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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 data for any predictable variation in BCA due to changes in species composition, mean annual temperature, or elevated CO₂ in existing Free Air CO₂ 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 CO₂ 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 by terrestrial vegetation through photosynthesis, with most this gross carbon flux occurring in ecosystems with trees (Grace and Rayment 2000) On an annual time step,. 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). However, 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. Specifically, efforts to test these 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 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 rates of processes depending on factors such as species composition, nutrient and water supplies, and temperature. These factors influence BCA, and will determine the overall response to global-scale changes. 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 grasses in agricultural systems, perennial grasses and forbs in managed pastures, and long-lived trees in forest plantations.

Agricultural land use changes soil, most of which are negative with regards to nutrient availability and 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 these 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 CO₂ 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 such as ozone (O₃) inhibit productivity (Reich 1983, Reich and Amundson 1985, King et al. 2001, Karnosky et al. 2003). Elevated CO₂ and other greenhouse gases also indirectly affect plants and ecosystems by changing global climate. Taken together, these direct and indirect effects on plants will likely impact BCA but above to belowground links (Figure 1) remain poorly quantified.

Rising greenhouse gases are likely to warm the biosphere, and micro to meso-scale studies often show strong temperature and moisture effects on plants and microbes (Uselman et al. 2000, 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 CO₂ 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 CO₂ 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 CO₂ efflux within days of eliminating phloem transport of carbon to roots and mycorrhizae through girdling. Giardina et al. (2004) used ¹³C isotopic methods to calculate that 90% of soil surface CO₂ 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 CO₂ enrichment (FACE) experiment in Rhinelander, Wisconsin is especially important because three tree communities have been fumigated, singly and in combination, with gases that stimulate (CO₂) or reduce (O₃) 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 span the full spectrum of BCA studies. 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 the 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 on BCA of species, temperature and elevated CO₂. High rates of N deposition almost certain impact on 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 portions. 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 coarse root to above ground biomass relationships that relates to stand age (Figure 5; from Ovington 1957) can perhaps 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: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 5 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 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 appear to 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 after three years of treatment, and root:shoot allometry was constant despite a doubling of leaf area index and biomass in the fertilization and irrigation treatments (Figure 4). 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 4). Given that larger trees tend to have larger root systems, a key issue is whether the relationship between root and shoot biomass has Y-intercept of 0 (as in the loblolly pine case in Figure 4), or not (as in the *Eucalyptus* case study in Figure 4). 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 5) could not be ruled out.

Given the importance of large roots for supporting tall trees, King et al. (1999a) suggested that for a given site and species, coarse-root standing stock to aboveground biomass allometry may be less sensitive to changes in environmental conditions than root to shoot ratios of forbs (McConnaughay and Coleman 1999), or young seedlings (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 CO₂ or global climate, though changes in moisture may alter allometry (Stape et al. 2004). This assertion is supported by King et al. (1999) where above to belowground allometry of *P. taeda* and

P. ponderosa exposed to treatments of elevated temperature, CO₂ and nutrients, showed little effect of the treatments.

Even if changes in environmental conditions have little effect on root:shoot allometry, changes may still be driven by alteration of site fertility, stand age, and 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 CO₂ or temperature may accelerate maturation and age-related changes coarse-root standing stock to aboveground allometry (Figure 5), 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; trees 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:

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

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 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 (Usselman et al. 2000).

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 (Pobilcover 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 precision of measuring very small roots, especially in the surface few mm of soil (Vos and Groenwold 1987), installation of viewing windows may disturb root growth (Coleman et al. 2000; Joslin and Wolfe 1999), and scaling from 2-dimensional area to the mass in a volume of soil is challenging.

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 estimates 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 longevity. 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 sampling approach and quantification challenge are: 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 the very 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, quantifying BNPP is still extremely

difficult. First, girding or more destructive approaches cannot separate root respiration (autotrophic) from exudation (heterotrophic) derived CO₂. Second, CO₂ 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 saprophytic), 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 heterotrophic and autotrophic respiration, the heterotrophic sources of soil respiration are themselves very complex and difficult 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 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 4; 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 5) due to treatments such as irrigation, mineral nutrients and elevated CO₂ (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 (Figures 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.

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 CO₂, temperature, moisture and the interactions among these factors. Data are limited, but some species have shown repeatable patterns, and experimental manipulations have yielded predictable 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 line 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 environments 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 confounded by variation 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 temperature. We suggest that greater attention to distinguishing root classes and characterizing site and stand characteristics will be particularly valuable.

Elevated CO₂: Free Air CO₂ Exposure experiments.

Elevated CO₂ 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 CO₂ exposure (FACE) experiments provide some information. Several *Populus* species in the PopFACE experiment in Italy responded to three years of elevated CO₂ 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 CO₂ (Figure 10). Further, elevated CO₂ shifted the partitioning of primary productivity from ANPP to BNPP, indicating that belowground resource demand increased with elevated CO₂. Tissue quality and nitrogen cycling were not reduced by elevated CO₂ 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; Pritchard et al. 2001). For both US FACE studies, the effect of elevated CO₂ 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 CO₂. Elevated CO₂ 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 CO₂, 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 CO₂ when compared to ambient conditions, but increased 4.8 fold in elevated CO₂ + O₃ treatments compared to elevated O₃ (Lilleskov, unpublished), indicating a strong interaction of the effects of CO₂ and O₃ 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 cause distinct 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 temperature 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 CO₂, 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 CO₂ and temperature both increased root production and growth (Wan et al. 2004). In an earlier study, elevated CO₂ increased root biomass of *P. taeda* and *P. ponderosa*, with temperature interacting with CO₂ in *P. taeda* (King et al. 1996). Root growth may increase in response to combined elevated CO₂ 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 CO₂ 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 this 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 CO₂ 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 soil respiration must equal C inputs:

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

The utility of TBCA estimates differs from that of soil respiration in several important ways. Soil surface CO₂ 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 CO₂ efflux (F_S) can be described by the following equation:

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

where F_R is the flux of CO₂ from respiring roots, F_M is the flux of CO₂ from respiring mycorrhizae, F_{AL} and F_{BL} are fluxes of CO₂ from decomposing above and belowground litter (including root and mycorrhizal exudation and turnover), and F_{SOC} is the flux of CO₂ from decomposing organic C stored in mineral soil (microbial biomass, low-quality remains and by-products of litter decomposition). F_R represents CO₂ of autotrophic origin while F_M , F_{AL} , F_{BL} and F_{SOC} represent CO₂ released by heterotrophic organisms, though F_M has been described as autotrophic (Gower et al. 2001a).

Quantifying the individual components of soil surface CO₂ 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 CO₂ 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 CO₂ 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 CO₂ efflux. Given the potential for offsetting effects, changes in soil surface CO₂ 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 CO₂ 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 CO₂ efflux at low efflux rates (400 g C m⁻² yr⁻¹) to 31% of soil surface CO₂ efflux at high efflux rates (1500 g C m⁻² yr⁻¹). By difference, the belowground sources of CO₂ (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 CO₂ efflux.

In an effort to limit 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:

$$TBCA = F_S - F_A + \Delta [C_S + C_R + C_L] \quad (4)$$

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. First, 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 later measures are straight forward (Giardina and Ryan 2002), but soil respiration estimates can be sensitive (+/- 20%) to choice of equipment, frequency of measurement, soil moisture and temperature, 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 CO₂ efflux and aboveground litterfall are available to examine the effects of elevated CO₂ on TBCA. Finally, building on the relationship between TBCA and litterfall described by Raich and Nadelhoffer (1989) and Davidson et al. (2002), we examine large scale patterns of TBCA to ANPP with the goal of understanding whether TBCA can be predicted from ANPP.

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 CO₂ 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°C was on average 1.8 times greater than TBCA at a site with an MAT of 10°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 isolate the effects of species or climate on belowground processes may be compromised by legacy effects from earlier vegetation or 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°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.

The effect of elevated CO₂ on TBCA

No data are available on how TBCA responds to changes in atmospheric CO₂. Several FACE studies have reported an increase in soil respiration under elevated CO₂, 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 for both sites with elevated CO₂ (Figure 13). The increase in fine root NPP reported by Norby et al. (2004) for elevated CO₂ was similar to the increase in TBCA for the same plots, indicating that most of increase in TBCA was allocated to fine root production.

Conclusions

A key frontier in global change science involves our limited understanding of the controls on belowground carbon allocation and cycling. Confidence in measurement techniques is constrained 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 can serve as the basis to efficiently address the complex interactions of species, site, and global change factors.

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

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1. How does BCA vary by species?
 2. Are moisture and temperature the ultimate drivers of BCA?
 3. Alternatively, are stand characteristics such as species and stand age the ultimate drivers of BCA?
 4. How will CO₂, nutrient deposition and climate interact to impact BCA?
 5. How will these the impacts of CO₂ and climate 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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Literature Cited

- Adams, M.A., P. Ineson, D. Binkley, G. Cadisch, N. Tokuchi, M. Scholes, K. Hicks, and M. Chadwick. 2004. Soil functional responses to excess N inputs at global scales. *Ambio*, *in press*.
- Albaugh, T. J., H. L. Allen, P. M. Dougherty, L. W. Kress, and J. S. King. 1998. Leaf area and above- and belowground growth responses of loblolly pine to nutrient and water additions. *Forest Science* **44**:317-328.
- Allison, P.D. 1995. Survival analysis using SAS: A practical guide. SAS Institute Inc., Cary, NC, USA. 292 p.
- Bashkin, M. A., and D. Binkley. 1998. Changes in soil carbon following afforestation in Hawaii. *Ecology* **79**:828-833.
- Berntson, G.M. and F.A. Bazzaz 1996. The allometry of root production and loss in seedlings of *Acer rubrum* (*Aceraceae*) and *Betula papyrifera* (*Betulaceae*): Implications for root dynamics in elevated CO₂. *American Journal of Botany*. **83**:608-616.
- Bolte A, Rahmann T, Kuhr M, Pogoda P, Murach D and v. Gadow, K. 2004. Relationships between tree dimension and coarse root biomass in mixed stands of European beech (*Fagus sylvatica* L.) and Norway spruce (*Picea abies* [L.] Karst.) *Plant and Soil* **264**:1-11.

- Bond-Lamberty, B. W. Chuankuan, S.T. Gower. 2004. A global relationship between the heterotrophic and autotrophic components of soil respiration? *Global Change Biology* 10: 1756-1766.
- Bonfante-Fasolo, P. 1986. Anatomy and morphology of VA mycorrhizae, in: Powell, C., Bagyaraj, D. (Eds.), *VA Mycorrhiza*. CRC Press, Boca Raton, FL, pp.2-33.
- Burke, M.K. and J. L. Chambers. 2003. Root dynamics in bottomland hardwood forests of the southeastern United States Coastal Plain. *Plant and Soil* 250:141-153.
- Burton, A.J., and K.S. Pregitzer. 2003. Field measurements of root respiration indicate little to no seasonal temperature acclimation for sugar maple and red pine. *Tree Physiol.* 23:273-280.
- Burton, A.J., K.S. Pregitzer, R.W. Ruess, R.L. Hendrick, and M.F. Allen. 2002. Root respiration in North American forests: effects of nitrogen concentration and temperature across biomes. *Oecologia* 131:559-568.
- Binkley, D., and S. C. Resh. 1999. Rapid changes in soils following *Eucalyptus* afforestation in Hawaii. *Soil Science Society of America Journal* 63:222-225.
- Caldwell MM, Virginia RA. 1991. Root systems. Pages 367-398 in Pearcy RW, Ehleringer J, Mooney HA, Rundel PW, editors. *Plant physiological ecology: field methods and instrumentation*. Chapman and Hall, London.
- Cannell MGR, Dewar RC. 1994. Carbon allocation in trees: a review of concepts for modeling. *Advances in Ecological Research* 25: 59-104.
- Clark DA, Brown S, Kicklighter DW, Chambers JQ, Thomlinson JR, Ni J, Holland EA. 2001a. Net primary production in tropical forests: an evaluation and synthesis of existing field data. *Ecological Applications* 11:371-384.
- Clark, D. A., S. Brown, D. W. Kicklighter, J. Q. Chambers, J. R. Thomlinson, J. Ni, and E. A. Holland. 2001b. NPP Tropical Forest: Consistent Worldwide Site Estimates, 1967-1999. Data set. Available on-line [<http://www.daac.ornl.gov>] from the Oak Ridge National Laboratory Distributed Active Archive Center, Oak Ridge, Tennessee, U.S.A.
- Coleman, M.D., R.E. Dickson and J.G. Isebrands 2000. Contrasting fine-root production, survival and soil CO₂ efflux in pine and poplar plantations. *Plant and Soil*. 225:129-139.
- Coleman, M.D., A.L. Friend and C.C. Kern 2004. Carbon Allocation and Nitrogen Acquisition in a Developing *Populus deltoides* Plantation. *Tree Physiology*. in press.
- Coyle, D. and M. Coleman. *In press*. Forest production responses to fertilization and irrigation are not explained by shifts in allocation. *Forest Ecology and Management*.
- Crookshanks, M., G. Taylor and M. Broadmeadow 1998. Elevated CO₂ and tree root growth: contrasting responses in *Fraxinus excelsior*, *Quercus petraea* and *Pinus sylvestris*. *New Phytologist*. 138:241-250.
- Davidson EA, Savage K, Bolstad P, Clark DA, Curtis PS, Ellsworth DS, Hanson PJ, Law BE, Luo Y, Pregitzer KS, Randolph JC, Zak D. 2002. Belowground carbon allocation in forests estimated from litterfall and IRGA-based soil respiration measurements. *Agricultural and Forest Meteorology* 113:39-51
- Dickson R.E., J.G. Isebrands. 1993. Carbon allocation terminology: should it be more rational? *Bulletin of Ecological Society of America* 74:175-177.

- Eissenstat, D.M., C.E. Wells, R.D. Yanai and J.L. Whitbeck 2000. Building roots in a changing environment: implications for root longevity. *New Phytologist*. 147:33-42.
- Eissenstat, D.M. and R.D. Yanai. 1997. The ecology of root lifespan. *Advances in Ecological Research*. 27:1-60.
- Enquist, B.J. 2002. Universal scaling in tree and vascular plant allometry: Toward a general quantitative theory linking plant form and function from cells to ecosystems. *Tree Physiology*. 22:1045-1064.
- Enquist, B.J. and K.J. Niklas 2002. Global allocation rules for patterns of biomass partitioning in seed plants. *Science*. 295:1517-1520.
- Finer, L. and J. Laine. 1998. Root dynamics at drained peatland sites of different fertility in southern Finland. *Plant and Soil* 201:27-36.
- Fitter AH, Self GK, Brown TK, Bogie DS, Graves JD, Benham D, Ineson P (1999) Root production and turnover in an upland grassland subjected to artificial soil warming respond to radiation flux and nutrients, not temperature. *Oecologia*, **120**, 575-581.
- Fogel, R. and G. Hunt 1983. Contribution of mycorrhizae and soil fungi to nutrient cycling in a Douglas-fir ecosystem. *Canadian Journal of Forest Research*. 13:219-232.
- Gaudinski, J.B., S.E. Trumbore, E.A. Davidson, A.C. Cook, D. Markewitz and D.D. Richter 2001. The age of fine-root carbon in three forests of the eastern United States measured by radiocarbon. *Oecologia*. 129:420-429.
- Gebauer, R.L.E., J.F. Reynolds and B.R. Strain 1996. Allometric relations and growth in *Pinus taeda*: the effect of elevated CO₂ and changing N availability. *New Phytologist*. 134:85-93.
- Giardina, C., and C. Rhoades. 2001. Clear cutting and burning affect nitrogen supply, phosphorus fractions and seedling growth in soils from a Wyoming lodgepole pine forest. *Forest Ecology and Management* **140**:19-28.
- Giardina CP, Ryan MG (2000a) Evidence that decomposition rates of organic carbon in mineral soil do not vary with temperature. *Nature*, **404**, 858-861.
- Giardina, C., and M. Ryan. 2002. Soil surface CO₂ efflux, litterfall, and total belowground carbon allocation in a fast growing *Eucalyptus* plantation. *Ecosystems* **5**: 487-499.
- Giardina CP, *et al.* (2003) Primary production and carbon allocation in relation to nutrient supply in an experimental tropical forest. *Global Change Biol* **9**: 1438-1450.
- Giardina, C., D. Binkley, M. Ryan, and J. Fownes. 2004. Belowground carbon cycling in a humid tropical forest decreases with fertilization. *Oecologia* **139**: 545-550.
- Gill, R.A. and R.B. Jackson 2000. Global patterns of root turnover for terrestrial ecosystems. *New Phytologist*. 147:13-31.
- Godbold, D.L., G.M. Berntson and F.A. Bazzaz 1997. Growth and mycorrhizal colonization of three North American tree species under elevated atmospheric CO₂. *New Phytologist*. 137:433-440.
- Gower, S.T., O. Krankina, R.J. Olson, M. Apps, S. Linder and C. Wang 2001a. Net primary production and carbon allocation patterns of boreal forest ecosystems. *Ecological Applications*. 11:1395-1411.
- Gower, S. T., O. Krankina, R. J. Olson, M. Apps, S. Linder, and C. Wang. 2001b. NPP Boreal Forest: Consistent Worldwide Site Estimates, 1977-1994. Data set.

- Available on-line [<http://www.daac.ornl.gov>] from the Oak Ridge National Laboratory Distributed Active Archive Center, Oak Ridge, Tennessee, U.S.A.
- Grace J, and Rayment M (2000) Respiration in the balance. *Nature*, **404**, 819-820.
- Grandmougin-Ferjani A, Dalpe Y, Hartmann MA, Laruelle F, Sancholle M (1999) Sterol distribution in arbuscular mycorrhizal fungi. *Phytochemistry* **50**:1027-1031
- Grier, C.C., K.A. Vogt, M.R. Keyes and R.L. Edmonds 1981. Biomass distribution and above- and below-ground production in young and mature *Abies amabilis* zone ecosystems of the Washington Cascades. *Canadian Journal of Forest Research*. **11**:155-167.
- Haynes, B.E. and S.T. Gower 1995. Belowground carbon allocation in unfertilized and fertilized red pine plantations in northern Wisconsin. *Tree Physiology*. **15**:317-325.
- Hendrick, R.L. and K.S. Pregitzer 1992. The demography of fine roots in a northern hardwood forest. *Ecology*. **73**:1094-1104.
- Hendrick, R.L. and K.S. Pregitzer 1993. Patterns of fine root mortality in two sugar maple forests. *Nature*. **361**:59-61.
- Hendrick, R.L. and K.S. Pregitzer 1996. Temporal and depth-related patterns of fine root dynamics in northern hardwood forests. *Journal Of Ecology*. **84**:167-176.
- Hendrick, R.L. and K.S. Pregitzer 1997. The relationship between fine root demography and the soil environment in northern hardwood forests. *Ecoscience*. **4**:99-105.
- Högberg, P. *et al.* 2001. Large-scale forest girdling shows that current photosynthesis drives soil respiration. *Nature* **411**, 789-792.
- Holland EA, Neff JC, Townsend AR, McKeown B. 2000. Uncertainties in the temperature sensitivity of decomposition in tropical and subtropical ecosystems: Implications for models. *Global Biogeochemical Cycles*, **14**, 1137-1151.
- Holland, E. A. *et al.* 1997. Variation in the predicted spatial distribution of atmospheric nitrogen deposition and their impact on terrestrial carbon uptake. *Journal of Geophysical Research* **102**:15849-15866.
- Horwath, W.R., K.S. Pregitzer, E.A. Paul. ¹⁴C allocation in tree soil systems. *Tree Physiology* **14**: 1163-1176.
- Jackson, RB, JL Banner, EG Jobbágy, WT Pockman, DH Wall. 2002. Ecosystem carbon loss with woody plant invasion of grasslands. *Nature* **418**:623-626
- Janssens IA, *et al.* (2001) Productivity overshadows temperature in determining soil and ecosystem respiration across European forests. *Global Change Biology*, **7**, 269-278.
- Johnson, M.G., D.T. Tingey, D.L. Phillips and M.J. Storm 2001. Advancing fine root research with minirhizotrons. *Environmental and Experimental Botany*. **45**:263; 289.
- Joslin, J.D. and M.H. Wolfe 1999. Disturbances during minirhizotron installation can affect observation data. *Soil Science Society of America Journal*. **63**:218-221.
- Joslin, J.D., M.H. Wolfe and P.J. Hanson 2001. Factors controlling the timing of root elongation intensity in a mature upland oak stand. *Plant and Soil*. **228**:201; 212.
- Karnosky, D.F., *et al.* 2003. Tropospheric O₃ moderates responses of temperate hardwood forests to elevated CO₂: a synthesis of molecular to ecosystem results from the Aspen FACE project. *Functional Ecology* **17**, 289-304.
- Kaspar, T.C. and W.L. Bland 1992. Soil-Temperature and Root-Growth. *Soil Science*. **154**:290-299.

- Kern, C.C., A.L. Friend, J.M.-F. Johnson and M.D. Coleman 2004. Fine-root dynamics in a developing *Populus deltoides* plantation. *Tree Physiology*. 24:651-660.
- Keyes, M.R. and C.C. Grier 1981. Above- and below-ground net production in 40-year-old Douglas-fir stands on low and high productivity sites. *Canadian Journal of Forest Research*. 11:599-605.
- King, J.S., T.J. Albaugh, H.L. Allen and L.W. Kress 1999a. Stand-level allometry in *Pinus taeda* as affected by irrigation and fertilization. *Tree Physiology*. 19:769-778.
- King, J.S., K.S. Pregitzer and D.R. Zak 1999b. Clonal variation in above- and below-ground growth responses of *Populus tremuloides* Michaux: Influence of soil warming and nutrient availability. *Plant and Soil*. 217:119-130.
- King, J.S. *et al.* 2001. Fine-root biomass and fluxes of soil carbon in young stands of paper birch and trembling aspen as affected by elevated atmospheric CO₂ and tropospheric O₃. *Oecologia* 128, 237-250.
- King, J.S., P.J. Hanson, E. Bernhardt, P. DeAngelis, R.J. Norby and K.S. Pregitzer 2004. A multiyear synthesis of soil respiration responses to elevated atmospheric CO₂ from four forest FACE experiments. *Global Change Biology*. 10:1027-1042.
- King, J.S., R.B. Thomas and B.R. Strain 1996. Growth and carbon accumulation in root systems of *Pinus taeda* and *Pinus ponderosa* seedlings as affected by varying CO₂, temperature and nitrogen. *Tree Physiology*. 16:635-642.
- Landsberg, J.J. and S.T. Gower 1997. Applications of physiological ecology to forest management. Academic Press, New York. 354 p.
- Law BE, Ryan MG, Anthoni PM. 1999. Seasonal and annual respiration of a ponderosa pine ecosystem. *Global Change Biology* 5:169-182.
- Li, Z., W.A. Kurz, M.J. Apps and S.J. Beukema 2003. Belowground biomass dynamics in the Carbon Budget Model of the Canadian Forest Sector: recent improvements and implications for the estimation of NPP and NEP. *Canadian Journal of Forest Research-Revue Canadienne De Recherche Forestiere*. 33:126-136.
- Lilleskov EA, Fahey TJ, Lovett GM (2001) Ectomycorrhizal fungal aboveground community change over an atmospheric nitrogen deposition gradient. *Ecological Applications* 11: 397-410.
- Lilleskov EA, Fahey TJ, Horton TR, Lovett GM (2002) Belowground ectomycorrhizal fungal community change over a nitrogen deposition gradient in Alaska. *Ecology* 83(1): 104-115.
- Litton CM, MG Ryan, and J. Raich. *In Review*. Carbon allocation in forest ecosystems. *Oecologia*.
- Litton CM, MG Ryan, DB Tinker, and DH Knight. 2003. Below- and aboveground biomass in young post-fire lodgepole pine forests of contrasting tree density. *Canadian Journal of Forest Research* 33:351-363.
- Litton CM, MG Ryan, and DH Knight. 2004. Effects of tree density and stand age on carbon allocation patterns in a postfire lodgepole pine ecosystem. *Ecological Applications* 14: 460-475.
- Livingston, D.A., 1968. Some inter-stadial and post-glacial pollen diagrams from eastern Canada. *Ecological Monographs* 38:87-125.

- Loya, W., K. Pregitzer, N. Karberg, J. King, and C. Giardina. 2003. Reduction of soil carbon formation by tropospheric ozone under elevated carbon dioxide. *Nature* 425: 705-707.
- Lukac, M., C. Calfapietra and D.L. Godbold 2003. Production, turnover and mycorrhizal colonization of root systems of three *Populus* species grown under elevated CO₂ (POPFACE). *Global Change Biology*. 9:838-848.
- Luo, Y.Q. 2003. Uncertainties in interpretation of isotope signals for estimation of fine root longevity: theoretical considerations. *Global Change Biology*. 9:1118-1129.
- Lyr, H. and G. Hoffmann 1967. Growth rates and growth periodicity of roots. *International Review of Forestry Research*. 2:181-236.
- Matamala, R., M.A. Gonzalez-Meler, J.D. Jastrow, R.J. Norby and W.H. Schlesinger 2003. Impacts of fine root turnover on forest NPP and soil C sequestration potential. *Science*. 302:1385-1387.
- Matamala, R. and W.H. Schlesinger 2000. Effects of elevated atmospheric CO₂ on fine root production and activity in an intact temperate forest ecosystem. *Global Change Biology*. 6:967-979.
- McClaugherty, C.A., J.D. Aber, J.M. Melillo. The role of fine roots in the organic matter and nitrogen budgets of forested ecosystems. *Ecology* 63 1481-1490.
- McConnaughey, K.D.M. and J.S. Coleman 1999. Biomass allocation in plants: Ontogeny or optimality? A test along three resource gradients. *Ecology*. 80:2581-2593.
- McDowell, N.G., N.J. Balster, J.D. Marshall. 2001. Belowground carbon allocation of Rocky Mountain Douglas-fir. *Can. J. For. Res.* 31: 1425-1436.
- McElrone, AJ, WT Pockman, J Martinez-Vilalta, RB Jackson. 2004. Variation in xylem structure and function in stems and roots of trees to 20 m depth. *New Phytologist* 163: 507-517.
- McKevlin, M.R., D.H. Hook, A.A. Rozelle. Adaptations of plants to flooding and soil waterlogging. Pg. 173-204 In: M.G. Messina and W.H. Conner (eds), *Southern Forested Wetlands – Ecology and Management*. Lewis Press. Boca Raton, FL.
- Minkinen, K. and J. Laine. 1998. Long term effect of forest drainage on peat carbon stores of pine mires in Finland. *Can. J. For. Res.* 28:1267-1275.
- Melillo, J. et al. 2002. Soil warming and carbon-cycle feedbacks to the climate system. *Science* 298, 2173-2176.
- Nadelhoffer, K.J, J.W. Raich. 1992. Fine root production estimates and belowground carbon allocation in forest ecosystems. *Ecology* 73, 1139-1147.
- Nepstad, D.C., C.R. de Carvalho, E.A. Davidson, P.H. Jipp, P.A. Lefebvre, G.H. de Negreiros, E.D. da Silva, T.A. Stone, S.E. Trumbore, and S. Vieira. 1994. The role of deep roots in the hydrological and carbon cycles of Amazonian forests and pastures. *Nature* 372(6507):666-669.
- Norby, R.J. and R.B. Jackson 2000. Root dynamics and global change: seeking an ecosystem perspective. *New Phytologist*. 147:3-12.
- Norby, R.J. and Y. Luo. 2004. Evaluating ecosystem responses to rising atmospheric CO₂ and global warming in a multi-factor world. *New Phytologist*. 162: 281-293.
- Norby, R.J., C.A. Gunderson, S.D. Wullschlegler, E.G. O'Neill and M.K. McCracken 1992. Productivity and compensatory responses of yellow-poplar trees in elevated CO₂. *Nature*. 357:322-324.
- Norby, R.J., J. Ledford, C.D. Reilly, N.E. Miller and E.G. O'Neill 2004. Fine-root production dominates response of a deciduous forest to atmospheric CO₂

- enrichment. Proceedings of the National Academy of Sciences of the United States of America. 101:9689-9693.
- Norby, R.J. et al. 2002. Net primary production of a CO₂-enriched deciduous forest and the implications for carbon storage. *Ecological Applications* 12: 1261-1266.
- Olsson PA, Larsson L, Bago B, Wallander H, van Aarle IM (2003) Ergosterol and fatty acids for biomass estimation of mycorrhizal fungi. *New Phytol* 159:7-10
- Ovington JD. 1957. Dry-matter production by *Pinus sylvestris* L. *Ann. Bot (N.S.)* 82, 288-314.
- Paul E, Clark F (1996) *Soil Microbiology and Biochemistry* (Academic Press, New York).
- Paul, K.I., P.J. Polglase, J.G. Nyakuengama, P.K. Khanna. 2002. Change in soil carbon following afforestation. *Forest Ecology and Management*. 168: 241-257.
- Pendall, E., S. Bridgman, P.J. Hanson, B. Hungate, D.W. Kicklighter, D.W. Johnson, B.E. Law, Y.Q. Luo, J.P. Megonigal, M. Olsrud, M.G. Ryan and S.Q. Wan 2004. Below-ground process responses to elevated CO₂ and temperature: a discussion of observations, measurement methods, and models. *New Phytologist*. 162:311-322.
- Pregitzer, K.S. 2002. Fine roots of trees – a new perspective. *New Phytologist*. 154:267-273.
- Pregitzer, K.S. 2003. Woody plants, carbon allocation and fine roots. *New Phytologist*. 158:421-424.
- Pregitzer, K.S., J.S. King, A. J. Burton, S.E. Brown. 2000a. Response of tree fine roots to temperature. *New Phytologist* 147: 105-115.
- Pregitzer, K., D. Zak, P. Curtis, M. Kubiske, J. Teeri and C. Vogel. 1995. Atmospheric CO₂, soil nitrogen, and turnover of fine roots. *New Phytologist* 129:579-585.
- Pregitzer, K.S., D.R. Zak, J. Maziasz, J. DeForest, P.S. Curtis and J. Lussenhop 2000b. Interactive effects of atmospheric CO₂ and soil-N availability on fine roots of *Populus tremuloides*. *Ecological Applications*. 10:18-33.
- Pritchard, S.G., H.H. Rogers, M.A. Davis, E. Van Santen, S.A. Prior and W.H. Schlesinger 2001. The influence of elevated atmospheric CO₂ on fine root dynamics in an intact temperate forest. *Global Change Biology*. 7:829-837.
- Pubilcover, D.A. and K.A. Vogt 1993. A comparison of methods for estimating forest fine root production with respect to sources of error. *Canadian Journal of Forest Research*. 23:1179-1186.
- Raich, J. W., and K. J. Nadelhoffer. 1989. Belowground carbon allocation in forest ecosystems: Global trends. *Ecology* 70:1346-1354.
- Reich, P.B. 1983. Effects of low concentrations of O₃ on net photosynthesis, dark respiration, and chlorophyll contents in aging hybrid poplar leaves. *Plant Physiology* 73, 291-296.
- Reich, P.B., and Amundson, R.G. 1985. Ambient levels of O₃ reduce net photosynthesis in tree and crop species. *Science* 230, 566-570.
- Reich, P., and P. Bolstad. 2001. Productivity of evergreen and deciduous temperate forests. Pages 245-283 in Roy, J., Saugier, B. & Mooney, H. A, editors. *Terrestrial Global Productivity*. Academic Press, San Diego, USA.
- Reuss, R. W., R.L. Hendrick, A.J. Burton, K.S.Pregitzer, Bjartmar Sveinbjornsson, M.F. Allen, and G.E.Maurer. 2003. Coupling fine root dynamics with ecosystem carbon cycling in black spruce forests of interior Alaska. *Ecological Monographs* 73: 643-662.

- Ryan MG, Linder S, Vose JM, Hubbard RM. 1994. Dark respiration in pines. Pages 50-63 in Gholz HL, Linder S, McMurtrie RE, editors. Pine Ecosystems. Ecological Bulletins 43, Uppsala.
- Ryan, M. G., R. M. Hubbard, S. Pongracic, R. J. Raison, and R. E. McMurtrie. 1996. Autotrophic respiration in *Pinus radiata* in relation to nutrient status. *Tree Physiology* 16:333-343.
- Ryan MG, Lavigne MB, Gower ST. 1997. Annual carbon cost of autotrophic respiration in boreal forest ecosystems in relation to species and climate. *Journal of Geophysical Research* 102(D24):28871-28884.
- Rygiewicz PT, Johnson MG, Ganio LM, Tingey DT, Storm MJ (1997) Lifetime and temporal occurrence of ectomycorrhizae on ponderosa pine (*Pinus ponderosa* Laws) seedlings grown under varied atmospheric CO₂ and nitrogen levels. *Plant and Soil* 189:275-287
- Santantonio, D. and R.K. Hermann 1985. Standing crop, production and turnover of fine roots on dry, moderate, and wet sites of mature Douglas-fir in western Oregon. *Annales des Sciences Forestieres*. 42:113-142.
- Sarmiento J (2000) That sinking feeling. *Nature*, 408, 155-156.
- Shaver, G.R., and S. Jonasson. 2001. Productivity of Arctic Ecosystems. Pages 189-210 in Roy, J., Saugier, B. & Mooney, H. A, editors. Terrestrial Global Productivity. Academic Press, San Diego, USA.
- Schimel, D.S., B.H. Braswell, E.A. Holland, R. McKeown, D.S. Ojima, T.H. Painter, W.J. Parton, and A.R. Townsend. 1994. Climatic, edaphic, and biotic controls over storage and turnover of carbon in soils. *Global Biogeochem. Cycles* 8:279-293.
- Six, J., P. Callewaert, S. Lenders, S. Degryze, S.J. Morris, E.G. Gregorich, E.A. Paul and K. Paustian. 2002. Measuring and understanding carbon storage in afforested soils by physical fractionation. *Soil Sci. Soc. Am. J.* 66:1981-1987.
- Smith SE, Read DJ (1997) Mycorrhizal Symbiosis. London, UK: Academic Press
- Smith FW, Resh SC. 1999. Age-related changes in production and below-ground carbon allocation in *Pinus contorta* forests. *Forest Science* 45:333-341.
- Stape JL, D Binkley, and MG Ryan. 2004. Eucalyptus production and the supply, use and the efficiency of use of water, light and nitrogen across a geographic gradient in Brazil. *Forest Ecology and Management* 193:17-31.
- Steele SJ, Gower ST, Vogel JG, Norman JM. 1997. Root mass, net primary production and turnover in aspen, jack pine and black spruce forests in Saskatchewan and Manitoba, Canada. *Tree Physiology* 17:577-587.
- Stevens, G.N., R.H. Jones and R.J. Mitchell 2002. Rapid fine root disappearance in a pine woodland: a substantial carbon flux. *Canadian Journal of Forest Research- Revue Canadienne De Recherche Forestiere*. 32:2225-2230.
- Teskey, R.O. and T.M. Hinckley 1981. Influence of temperature and water potential on root growth of white oak. *Physiologia Plantarum*. 52:363-369.
- Tierney, G.L., T.J. Fahey, P.M. Groffman, J.P. Hardy, R.D. Fitzhugh, C.T. Driscoll and J.B. Yavitt 2003. Environmental control of fine root dynamics in a northern hardwood forest. *Global Change Biology*. 9:670-679.
- Townsend, A. R., B. H. Braswell, E. A. Holland, and J. E. Penner. 1996. Spatial and temporal patterns in terrestrial carbon storage due to deposition of fossil fuel nitrogen. *Ecological Applications* 6:806-814.

- Treseder, K. K. 2004. A meta-analysis of mycorrhizal responses to nitrogen, phosphorus, and atmospheric CO₂ in field studies. *New Phytologist* 164(2): 347-355
- Trettin, C.C. and M.F. Jurgensen. 2003. Carbon cycling in wetland forest soils. P. 311-328 In: J. Kimble, R. Birdsie, R. Lal. *The Potential of U.S. Forest Soils to Sequester Carbon and Mitigate the Greenhouse Effect*. CRC Press. Boca Raton, FL.
- Uselman, S.M., R.G. Qualls, and R.B. Thomas. 2000. Effects of increased atmospheric CO₂, temperature, and soil N availability on root exudation of dissolved organic carbon by a N-fixing tree (*Robinia pseudoacacia* L.). *Plant and Soil*. 222: 191-202.
- VEMAP members (1995) Vegetation/ecosystem modeling and analysis project: Comparing biogeography and biogeochemistry models in a continental-scale study of terrestrial ecosystem response to climate change and CO₂ doubling. *Global Biogeochemical Cycles*, 9, 407-437.
- Vos, J. and J. Groenwold 1987. The relation between root growth along observation tubes and in bulk soil. In *Minirhizotron observation tubes: Methods and applications for measuring rhizosphere dynamics* Ed. H.M. Taylor. American Society of Agronomy, Inc., Madison, WI, pp. 39-49.
- Wan, S.Q., R.J. Norby, K.S. Pregitzer, J. Ledford and E.G. O'Neill 2004. CO₂ enrichment and warming of the atmosphere enhance both productivity and mortality of maple tree fine roots. *New Phytologist*. 162:437-446.
- Wells, C.E. and D.M. Eissenstat 2001. Marked differences in survivorship among apple roots of different diameters. *Ecology*. 82:882-892.
- Wells, C.E., D.M. Glenn and D.M. Eissenstat 2002a. Changes in the risk of fine-root mortality with age: A case study in peach, *Prunus persica* (Rosaceae). *American Journal of Botany*. 89:79-87.
- Wells, C.E., D.M. Glenn and D.M. Eissenstat 2002b. Soil insects alter fine root demography in peach (*Prunus persica*). *Plant Cell and Environment*. 25:431-439.
- Withington, J.M., A.D. Elkin, B. Bulaj, J. Olesinski, K.N. Tracy, T.J. Bouma, J. Oleksyn, L.J. Anderson, J. Modrzyński, P.B. Reich and D.M. Eissenstat 2003. The impact of material used for minirhizotron tubes for root research. *New Phytologist*. 160:533-544.
- Wallander H, Nilsson LO, Hagerberg D, Baath E. 2001. Estimation of the biomass and seasonal growth of external mycelium of ectomycorrhizal fungi in the field. *New Phytologist* 151:753-760
- Zak, D.R., K.S. Pregitzer, J.S. King, W.E. Holmes. 2000a. Elevated atmospheric CO₂ and the composition and function of soil microbial communities. *Ecological Applications*. 10: 47-59.
- Zak, D.R., K.S. Pregitzer, P.S. Curtis, W.E. Holmes. 2000b. Atmospheric CO₂, fine roots and the response of soil microorganisms: a review and hypothesis. *New Phytologist*. 147: 201-222.

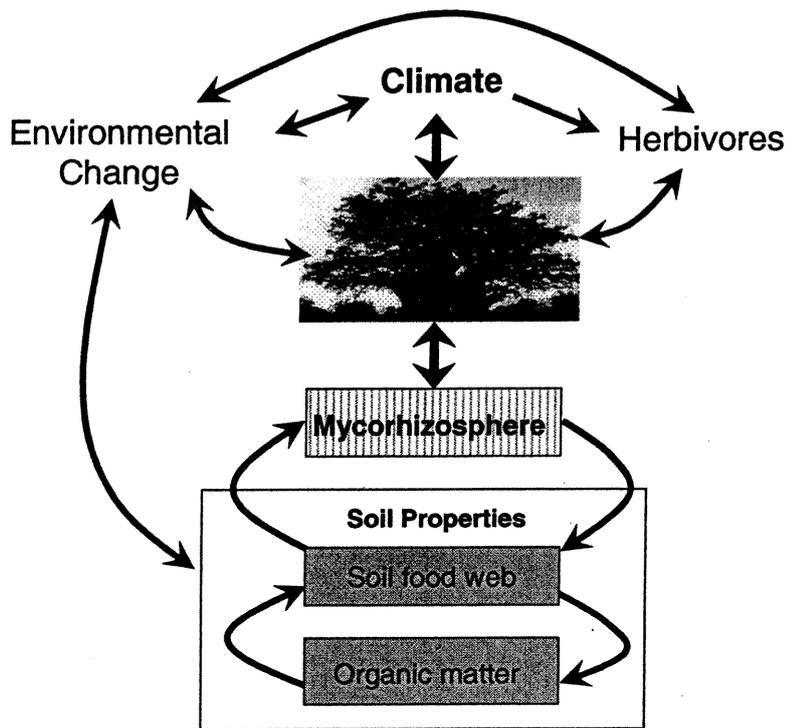


Figure 1. Diagram of the direct and indirect effects of environmental change (e.g., increased CO₂, O₃ and other greenhouse gases, change in fire regimes, elevated nutrient deposition rates, altered species succession, species invasion, etc.) on belowground carbon allocation through changes in canopy function, aboveground herbivore communities and soil properties (soil carbon and nutrient quality, soil food web including belowground herbivores).

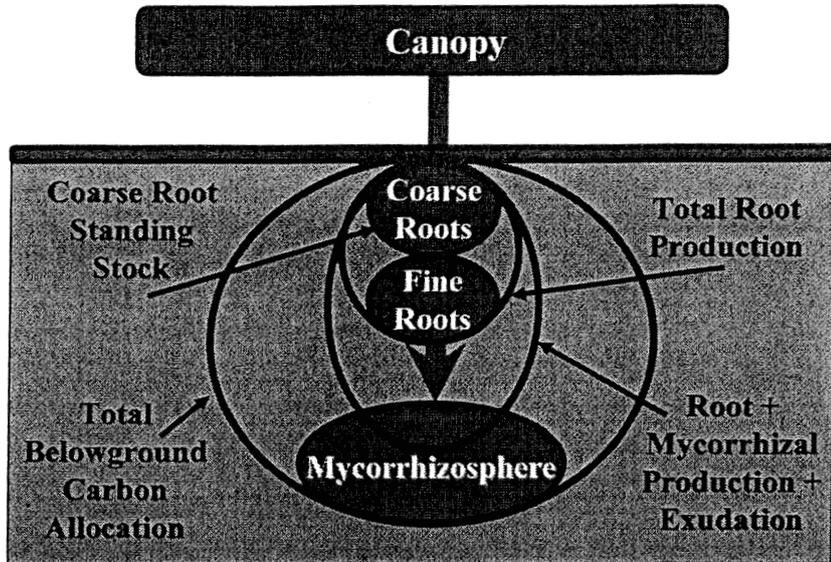


Figure 2. Various approaches to examining BCA in forests. Coarse root standing stock is a pool of carbon in soil measured by excavation and weighing at a single point in time. Total root production is comprised of coarse and fine root NPP, but typically excludes exudation or mycorrhizal production. Root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

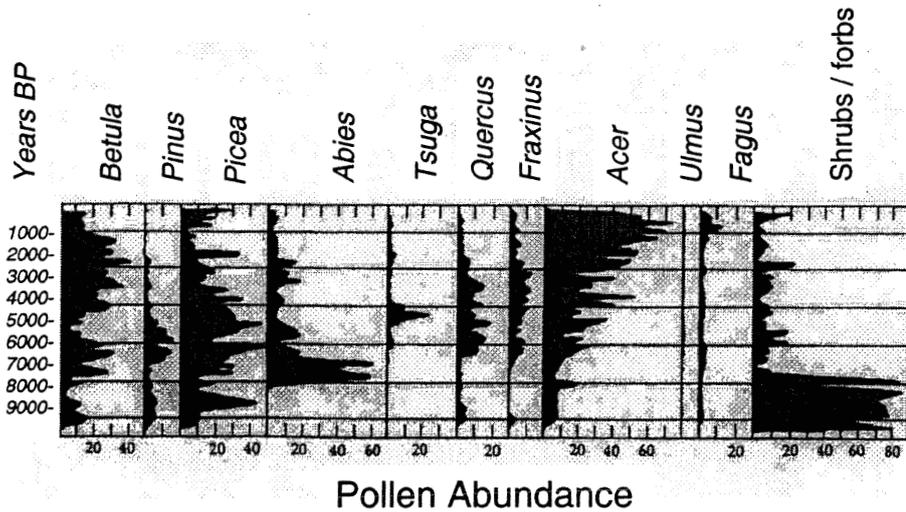


Figure 3. 9000 year record of change in vegetation (years before present) in Nova Scotia, as captured by a change in the quantity of pollen from different genera of common North American trees (Adapted from Livingston, 1968).

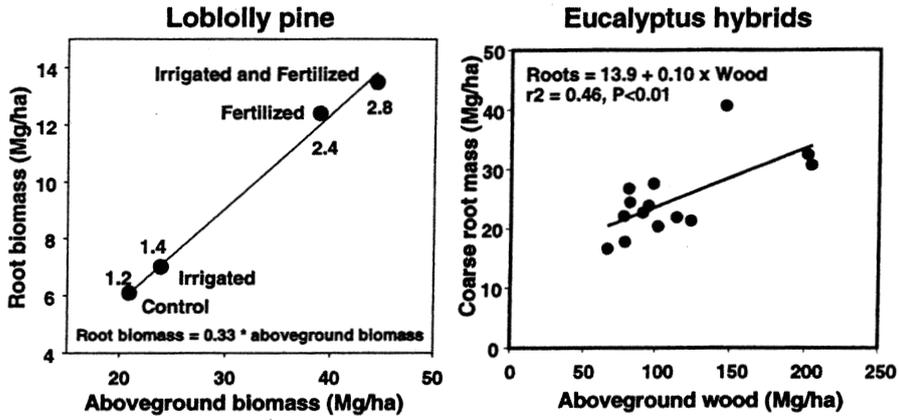


Figure 4. The root:shoot ratio was relatively constant for loblolly pine across treatments on a poor site ($R^2 = 0.95, P < 0.01$; data from Albaugh et al. 1998), whereas a non-zero intercept in the same relationship across 14 sites along a productivity gradient with *Eucalyptus* in Brazil had a positive Y intercept, indicating that root mass increased as a proportion of aboveground mass from 16% on fertile sites to 32% on infertile sites (from data in Stape et al. 2004).

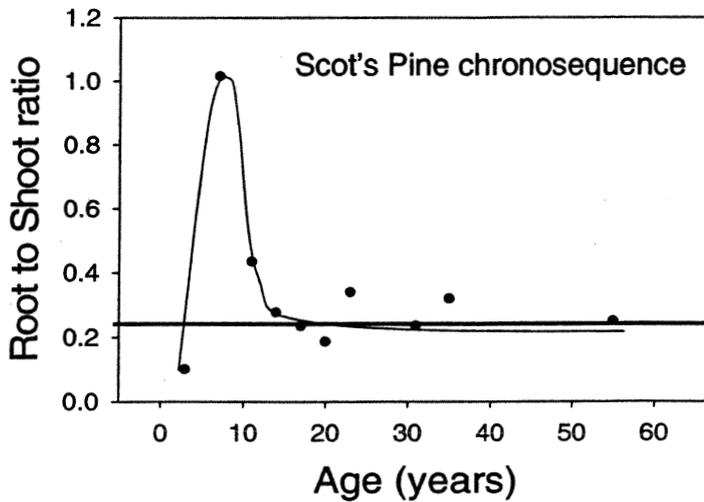


Figure 5. Data for Scots pine from Ovington (1957) and the root to shoot ratio (identified by the line at a root to shoot ratio of 0.22) used by Li et al. (2003) for estimating coarse root standing stocks for pine forests in Canada. The value of 1.0 for the 8-year-old stand of Ovington either indicates a major difference between the two studies, or it is an outlier and the studies support similar conclusions.

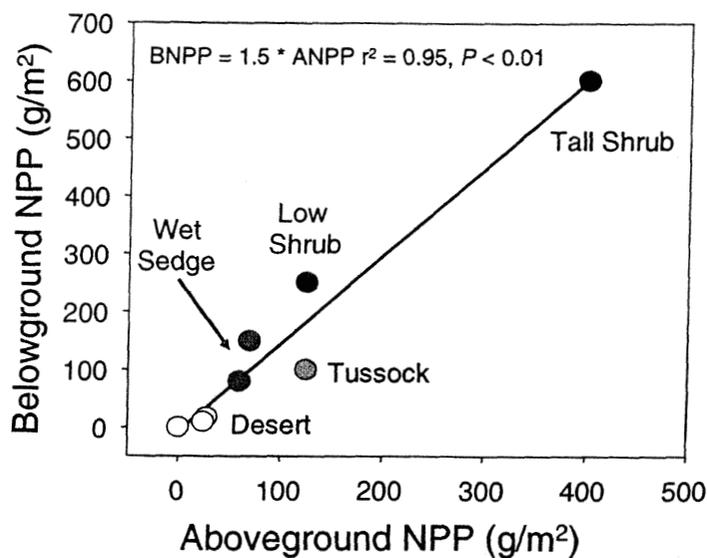


Figure 6. Data from review by Shaver and Jonasson (2001), showing the stability of BNPP to ANPP for Arctic ecosystems in North America.

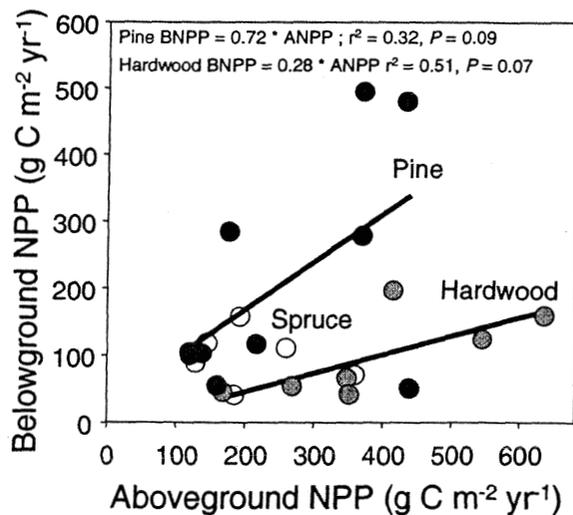


Figure 7. Data from review by Gower et al. (2001a), showing the stability of BNPP to ANPP for boreal hardwood and pine species across Russia and North America. There was no relationship between BNPP and ANPP for spruce species across sites, but the relationship appears negative.

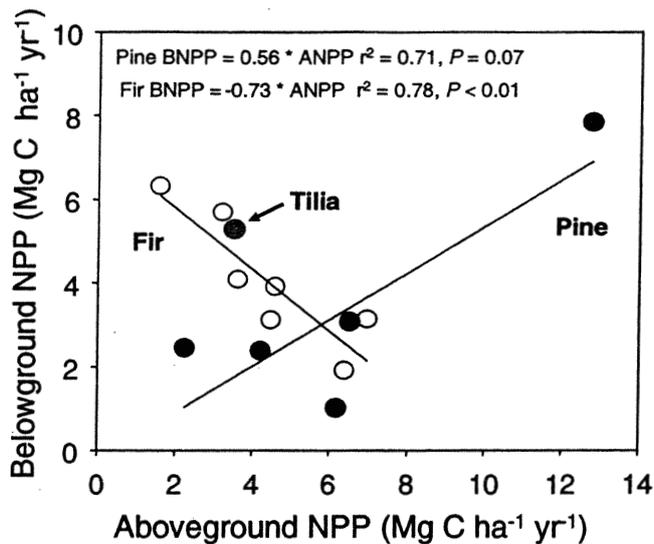


Figure 8. Data from review by Reich and Bolstad (2001) showing strong but opposing relationships between ANPP and BNPP for temperate pine and fir (true fir and Douglas-fir) species across sites. Only one point is reported for a hardwood point so no relationship is given.

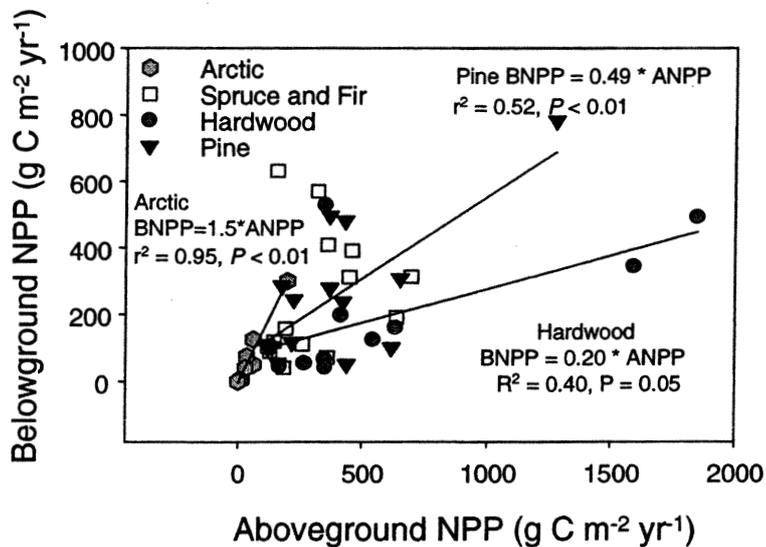


Figure 9. Global scale relationship between ANPP and BNPP with data from Figures 6 through Figure 8. The global relationships confirm regional patterns for pine and deciduous vegetation (hardwood trees plus larch, shrubs, forbs, and grasses), but fail to support patterns for spruce, fir, and Douglas-fir.

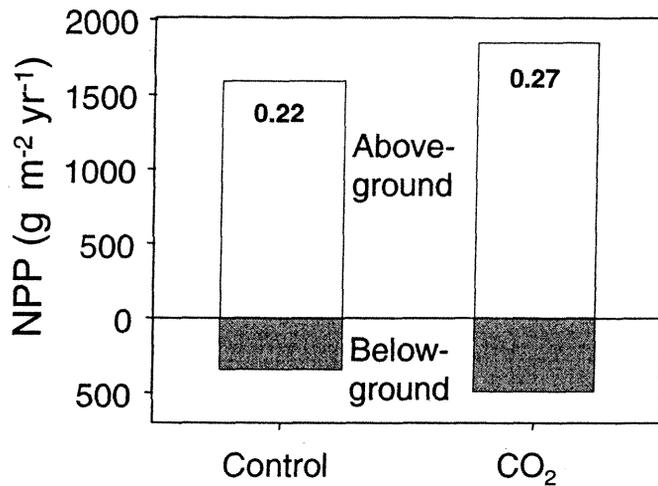


Figure 10. Data from Norby et al. 2002 showing the positive effects of elevated CO₂ on above and belowground NPP. Elevated CO₂ in this FACE experiment increased ANPP and BNPP, and also increased the BNPP to ANPP ratio (the ratio is identified in each bar).

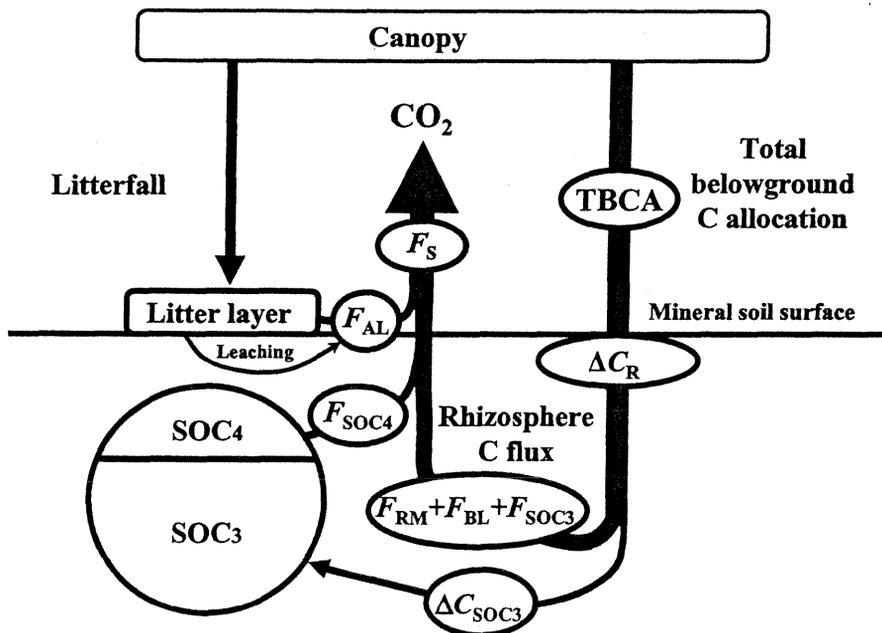


Figure 11. Conceptual model for how TBCA and stable isotopes can be used to examine the belowground fate of TBCA and the component sources of soil respiration (adapted from Giardina et al. 2004).

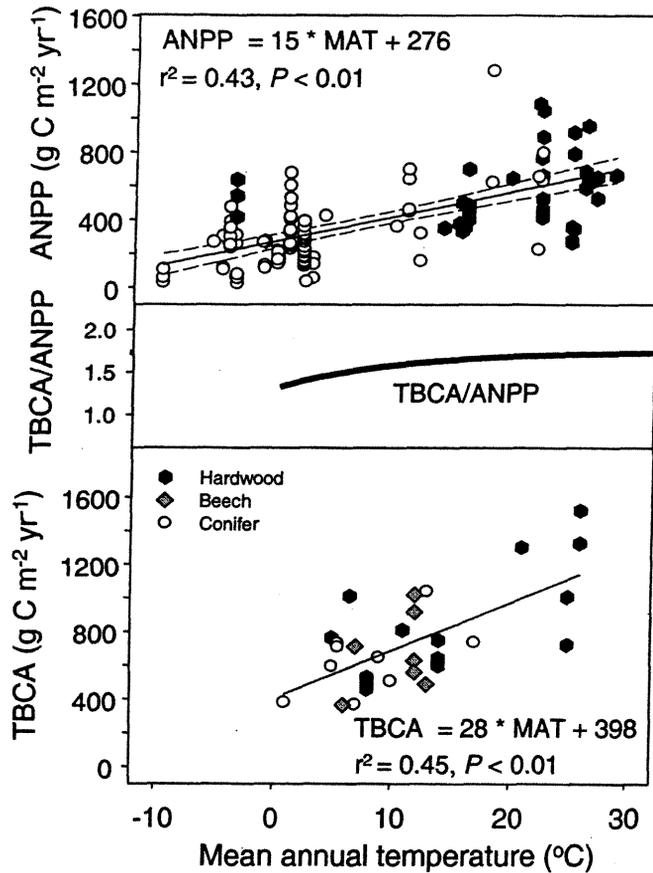


Figure 12. Global scale relationship between mean annual temperature (MAT) and ANPP (Top Panel), between MAT and TBCA (Bottom Panel) and the ratio of TBCA to ANPP derived from the equations describing the two relationships. Points are unfertilized forests across a global scale gradient in MAT. Data are 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.

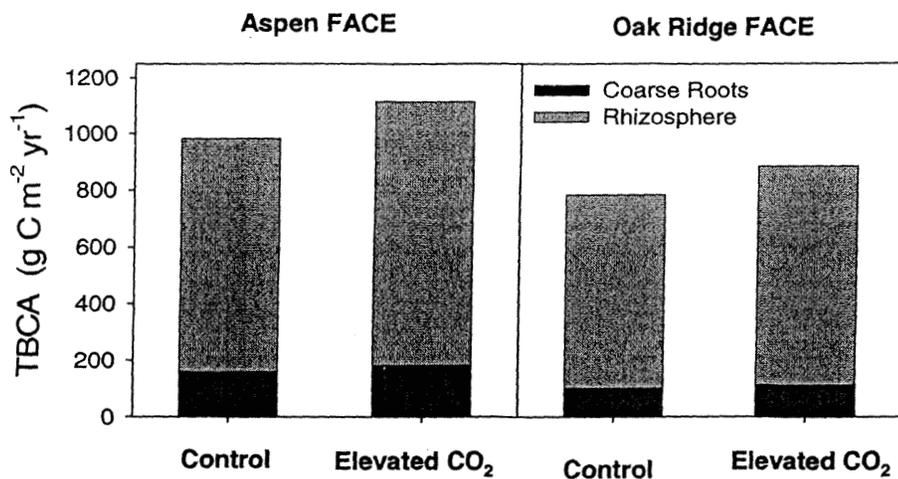


Figure 13. The effects of elevated CO₂ on mixed aspen-birch stands in Rhinelander Wisconsin (Left panel) and sweetgum (*Liquidambar styraciflua*) in Oak Ridge, Tennessee. Data for calculations of TBCA are from King et al. (2004), Norby et al. (2002), Norby et al. (2004), and unpublished data of the authors..

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