multiple global change variables (Karnosky et al. 2003). The free air CO_2 enrichment (FACE) experiment in Rhinelander, Wisconsin is especially important because three tree communities have been fumigated, singly and in combination, with gases that stimulate (CO_2) or reduce (O_3) plant primary production. The high cost of such replicated and multi-factorial ecosystem-scale experiments limits the number of interacting factors that can be examined. Consequently, the numerous interacting feedbacks originating both above and belowground will likely have to be examined through a combination of one and two-factor experiments, natural gradient studies, and modeling (Norby and Luo 2004).

A final feature of BCA complexity involves definitions of BCA. The terminology employed to describe carbon allocation within plants has been described as "varied, inconsistent, confusing, and often inadequate for understanding and integrating research results" (Dickson and Isebrands 1993). A similar lack of clarity continues to exist in BCA studies (Figure 2). BCA is often defined as fine or coarse root biomass standing stock (defined as partitioning by Dickson and Isebrands 1993), fine root production, total root production (coarse plus fine), total root production plus exudation and mycorrhizal production (which equals BNPP), and total belowground carbon allocation (which equals TBCA). Further, important components of BNPP are sometimes ignored in efforts to estimate whole stand or large scale patterns of NPP. While methodological ambiguities are not uncommon in ecological studies, the implications with respect to BCA are sizable because estimates are often scaled to entire regions or continents (e.g., Schimel et al. 1994, VEMAP 1994, Li et al. 2003), with uncertainties seriously impeding efforts to model climate change impacts on the global carbon cycle (Holland et al. 2000, Sarmiento 2000).

In this synthesis of global changes and the response of belowground production, we examine three BCA methods: coarse root standing stock, belowground net primary production (BNPP), and total belowground carbon allocation (TBCA). These three categories are methodologically and conceptually distinct, spanning the full spectrum of BCA studies (Figure 2). Coarse root standing stock is a pool determined through excavation and weighing, with quantification occurring at a single point in time. Changes over time are often inferred to be proportional to changes in aboveground biomass. BNPP is a flux arrived at by summing individually and periodically measured components, including period application of carefully determined allometries. Total belowground carbon allocation is a mass-balance approach that estimates a flux through periodic measurement of losses and changes in carbon storage. We examine available information on these three approaches to identify key features of each methodology, caveats, and data availability for examining BCA response to global change. Specifically, we examine whether aboveground measures can be used to predict BCA, and the likely magnitudes of BCA in relation to species, temperature and elevated CO_2. High rates of N deposition will likely impact BCA (Adams et al. 2004), but this was beyond the scope of our review. We finish the chapter with a list of the most pressing questions in the science of belowground carbon allocation.

COARSE ROOT STANDING STOCK

Quantifying root to shoot ratios has a long history in ecology (Ovington 1957, Cannel and Dewar 1994), with most studies measuring plant root standing stocks in non-woody plants or tree seedlings where roots serve primarily up-take and transport functions (McConnaughay and Coleman 1999, Giardina et al. 2001). The roots of older trees may be extensive for scavenging for resources, and very large to support massive aboveground structures. For example, Nepstad and colleagues (1994) showed that coarse roots can extend 7 m or more into soil in a seasonally dry tropical forest. Similarly, exploratory studies in riparian systems have shown that roots of trees including obligate phreatophytes (e.g., *Populus fremontii*) can extend many meters into soil to capture fluctuating groundwater (McElrone et al. 2004). As a result, accurately measuring the coarse-root standing stock of even young forests is challenging. Size is also complicated by variation in the horizontal distribution of coarse roots, with many studies sampling coarse roots between stumps to avoid digging up whole trees and their underlying tap roots. Because the largest mass of roots is located underneath the stump, this sampling bias renders between-tree coarse root mass estimates difficult to interpret. Variation in mass relating to species, fertility, or age also complicates efforts to estimate coarse-root standing stock, especially in mixed age / mixed species stands typically encountered in nature.

Variation in root to shoot relationships that relates to stand age (Figure 4; from Ovington 1957) can be dynamic, but process models typically use a single ratio to predict coarse-root standing stock, often set as a fixed proportion of aboveground biomass. For example, the function incorporated into a Canadian empirical model for pine relies on a single ratio of 0.22 (Li et al. 2003); this might be adequate for very broad assessments, but would miss important local detail if applied to individual stands. Recent papers demonstrating differences in root to shoot for conifers and hardwoods include Li et al. (2003) and Bolte et al. (2004).

The source of the discrepancy between model assumptions (e.g., Li et al. 2003) and the Ovington (1957) data presented in Figure 4 is unknown. However, if Ovington's Scots pine data are accurate, then process models may be under-predicting coarse-root standing stock in younger age classes of pine. Similar coarse-root standing stock data are available for hardwoods (Li et al 2003), but again age, species or site related patterns are poorly quantified. In general, uncertainty of coarse-root standing stock estimates has important implications for global C budgets. For example, of the estimated 60 Pg C allocated belowground by plants to roots and mycorrhizae, at least half occurs in wooded ecosystems (Grace and Rayment 2000). Based on limited

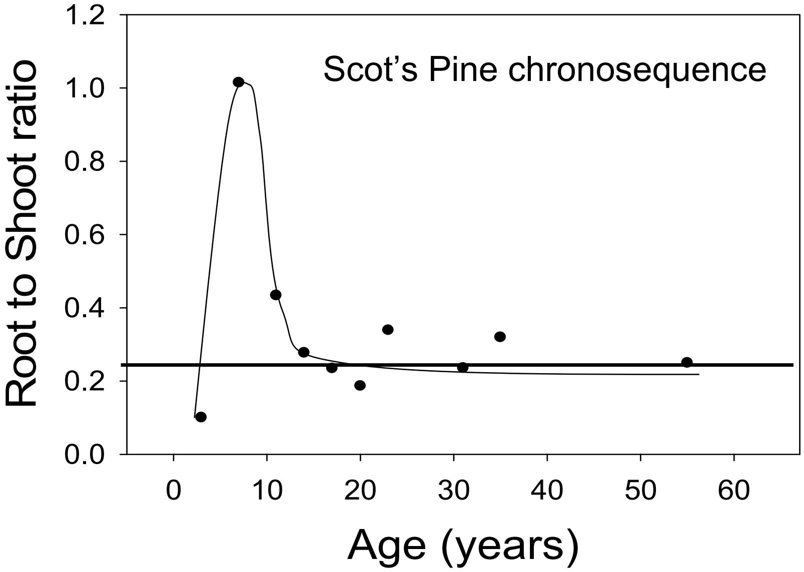

Figure 4. Data for Scots pine from Ovington (1957) and the root to shoot ratio (identified by the line at a root to shoot ratio of 0.22) used by Li et al. (2003) for estimating coarse root standing stocks for pine forests in Canada. The value of 1.0 for the 8-year-old stand of Ovington either indicates a major difference between the two studies, or it is an outlier and the studies support similar conclusions.

knowledge of how TBCA is partitioned belowground (Giardina and Ryan 2002, Giardina et al. 2004), approximately 10% of the C allocated belowground in wooded systems (3 Pg C) is allocated to coarse root production. If current coarse root allometries under-estimate coarse root to aboveground biomass ratios by an average error of 20%, then globally about 0.6 Pg C of coarse root NPP in forested ecosystems would be missed in current model estimates. To put this error in context, 0.6 Pg C is approximately 10% of annual global fossil fuel emissions.

While variation in coarse-root standing stock in relation to climate and species is poorly quantified, and errors in coarse root assumptions limit our ability to generalize about species or site differences, there appears to be some confidence that coarse root allometries within a species or climate zone are relatively insensitive to changes in fertility (King et al. 1999, Enquist et al. 2001, Giardina and Ryan 2002, Coleman et al. 2004, Coyle and Coleman 2005). Albaugh et al. (1998) harvested *Pinus taeda* trees from control, fertilized, irrigated and fertilized+irrigated stands after three years of treatment, and root to shoot allometry was constant despite a doubling of leaf area index and biomass in the fertilization and irrigation treatments (Figure 5). In contrast, Stape et al. (2004) observed a decrease in root to shoot ratios from 0.32 to 0.16 in *Eucalyptus* plantations with increasing moisture (Figure 5). Given that larger trees tend to have larger root systems, a key issue is whether the relationship between root and shoot biomass has a Y-intercept of 0 (as in the loblolly pine case in Figure 5), or not (as in the *Eucalyptus* case study in Figure 5). Litton et al. (2003) observed that root to shoot biomass ratio of young lodgepole pine trees increased with stand density but decreased with average stand basal area. In both cases, tree size varied with the treatment, such that ontogeny related effects (Figure 4) could not be ruled out.

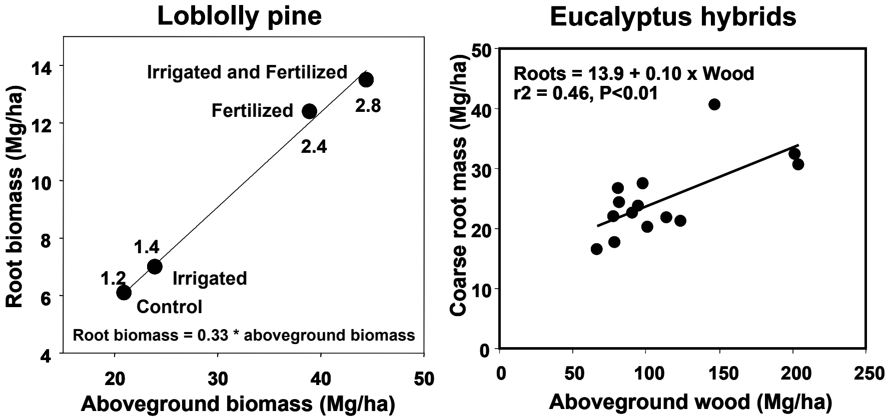

Figure 5. The root to shoot ratio was constant for loblolly pine (0.33) across treatments on a poor site ($R^2 = 0.98$, $P < 0.01$; from Albaugh et al. 1998). The same relationship for *Eucalyptus* in Brazil across 14 sites of varying productivity had a positive Y intercept, indicating that root mass increased as a proportion of aboveground mass from 16% on fertile sites to 32% on infertile sites (from Stape et al. 2004). Leaf area index (m^2/m^2) indicated next to loblolly points.

Given the importance of large roots for supporting large trees, King et al. (1999a) suggested that for a given site and species, root to shoot allometry should be less sensitive to changes in environmental conditions than root to shoot ratios of forbs (McConnaughay and Coleman 1999), or seedlings of woody plants (Gebauer et al 1996, King et al. 1999a, Giardina and Rhoades 2001). Specifically, these findings point to the important possibility that whole tree allometry may not change in response to anticipated changes in atmospheric CO_2 or global climate, though changes in moisture may alter allometry (Stape et al. 2004). This assertion is supported by King et al. (1999) where root to shoot allometry of *P. taeda* and *P. ponderosa* exposed to treatments of elevated temperature, CO_2 and nutrients, showed little effect of the treatments.

Even if changes in environmental conditions have little effect on root to shoot allometry, allometry may still change in response to alteration of site fertility, stand age, or species composition (King et al. 1996, King et al. 1999a, Bolte et al. 2004, Coleman et al. 2004, Coyle and Coleman, 2005). For example, increased tree growth due to elevated CO_2 or temperature may accelerate maturation and age-related changes coarse-root standing stock to aboveground allometry (Figure 4), and these changes could be misinterpreted as direct treatment effects on whole tree allometry rather than indirect effects of the treatments on allometry through accelerated ontogeny (see McConnaughay and Coleman 1999). We also note that changes in vegetation types may also occur; tree invasion of grasslands altered root architecture, BCA, and soil C storage (Jackson et al. 2002).

BELOWGROUND NET PRIMARY PRODUCTION

BNPP defined

Belowground net primary production (BNPP) has been defined as the mass of roots produced plus any root mortality occurring over a specified period of time. Increasingly, BNPP is defined as including all carbon allocated belowground by plants and not used for autotrophic respiration:

$$\mathrm{BNPP} = \Delta B + D + H + E + M \qquad (1)$$

where ΔB is the change in root biomass, D is detritus generated, H is losses to herbivory, E is exudation from the roots, and M is C flowing to mycorrhizae. Change in biomass (ΔB) includes the increment in tap roots, structural roots and feeder-root tissue, measured over some increment of time (typically one year). Detritus (D) includes root mortality, root tissue loss, and mycorrhizal turnover during the year. Fine roots have received the most attention because the equivalent of their entire mass may be replaced (turnover) in one year or less (Eissenstat and Yanai 1997). Although the fraction of root tissue found in feeder roots at any time may be only 5% to 10% of belowground biomass, the rapid turnover rate makes this an important fraction of BNPP. Tap and coarse root mortality is typically low for healthy trees, but tree mortality is a normal component of forest development (and harvesting and fire!), and this would lead to significant mortality of tap roots and coarse roots. The loss of root cortical tissue during secondary thickening of feeder roots and sloughing of periderm tissue in large coarse roots should also be included as a component of BNPP, but it is unlikely to be a large fraction of total BNPP. The magnitude of insect herbivory on roots (*H*) remains poorly known, but may be large in some cases. The C allocated to mycorrhizae (*M*) has long been known to be a large component of BNPP, and probably remains the largest poorly quantified component of BNPP (Fogel and Hunt 1983, Eissenstat et al. 2000; Stevens et al. 2002; Wells et al. 2002b). Exudation (*E*) of organic compounds supporting rhizosphere organisms is difficult to quantify, yet *E* also may be a significant component of the BNPP budget and an important flux of labile carbon to soil (Uselman et al. 1999).

Methods of measuring BNPP

Each technique for measuring BNPP has advantages and disadvantages, and no perfect method is available to gauge the accuracy of other methods. Most effort has gone into assessing fine-root growth in part because of the importance of these tissues for nutrient uptake but also because they are the easiest component of BNPP to measure. Net production of fine roots has been studied using sequential coring, root in-growth cores or screens (Caldwell and Virginia 1991). Fine root biomass has also been estimated by coupling repeated soil coring with images from mini-rhizotrons (Hendrick and Pregitzer 1992).

With sequential coring, fine-root production and mortality are determined from changes in standing crops of live and dead fine-roots harvested from cores collected periodically throughout the year (e.g. Grier et al. 1981). The method assumes that incremental increases in live roots represent production and incremental increases in dead-roots represent mortality (Santantonio and Hermann 1985). The method also assumes that arbitrary size classes (e.g., < 2.0 mm) accurately reflects the dynamic portion of the root system over the time steps of interest, that recovery of roots is unbiased, and that pools of live and dead roots are near steady-state, none of which may be necessarily true (Pregitzer 2002). Finally, sequential coring methods assume production and mortality do not occur simultaneously, and therefore the method can underestimate fine-root turnover and production (Publicover and Vogt 1993). The method can also overestimate root turnover if random variation in fine root estimates are mistaken for real gains and losses between sampling periods.

Viewing methods use rhizotrons, which involve using transparent viewing surfaces placed against the soil to allow measurement of the appearance, disappearance and lifespan of individual roots (Keyes and Grier 1981). Converting recorded images into estimates is labor intensive, but specialized software may be useful for image processing (Hendrick and Pregitzer 1992). Viewing methods simultaneously quantify fine-root production and loss, and rhizotron-based methods coupled with survival analysis techniques (Allison 1995; Wells and Eissenstat 2001) are leading to new insights into how environmental, developmental and phenological factors control fine-root turnover, especially when coupled with soil coring methods (Hendrick and Pregitzer 1996; Kern et al. 2004; Reuss et al. 2003; Wells et al. 2002a). Potential sources of error with rhizotron-based approaches to estimating BNPP include any effect of the observation window on root longevity (Withington et al. 2003), the difficulty of precisely measuring very small roots, especially in the surface few mm of soil (Vos and Groenwold 1987), disturbance effects of viewing windows installation on root growth (Coleman et al. 2000; Joslin and Wolfe 1999), and scaling from 2-dimensional area to the mass in a volume of soil.

Fine root lifespan can also be quantified with radiocarbon and stable carbon isotope depletion methods. These methods use bomb ^{14}C released during nuclear testing (Gaudinski et al. 2001) or a change in ^{13}C label from elevated CO_2 experiments (Matamala et al. 2003) to determine the mean residence time of root carbon. These isotope-based methods examine the isotopic composition of the total root pool at the end of some measurement interval. Notably, the survivorship of roots in soil is highly skewed, with a small portion living for a long period of time. However, short-lived roots that form and die within the measurement interval, perhaps the majority of roots in soil, will not be measured so that root longevity and turnover time may be overestimated. In fact, isotope based estimates appear to be many months to years longer than rhizotron-based approaches that track the birth and death of individual roots, although some of the discrepancy may derive from differences in size classes of roots of varying longevities. Roots that grow and die between measurement periods will not be sampled by isotope or rhizotron methods. With isotope methods, assumptions about the shape of the depletion curve and internal cycling of carbon also have strong effect on root lifespan estimates (Gaudinski et al 2001, Luo 2003). Reconciling discrepancies between rhizotron and isotope methods will almost certainly improve confidence in estimates of fine root NPP.

No validated estimates of mycorrhizal contribution to BNPP are available, in part because there are enormous challenges involved in trying to ascertain mycorrhizal fungal biomass, production and turnover. Three pools of mycorrhizal fungal biomass that differ in sampling approach and quantification include: reproductive sporocarps (mushrooms and spores), mycorrhizal roots and mycorrhizal mycelium in soil.

Saprotrophic sporocarps and their spores can be reliably distinguished from the sporocarps of mycorrhizal species, so sporocarp production is the most easily quantifiable component of mycorrhizal contribution to BNPP. Not all mycorrhizae fruit aboveground (e.g., truffles and truffle-like fungi), so quantification would require raking for hypogeous sporocarps of ectomycorrhizal fungi, and soil coring and extraction of the large spores of AM fungi. In Mediterranean climates, hypogeous sporocarps can be a large component of mycorrhizal sporocarp production. Sporocarp quantification also requires intensive sampling throughout the growing season, as fruiting can take place through the growing season

For mycorrhizal roots, visual estimates of fungal abundance (for AM fungi) and biochemical markers (for ectomycorrhizal fungi) are the primary approaches for estimating biomass. The arbuscular mycorrhizal (AM) component can be quantified by clearing and staining combined with some estimate of internal hyphal colonization. The ectomycorrhizal (EM) component can be quantified by using ergosterol, a sterol unique to fungi that has been used to quantify fungal biomass for basidiomycetes and ascomycetes, but appears to be absent in AM fungi (e.g., Grandmougin-Ferjani et al. 1999, Olsson et al 2003). Even when present, ergosterol concentrations in fungal tissue can vary several-fold, leaving large uncertainties in biomass conversions. Specific phospholipid fatty acids (PLFAs) are used as fungal biomarkers, but their concentrations are even more variable than that of ergosterol, making their use as biomass estimators untenable (Olsson et al. 2003). Production and turnover of mycorrhizal root tips can be estimated using minirhizotron systems, though we know of no such production estimates.

The quantification of mycorrhizal biomass and production in soils is probably the greatest challenge in estimating BNPP. Distinguishing between mycorrhizal and free-living heterotrophic fungi in soil is problematic, as dominant ectomycorrhizal and saprotrophic fungi are not taxonomically distinct, both being comprised primarily of Basidiomycetes and Ascomycetes. Recently natural abundance isotopes have been used in combination with ingrowth fine mesh bags with and without trenching, to estimate ectomycorrhizal fungal biomass production (e.g., Wallander et al 2001).

It is somewhat easier to distinguish AM fungi from saprotrophs, because the fungi that form arbuscular mycorrhizae are Glomeromycetes (formerly Glomales), which are taxonomically, morphologically and biochemically distinct from the Basidiomycetes and Ascomycetes (Smith and Read 1997). Given the uncertainties associated with biochemical markers described above, the best current method for biomass estimation is using microscopic methods (Bonfante-Fasolo 1986). Combined with in-growth fine mesh bags, some estimates of net production could be made.

Constraints of terminology on BNPP

Understanding the response of BNPP to global change is hindered by terminology. The estimates of BNPP in Figures 6, 7 and 8 were all termed BNPP, but none included all components of BNPP (Equation 1). Exudation is commonly excluded, or a "best guess" is used to constrain the magnitude of this component. Depending on how mycorrhizae are defined (heterotrophic or autotrophic), mycorrhizal contributions to BNPP are also poorly quantified.

These problems exist because it is extremely difficult to separate autotrophic from heterotrophic components of the total belowground C allocation (TBCA) at scales of stand and years. Attempts at quantifying BNPP have had to ignore key components to arrive at estimates, or have wrestled with the challenge of separating autotrophic and heterotrophic components. Recent advances have been made in the effort to separate heterotrophic from autotrophic components of soil respiration using trenching, stem girdling, or a components approach, but quantifying BNPP is still very difficult. First, girdling or more destructive approaches cannot separate root respiration (autotrophic) from exudation (heterotrophic) derived CO_2. Second, CO_2 from mycorrhizal respiration (autotrophic?) and mycorrhizal turnover (heterotrophic) cannot be separated (conceptually or physically) from root processes. Biologically, mycorrhizae are heterotrophic (and in some cases partially saprotrophic), but functionally they extend a plant's root system and therefore may be viewed as being autotrophic. For example, Gower et al. (2001a) identified mycorrhizae as a significant part of BNPP, and therefore autotrophic. Finally, even if soil respiration could be precisely divided into

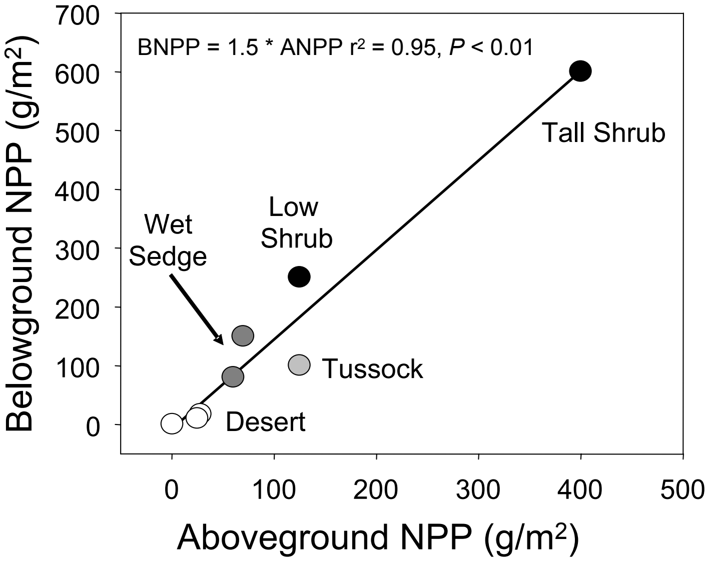

Figure 6. Data from review by Shaver and Jonasson (2001), showing the stability of BNPP to ANPP for Arctic ecosystems in North America.

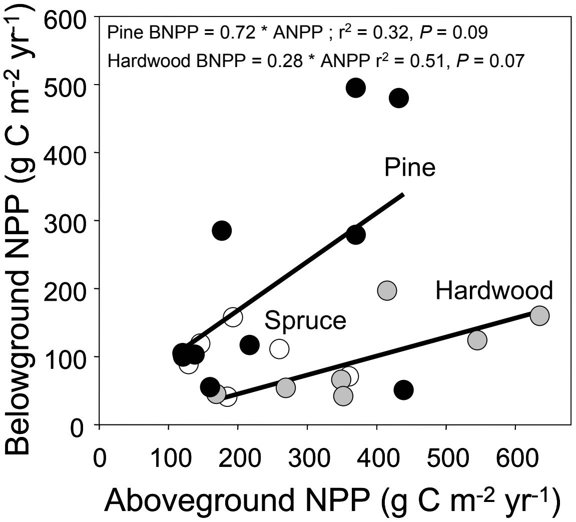

Figure 7. Data from review by Gower et al. (2001a), showing the stability of BNPP to ANPP for boreal hardwood and pine species across Russia and North America. There was no relationship between BNPP and ANPP for spruce species across sites, but the relationship appears negative.

heterotrophic and autotrophic respiration, the heterotrophic sources of soil respiration are themselves very complex and perhaps impossible to separate (Bond-Lamberty et al. 2004). For example, a significant but seasonally variable fraction of soil respiration is derived from aboveground litterfall carbon (Raich and Nadelhoffer 1989), but because leaves may be comminuted and transported within the soil by animals, heterotrophic decomposition of the leaf material may occur anywhere in the soil profile.

Other terminology issues complicate comparison among studies. Terms such as "root turnover" and "fine root" have been defined inconsistently. Root turnover is the rate at which roots are produced or lost during a specified period (based on mass or length) divided by the average standing crop during that period. Results are typically expressed in units of g g^{-1} d^{-1} or simply d^{-1}, which is the inverse of median root lifespan. The numerator may include production, mortality or the average of the two. The denominator may include maximum, minimum or average standing crop. For root systems at steady state, production and mortality should be equal, such that the choice of the parameter for the numerator is of little importance. However, steady-state conditions are rare within a season or through the development of a stand over years (Haynes and Gower 1995, Kern et al. 2004; Pregitzer et al. 2000b). Under non-steady-state conditions production and mortality differ and the choice of denominator or even numerator used for turnover calculations will influence estimates. For example, because turnover is the inverse of median lifespan, using lifespan emphasizes the importance of mortality rate. The use of survival or proportional hazard analysis provides powerful statistical tools for testing controls of turnover rate. Evaluating lifespan using root viewing or

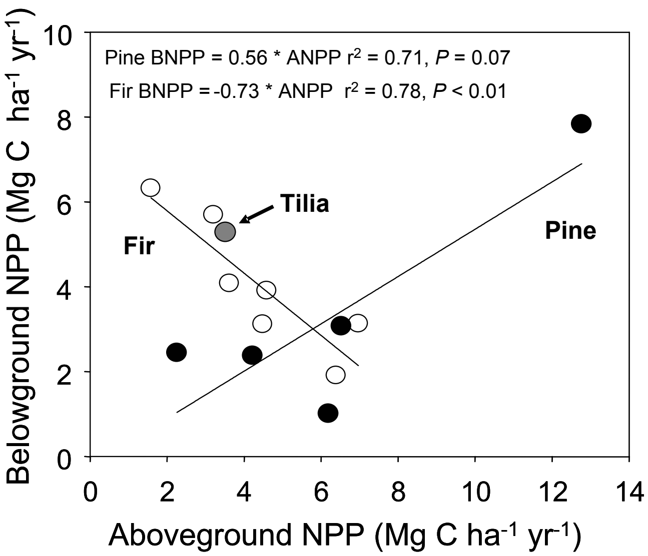

Figure 8. Data from review by Reich and Bolstad (2001) showing strong but opposing relationships between ANPP and BNPP for temperate pine and fir (true fir and Douglas-fir) species across sites. Only one point is reported for a hardwood point so no relationship is given.

isotopes methods are important techniques for determining lifespan that are free from choices of rate and standing crop.

Finally, definitions that include size classes can complicate comparisons. Fine roots are commonly distinguished based on their diameter, with definitions ranging from <1 mm to <5 mm. However, much of the perennial root system ranges between 1 and 5 mm, and ephemeral, small-diameter feeder roots increase in specific root length, nitrogen concentration, rate of root respiration, and risk of mortality from the proximal to distal end of the root system (Pregitzer, 2002, 2003). Accurately describing the range of individual root lifespan and primary function will require adopting terminology that more precisely describes the actual function of the branching root system and recognizes that root systems integrate a complex assembly of functionally distinct individuals (Pregitzer et al. 2000b, Pregitzer 2003).

Aboveground factors as predictors of BNPP

Can aboveground measures be used to predict belowground measures of BNPP? The answer depends in part on which components of BNPP are considered. Coarse root biomass often correlates highly with stem biomass (Figure 5; see above discussion; also Enquist 2002; Enquist and Niklas 2002). Our understanding of environmental controls on this ratio is also improving (Albaugh et al. 1998, Litton et al. 2003, Stape et al. 2004). These initial findings suggest that measures of aboveground stem biomass increment may adequately predict coarse root NPP (e.g. King et al. 1999a, Coleman et al.

2004). Yet, variation due to species and environmental factors may be large; some of this variation may be explained by accelerating development (Figure 4) due to treatments such as irrigation, mineral nutrients and elevated CO_2 (Gebauer et al. 1996, King et al. 1999a, McConnaughay and Coleman 1999, Coleman et al. 2004, Coleman et al. 2005), but the limited number of studies available to identify let alone quantify controls on above to belowground biomass relationships limits our ability to generalize across studies.

Relationships between aboveground measures and more complete measures of BNPP are more variable (Litton et al., *in review*). Shaver and Jonasson (2001) showed a strong correlation between BNPP and ANPP for arctic ecosystems (Figure 6). Gower et al. (2001a) also found strong correlations for boreal pine and hardwood forests (Figure 7), though the relationship was poor for boreal spruce. Gower et al. (2001a) suggested that total NPP (and BNPP by difference), could be predicted from commonly available forest inventory data. This assertion was supported by temperate forest data compiled by Reich and Bolstad (2001), showing a strong positive relationship for pine (Figure 8). Reich and Bolstad (2001) also found an inverse relationship between ANPP and BNPP for fir and Douglas-fir (Figure 8), in line with the inverse trend reported by Gower et al. (2001a) for spruce. Overall, a general relationship between ANPP and BNPP (Figure 9) is encouraging given variation in climatic conditions and soils. However, variation is substantial and meaningful generalizations about central tendencies, especially across species and biomes, will require more data.

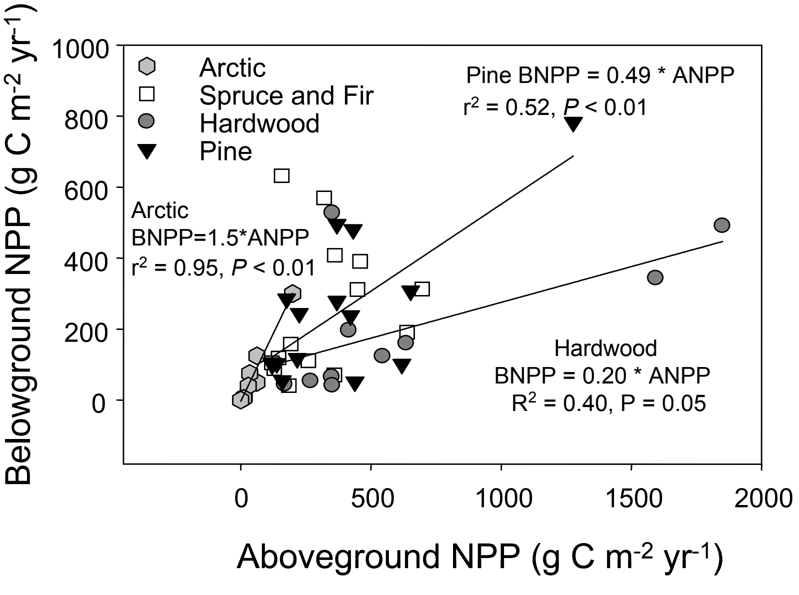

Figure 9. Global scale relationship between ANPP and BNPP with data from Figures 6 through Figure 8. The global relationships confirm regional patterns for pine and deciduous vegetation (hardwood trees plus larch, shrubs, forbs, and grasses), but fail to support patterns for spruce, fir, and Douglas-fir.

The use of aboveground factors to estimate BNPP requires that any measure integrates factors influencing both fine-root production and mortality. The response of root mortality to a range of environmental factors, along with the seasonal separation in fine-root production and mortality suggests that these processes might respond independently, and independence of two major components of BNPP would suggest that predicting the ephemeral root fractions from aboveground measures will remain difficult (Landsberg and Gower 1997; Pregitzer et al. 2000a, Pregitzer et al. 2000b).

Global change factors affecting BNPP

Global change factors that could influence BNPP include shifts in species composition with climatic change, elevated CO_2, temperature, moisture and the interactions among these factors. Data are limited, but some species have shown repeatable patterns, and experimental manipulations have yielded predicable changes in BNPP.

Species composition

Global change could affect species distribution by altering site temperature, precipitation and nitrogen deposition. Tree root systems acquire limiting resources from the soil, and changes in these resources will lead to changes in the belowground processes controlling species distribution (Norby and Jackson 2000). Both theory and paleoclimatic evidence indicate that global change will change species cover and distributions, but the interactions between species and sites with BNPP will likely be complex. Combining data from Figures 6, 7 and 8 into a single figure shows that arctic, pine and hardwood vegetation each fall on significant regression lines for aboveground-to-belowground NPP (Figure 9). In contrast, no relationship emerges for spruce and fir. There appears to be some variation among biome and species, but the two cannot be disentangled. Further, species segregate across landscapes in response to variation in site conditions, with both species and site altering BNPP. For example, arctic ecosystems appear to allocate substantially more carbon to BNPP relative to ANPP when compared with boreal and temperate pine ecosystems, but differences in species, hydrology, other site variables or methodology also could explain these patterns. In turn, pine species appear to allocate more carbon to BNPP relative to ANPP when compared with boreal and one temperate hardwood species, but again, the cause of the difference is difficult to ascertain. No apparent differences emerge across climate types for pine and hardwoods, and the variation is high. Forested wetlands tend to fall between the pine and hardwood trends (Figure 9), but these forests also exhibit considerable variation among species (Burke and Chambers, 2003) and site conditions (Finer and Laine, 1998). The absence of a clear trend for spruce, fir and Douglas-fir indicates that controls on variation in allocation patterns are still poorly understood within and across species.

Gill and Jackson (2000) considered available belowground data sets and found that fine root turnover generally increased with mean annual temperature. This pattern was consistent between various life forms and implies that warmer sites require greater production to maintain similar amounts of root biomass. In contrast to Figure 9, Gill and Jackson (2000) found no differences between temperate conifer and broadleaved tree species in either mean or temperature weighted root turnover, and that forest type in general explained little of the variation in turnover rates. Using similar data, Li et al. (2003) also found no differences for BNPP between hardwood and conifer forest types, despite differences in coarse-root standing stock.

Common garden studies present the most direct test of how species impact carbon allocation patterns in forests, and several studies have distinguished differences among evergreen conifers and deciduous hardwoods. In a planted species trial, Coleman et al. (2000) used minirhizotron methods, and estimated that fine-root production by *Pinus resinosa* was only 6% of that of *Populus* hybrid. Steele et al. (1997) used both sequential coring and minirhizotron techniques to show greater fine-root production for *Populus tremuloides* compared with *Pinus banksiana*, especially when adjusted for soil temperature.

These results agree with indirect nitrogen budget technique results, where evergreen conifers have lower annual fine-root biomass production than deciduous hardwoods across broad gradients in environment rather than in common gardens (Aber et al., 1985). However, most studies of conifers and hardwoods across wide ranges of site conditions have not found differences in fine root NPP (McClaugherty et al., 1982; Nadelhoffer and Raich, 1992). Interpreting discrepancies between comparisons of species at a single site versus across diverse sites is confounded by variation in site characteristics (and methodology). For example, pines might occupy nutrient poor sites, where high allocation to roots is required to meet water or nutritional needs, while hardwoods might occupy higher quality sites, where a greater allocation to ANPP is permitted. When pines and hardwoods are grown on the same site, as in a common garden, allocation patterns often change in response to altered soil conditions (Cannel and Dewar 1994, Giardina et al. 2003). Common garden studies are limited because patterns found between species at a single site may not represent the patterns that would be found across other sites (see Binkley and Menyailo, this volume).

Overall, new thinking is required to accurately predict climate change impacts on belowground productivity and allocation patterns in relationship to species and species interactions with climate. We suggest that greater attention to distinguishing root classes and characterizing site and stand characteristics will be particularly valuable.

Elevated CO_2: Free Air CO_2 Enrichment experiments.

Elevated CO_2 commonly increases BNPP in experiments with seedlings in growth chambers and in open-top chambers in the field (Berntson and Bazzaz 1996; Crookshanks et al. 1998; Godbold et al. 1997; King et al. 1996; Norby et al. 1992). The response of intact forest stands remains poorly known, but free air CO_2 enrichment (FACE) experiments provide some information. Several *Populus* species in the PopFACE experiment in Italy responded to three years of elevated CO_2 by increasing root production by 42 to 88% (Lukac et al. 2003). Minirhizotron observations of sweetgum forest stands during 6 years of treatment at the US Department of Energy Oak Ridge National Lab FACE facility increased root production, mortality and standing crop (Norby et al. 2002, 2004); the magnitude of these BNPP changes were large enough to account for the entire NPP response to elevated CO_2 (Figure 10). Further, elevated CO_2 shifted the partitioning of primary productivity from ANPP to BNPP, indicating that belowground resource demand increased with elevated CO_2. Tissue quality and nitrogen cycling were not reduced by elevated CO_2 relative to control plots, so the mechanism for the shift in allocation is not clear.

Similar trends have been reported for the loblolly pine FACE at Duke University in North Carolina, but the trends have not been significant. During one year of observation, fine root production increased 26%, fine root mortality increased 46%, and fine root standing crop increased 16 to 68% depending on the method of measurement (Matamala and Schlesinger 2000;

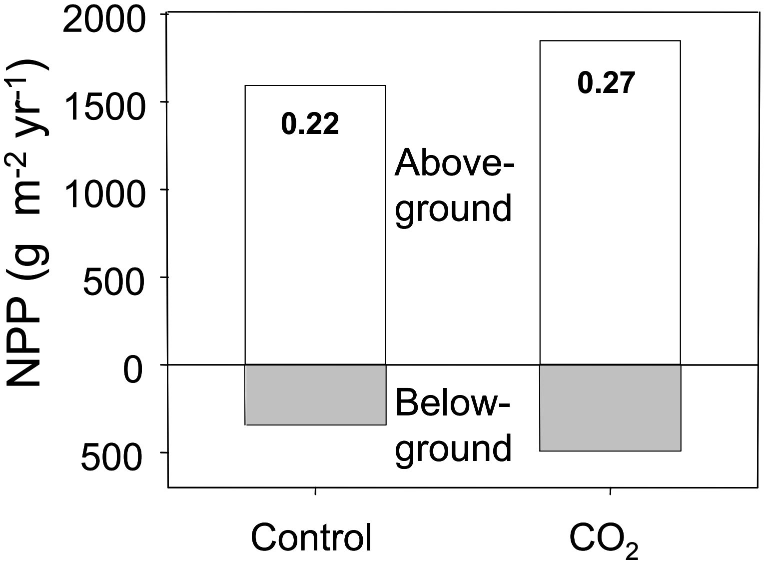

Figure 10. Data from Norby et al. 2002 showing the positive effects of elevated CO_2 on above and belowground NPP. Elevated CO_2 in this FACE experiment increased ANPP and BNPP, and also increased the BNPP to ANPP ratio (the ratio is identified in each bar).

Pritchard et al. 2001). For both US FACE studies, the effect of elevated CO_2 on fine-root turnover rate was limited, as observed in minirhizotrons and confirmed by carbon isotope depletion method in an inter-site comparison (Matamala et al. 2003).

Given numerous challenges associated with quantifying mycorrhizal fungal biomass, production and turnover, it is not surprising that there is limited information on the response of the mycorrhizal component of BNPP to elevated CO_2. Elevated CO_2 leads to an increase in mycorrhizal parameters (mostly measured as percent root colonization) of approximately 1.5 fold in field studies (Treseder, 2004). In the POPFACE study, root colonization after three years by arbuscular mycorrhizal and ectomycorrhizal fungi increased for two of three and one of three *Populus* species, respectively (Lukac et al 2003). The mycorrhizae of the hybrid *P. x euroamericana* did not respond to CO_2, despite strong responses in standing root biomass and fine root production. At the FACE site in Rhinelander, WI, mycorrhizal fungal sporocarp biomass production rates increased approximately 1.25 fold in elevated CO_2 when compared to ambient conditions, but increased 4.8 fold in elevated $CO_2 + O_3$ treatments compared to elevated O_3 (Lilleskov, unpublished), indicating a strong interaction of the effects of CO_2 and O_3 on this component of fungal production. Much more information will be needed to characterize the variety of mycorrhizal responses that alter BNPP.

Soil Temperature

Tree root growth commonly increases with soil temperature (Kaspar and Bland 1992; Lyr and Hoffmann 1967; Teskey and Hinckley 1981). Warm soil temperatures can increase fine-root production and decrease root longevity (Eissenstat and Yanai 1997; Hendrick and Pregitzer 1993; King et al. 1999b; Wan et al. 2004), with some studies showing that inter-annual variation in fine-root production relates strongly to inter-annual temperature fluctuations (Coleman et al. 2000; Tierney et al. 2003). Other studies have shown weak effects of temperature (Hendrick and Pregitzer 1997; Joslin et al. 2001). Seasonal changes in soil temperature are usually associated with seasonal changes in root growth, but this covariation confounds any temperature effect with normal seasonal phenology of plants (Pregitzer et al. 2000b). Other environmental factors such as drought, soil solution nutrient concentrations or freezing temperatures can also exert control over both production and mortality (Joslin et al. 2001; Tierney et al. 2003).

Soil Moisture – Flooding

Hydric soil conditions can cause root morphological and physiological adaptations to saturated conditions (McKevlin et al., 1998), but the influence of hydric conditions on BNPP remain largely unexplored. Trettin and

Jurgensen (2003) reviewed the state of knowledge for wetland forests, and found that BNPP in boreal bogs and fens was approximately 50% of ANPP, with that proportion declining in boreal swamps (30%) and temperate bottomland hardwoods (25%). This trend across sites may not be matched by responses to hydrologic regime within single sites. For example, studying BNPP in drained peatlands, Finer and Laine (1998) reported increased belowground allocation with increased temperature and aeration, and that the BNPP of the tree and shrub strata do not necessarily respond similarly to site conditions. Burke and Chambers (2003) found large differences among *Quercus* species responses to flooding in a southern bottomland forest, and that alternated aeration led to an increase in BNPP as trees adjusted to the variable soil conditions. Understanding BNPP dynamics in wetland soils is particularly important as they contain a disproportionate amount of the global terrestrial C (approximately 30%) and changes in the water cycle is a likely consequence of most global change scenarios (Trettin and Jurgensen, 2003).

Interactions among CO_2, forest type, and temperature

Interactions among global change factors are often the most intriguing and important responses of forests to multiple variables, highlighting the complex nature of environmental controls. In a study of two *Acer* species elevated CO_2 and temperature both increased root production and growth (Wan et al. 2004). In an earlier study, elevated CO_2 increased root biomass of *P. taeda* and *P. ponderosa*, with temperature interacting with CO_2 in *P. taeda* (King et al. 1996). Root growth may increase in response to combined elevated CO_2 and temperature, but there negative responses are also possible due to increased respiration, higher root N concentration, and altered soil microbial activity (Pendall et al. 2004). A multi-factored study of root exudation in *Robinia pseudoacacia* mesocosms found that elevated CO_2 did not influence exudation, whereas elevated temperature and additions of nitrogen stimulated exudation (Uselman et al. 1999). Overall, the lack of appropriate data from across species and for adult trees prevents any generalization about how forests will respond to multiple factors.

TOTAL BELOWGROUND CARBON ALLOCATION

Total belowground carbon allocation (TBCA) is defined as that carbon allocated belowground by plants to produce coarse and fine roots, root respiration, and root exudates and mycorrhizae (Figure 2). Belowground C allocation can be a large fraction of gross primary production (Ryan et al. 1994, 1997a; Giardina et al. 2003), sometimes exceeding aboveground net primary production (Law et al. 1999). Our understanding of the factors that control TBCA is poor, though increases in the numbers of experiments will help clarify the major role of TBCA in the C balance of terrestrial ecosystems, (Giardina et al. 2004).

Raich and Nadelhoffer (1989) originally proposed a mass-balance approach to quantify the total quantity of carbon allocated belowground by trees on an annual time step. This approach relies on mass balance to estimate TBCA with quantifiable uncertainty for all fluxes (unlike the BNPP methods described above). Plants send fixed C to roots. This C must either be respired by microbes or roots (measured as soil-surface CO_2 efflux or F_S) or stored in soil as organic matter, in the litter layer, or in living and dead roots. If C storage in soil, roots, or the litter layer does not change over the measurement period of interest, and leaching and erosion losses are negligible, then conservation of mass dictates (i.e., any soil carbon that is formed from TBCA will be offset by older carbon that is released through decomposition) that TBCA must equal F_S minus C inputs from aboveground litter (F_{AL}):

$$\mathrm{TBCA} = F_S - F_{AL} \qquad (2)$$

The utility of TBCA estimates differs from that of soil respiration in several important ways. Soil surface CO_2 efflux ('soil respiration') is an integrator of the key components of the belowground C cycle, and consequently has been viewed as an index of belowground C cycling rates. From established information on soils, roots, and organisms inhabiting soils and the rhizosphere, soil surface CO_2 efflux (F_S) can be described by the following equation:

$$F_S = F_R + F_M + F_{AL} + F_{BL} + F_{SOC} \qquad (3)$$

where F_R is the flux of CO_2 from respiring roots, F_M is the flux of CO_2 from respiring mycorrhizae, F_{AL} and F_{BL} are fluxes of CO_2 from decomposing above and belowground litter (including root and mycorrhizal exudation and turnover), and F_{SOC} is the flux of CO_2 from decomposing organic C stored in mineral soil (microbial biomass, low-quality remains and by-products of litter decomposition). F_R represents CO_2 of autotrophic origin while F_M, F_{AL}, F_{BL} and F_{SOC} represent CO_2 released by heterotrophic organisms, though F_M has been described as autotrophic (Gower et al. 2001a).

Quantifying the individual components of soil surface CO_2 efflux is challenging because belowground C processes are intimately associated with the soil matrix. Sampling for individual components is often labor intensive (e.g., root excision to estimate F_R or trenching to estimate F_{SOC}), and estimates of the components of soil surface CO_2 efflux are often limited to a snapshot or a small area. Roots, mycorrhizae and soil are intimately connected, so these studies may not accurately represent belowground processes as they would occur in undisturbed soil (Högberg et al. 2001). Finally, the belowground (roots, microbes) and aboveground (leaf and branch litterfall) components of soil surface CO_2 efflux may not respond similarly to changes in the environment (e.g., Giardina et al. 2004). Where more than one variable is changing (e.g., temperature, moisture and nutrient supply), ecosystem responses to these multiple changes may be quite complex (Figure 1). Warming may increase decomposition rates, but associated increases in nutrient mineralization rates may alter plant allocation strategies, perhaps shifting C allocation away from roots to aboveground parts, lowering soil surface CO_2 efflux. Given the potential for offsetting effects, changes in soil surface CO_2 efflux are difficult to interpret, especially with respect to how component fluxes are altered.

The TBCA approach has the advantage of non-invasive, integrative over time and space, and bounded by mass balance. As conceived by Raich and Nadelhoffer (1989), the approach relies only on direct measures of soil surface CO_2 efflux and litterfall. Using Equation 2 and assuming that leaching losses of C are negligible, and that soil, forest floor and root C storage were in steady state, Raich and Nadelhoffer (1989) estimated TBCA for a wide variety of mature forests from published measurements of soil respiration and litterfall. They found that aboveground litter (F_{AL}) contributed 23% of soil surface CO_2 efflux at low efflux rates (400 g C m^{-2} yr^{-1}) to 31% of soil surface CO_2 efflux at high efflux rates (1500 g C m^{-2} yr^{-1}). By difference, the belowground sources of CO_2 (i.e., TBCA which equals [$F_R + F_M + F_{BL} + F_{SOC}$] when soil C is in steady state) contributed 69% to 77% of soil surface CO_2 efflux.

In an effort to address uncertainties resulting from steady state assumptions, Giardina and Ryan (2002) outlined a similar approach that accounts for changes in belowground and forest floor C storage:

$$\text{TBCA} = F_S - F_{AL} + \Delta\,[C_S + C_R + C_L] \tag{4}$$

where C_S = carbon content of mineral soil, C_R = carbon content of root (coarse + fine) biomass, and C_L = carbon content of the litter layer. Increases in C storage will decrease soil respiration, while decreases in storage will increase soil respiration. This approach to estimating TBCA still requires that losses of C to leaching or erosion are negligible, but this will be true for most forests on level topography (Giardina and Ryan 2002).

An important finding of Giardina and Ryan (2002) was that litterfall was a poor predictor of TBCA across their treatments. More importantly, they found that changes in soil or forest floor carbon storage, while dynamic, contributed little to the TBCA budget in a young, fast growing plantation forest. They concluded that non-steady state conditions may not be a concern as long as both soil respiration and litterfall are measured; the failure of litterfall to predict TBCA did not relate to violation of steady state assumptions, but to the dynamic variation in the relationship between TBCA and litterfall.

The TBCA approach has limitations. Estimates of TBCA cannot be used to quantify BNPP, though TBCA can anchor BNPP estimates derived from other methods within a total belowground budget (Ryan et al. 1996, McDowell et al. 2001). Further, TBCA estimates as with all ecological measures rely on accurate estimates of soil respiration and litterfall, and in Equation 3, soil carbon, forest floor carbon and coarse roots. The latter measures are straight forward (Giardina and Ryan 2002), but soil respiration estimates can be sensitive (+/- 20%) to choice of equipment, frequency of measurement, and other factors.

The effects of species and temperature on TBCA

Here we summarize available TBCA data and examine relationships with mean annual temperature (MAT) and species. We also estimate TBCA for sites where soil CO_2 efflux and aboveground litterfall data are available to examine how elevated CO_2 affects TBCA. Finally, building on previously described relationship between TBCA and litterfall (Raich and Nadelhoffer 1989, Davidson et al. 2002), we examine large-scale patterns of TBCA to ANPP with the goal of understanding whether ANPP predicts TBCA.

TBCA is sensitive to changes in tree age (Giardina and Ryan 2002) and site fertility (Ryan et al. 1996). Tree age exerts a large influence on TBCA, with TBCA declining by as much as 30% from maximum rates at canopy closure (Smith and Resh 1999, Giardina and Ryan 2002). Reported responses of TBCA to fertilization also have been large. In plantations of *P. radiata* and *Eucalyptus saligna*, fertilization reduced TBCA by 28% and 12%, respectively. Using the TBCA approach in conjunction with stable C isotope measurements of SOC, Giardina et al. (2004) examined the belowground fate of TBCA in a *Eucalyptus* plantation, including the efficiency with which TBCA is retained in soil as new soil C, and the fraction returned to the atmosphere as soil surface CO_2 efflux (Figure 11). Increased nutrient supply shifted the allocation of carbon from fine roots and mycorrhizae to coarse roots and aboveground leaf and wood production, but did not alter the efficiency with which TBCA was converted into new soil carbon.

It would be difficult to extrapolate responses from stand age or fertilization studies to scenarios of global warming or species change. However, there are no experimental studies of TBCA response to changes in these variables. We addressed this information gap by assembling TBCA estimates for widely ranging forests, and examining how much variation across sites could be ascribed to mean annual temperature or species (Figure 12). TBCA at a site with an MAT of $20^{O}C$ was on average 1.8 times greater that TBCA at a site with an MAT of $10^{O}C$, yielding a Q_{10} of 1.8 for TBCA across sites. The relationship was robust ($R^2 = 0.47$; $P < 0.01$) considering the wide diversity of soil and vegetation types, methods and studies.

As with any natural gradient study, confounding factors may complicate interpretation of results. For example, temperature may co-vary with soil development, and as discussed above for BNPP, TBCA can change in response to differences in soil characteristics (Giardina et al. 2004). Efforts to

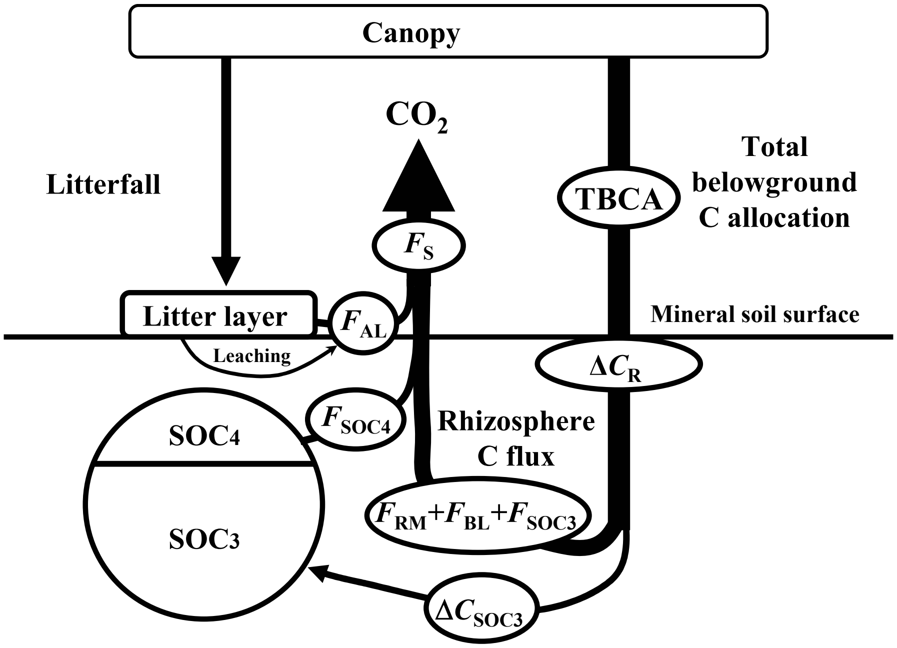

Figure 11. Conceptual model for how TBCA and stable isotopes can be used to examine the belowground fate of TBCA and the component sources of soil respiration (adapted from Giardina et al. 2004).

isolate the effects of species or climate on belowground processes may be compromised by legacy effects from earlier vegetation or by sampling periods that are too short to capture lags associated with yearly variations in above and belowground processes (Davidson et al. 2002). The lack of species effects on TBCA conflicts somewhat with data presented in Figure 9 but is consistent with several studies described above in the BNPP section. Further, across large scales, TBCA to ANPP ratios average about 1.5, indicating that TBCA generally represents a larger sink for GPP than does ANPP. While a TBCA to ANPP ratio of 1.5 represents a general central tendency of the data set, the variation in ANPP to MAT and TBCA to MAT relationships translates into some uncertainty about this tendency. When the 95% confidence intervals for both relationships are considered, a 95% confidence interval for a TBCA to ANPP ratio at $10^{O}C$ of 1.56 would include 1.33 to 1.82. Confidence would be lower at cooler or warmer ends of the relationship.

The fraction of TBCA that is BNPP is poorly quantified, but may be critical to correctly modeling ecosystem carbon cycling and the belowground carbon cycle. Because little data are available, it has been largely assumed that approximately 50% of TBCA is BNPP (Law et al. 1999, Giardina et al. 2003). It is noteworthy that comparing Figures 9 and 12, TBCA varied from 400 to 1500 g C m^{-2} yr^{-1}, while over a similar ANPP range, BNPP varied from 150 to 800 g C m^{-2} yr^{-1}, indicating that, despite high variance, BNPP is on average about 50% of TBCA.

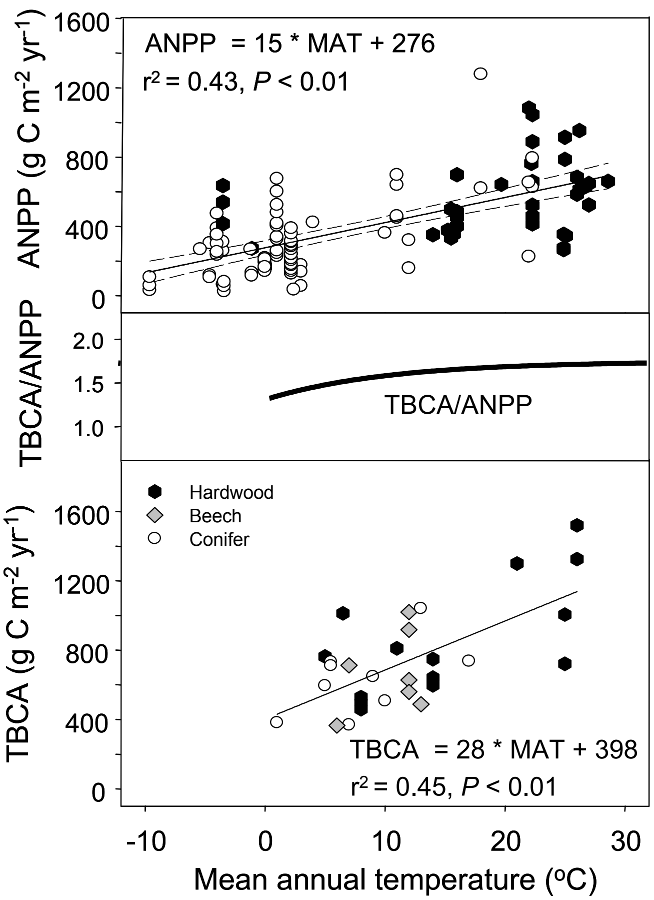

Figure 12. Global scale relationship between mean annual temperature (MAT) and ANPP (Top Panel), between MAT and TBCA (Bottom Panel) and the ratio of TBCA to ANPP derived from the equations describing the two relationships. Points are unfertilized forests across a global scale gradient in MAT. Data are from Ryan et al. (1996), Clark et al. (2001b), Gower et al. (2001b), McDowell et al. (2001), Reich and Bolstad (2001), Davidson et al. (2002), Giardina et al. (2003), and Litton et al. (2004). ANPP data were screened to include studies that reported at least wood and leaf NPP. If reported, branch NPP was included. We did not include forests growing on new or very young soils.

The effect of elevated CO_2 on TBCA

No data are available on how TBCA responds to changes in atmospheric CO_2. Several FACE studies have reported an increase in soil respiration under elevated CO_2, and these increases have been ascribed to increased fine root NPP (Norby et al. 2004), and increased exudation or litterfall (King et al. 2004). We modified the approach outlined by Giardina and Ryan (2002) to examine how FACE treatments altered TBCA at the Rhinelander and Oak Ridge FACE sites. We used these data to estimate TBCA as soil respired C *plus* coarse root increment C *minus* litterfall C. TBCA was 10 to 15% higher

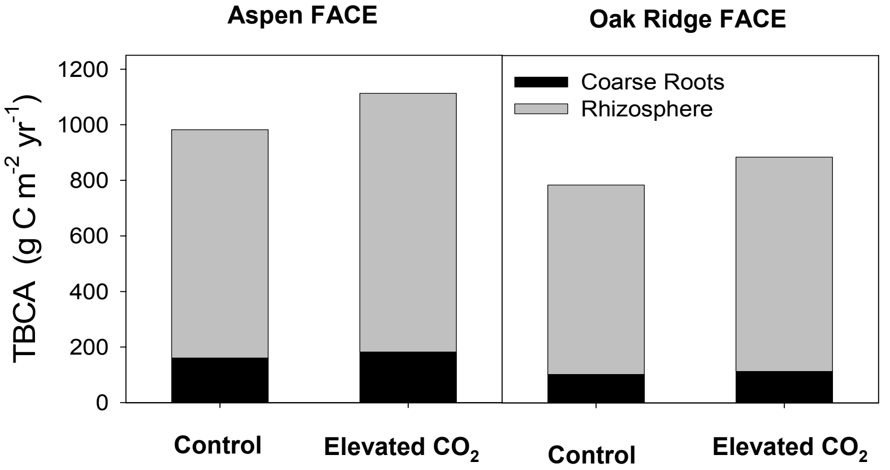

Figure 13. The effects of elevated CO_2 on mixed aspen-birch stands in Rhinelander Wisconsin (Left panel) and sweetgum (*Liquidambar styraciflua*) in Oak Ridge, Tennessee. Data for calculations of TBCA are from King et al. (2004), Norby et al. (2002), Norby et al. (2004), and unpublished data of the authors.

for both sites with elevated CO_2 (Figure 13). The increase in fine root NPP reported by Norby et al. (2002) for elevated CO_2 was similar to the increase in TBCA for the same plots, indicating that most of the increase in TBCA was allocated to fine root production.

CONCLUSIONS

A key frontier in global change science involves understanding the controls on belowground carbon allocation, processing and retention. Confidence in measurement techniques is constrained by our inability to directly measure the carbon flows of interest: how can we be sure that our data accurately represent a process of interest? Measurements of TBCA come closest to direct measurements, but this aggregated measure provides the least insight on the details of all the processes that comprise BCA. The perceived and actual sensitivity of the flux measurements to changes in the environment, and the sensitivity of these measures to artifacts, vary widely across methods. With these warnings in mind, we suggest three important generalizations:

- Changes in BCA will vary in concert with changes in aboveground productivity, because overall, BCA and ANPP vary in concert. While the fraction of GPP for each may change under global change, BCA and ANPP in general are closely linked.
- Greater integration of available data across biomes and species is needed to test what appear to be reasonable generalizations within a biome or species.

- The complete suite of BCA components needs to be measured for more forests, with explicitly defined populations (soil types, species, or gradients where both vary). These C budgets also need to be explicitly connected to experimental manipulations of resources and species within sites, to provide a gauge of the value of cross-site comparisons for predicting within-site responses.

Overall, BNPP studies have greatly advanced our understanding of how forest ecosystems function and will respond to global change. Studies that now combine isotopes of carbon with BNPP observations and mass balance approaches are building on these ground breaking BNPP studies. Future studies that combine TBCA, BNPP and isotope-based methods will lead to greater insights into how the belowground carbon cycle will respond to a changing world. A myriad of important questions remain unanswered about belowground carbon cycling (Table 1). It is our challenge to apply these new methods while continuing to develop new techniques for assessing belowground processes. We also need to prioritize these questions, as funding resources are limited and the potential combinations of conditions and factors are enormous. We feel that temperature and moisture gradients and manipulations of species and nutrients will serve as the basis to efficiently address the complex interactions of species, site, and global change factors.

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Table 1. Our list of pressing questions in the science of belowground carbon cycling.

1. How does BCA vary by species?
2. Are stand characteristics such as species and stand age the ultimate drivers of BCA?
3. Alternatively, are moisture and temperature the ultimate drivers of BCA?
4. How will elevated CO_2, climate, and nutrient deposition interact to impact BCA?
5. How will the impacts of CO_2, climate and nutrients interact with species and site?
6. What is the efficiency with which BCA is converted into new soil carbon?
7. Does conversion efficiency vary by species, site or climate?
8. In a warmer world, will increases in BCA offset reductions in the conversion efficiency of BCA into soil organic matter, maintaining historic rates of formation of soil organic matter?

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