FINE ROOT DYNAMICS FOR THREE DISTINCT NORTHERN ONTARIO FORESTS: A COMPARISON OF APPROACHES USED TO ESTIMATE FINE ROOT BIOMASS, PRODUCTIVITY, AND TURNOVER

By

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ABSTRACT

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There is increasing interest to develop quantitative approaches to carbon accounting and determine carbon sequestration potential at both the site and landscape scales. Currently, our lack of understanding of fine root dynamics in northern temperate forest systems has hampered efforts to accurately parameterize any of the existing C budget models (*e.g.*, CBM-CFS3). The objectives of this study were to: 1) describe the various approaches most commonly used to estimate fine root biomass, highlighting their strengths and limitations, 2) develop species- and diameter class-specific standard root lengths (*i.e.*, factor for converting measured root lengths to biomass when using minirhizotron technologies) for selected northern temperate tree species, and 3) compare/contrast the estimates of fine root biomass, productivity, and turnover rates derived from the commonly applied indirect (*i.e.*, used in most carbon accounting models) and direct (*i.e.*, *in situ* stand-level measurements) methods. This study was conducted for three distinct northern temperate forest/stand types (*i.e.*, northern hardwoods – sugar maple ; northern coniferous – jack pine ; and boreal mixedwood – aspen, spruce, balsam fir).

It was determined that there was a significant difference in fine root standard root length (SRL) between northern temperate tree species and diameter classes. Angiosperms tended to had significantly higher average SRLs compared to gymnosperms. Within these species groupings, tolerant, late successional species had higher SRLs compared to their intolerant, early successional counterparts. Standard root lengths dropped significantly (>200 %) as diameter class increased. The results suggest that the development of species- and diameter class-specific SRLs should provide better estimates of fine root biomass and productivity, leading to a better understanding of temperate forest C dynamics. The results from the comparison of the indirect and direct methods to estimate fine root biomass, productivity, and turnover accentuate the potential challenges associated with incorporating site specific fine root research into broader generalizations applied to large land masses. These constructs inevitably incorporate inherent errors associated with aboveground biomass estimates, conversions factors to belowground estimates (indirect methods), high within-site variability associated with direct measurements, and all the numerical accounting methods and assumptions needed to arrive at fine root estimates. A more thorough examination of fine root dynamics is required in order to ensure wider scientific acceptance of broad-based models and their ability to predict the impacts of forest management activities or climate change.

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1.0 INTRODUCTION

A contemporary challenge facing scientists, legislators and forest managers is understanding ecosystem carbon (C) dynamics. Ecosystem C models can be used to determine a site's (or nation's) C reserves and C sequestering potential (productivity), and turnover. Adequate measurements of C sources and sinks, however, are required in order to determine what effect a management treatment may have on C storage and cycling. It is generally accepted that forests can be considered either a source or a sink depending on the biotic and abiotic conditions that surround it. Forests are considered C sinks when they are net importers of C, and C sources when they are net exporters of C. Figure 1.1 is a generalized depiction of a global C budget model developed by Schimel (1995). In this scenario, C found in vegetation and soils compose 5% of global C reserves, and are responsible for 40% of the flux of C between the atmosphere and the earth's surface.

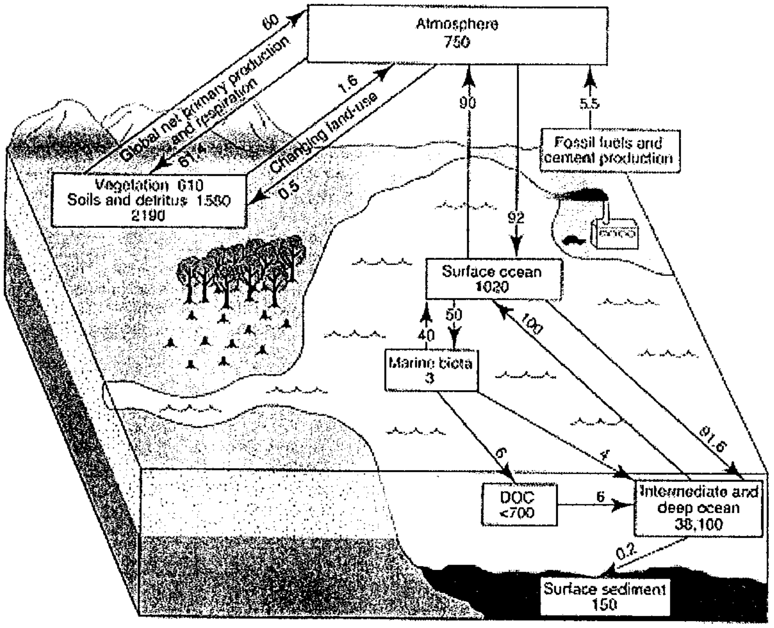

Figure 1.1. Global reservoirs and fluxes of C, expressed in gigatonnes C per year (Schimel 1995).

Malhi *et al.* (1999) also remarked that the world's forests, particularly those situated at northern latitudes, contain large reservoirs of biomass and soil C.

The Kyoto Protocol of the United Nations Framework Convention on Climate Change (UNFCCC) was the first global attempt to limit the amount of CO_2 emissions and other greenhouse gasses from entering the atmosphere and thereby reduce its potential effects on climate change (Grace 2004). The text of the protocol was adopted and signed by a number of nations at the third Conference of the Parties (COP3) in Kyoto on December 11th, 1997 (Grace 2004). This was done in response to dire warnings that the current level of CO_2 in the atmosphere would lead to an increase of the global mean temperature by 2.4°C by 2100. This increase in atmospheric C is believed to account for 60% of all global warming (Grace 2004, and, Malhi *et al.* 1999). Grace (2004) and Malhi *et al.* (1999) have stated that global climate change is a direct result of deforestation and the burning of fossil fuels. Most of this warming is to occur over the land (3 to 4°C) rather than over the oceans (0 °C to 2°C) (Malhi *et al.* 1999). Almost all the models indicate that there will be a warming of 4 °C to 6°C, predominantly in the autumn and winter, in boreal ecosystems. Global atmospheric CO_2 concentrations have risen from 260 ppm, reached 8200 years ago, to approximately 360 ppm today (Indermuhle *et al.* 1999). Mean global precipitation is also predicted to increase with climate change.

Certain countries (Annex 1) in the protocol have been asked to count a number of practices, known as "flexible mechanisms" that can be used toward their emissions reduction (Grace 2004). These practices include: 1) planting of new forests creating C sinks, and, adopting new agricultural practices to reduce emissions, 2) C trading and, 3) developing "clean" projects that help reduce emissions, and, in turn, create additional C sinks (Grace 2004). Even though many of the Annex 1 countries agreed to the protocol, there is still considerable controversy as to whether these flexible mechanisms will, indeed, improve C sequestration on a global level (Grace *et al.* 2003).

Temperate forests have been identified as playing a key role in the global C cycle (Fan *et al.* 1998). McGuire *et al.* (1995) explained that even though high latitude forest ecosystems account for ~24% of the world's land area, they store ~40% of the reactive C. Raich and Potter (1995) provided an estimate of 60 gigatonnes of C that is sequestered by plants on an annual basis. In forested ecosystems a significant portion of C is stored in aboveground and fine root (< 0.2mm diameter) biomass. As a result, a thorough examination of the net primary productivity (NPP) of ecosystem compartments is essential in understanding the C sequestration capacity of Canada's forests (Coleman *et al.* 2000). Fine root NPP often exceeds aboveground NPP in forest ecosystems even though fine root biomass only constitutes a small fraction of total stand biomass (Gower *et al.* 1992, Hendrick and Pregitzer 1993a,b).

Jackson *et al.* (1997) reported that as much as 33% of global NPP is used for the production of fine roots. Fine root productivity, however, is largely an unknown variable in C budget modeling. To date, few studies have been completed to examine what effect competition for water and nutrients may have on fine root growth and development (Ford and Deans 1978, Green 2004), and even fewer studies have been published examining the relationships between fine root productivity and aboveground productivity (Ruess *et al.* 1996, Green 2004). Altering stand composition and architecture through silvicultural treatments may influence the C allocation patterns, C flux and NPP of a given site. Therefore, increased understanding of these processes could lead to improved decisions regarding scheduling and harvesting. This could assist in increasing fiber production efficiency, reducing rotation time and/or increasing the C sequestering abilities of managed stands (Comeau and Thomas 1996, Coleman et al. 2000). A tree's aboveground NPP may be affected by fine root dynamics and the energy required for their maintenance. New reliable information on ecophysiology of temperate forests will assist forest managers in making decisions to optimize the use of forest resources and resolve future wood supply shortages (OMNR 2004).

The objectives of this study were to: 1) describe the various approaches most commonly used to estimate fine root biomass, highlighting their strengths and limitations, 2) develop species- and diameter class-specific standard root lengths (*i.e.*, factor for converting measured root lengths to biomass when using minirhizotron technologies) for selected northern temperate tree species, and 3) compare/contrast the estimates of fine root biomass, productivity, and turnover rates derived from the commonly applied indirect (*i.e.*, used in most carbon accounting models) and direct (*i.e.*, *in situ* stand-level measurements) methods. This study was conducted for three distinct northern temperate forest/stand types (*i.e.*, northern hardwoods – sugar maple ; northern coniferous – jack pine ; and boreal mixedwood – aspen, spruce, balsam fir).

2.0 METHODOLOGICAL APPROACHES USED WHEN CONDUCTING FINE ROOT BIOMASS EXPERIMENTS

2.1 INDIRECT METHODS

The most common indirect method is to derive a relationship between aboveground stand-level parameters (*i.e*, aboveground biomass – $T \cdot ha^{-1}$, basal area – m^2

$\cdot$ ha^{-1}) and belowground root biomass. The equations and associated coefficients for calculating total root (coarse + medium + small + fine) and fine root biomass are summarized in Table 2.1. In the case of Kurz *et al.* (1996) and Li *et al.* (2002), they calculated total root biomass, total fine root biomass, and fine root turnover based on aboveground biomass. In these models, different multipliers for conifer (gymnosperms) versus hardwoods (angiosperms) were applied to the aboveground biomass estimates.

Table 2.1. Equations used to estimate fine root biomass for three indirect methods evaluated in this study.

Equation	Reference
$RB_s = 0.2317ABD_s$ $RB_h = 1.432ABD_h^{0.639}$ $FRP = 2.737 \times RB^{-0.841}$ * all proportions > 0.9 are reduced to 0.9 $FRBD = RB \times FRP$	Kurz et al. (1996) CBM / CFS II
$RB_s = 0.222ABD_s$ $RB_h = 1.576ABD_h^{0.615}$ $FRP = 0.072 + 0.354e^{-0.060RB}$ * all proportions > 0.9 are reduced to 0.9 $FRBD = RB \times FRP$	Li et al. (2002) CBM / CFS II
$STFRB = 1 / 1 + (8.9483BA^{-0.9896})$ $SITNFRB = 1 / 1 + (39.683BA^{-1.0818})$ $SITBFRB = 1 / 1 + (262998.4BA^{-3.6199})$	Chen et al. (2004)

FRBD = Fine Root Biomass Density; ABD = Aboveground Biomass Density; RB_s = Softwood Root Biomass; RB_h = Hardwood Root Biomass; FRP = Fine Root Proportion; STFRB = Shade Tolerant Fine Root Biomass; SITNFRB = Shade Intolerant Needlebearing Fine Root Biomass; SITBFRB = Shade Intolerant Broadleaf Fine Root Biomass

As an alternative approach, Chen *et al.* (2004) derived a relationship to calculate fine root biomass at the individual tree level using basal area, then summed these on a per hectare basis. In their model, they differentiated between shade tolerant species, shade intolerant (needleleaf), and shade tolerant (broadleaf) (Table 2.1). Chen *et al.* (2004) developed a multi-parameter equation to estimate fine root turnover that incorporates the previously calculated fine root biomass, as well as stand age and mean annual air temperature.

These indirect approaches generate modeled estimates of fine root biomass and fine root NPP that, in themselves, are based on modeled estimates of aboveground biomass (see Baskerville (1972) for a description of this common double sampling approach). In most cases, model validations have not been published. Furthermore, there are very few published aboveground to belowground relationships available for northern temperate species. Past studies have also been limited in stand age, soil type, species, species associations, habitat, form, site index, live crown size, volume, and height suggesting broader application to be limited. Studies examining these factors are required in order to better ascertain site-specific fine root biomass and fine root NPP potential.

2.2 DIRECT METHODS

In recent years, a number of techniques have been developed to quantify fine root biomass, productivity, and turnover. Techniques and equipment that have historically been used for the assessment of fine root research include rhizotrons (glass wall method), minirhizotron tubes, soil pits, ingrowth bags, sequential soil coring, containers, and, full tree / site excavation. Many of these methods have proven to be labour intensive and require destructive augmentation of the site which tends to limit the duration of fine root experiments and their spatial extent (Bohm 1979).

Fine root measurements typically include mass, length, diameter, density, tissue density, number of roots tips, rooting depth, phenolics (tannin and suberin), carbohydrate and N concentration (Pregitzer *et al.* 2002, McMichael and Taylor 1987). In addition, fine roots are often assessed for mycorrhizal inoculation rates. Fine root biomass density, production, net primary productivity and turnover can be measured by sampling a given area / volume of soil through a variety of methods, and by applying a medley of mathematical permutations. Fine root measurements are normally summarized and reported on a length per unit area ($m \cdot m^{-2}$), biomass per unit area ($kg \cdot m^{-2}$), biomass per unit volume ($kg \cdot m^{-3}$), and standard root length ($mm \cdot g^{-1}$) basis. NPP ($kg \cdot m^{-2} \cdot yr^{-1}$), fine root turnover ($kg \cdot m^{-2} \cdot yr^{-1}$), and C allocation ($kg \cdot m^{-2} \cdot yr^{-1}$) can be calculated by utilizing various equations that can be found in the scientific literature. These will be discussed later in this chapter.

The proportion of photosynthate allocated has been shown to be highly variable, ranging from 4 to 69 % of total annual C fixed. (Vogt *et al.* 1996). Fine root morphological features, including root grafting and resource sharing, are seldom taken into account when assessing their biomass density, NPP or turnover (Vogt and Persson 1991).

2.2.1 Soil Coring Technique

Soil coring has been the most common approach in the determination of belowground fine root biomass density and fine root NPP (Allen *et al.* 2000, Vogt and

Persson 1991). Multiple samples are taken from randomly selected locations in each plot on a monthly basis. Samples are normally dissected into varying soil depths (Joslin and Wolfe 1999). Extracted cores are placed in polyethylene bags and refrigerated at 4.0° C or frozen until they are processed (Joslin and Wolfe 1998, Tierney and Fahey 2001, Ruess *et al.* 1996). Certain features, such as gleying, mottling, and horizon depths can be spotted readily using this method (Deans 1979, Baker *et al.* 2001). Additional soil properties (*e.g.*, pH, macro- and micronutrient concentrations) can also be determined from the extracted core.

In the laboratory, the cores are gently washed to separate the roots from the soil, which are then categorized by size, colour, strength and flexibility to determine age classification (Allen *et al.* 2000, Baker *et al.* 2001, Tierney and Fahey 2001). Dead fine roots are considered to be roots that are dark brown or black in colour (Allen *et al.* 2000), flaccid or have a cortex that is easily pulled away from the stele (Coleman *et al.* 2000). White and yellow fine roots are considered to be alive. Fine roots are occasionally separated into size classes: < 1.0 mm, 1.0 mm – 2.0 mm, 2.0 mm + (Joslin and Wolfe 1998). Root samples are then dried at 60° C until constant weight is achieved. Standard root length and root length per surface area ($mm \cdot m^{-1}$) are then determined by digitizing the fine root sample (Coleman *et al.* 2000). Occasionally fresh mass is assessed prior to drying (Dawson 1993).

Ashing roots at 525° C provides an estimate of organic matter content of the sample (Coleman *et al.* 2000), with % C assumed to be 50% of organic matter content. Nutritional levels or ratios (*e.g.*, N:P, N:K, N:Ca, N:Mg, N:Bo) are typically calculated for both above and below ground portions of the plant to determine resource allocation and partitioning (Pregitzer *et al.* 1995, Baker *et al.* 2001, King *et al.* 2002).

The main disadvantage of the soil coring technique is the high labour costs associated with processing of the cores. In addition, developing a procedure or root key to determine species or root viability (live vs. dead) can be tedious, and accuracy questionable as it is extremely difficult to visually assess fine roots (Kurz and Kimmins 1987, Publicover and Vogt 1992, Vogt *et al.* 1986). Staining procedures can be used to aid in the assessment but requires additional time and resources (Vogt *et al.* 1998).

Soil core data can be used to provide an estimate of fine root biomass density ($g \cdot m^{-3}$), fine root length density ($mm \cdot m^{-3}$), fine root NPP, and fine root turnover, at all diameter classes and depths. Fine root biomass density is calculated as the total sum mass of all living root material in a particular soil core or layer on a sample date per unit area (Hendrick and Pregitzer 1993a, Joslin and Wolfe 1999). Fine root biomass density can also be expressed as the total sum length of living root material present within a soil core or layer on a sample date per unit volume (Johnson *et al.* 2001). Fine root length density can be quickly calculated by dividing the fine root biomass density by the standard root lengths of various fine root diameter classes. Fine root NPP is calculated as the difference between maximum and minimum live fine root biomass attained within one growing season (Harris *et al.* 1977, Persson 1978, Ruess *et al.* 1996). Fine root turnover is calculated as the ratio of annual root production to fine root biomass (Ostonen *et al.* 2005, Ruess *et al.* 1996).

Ostonen *et al.* (2005) reported that fine root NPP calculations were greater in sites sampled with soil cores than minirhizotrons or ingrowth bags. Kurz and Kimmins

(1987), Persson (1978), Publicover and Vogt (1993) and Vogt *et al.* (1986) suggested that soil coring methods could over or underestimate fine root biomass and fine root NPP depending on the circumstance, in relation to all other methods. Burke and Raynal (1994) suggested that over or underestimates of fine root NPP are probably associated with significant random errors that are derived from the creation of those estimates. Soil compaction during the coring operation may also affect the volumetric calculations.

2.2.2 Minirhizotron Technique

Minirhizotrons have been increasingly used as an important tool in the study of fine root dynamics in forestry (Hendrick and Pregitzer 1996). Typically, several nests of 50 mm diameter, 2 metre long butyrate or acrylic tubes are installed into the forest soil at angles between 30° and 45° (Tierney and Fahey 2001, King *et al.* 2002, Joslin and Wolfe 1998). Coarse fragment content and compacted soil layers commonly limit installation depths to between 60 to 90 cm (Joslin and Wolfe 1999). The minirhizotron tubes are normally inserted into pre-augered holes using a Lexan plastic sleeve to prevent the outside surface of the tube from being scratched by protruding rocks which would hamper visibility (Joslin and Wolfe 1999). Scratching, smearing, the creation of water channels, and decomposing debris can all effect final results (Van Noorwijk *et al.* 1985). It is therefore recommended that minirhizotron tubes be installed during the dry season to avoid inadequate seating and instability (Baker *et al.* 2001). Once the tubes are inserted, the aboveground portion of the tube is insulated and painted to prevent light and heat from affecting the subsurface of the soil (Joslin and Wolfe 1999, Baker *et al.* 2001, Tierney and Fahey 2001). The tube is then capped with a rubber stopper (Joslin and Wolfe 1999, Baker *et al.* 2001).

Only the upper surfaces of the tubes are monitored up to insertion depth which is typically 30 to 50 cm. Snapshots of frames are usually taken by using a root camera, laptop and Adobe Premiere 5.1 ® software. This procedure utilizes a periscope video camera that is equipped with a locking shaft to permit consistent, incremental lowering of the camera through the tube (Baker *et al.* 2001). The video images are then transformed into .PICT format. Images of fine roots within frames can then be digitized and categorized according to life stage by using RooTracker, an interactive Mac-based computer program (Craine and Tremmel 1995). Images can also be digitized using MSU-ROOTS software (Enslin *et al.* 1994) or Snappy Video Snapshot along with other image analysis software (Baker *et al.* 2001). The use of automated image assessment software has been attempted but due to the nature and size of some of the "finer roots" not all roots are normally processed (Baker *et al.* 2001). Occasionally poor image quality becomes a factor due to increased soil moisture or heaving (Baker *et al.* 2001).

Life stages include: new, live and dead fine roots, according to Pregitzer *et al.* (1995). Using this approach, fine root development can be tracked on a chronological basis to determine changes in size, morphology, length production, depth, and mortality (Tierney and Fahey 2001, Joslin and Wolfe 1999). Fine roots classified as "Dead" are identified by colour (*i.e.*, dark brown or black), or due to length reductions. Dead roots need to be subsequently tracked to determine if their "dead" status was indeed true and not a result of poor image quality (Tierney and Fahey 2001). It has been argued by Comas *et al.* (2000), that fine root death is a gradual process and that even though a root may appear to be "dead" this may be due to patches of dead cells coexisting with patches of live cells within the same root. Dates for "birth" and "death" are assumed to be the mid-point between image dates before and after the event (Tierney and Fahey 2001).

Minirhizotron data is normally collected on a monthly basis (Hendrick and Pregitzer 1992, 1993a). Baker *et al.* (2001) suggested that sampling intervals may have to be adjusted in seasonally or periodically flooded environments to ensure that all fine root NPP pulses are captured. Joslin and Wolfe (1998) suggested that optimum sampling periods should start two weeks prior to bud break and then proceed every two weeks after that. Minirhizotron fine root data can be summarized as longevity, fine root length density (squared or cubed), fine root biomass, fine root turnover, fine root tip density, and completeness, for all life stages, diameter classes and depths. As noted previously standard root length values can be converted into a biomass value.

Even though monitoring fine root lengths and demography with minirhizotron is quite simple, conversions to biomass or C fluxes in bulk soil can be problematic (King *et al.* 2002). Assumptions must be made regarding root mass to length relationships (standard root length), even though standard root lengths of fine roots often vary with diameter and state of maturity (Pregitzer *et al.* 1997, Tierney and Fahey 2001). In all cases it is assumed that the measured fine roots have a defined standard root length unless each root is exhumed and massed.

Fine root longevity can be calculated for an individual cohort of roots (Hendrick and Pregitzer 1992) or for multiple cohorts of roots born throughout the year (Majdi and Kangas 1997, Fahey *et al.* 1999), and can be expressed on a root number, root length or root mass basis (Tierney and Fahey 2001).

Fine root length density is calculated as the total sum length of all living root material present on a particular frame or with the field of view on a sample date (Hendrick and Pregitzer 1993a, Joslin and Wolfe 1999). The field of view for a minirhizotron frame has been quantified as 2.0 mm (Klepper *et al.* 1973, Taylor and Klepper 1973, and Steele *et al.* 1995) to 3.0 mm (Taylor *et al.* 1970, Sanders and Brown 1978, and Itoh 1985). Fine root biomass density, in turn, is calculated by dividing the fine root length density by the standard root lengths of the various fine root diameter classes. An alternative approach to calculating fine root length density and fine root biomass density used root numbers instead of lengths (Taylor *et al.* (1970), Sanders and Brown (1978), and Moore (1981), which involved the. This approach, however, required theoretical conversion factors which assume a mean expected root length (Merrill and Upchurch 1994).

A number of approaches have been applied to provide estimates of fine root NPP (Table 2.2). When fine root standard root lengths by diameter class are available, fine root NPP is determined for each diameter class and then summed (Tierney and Fahey 2001). Vogt *et al.* (1986) used maximum fine root biomass density minus minimum fine root biomass density to calculate fine root NPP. Santantonio and Grace (1987) calculated fine root NPP as the biomass of new fine roots minus biomass of dead fine roots in a particular minirhizotron tube. Majdi and Ohrvik (2004) calculated fine root NPP as the total new fine root biomass over a particular time period. Hendrick and Pregitzer (1993a) calculated fine root NPP as the initial fine root biomass density multiplied by the total new fine root biomass density subtracted by the total dead fine root biomass density divided by the initial fine root biomass density. Tierney and Fahey (2001) used initial fine root biomass divided by the median fine root longevity to produce fine root NPP. Bernier and Robitaille (2004) proposed a method that could be used to calculate fine root NPP in two and three dimensions by using total radius of root intersections found on minirhizotron images, standard root length, the coarse fraction of the soil, tube angle, ground angle minirhizotron frame size and viewing depth. This method could be applied to all new, mature and dead roots being observed at a given time.

Table 2.2. Fine root NPP equations applied to fine root biomass data obtained from minirhizotrons.

Equation	Reference
$\text{FR NPP} = \text{FRBD}_{t+1} - \text{FRBD}_t / t$	Hendrick and Pregitzer (1992) Joslin and Wolfe (1999) Persson (1978)
$\text{FR NPP} = \text{FRBD}_{\text{max}} - \text{FRBD}_{\text{min}}$	Harris *et al.* (1977) Persson (1978) Vogt *et al.* (1986) Ruess *et al.* (1996)
$\text{FR NPP} = \text{FRBD}_{\text{new}} - \text{FRBD}_{\text{dead}} / t$	Santantonio and Grace (1987) Ruess *et al.* (2003)
$\text{FR NPP} = \Sigma\ \text{FRBD}_{\text{new}} / t$	Majdi and Ohrvik (2004) Burton *et al.* (2000)
$\text{FR NPP} = 10^6\ \text{SRL}(1 - \text{FC})\text{V} \times \text{A}(\text{Zcos}y/\text{WLD})$	Bernier and Robitailles (2004)

FR NPP = Fine Root Net Primary Productivity; t = time; FRBD = Fine Root Biomass Density; SRL = Standard Root Length; FC = Course Fraction; V = volume; A = area; Z = insertion depth; y = insertion angle; W = frame width; L = frame length; D = frame depth

Fine root turnover is defined as the C flux derived from fine roots divided by C pool size, or the proportion of fine root biomass that dies annually (Norby and Jackson 2000). Hendrick and Pregitzer (1992) and Publicover and Vogt (1992) calculated fine root turnover as annual fine root death. Estimates of fine root turnover have been calculated by dividing the average annual fine root NPP by the live fine root standing crop (Burton *et al.*, 2000). Alternatively, Ruess *et al.* (1996) and Ostenen *et al.* (2005) calculated fine root turnover as the NPP of fine roots divided by the mean annual fine root biomass density.

It has been suggested that minirhizotrons can provide accurate accounts of biomass and production (Aerts *et al.* 1989, Steele *et al.* 1997) if the annual average root biomass is near an equilibrium state (Hendrick and Pregitzer 1992) and fine root longevity is not altered (Fahey *et al.* 1999, Tierney and Fahey 2001). To date no method of calculating fine root longevity has been evaluated as to its accuracy (Fahey *et al.* 1999). Thus, all fine root longevity data based on a single cohort should be interpreted with caution.

Concerns have been raised that the installation of minirhizotron tubes may stimulate fine root production because of unintentional pruning, and nutrient release (Joslin and Wolfe 1998, Tierney and Fahey 2001). Installation of minirhizotron tubes in ecosystems that are dominated by perennial plants would cause fine root pruning which would augment translocation of hormones and carbohydrates to the pruned area, causing a root proliferation effect (Hendrick and Pregitzer 1996, Burton *et al.* 2000, King *et al.* 2002, Tierney and Fahey 2001). Installation also creates a root free zone ideal for colonization and the physical characteristics (bulk density, temperature, moisture) inherent to the surrounding soil may be changed particularly in the first 30 cm of the soil (Joslin and Wolfe 1998, Teirney and Fahey 2001, Vogt *et al.* 1998). It has also been suggested that the butyrate tube - soil interface may also have an effect on fine root growth by creating either a favourable (Joslin and Wolfe 1998, Tierney and Fahey 2001) or unfavourable environment (Withington *et al.* 2003). A number of authors have argued that minirhizotron technology does not provide reliable estimates because of the temporary soil disturbance during the installation of tubes, and, because of the subsequent compaction of soil around the tubes causing increased bulk density, poor soil contact, or increasing N availability (Joslin and Wolfe 1999). Installation effects can increase production from 300 to 400 % (Joslin and Wolfe 1999). To minimize this effect, it has been suggested that minirhizotron tubes should be left in the ground for one year without sampling to allow for installation effects to subside (Joslin and Wolfe 1999, Hendrick and Pregitzer 1996). Data in subsequent years tend to be compared to those obtained using other traditional fine root biomass data (Joslin and Wolfe 1999). Coleman et al. (2000), however, have suggested that installation effects can last several years suggesting that longer term studies are required for fine roots to properly colonize minirhizotron tubes.

2.2.3 Ingrowth Bag Method

Fine root ingrowth bags have been used for studying fine root dynamics in forested ecosystems, and in short term studies, the root ingrowth bag method is capable of giving an index of fine root growth potential (Vogt *et al.* 1998). The ingrowth bag method is thought to be a better method of calculating fine root production than the sequential coring method (Vogt and Persson 1991).

Root bags are constructed of 0.2 - 0.6 mm fiberglass mesh nylon and filled with soil with a bulk density representative of the site (Finer and Laine 2000). The soil would also be sieved and free from roots or other litter (Lassoie and Hickley 1991). The ingrowth bags would then be inserted into the ground at specified depths, ensuring that no air pockets are formed (Lassoie and Hickley 1991). Ingrowth bags are sampled periodically and treated similarly to the sequential coring method (Finer and Laine 2000). New growth is calculated by measuring the mass of roots and subtracting the mass of the live roots from the previous sampling (Allen *et al.* 2000). Fine root NPP can then be calculated by dividing by the number of days between sampling periods.

The ingrowth bag method has been criticized by Madji (1996) and Vogt *et al.* (1998) because the soil within the bag usually has higher nutrients values, lower bulk density, and a less competitive environment than surrounding, undisturbed soil.

3.0 STANDARD ROOT LENGTH VARIANCE AMONG NORTHERN TEMPERATE SPECIES

3.1 INTRODUCTION

Standard root length (SRL) is the ratio of root length to mass (Pregitzer *et al.* 2002) and is required when using minirhizotron systems. Manipulations of standard root length values and how they are applied can have significant implications on fine root biomass, NPP, and turnover estimates.

Increased knowledge of fine root SRLs is necessary to provide more accurate estimates of fine root biomass, NPP, and C allocation in northern temperate forests when using minirhizotrons. Few studies have been completed to examine the effect SRL or size classifications would have on current estimates of fine root dynamics. Virtually all studies using minirhizotrons to develop estimates of fine root biomass, NPP and turnover only calculate SRL values for a single diameter class (*e.g.*, < 2.0 mm). The objective of this study was to examine differences in SRLs between 4 northern temperate species (sugar maple - *Acer saccharum* Marsh., jack pine - *Pinus banksiana* Lamb., boreal deciduous – trembling aspen - *Populus tremuloides* Michx. / white birch - *Betula papyrifera* Marsh., boreal conifer – white spruce - *Picea glauca* Moench / balsam fir - *Abies balsamea* (L.) Miller.), and to determine if these SRLs changed between increasing diameter classes (0.2 – 0.4 mm, 0.4 – 0.6 mm, 0.6 – 0.8 mm, 0.8 – 1.0 mm, 1.0 – 2.0 mm, 2.0 mm+).

3.2 MATERIALS AND METHODS

3.2.1 Study Site Description

Three study sites were selected as part of the SRL Study: Turkey Lakes Watershed – sugar maple, Nimitz – jack pine, and Groundhog River – boreal mixedwood, located in northeastern Ontario (Figure 3.1).

The Turkey Lakes Watershed is located in Wishart Township, Ontario, approximately 60 km north of Sault Ste. Marie, Ontario and 13 km inland from Batchewana Bay, Lake Superior (lat. 47°02'N, long. 84°24'W, elevation 393 – 407 m). The site is situated in the Algoma Section of the Great Lakes – St. Lawrence Forest Region (Rowe 1972) in the Superior Climatic Region. The average length of the growing season is approximately 175 days, from May until September (Barrie *et al.* 1984). The

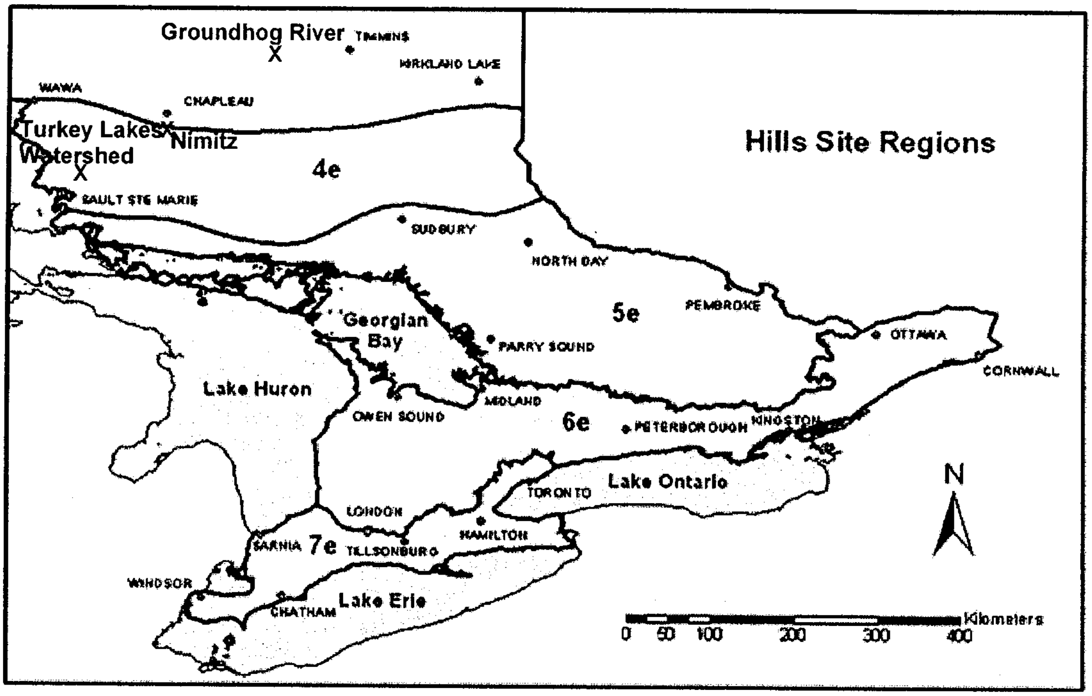

Figure 3.1. The location of the three study sites situated within Hills' Site region 3e and 4e (Hills 1959).

area receives approximately 1224 mm of precipitation annually, with 65 % of this falling during the growing season (Buttle *et al.* 2000). The dominant soil profile on the watershed is an orthic humo-ferric podzol, with well defined L and F horizons (Agriculture Canada Expert Committee on Soil Survey 1987). Species composition of the forest consisted of >90 % mature to overmature sugar maple, 250+ years old, with scattered (<10%) yellow birch (*Betula alleghaniensis* Britton.) and patchy understory shrubs and herbs.

The Nimitz study site is located approximately 28 km south of Chapleau, Ontario on Highway 129 (lat. 47°38'N, long. 83°15'W, elevation 454 m). The site is situated in the Misinaibi – Cabonga section of the Boreal Forest Region (Rowe 1972) in the Height of Land Climatic Region (Chapman 1953). The average length of the growing season is approximately 161 days, from May until September (Morrison *et al.* 1993). Precipitation totals were measured from the nearest weather station (Chapleau, Ontario) and was calculated as 834 mm annually with 53 % of this falling during the growing season (Environment Canada 1982). The soil profile is a very rapidly drained, orthic dystric brunisol (Agriculture Canada Expert Committee on Soil Survey 1987), and is overtopped by 8cm of humifibrimor humus. The area was dominated by an undisturbed, natural pinery (100 % jack pine), dating from a 1922 fire (Morrison *et al.* 1993).

The Groundhog River study site is located in Reeves Township, Ontario, approximately 80 km southwest of Timmins, Ontario (lat. 48°16'N, long. 82°09'W, elevation 315 - 345 m) on the east side of the Groundhog River, north of Highway 101. The study site is in the Missinaibi - Cabonga forest located in the Boreal Forest Region (Rowe 1972). The normal average length of the growing season is approximately 160 days, from May until September (Environment Canada 2007). Normal precipitation totals were calculated as 831 mm annually with 50 % of this falling during the growing season (Environment Canada 2007). The study site is situated on the Groundhog River flood plain. The soil profile is classed as an orthic humo-ferric podzol with a thick (15 cm) organic (fibrihumimor) layer and a deep, coarse fragment free, silty to clayey B horizon. The forest is characteristic of a typical mature, boreal mixedwood forest consisting of various ratios of trembling aspen, black spruce (*Picea mariana* (Mill.) BSP.), white spruce, white birch, and balsam fir. The area was harvested for white spruce in the 1930s, with no follow-up silviculture or renewal efforts (McCaughey *et al.* 2006).

3.2.2 Soil Core Collections

In the sugar maple stand at the Turkey Lakes site, three random soil cores per 12 – 10m x 10m plots were taken in September, 1999 for a total of 36 cores. A soil corer with a total length of 45 cm and an inside diameter of 48 mm was used. A hinged plastic sleeve was placed inside the corer to ensure that the cores would remain intact when they were being extracted. Forest floor hummocks and depressions were avoided during sampling.

Root samples were returned to the laboratory within six hours and stored in a refrigerator at 4° C. Roots collected were washed using a Mini-Hydroelute Rootwashing Unit (Gillisons Variety Fabrication Inc., Benzonia, Michigan). The Mini-Hydroelute Rootwashing Unit uses forced air and water to break up soil cores. Roots are then strained from the muddy water by using 1 mm and 0.5 mm sieves. All roots were separated from any remaining litter. Roots appearing to be dead or were > 5.0 mm in diameter were discarded. All live tree root samples were keyed out as sugar maple. Although some material may have been incorrectly keyed to sugar maple rather than yellow birch, it was felt that this would have been a rare occurrence as yellow birch was only a scattered component of the stand. Other plant roots (*i.e.*, shrubs and herbs) were discarded. Roots were then dried at 70° C to constant weight and weighed (Johnson *et al.* 2001, Tierney and Fahey 2001). Roots less than 2.0 mm in diameter were scanned (digitized) using a Hewlett Packard Scanjet Scanner with associated software in 2 bit black and white format, and, measured using the MacIntosh OS9 based NIH Image 1.62 software (http://rsb.info.nih.gov/nih-image/).

At the Nimitz study site, three random soil cores were taken from each of 9 – 10m x 10m plots in the summer of 2004. Fine roots were separated from the soil using high pressure tap water and a fine mesh screen, and evaluated by diameter class (0.2 – 0.4 mm, 0.4 – 0.6 mm, 0.6 – 0.8 mm, 0.8 – 1.0 mm, 1.0 – 2.0 mm, 2.0 mm+). All tree root samples were keyed out to jack pine. The image analysis software used was ImageJ. All changes to image parameters were accounted for.

At the Groundhog River study site, four random soil cores were taken from each of 3 plots. At this site only, 5 repeat sampling sets were collected (August and September, 2005 and May, June and July, 2006) to examine seasonal variability in SRL. In addition, the individual cores were separated into 3 sections: Organic horizon, upper 0-10 cm of mineral horizon, and >11 cm (core length was typically 35 to 50 cm depending on compaction). Processing of roots was similar to that was done for the cores collected from the Nimitz site, including partitioning into diameter classes. In this case, roots were keyed out to deciduous (intolerant: aspen or birch) or conifer (tolerant: spruce or balsam fir). Deciduous species were considered to be those that were generally, thinner, lighter in colour, less gnarled, with longer intermodal lengths. Coniferous species were considered to be those that were generally, thicker, darker in colour (thicker epidermis), gnarled, with shorter intermodal lengths. Further information can be found in Pregitzer et al. (2002).

3.2.3 Statistical Approach

The changes in the fine root sampling and processing procedures from the initial sampling in 1999 at the Turkey lakes site (sugar maple stand) to the final sampling in 2005/6 at the Groundhog River site (boreal mixedwood stand) required a tiered approach to the analysis. The full, 4-factor ANOVA model (species, diameter class, soil depth, sampling date) could only be run on the SRL data collected from the boreal mixedwood site. Based on the results from this analysis and the non-significance of soil depth and sampling date and all interactions (see Appendix 1), a 2-factor ANOVA model (species and diameter class) was applied to a combined data set for Nimitz (jack pine stand) and Groundhog River (boreal mixedwood). Finally, a 1-way ANOVA model (species) was performed on the complete data set. The PROC GLM procedure of SAS/STAT software (version 9.1) was used to perform the ANOVAs. Post-hoc examination of significant factors was done using the Student-Newman-Kuels (SNK) multiple range test.

3.3 RESULTS

Table 3.1 summarizes the average SRLs by species and diameter class. Based on the results from the full (4-factor) ANOVA model completed on the Groundhog River data, neither soil depth (F ratio = 2.51, $p = 0 .082$) or sampling date (F ratio = 1.64, $p = 0.162$) had a significant influence on SRLs (see Appendix 1 for complete ANOVA results). In addition, all interactions had p values > 0.10, with many greater than 0.9. As a result, the reduced (2-factor) model was applied to the combined dataset to focus on the two main factors of interest: species and diameter class. Both main factors had highly significant influences on SRL values (species: F ratio = 5.50, $p = .0042$; diameter class: F ratio = 94.13, $p < 0.0001$). Their interaction term, however was not significant (F ratio= 1.27, $p = 0.240$), allowing for a further (post-hoc) examination of these two main factors.

Table 3.1. Average standard root lengths (standard errors in brackets) for the species and diameter classes included in this study. Units are in m · g^{-1}.

Diameter Class	Species Ms (tolerant)	 Deciduous (intolerant)	 Conifer (tolerant)	 Pj (intolerant)
0.2 – 0.4	na	12.71 (1.078)	9.86 (.368)	7.99 (3.534)
0.4 – 0.6	na	5.64 (.272)	6.28 (.825)	3.62 (1.433)
0.6 – 0.8	na	4.41 (.388)	3.48 (.179)	2.83 (1.021)
0.8 – 1.0	na	3.04 (.178)	3.23 (.230)	3.13 (1.507)
1.0 – 2.0	na	1.90 (.114)	1.67 (.157)	0.78 (.209)
> 2.0	na	0.57 (.051)	0.66 (.066)	0.37 (.212)
All Classes	10.11 (.566)	5.44 (.200)	5.24 (.281)	2.45 (1.025)

Figure 3.2 illustrates the differences in average SRL values obtained for the range of tree species (or species groups) included in this study. A considerable range (10.11 to 2.45 m · g^{-1}) in SRL was detected with sugar maple having a significantly higher SRL than all other species considered in this study. It is notable that within the hardwood and conifer groupings, the tolerant species had significantly higher SRLs when compared to its intolerant counterpart.

As might be expected, there was a clear downward trend in SRLs as fine root diameter increased (Figure 3.3). The SRL dropped rapidly from over 11 m · g^{-1} for the smallest diameter class (0.2 – 0.4 mm) to under 6 m · g^{-1} for the second class (0.4 – 0.6

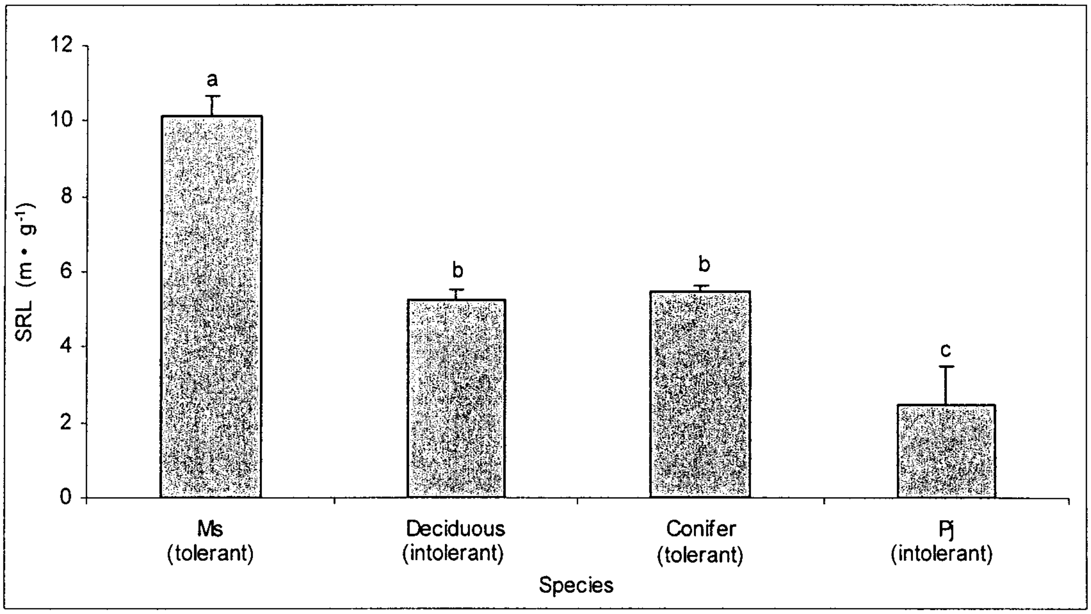

Figure 3.2. Average SRL values for the species included in this study. Species include: Ms – sugar maple, tolerant hardwood ; deciduous, intolerant hardwood (predominately trembling aspen) ; conifer, tolerant conifer (mixture of boreal spruce and balsam fir) ; Pj – jack pine, intolerant conifer.

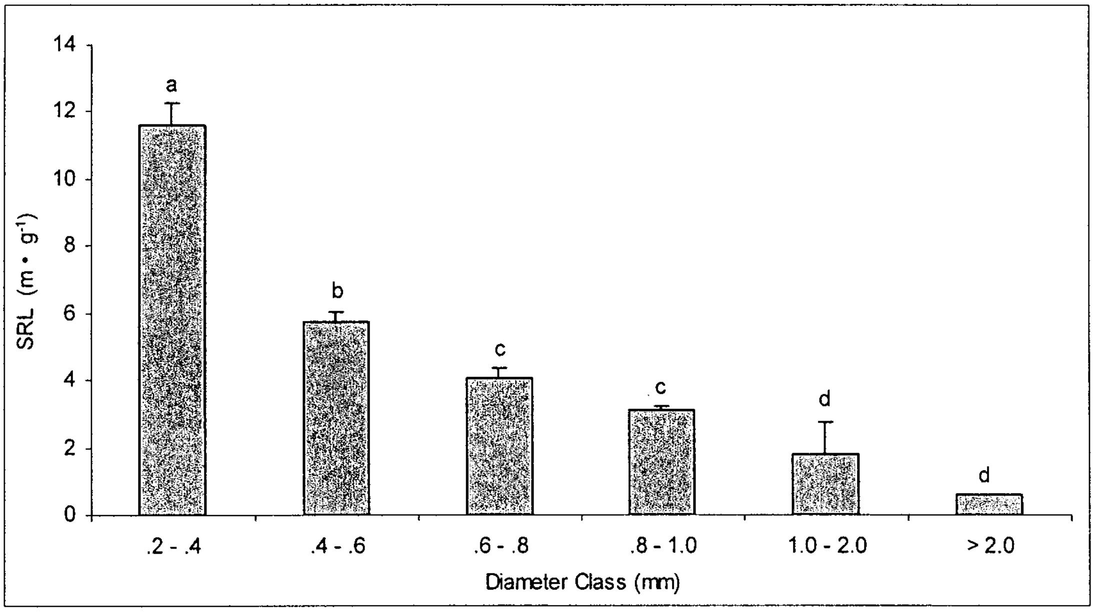

Figure 3.3. Average SRL values for the range of diameter classes included in this study.

mm), then continuing with a more gradual decline to under 0.6 m · g^{-1} in the final class (> 2.0 mm).

3.4 DISCUSSION

To date, few other studies have documented standard root lengths. Virtually all studies using minirhizotrons to develop estimates of fine root biomass, NPP and turnover only calculate SRL values for a single diameter class (*e.g.*, < 2.0 mm). A summary of other fine root standard root lengths for nine North American species was complied by Pregitzer *et al.* (2002) (Table 3.2). The range of SRLs reported by Pregitzer *et al.* (2002) are considerably higher than the values reported in the current study. For example, sugar maple values in Table 3.2 range from 45 to 65 m · g^{-1} compared to this study's average SRL (all diameter classes) of 10.1 m · g^{-1}. For the current study, fine root diameters up to and including 2.0 mm in size were included in the SRL measurements which would tend to lower the average SRL compared to those values reported in Table 3.2 (*i.e.*, only included fine roots less than 1.0 mm in diameter). In addition, it is possible that some of our numbers may have been low when too much fine root material was scanned at one time, resulting in low length estimates using the 2 Bit image analysis.

However, based on the relatively large number of samples processed and the small standard error bars depicted in Figure 3.2, we are confident in the values generated for the species and sites included in this study. Although speculative at this stage, it seems reasonable that other factors such as climate, soil type, and other local site factors may influence fine root length to biomass ratios for a given species based on findings regarding the effects of climate on wood density (Fritts *et al.* 1991).

Table 3.2. Summary of standard root lengths (SRLs) for nine North American species (adapted from Pregitzer *et al.* 2002).

Species	Site	Diameter Class	SRL
Acer saccharum	Michigan, U.S.A.	< 1.0 mm	~ 45 - 65
Liriodendron tulipifera	North Carolina, U.S.A.	< 1.0 mm	~ 10 - 15
Populus balsamifera	Alaska, U.S.A.	< 1.0 mm	~ 55 - 105
Quercus alba	Georgia, U.S.A.	< 1.0 mm	~ 30 - 90
Juniperus monosperma	New Mexico, U.S.A.	< 1.0 mm	~ 12 - 22
Picea glauca	Alaska, U.S.A.	< 1.0 mm	~ 20 - 35
Pinus edulis	New Mexico, U.S.A.	< 1.0 mm	~ 10 - 22
Pinus elliottii	Florida, U.S.A.	< 1.0 mm	~ 18 - 38
Pinus resinosa	Michigan, U.S.A.	< 1.0 mm	~ 22- 38

Although previous studies have suggested that fine root SRL varies seasonally (Tierney and Fahey 2001, Wang *et al.* 2006), as well as decreasing with increasing soil depth (Baker *et* al. 2001), the results from the current study did not detect any significant shifts in SRL for these sampling parameters. It should be noted, however, that the seasonal sampling was conducted only on the boreal mixedwood site (Groundhog River), and sampling spanned 2 years: late summer and fall sampling in 2005 and spring and early summer sampling in 2006. Thus, additional work, particularly for seasonal patterns is warranted.

Species-level differences in SRL have been reported in the literature, most commonly reporting that angiosperms (hardwoods) have significantly higher SRLs when compared to gymnosperms (Bauhus and Messier 1999, Pregitzer *et al.* 2002). Results from the current study are consistent with these reported findings. Angiosperm average

SRL (sugar maple ; boreal hardwoods – aspen, white birch) ranged from 10.1 to 5.4 m · g^{-1} compared to the values for the gymnosperms (tolerant conifers – spruce, balsam fir ; jack pine) at 5.2 to 2.5 m · g^{-1}. Studies have also reported that fast growing species that favour rapid soil exploration tend to have higher SRLs (Comas and Eissenstat 2004, Wardle *et al.* 2002, Nicotra *et al.* 2002). Although not explicitly stated, this would suggest that intolerant, early successional species would have high SRLs when compared to their tolerant, late successional counterparts. In the current study, the opposite was true, with the tolerant, late successional species having significantly higher SRLs for both the angiosperm (sugar maple – 10.1 m · g^{-1} vs intolerant hardwoods – 5.4 m · g^{-1}) and gymnosperm (tolerant conifers – 5.2 m · g^{-1} vs intolerant jack pine – 2.5 m · g^{-1}) groupings. This, in turn, would suggest that these species have developed rooting traits (*i.e.*, high length to mass ratios) to more fully access the soil profile which would provide for greater competitive abilities particularly when growing in species mixtures.

For a given species, Pregitzer *et al.* (1997, 1998) suggested that fine root SRL decreases with increased diameter. In their studies, fine roots in the 0.2 to 0.4 mm class had average SRL values that were approximately ten times higher than those for the1.0 to 2.0 mm diameter class, and, twenty times for the those roots in a 2.0 to 5.0 mm diameter class. This decreasing pattern in SRL was consistent across the range of species included in the current study, dropping rapidly by the second diameter class (0.4 – 0.6 mm), in most cases by greater than 200 %, then a more gradual but significant decline through to the largest diameter class. The inclusion of a diameter class-based SRL would greatly approve our accounting efforts with respect to fine root biomass and belowground carbon, leading to a better understanding of the temperate forest C dynamics.

It is important to recognize that very little work has been done to determine species- or site-specific SRLs. Based on the limited understanding of fine root architecture (*i.e.*, quantity of fine roots with regards to radius and length) and biomass allocation, many of the current estimates of fine root biomass from minirhizotrons may be inaccurate and could have a significant effect on fine root C accounting. Common to fine root research studies that utilize minirhizotron systems is that they use published SRL values to covert their measured root lengths to biomass. If this approach was done for the current study fine root biomass would have been substantially underestimated. Depending on the species, this underestimate could be as much as a 5 fold difference for sugar maple (current study SRL – 10.1 $m \cdot g^{-1}$; published SRL – 45 to 65 $m \cdot g^{-1}$) and as much as a 10 fold difference for jack pine (current study SRL – 2.5 $m \cdot g^{-1}$; published SRL for pine – 22 to 38 $m \cdot g^{-1}$).

3.5 CONCLUSIONS

It was determined that there was a significant difference in fine root standard root length (SRL) between northern temperate tree species and diameter classes. Angiosperms tended to have significantly higher average SRLs compared to gymnosperms. Within these species groupings, tolerant, late successional species had higher SRLs compared to their intolerant, early successional counterparts. Standard root lengths dropped significantly (>200 %) as diameter class increased. The results suggest that the development of species- and diameter class-specific SRLs should provide better estimates of fine root biomass and productivity, leading to a better understanding of temperate forest C dynamics.

4.0 COMPARISON OF COMMONLY USED METHODS FOR ESTIMATING FINE ROOT BIOMASS, PRODUCIVITY, AND TURNOVER.

4.1 INTRODUCTION

Although numerous studies have examined aboveground biomass and productivity, very few studies have provided belowground estimates, particularly with respect to fine roots (Ruess *et al.* 1996). Studies on tree roots are often overlooked due to the time and cost associated with this type of sampling (Baker *et al.* 2001, King *et al.* 2002). As an alternative, many studies use indirect methods to obtain belowground estimates by applying theoretical equations to aboveground results to develop total site biomass relationships (Kurz *et al.* 1996, Li *et al.* 2002, Chen *et al.* 2004). Hendrick and Pregitzer (1993a), however, have suggested that both fine root biomass and productivity ratios can vary greatly depending on the methodology utilized to collect data. To date, there have been limited studies that use common forested plots to directly compare the fine root estimates generated from the different methods in northern forested ecosystems.

The objective of this study was to compare the most common indirect and direct methods used to estimate fine root biomass, fine root productivity, and fine root turnover using the same plots in both a hardwood-dominated (sugar maple) and a conifer-dominated (jack pine) stand. Specific research questions include: 1) do the various methods provide similar stand-level estimates of the fine root parameters considered, and 2) if not, are these estimates strongly correlated to each other (*e.g.*, consistently over- or under-estimated)?

4.2 MATERIALS AND METHODS

4.2.1 Study Site Descriptions

This method comparison study used the same sugar maple-dominated stand located on the Turkey Lakes watershed and the jack pine-dominated stand near Chapleau, ON. Described in Chapter 3.

4.2.2 Sampling Protocols for Direct and Indirect Measures of Fine Root Biomass, Productivity, and Turnover

Minirhizotron (MR) Tubes

In the sugar maple stand, three acrylic MR tubes (two metres in length with an inside diameter of 50 mm) were installed in six fixed area plots (18 tubes in total) in late November and early December of 1997. For the jack pine stand, five MR tubes were installed in a random circular fashion (diameter approximately 5 m) in each of 3 fixed area plots (15 tubes in total). The tubes were installed to a maximum depth of 40 cm at an angle of 45° (Figure 4.1). The tubes were sealed at one end with a rubber stopper and silicone sealant to prevent moisture from seeping into the tube. Tubes were painted black, overlain with white paint and capped with an aluminum can to reduce heat and light transfer to the soil. Wooden supports were also installed to reduce the likelihood of tubes breaking under heavy snow loads.

For the sugar maple stand, MR tubes were sampled (digital photos taken) monthly in 1998, 1999 and 2000 from May to October. A similar monthly sampling regime was used for the jack pine stand in 2001 through 2003. Images were collected at 13 mm intervals along the entire belowground length of the tube using the minirhizotron camera system and Smucker Indexing Rod (Bartz Technology Inc., Santa Barbara, California).

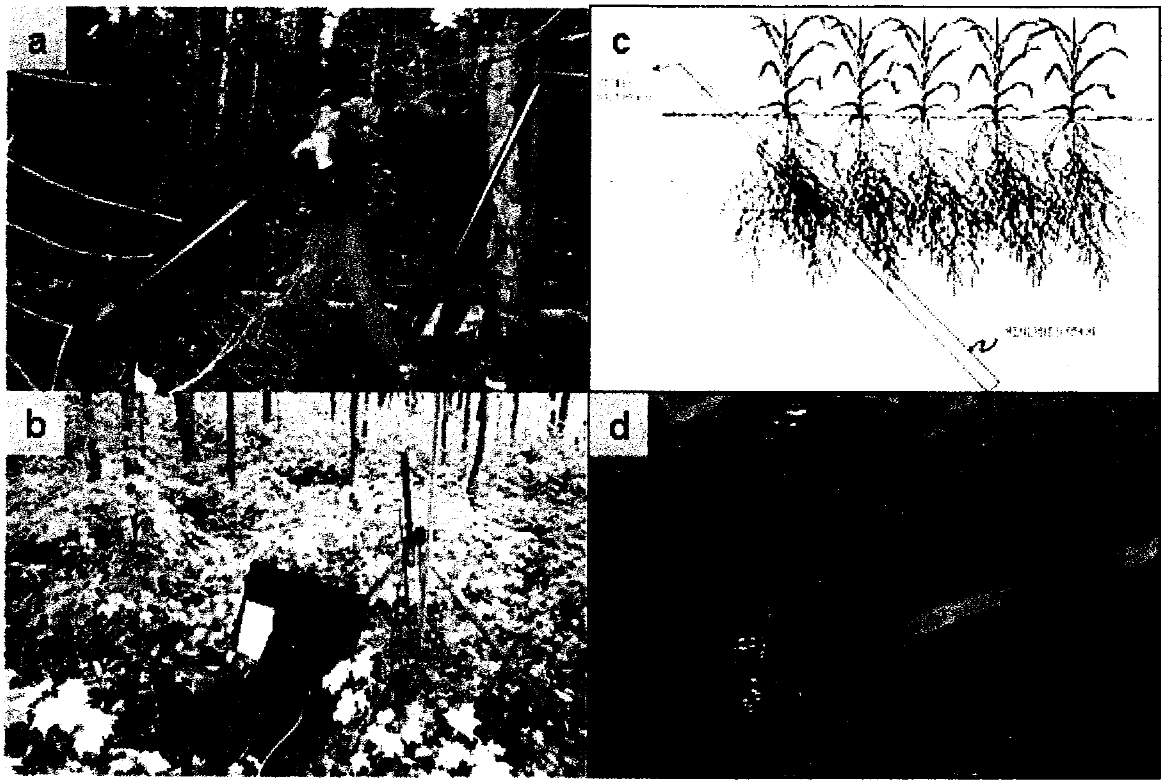

Figure 4.1. Photograph showing an *in situ* minirhizotron installation (A), setup (B), a schematic showing the camera placed within the rooting zone (C), and a digital root image (D).

In 1998 and 1999 images were recorded onto a Hi-8 video-cassette and then later transcribed into digital format using Adobe Premiere®5.1. From 2000 on, images were recorded directly into digital format using Adobe Premiere® 5.1 and a MacIntosh G3 laptop. Root images were subsequently analysed using RooTracker 2.0 (Duke University), an interactive MacIntosh based program (Craine and Tremmel, 1995). Fine root location, length, diameter, and age classification of all roots growing across the field of view were recorded. Changes in size, morphology and age (*i.e.*, new, mature or dead) of each root were tracked through successive sample intervals. Ultra-fine roots (< 0.2 mm) and roots of other non-target species were not recorded. Morphology and branching patterns were used to distinguish sugar maple and jack pine roots. "New" roots were those that had not been found in the same frame on previous sampling dates. Roots that continued to be found in subsequent frames were then described as "mature" or "dead". "New" roots were normally white in colour while "mature" roots tended to be light to dark brown depending on their age and degree of suberization. A "dead" root description was used when a root disappeared from the frame, was very faint, shrivelled or black in colour. The appearance date of a "new" root and "death" date for a dead root was assumed to be the mid-point between successive image dates.

Soil Cores

Three soil cores per plot (18 in total) were taken monthly in 1998 and 1999 at the beginning of each month from June to October in the sugar maple stand. Sampling in the jack pine stand was done only once during the summer of 2004, with 3 cores taken from each of three plots. The soil corer used in the sampling had a total length of 45 cm and an inside diameter of 48 mm. A hinged plastic sleeve was placed inside the corer to ensure that the cores would remain intact when they were being extracted.

Root samples were returned to the laboratory within six hours and stored in a refrigerator at 4° C. Roots were washed using a Mini-Hydroelute Rootwashing Unit (Gillisons Variety Fabrication Inc., Benzonia, Michigan). Roots are then strained from the muddy water by using 1.0 and 0.5 mm sieves. All roots were separated from any remaining litter. Roots appearing to be dead or were > 5.0 mm in diameter were discarded. Roots were then dried at 70° C for 24 hours and weighed.

In-growth Bags (sugar maple stand only)

In May, 1999, 216 in-growth bags (36 bags per plot) were installed to a depth of 40cm. The bags were made of nylon mesh (0.75mm x 1.0mm) and had a diameter of approximately 5.0cm. The in-growth bags were filled with dried soil collected from the sugar maple site. The soil was gently packed in the bag to approximate the undisturbed soil bulk density.

Sets of bags were extracted from the field on different dates: August 12, 1999 (78 days), October 20, 1999 (147 days), June 6, 2000 (375 days), and August 22, 2000 (452 days). Extracted in-growth bags were returned to the laboratory within six hours and stored in a refrigerator at 4°C. In-growth cores (*i.e.*, material in the bags) were processed and extracted using the same method applied to the soil cores.

Plot Inventory Data

The indirect methods of Kurz *et al.* (1996) and Li *et al.* (2002) require an estimate of aboveground biomass ($kg \cdot ha^{-1}$). All trees with a Dbh $\geq$ 9 cm were included in the tree inventory conducted on each fixed area plot (TLW: 6 – 15 m radius plots, Nimitz = 5 – 11.3 m radius, Groundhog: 3 -11.3 m radius plots). The individual tree diameters measured on each plot were applied to species-specific aboveground biomass equations reported in Ter-Mikaelian and Korzukhin (1997), then summed to provide plot-level estimates of aboveground biomass following the double sampling approach outlined by Baskerville (1972) .

Re-measurements of the plots five years later provided an estimate of aboveground NPP by subtracting biomass estimates for T1 from T2, and dividing by the

5 year time interval. Since these stands are mature (steady state), stand-level biomass increment was minimal and since litterfall was not collected, NPP would have been underestimated.

4.2.3 Data synthesis and analysis

Table 4.1 provides a summary of the conversion equations required by the two indirect methods (Kurz *et al.* 1996 and Li *et al.* 2002) to estimate fine root biomass, productivity, and turnover.

The processing of the minirhizotron images provides a measure of the total sum length of all living root material present on a particular frame or with the field of view on a given sample date (Hendrick and Pregitzer 1993, Joslin and Wolfe 1999). Fine root biomass density was then calculated by dividing the fine root length density by the standard root lengths for the particular species and diameter classes, as developed in Chapter 3. There have been four methods published in the literature to estimate fine root productivity using minirhizotron data. For the purpose of comparison, the sequential sampling method ($FR_{NPP} = FRBD_{t+1} - FRBD_t / t$), as described by Hendrick and Pregitzer (1992) and Joslin and Wolfe (1999) was used. Fine root turnover was calculated as the $\sum$ Dead / t (Hendrick and Pregitzer 1992, Publicover and Vogt 1992).

For the soil core method, fine root biomass density is calculated as the total sum mass of all living root material in a particular soil core or layer on a sample date per unit area (Hendrick and Pregitzer 1993a, Joslin and Wolfe 1999). Fine root productivity used the sequential coring method, as above. Turnover was the ratio of fine root productivity

Table 4.1. Equations used to estimate fine root biomass, NPP, and turnover for the two indirect methods evaluated in this study.

Parameter	Equation	Reference
FRBD - Kurz	$RB_s = 0.2317ABD_s$ $RB_h = 1.432ABD_h^{0.639}$ $FRP = 2.737 \times RB^{-0.841}$ $FRBD = RB \times FRP$	Kurz *et al.* (1996)
FRBD – Li	$RB_s = 0.222ABD_s$ $RB_h = 1.576ABD_h^{0.615}$ $FRP = 0.072 + 0.354e^{-0.060RB}$ $FRBD = RB \times FRP$	Li *et al.* (2003)
FRNPP – Kurz and Li	$FR\ NPP = FRBD_{t+1} - FRBD_t / t$	Hendrick and Pregitzer (1992)
FRT – Kurz	$FRT = 0.735FRBD$	Kurz *et al.* (1996)
FRT – Li	$FRT = 0.641FRBD$	Li *et al.* (2002)

FRBD = Fine Root Biomass Density; ABD = Aboveground Biomass Density; RB_s = Softwood Root Biomass; RB_h = Hardwood Root Biomass; FRP = Fine Root Proportion; FR NPP = Fine Root Net Primary Productivity; FRT = Fine Root Turnover

divided by the estimate of the fine root biomass pool (Ruess *et al.* 1996, Ostonen *et al.* 2005).

The ingrowth bag method provided an estimate of fine root productivity as the mass of all living "ingrown" root material, converted to a per annum basis.

A series of Paired Comparison T-tests were performed to determine if the estimates provided by the various methods were significantly different from each other (H_o: Difference = 0) for each of the stand types sampled. Pearson correlation coefficients (r) were generated to determine if the various methods consistently under- or over-estimated fine root biomass. A one-way ANOVA was performed on the ingrowth bag estimates of fine root productivity to determine if time of placement (# of days between installation and retrieval) influenced the productivity estimates. Post-hoc examination was performed using the Student-Newman-Kuels (SNK) multiple range test.

4.3 RESULTS AND DISCUSSION

4.3.1 Comparison of Fine Root Biomass Estimates

Figure 4.2 compares the stand-level average fine root biomass estimates obtained from the four sampling methods used in the study. For the most part, the indirect methods of Kurz *et al.* (1996) and Li *et al.* (2002) significantly under-estimated fine root biomass when compared to either of the direct methods (Table 4.2).

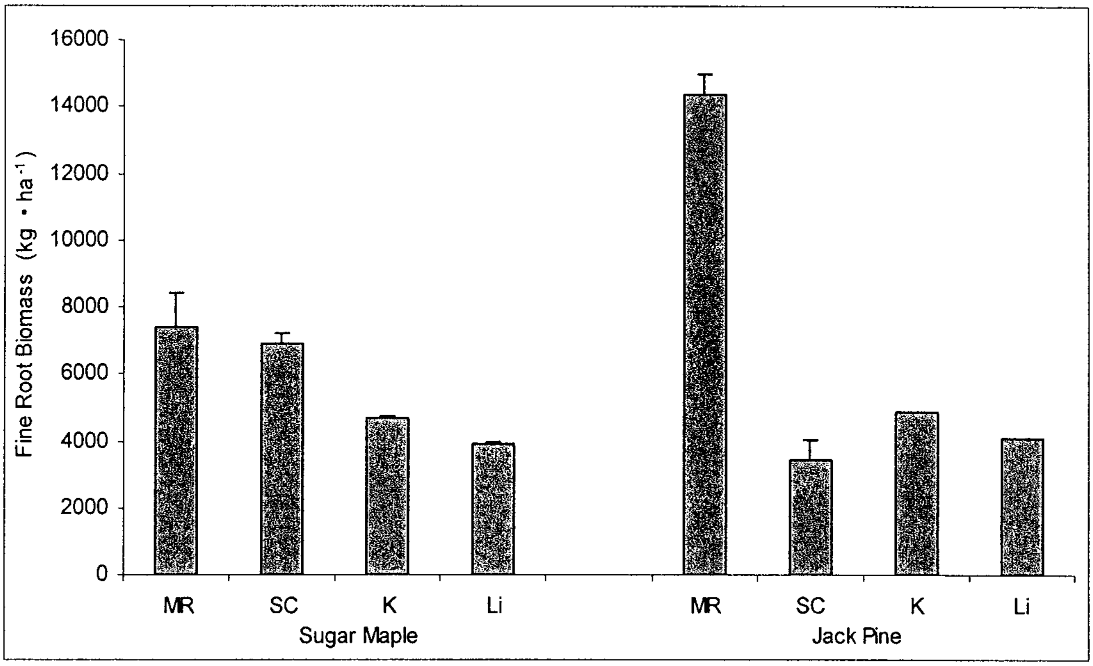

Figure 4.2. Fine root biomass estimates obtained from the two direct (MR – minirhizotron ; SC – soil core) and two indirect (K – Kurz ; Li) methods used in this study for the sugar maple and jack pine stands.

Table 4.2. Summary of paired t-test results comparing the fine root biomass estimates for the four sampling methods (MR – minirhizotron ; SC – soil core ; Kurz ; Li) included in this study.

Method Comparison	Mean Difference	T-Value	P > t
Sugar Maple			
MR – SC	482.6	0.51	0.6293
MR – Kurz	2692.1	2.75	**0.0404**
MR – Li	3479.4	3.58	**0.0159**
SC – Kurz	2209.6	6.68	**0.0011**
SC – Li	2996.8	9.05	**0.0003**
Kurz - Li	787.3	76.84	**<0.0001**
Jack Pine			
MR – SC	10967.3	43.58	**0.0005**
MR – Kurz	9217.6	10.30	0.0616
MR – Li	10028.1	11.20	0.0567
SC – Kurz	1965.2	-2.93	0.2092
SC – Li	1154.6	-1.72	0.3349
Kurz - Li	810.6	2315.9	**0.0003**

For example, in the sugar maple stand, the direct methods generated estimates of approximately 7,000 kg ha^{-1} and are within the range of published estimates from other sugar maple stands (Table 4.2: 3,200 to 9,500 kg · ha^{-1}). The mean difference between the estimates provided by the two direct methods (*i.e.*, minirhizotron versus soil coring) was less than 500 kg · ha^{-1} (Table 4.2). In comparison, the indirect methods produced estimates on the low end of the published estimates, and nearly 3,000 kg · ha^{-1} lower than the direct methods.

The methods comparison conducted in the jack pine stand yielded different results. In this case, the soil core along with the two indirect methods generated similar estimates (3,000 – 5,000 kg · ha^{-1}), which are comparable to other published values for conifer stands (Table 4.3). Table 4.3, however, only reports on fine root biomass data obtained from the soil core procedure. The estimates generated using the minirhizotron system provided considerably higher estimates of fine root biomass in excess of 14,000 kg · ha^{-1}.

Table 4.3. Summary of published fine root biomass estimates (kg · ha^{-1}) from other studies in northern forested ecosystems.

Forest Type	Location	Method	FRBD	Reference
Sugar Maple	Wisconsin, U.S.A.	soil core	3,230 – 4,280	Aber *et al.*, 1985
Sugar Maple	Michigan, U.S.A.	MR	6,887 - 9,530	Hendrick and Pregitzer (1993)
Oak - Maple	Massachusetts, U.S.A.	soil core	6,100	Aber *et al.*, 1985
Red Pine	Wisconsin, U.S.A.	soil core	4,020 - 4,410	Aber *et al.*, 1985
Red Pine	Massachusetts, U.S.A.	soil core	5,100	Aber *et al.*, 1985
Red Pine	Wisconsin, U.S.A.	soil core	590 – 4,310	Haynes and Gower (1995)
Scots Pine	Finland	soil core	2,750 - 4,000	Finer and Laine (2000)
White Pine	Wisconsin, U.S.A.	soil core	3,720	Aber *et al.*, 1985
White Pine	Wisconsin, U.S.A.	soil core	2,890	Aber *et al.*, 1985
Norway Spruce	Estonia	soil core	1,420	Ostonen *et al.* (2005)

There is considerable variability associated with the estimates of fine root biomass even with the direct methods. This expression of variability associated with the two indirect methods is exclusively a function of the between-plot variability in the aboveground biomass estimate. The direct measurements, which include multiple sampling points within each plot combined with repeated measures over time, incorporate expressions of within-plot, between-plot, and temporal variability thereby providing a better overall "stand-level" average. Figure 4.3 and 4.4 provide an illustration of these

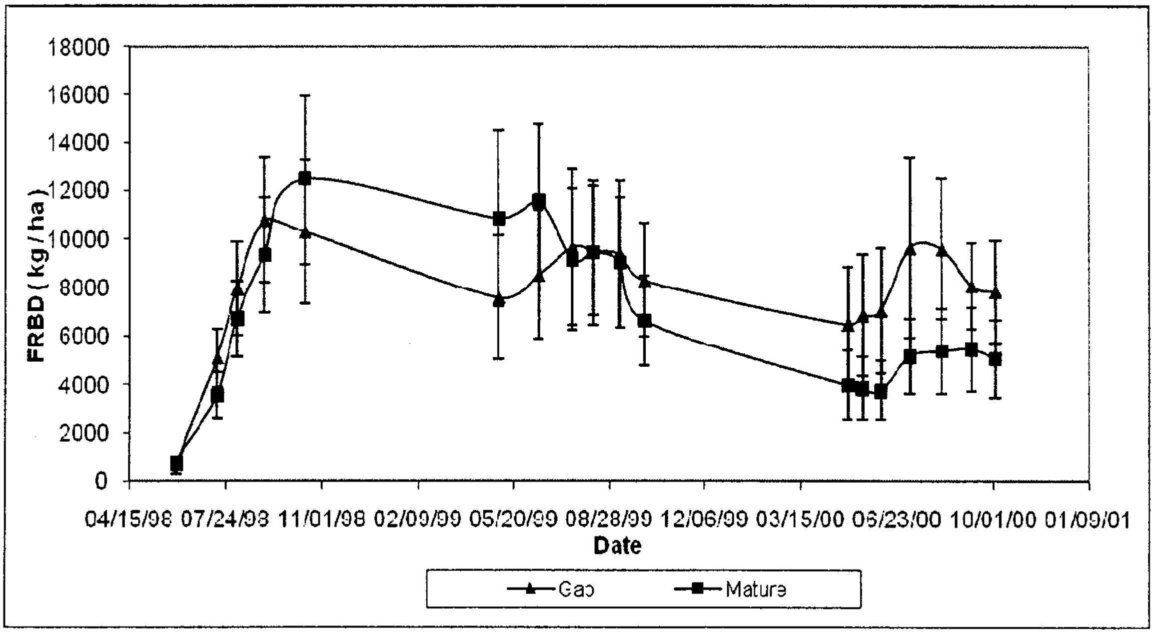

Figure 4.3.Temporal patterns, and associated variability depicted by standard error bars, in fine root biomass (FRBD) in the sampled sugar maple stand obtained from repeated measures of the minirhizotron tubes.

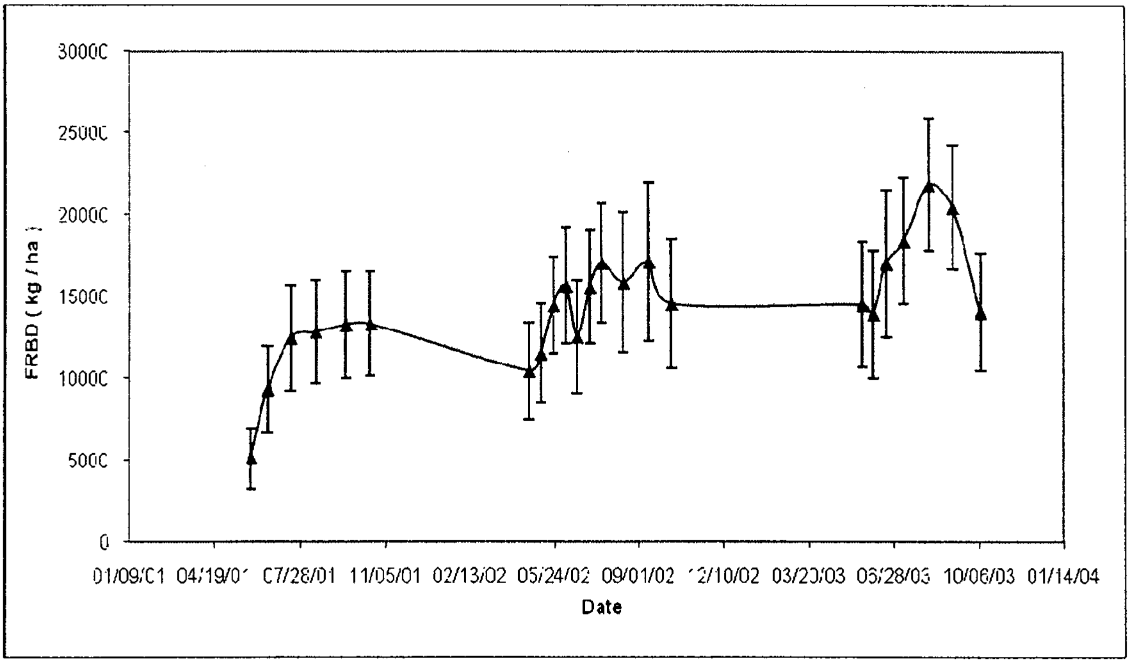

Figure 4.4.Temporal patterns, and associated variability depicted by standard error bars, in fine root biomass (FRBD) in the sampled jack pine stand obtained from repeated measures of the minirhizotron tubes.

patterns generated from the minirhizotron data for both the sugar maple and jack pine stands, respectively.

Joslin and Wolfe (1998), and Tierney and Fahey (2001) had concerns about minirhozotron estimates to be artificially high due to unintentional nutrient release and root pruning during tube installation. Hendrick and Pregitzer (1996), Burton *et al.* (2000), King *et al.* (2002), and Teirney and Fahey (2001) have suggested that roots may proliferate around a minirhizotron tube due to pruning as well. Installation also creates a root free zone ideal for colonization, and the physical characteristics (bulk density, temperature, moisture, etc.) inherent to the surrounding soil may be changed particularly in the first 30 cm (Joslin and Wolfe 1998, Teirney and Fahey 2001, Vogt *et al.* 1998). Joslin and Wolfe (1998), Tierney and Fahey (2001), Withington *et al.* (2003) suggested that the butyrate (minirhizotron) tube –soil interface may also have an effect on fine root growth and biomass density. Joslin and Wolfe (1999) have noted that minirhizotron estimates can exceed other estimates by as much as 300 to 400 %. They suggest that this may be a result of soil disturbance during tube installation, causing increased nitrogen availability (Joslin and Wolfe 1999). Also, since auger holes cannot be drilled through course soil material, tube installations may favour more productive sites. This may help explain why minirhizotron fine root biomass estimates are higher than soil core fine root biomass estimates in both the sugar maple and jack pine forests. It does not, however, explain why the jack pine fine root biomass estimates from the minirhizotrons greatly exceeded those reported for other pine forests (Table 4.3).

Several factors may have had an affect on the fine root biomass estimates. These factors include, but are not limited to: tree species, age, sampling date, climatic variables, mycorrhizal associations, vegetation associations, soil type, parent material, slope, aspect, microtopography, soil moisture and drainage, type and depth of forest humus, and disturbance history. How these factors affect carbon partitioning within trees is not well understood (Hendrick and Pregitzer 1996). Due to the diverse and dynamic nature of these factors it may be very difficult to calculate fine root biomass estimates accurately (*i.e.*, predictable estimates with a high level of certainty). Bohm (1979) echoes these statements, and emphasizes that due to the extreme labour intensity associated with this type of sampling, individual research projects tend to have highly restricted inference space (*i.e.*, estimates relevant only to the stand(s) sampled). Destructive augmentation of a site will also limit the duration of fine root experiments (Bohm 1979).

4.3.2 Comparison of Fine Root Productivity Estimates

For this evaluation, only the sugar maple stand was considered as it provided estimates for three direct and the two indirect methods. Based on the summary data presented in Figure 4.5, the estimates of fine root productivity (FR_{NPP}) appear to be more problematic than those for fine root biomass, particularly for the indirect methods. These methods simply rely on the measured change in aboveground biomass over a specified sampling period (*e.g.*, 5 year remeasurement period on permanent sample plots). Although this may be appropriate in aggrading systems, in the case of mature, steady state conditions as was used in this study, measured aboveground biomass increment (Δ live tree biomass + ingress – tree mortality) was minimal resulting in FR_{NPP} near zero. The direct methods provided comparable estimates of FR_{NPP} ($200 - 800$ kg $\cdot$ ha^{-1} $\cdot$ yr^{-1}), but was on the low end of the published values for sugar maple (Table 4.4).

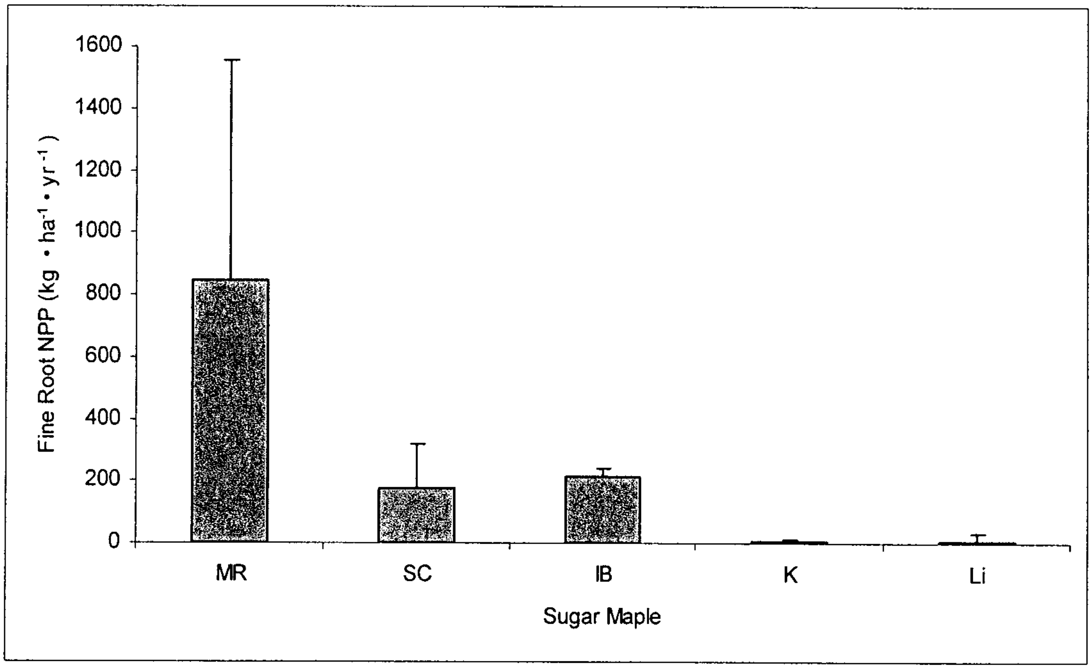

Figure 4.5. Fine root productivity estimates obtained from five methods (MR – minirhizotron ; SC – soil core ; IB – ingrowth bag ; K – Kurz ; Li) used in this study for the sugar maple stand only.

The FR_{NPP} estimate obtained from the minirhizotron measurements was higher than either the sequential soil coring or ingrowth bag method. This difference, however, was not statistically different, largely due to the high variability associated with this method, illustrated in Figure 4.6, where the repeated measures at both the individual tube and plot levels generated extreme positive to negative estimates.

The ingrowth bag method provided the most consistent stand-level estimates compared to the other two direct methods, and integrates the temporal variability into the design (*i.e.*, ingrowth bags are in place to capture the peaks and valleys of fine root growth over an annual basis). The one question frequently asked when considering this method, is: what is an appropriate time interval for *in situ* placement, particularly with

Table 4.4. Summary of published fine root productivity estimates (kg · ha^{-1} · yr^{-1}) from other studies in northern forested ecosystems.

Forest Type	Location	MR	Ingrowth	Reference
Birch	Wisconsin, U.S.A.	1560		Nadelhoffer *et al.* 1985
Red Maple	Wisconsin, U.S.A.	1100		Aber *et al.* 1985
Sugar Maple	Michigan, U.S.A.	7300 - 8100		Hendrick and Pregitzer 1993
Red Pine	Wisconsin, U.S.A.	4070		Gower *et al.* 1992
Red Pine	Wisconsin, U.S.A.	690 - 2500		Aber *et al.* 1985
Red Pine	Wisconsin, U.S.A.	940 - 2500		Haynes and Gower 1995
Red Pine	Massachusetts, U.S.A.	4100		McClaugherty *et al.* 1982
Scots Pine	Sweden	2170	2,260	Persson 1983
White Pine	Wisconsin, U.S.A.	970 - 1600		Aber *et al.* 1985
Alder - Balsam Poplar	Alaska, U.S.A.	6500 - 9700		Ruess *et al.* (1996)
Black Spruce	Alaska, U.S.A.	685		Ruess *et al.* (1996)
Norway Spruce	Estonia	2510	890 - 965	Ostonen *et al.* (2005)
White Spruce	Alaska, U.S.A.	4500 - 3700		Ruess *et al.* (1996)
Mixed hardwood	Massachusetts, U.S.A.	5400		McClaugherty *et al.* 1982
Northern Hardwood	New York, U.S.A.	1500		Burke and Raynal 1995

slower growing northern systems where root growth exhibits a strong seasonal pattern? The concern is that too short of a placement would not allow adequate root exploration into the mesh bag, whereas too long of a period could result in barriers to additional root inclusions due to blocked holes. Figure 4.7 compares the estimates of new fine root biomass "captured" in the ingrowth bag upon retrieval. The short placement times (78, 147 days), when adjusted to a per annum basis, produced significantly higher FR_{NPP} than the 12 and 15 month placements. These estimates (230 and 325 kg · ha^{-1} · yr^{-1}) are likely

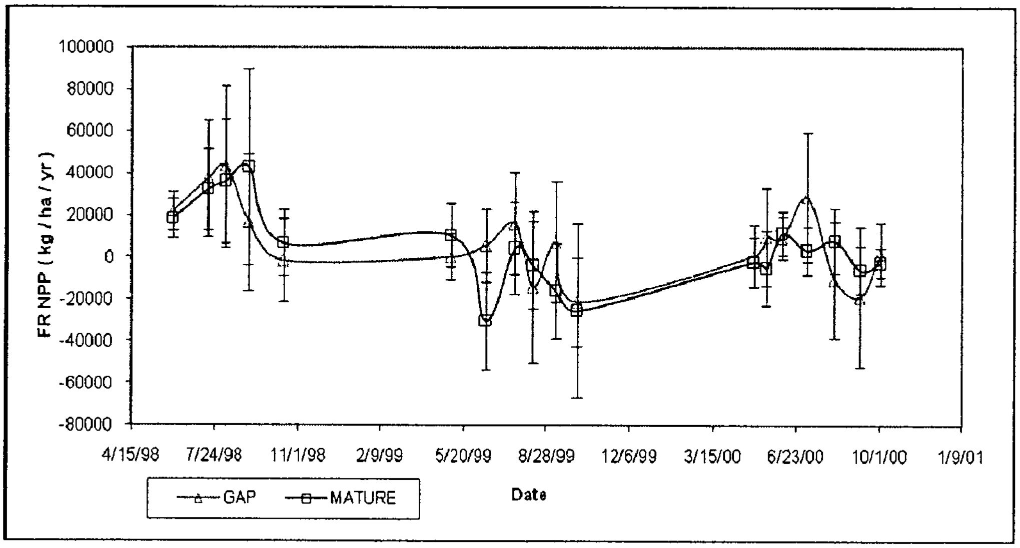

Figure 4.6.Temporal patterns, and associated variability depicted by standard error bars, in fine root productivity (FR_{NPP}) in the sampled sugar maple stand obtained from repeated measures of the minirhizotron tubes.

over-estimates as they only consider the growing season portion of the year (May – August, May – Oct). When comparing these two placements, however, it does suggest that the late summer/fall period is an active period of root development and growth for sugar maple. In this case, fine root productivity was at nearly 1.5 kg $\cdot$ ha^{-1} $\cdot$ day^{-1} compared to under 0.6 kg $\cdot$ ha^{-1} $\cdot$ day^{-1} for the spring/early summer period. The longer placement sets produced comparable estimates at approximately 150 kg $\cdot$ ha^{-1} $\cdot$ yr^{-1}, and likely provide a better annual estimate (*i.e.*, include both the growing and dormant seasons). These estimates are lower than the estimates reported by Persson (1983) for Scots pine (2260 kg $\cdot$ ha^{-1} $\cdot$ yr^{-1}) and Ostonen *et al.* (2005) for Norway spruce (890 – 965 kg $\cdot$ ha^{-1} $\cdot$ yr^{-1}). Ideally, it would have been better to leave the final set in place for a full two years (730 days) to include and compare the initial growing season to the second year's growth where roots had already become established with the ingrowth bag, thereby

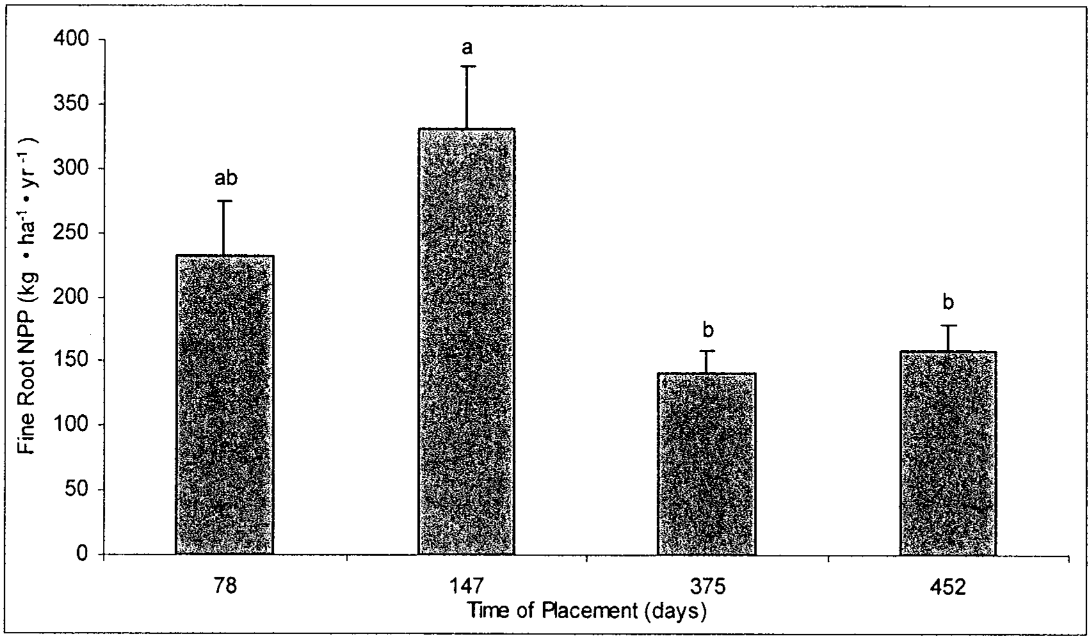

Figure 4.7. Fine root productivity (FR_{NPP}) estimates obtained from ingrowth bags placed in the soil in the sugar maple stand for different time periods.

providing a "full capacity" estimate from the bags (Finer and Laine 2000, Vogt *et al.* 1998).

4.3.3 Comparison of Fine Root Turnover Estimates

In the case of the fine root turnover estimates, a different pattern emerged when comparing the direct versus indirect methods (Figure 4.8). For sugar maple (hardwood), the indirect methods generated turnover rates (approximately 3000 kg · ha^{-1} · yr^{-1}) double those produced using the minirhizotron system (1500 kg · ha^{-1} · yr^{-1}). Although the indirect methods generated lower estimates than the minirhizotron method for jack pine

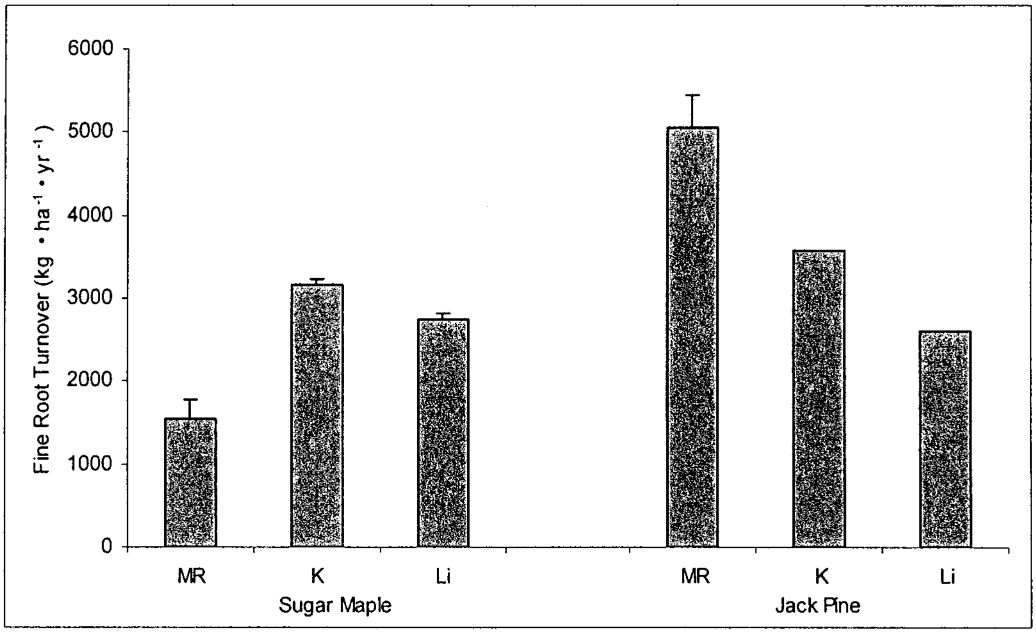

Figure 4.8. Fine root turnover estimates obtained from three methods (MR – minirhizotron ; K – Kurz ; Li) used in this study for the sugar maple and jack pine stands.

(conifer), these estimates still appear to be too high when compared to the fine root biomass pool these methods generated (Figure 4.2).

The fine root biomass : turnover ratios for both species using the indirect methods suggest that more than 70 % of the fine root biomass pool turns over annually, generating a mean residence time of 1.5 years. In contrast, the minirhizotron approach produces a more conservative estimate for these northern forest systems at 20 – 30 % and a mean residence time of 3 – 5 years. The latter estimates are consistent with other published estimates for northern hardwoods and conifers (Aber *et al.* 1985, Gower *et al.* 1992, Hendrick and Pregitzer 1993a, Ruess *et al.* 1996).

4.3.4 Implications for Carbon Modelling

Consistent with other fine root biomass studies (Van Cleve *et al.* 1991, Ruess *et al.* 1996, Green 2004), this study illustrated the high level of variability associated with the estimates of fine root biomass, productivity, and turnover. Eissenstat *et al.* (2000) suggested that a range of biotic and abiotic conditions can greatly influence the growth patterns of fine roots. Pregitzer *et al.* (2002) illustrated that fine roots can develop morphological and physiological adaptations unique to particular habitats, and emphasized the need for site-specific studies. Based on the current results, this site- and species-level variability can be further exacerbated depending on the method employed to estimate fine root biomass bringing into question the validity of broader scale modeling of fine root dynamics and the assessment of carbon stocks.

In Canada, carbon budgets are currently evaluated using the Carbon Budget Model of the Canadian Forest Sector 3 (CBM-CFS3) which continues to use indirect methods to estimate the fine root carbon pool. Based on the current study results, this approach, and the associated equations applied to either hardwood or conifer aboveground biomass stand-level data, would tend to under-estimate the fine root C pool, but over-estimate the turnover rate. A reasonable question that could be asked would be: are these under-estimates consistent across the range of species and plot conditions encountered in this study? If this is the case, an adjustment in the aboveground to belowground ratio should improve these indirect estimates, making them more consistent with the direct methods. Based on the results of the Pearson correlation analysis (Table

4.5), both the Kurz and Li estimates are significantly correlated with the minirhizotron estimates (r-values of 0.79 and 0.74, respectively).

Table 4.5. Summary of the Pearson correlation coefficients (r) comparing the four fine root sampling methods included in this study.

	MR	SC	Kurz	Li
MR	1.000	-0.698 **(0.036)**	0.788 **(0.020)**	0.736 **(0.037)**
SC		1.000	-0.712 **(0.047)**	-0.577 (0.134)
Kurz			1.000	0.983 **(<0.001)**
Li				1.000

Note: Values in parentheses are the p-values, with bolded values of p<0.05.

Working through the equations used in these indirect methods (see Table 4.1), the hardwood fine root biomass pool is 3.5 % of the aboveground biomass pool and the conifer pool is 3 %. When comparing the stand-level average aboveground biomass pools (sugar maple – 137.2 $T \cdot ha^{-1}$, jack pine – 162.9 $T \cdot ha^{-1}$) to the minirhizotron estimates of fine root biomass, the ratios are slightly higher at 5.4 % and 8.8 % for sugar maple and jack pine, respectively. A final question worth asking is: how would this adjustment (*i.e.*, using aboveground to belowground biomass ratios generated from species-specific direct measurements) influence the scaling up estimate of carbon stocks? As a simple example using the land class area data (hardwood - 2.3 million ha, conifer – 11.8 million ha, mixedwood – 9.7 million ha, assuming a 50/50 hardwood/conifer mix) provided in Ontario's State of the Forest Report 2006 (Anon 2006), scaled up estimates of the fine root C pool for Ontario's boreal forest were calculated using the current aboveground/fine root biomass ratios versus the ratios created in the current study using the minirhizotron estimates. Biomass to carbon conversion was assumed to be 50 %. The current CBM-CFS3 model would generate an estimate of Ontario's boreal forest fine root C pool at 48.4 M Tonnes, compared to 121.7 M Tonnes when applying the hardwood and conifer ratios estimated in the current study, representing difference of over 70 M Tonnes (3 fold difference) of fine root carbon.

Values generated as part of this scaling up exercise, however, should not be considered accurate, nor should the aboveground/fine root biomass ratios generated from this study be used in any future modeling calibration efforts due to the limited number of sites and species used in the study. The scaling up example, however, does clearly suggest that considerable more effort in estimating fine root biomass, productivity, and turnover in northern forest systems is warranted. Recently, Shaw *et al.* (2009) identified the ability to accurately estimate belowground carbon, particularly with respect to the fine root biomass pool as a current weakness in the CBM-CFS3 model. Shaw *et* al. (2009) indicated that there are plans to develop species-specific aboveground/belowground (fine root) biomass ratios using direct measures of fine root biomass.

4.4 CONCLUSIONS

The results from the current study accentuate the potential challenges associated with incorporating site specific fine root research into broader generalizations applied to large land masses. These constructs inevitably incorporate inherent errors associated with aboveground biomass estimates, conversions factors to belowground estimates (indirect methods), high within-site variability associated with direct measurements, and all the numerical accounting methods and assumptions needed to arrive at fine root estimates. This variability is further influenced by climate, hydrology, soil biota, and available soil nutrients, but the exact effect of each of these variables, or the associated interactive effects, has not yet been adequately examined.

Our poor understanding of belowground processes remains a weakness in current carbon models, particularly with respect to their predictive strength/reliability to scale up to regional, continental, or global scales. A more thorough examination of fine root dynamics is required in order to ensure wider scientific acceptance of broad-based models and their ability to predict the impacts of forest management activities or climate change.

5.0 SUMMARY

There is increasing interest to develop quantitative approaches to carbon accounting and determine carbon sequestration potential at both the site and landscape scales. Within the global context, northern temperate forests have been identified as playing a key role in the global C cycle, storing upwards of 40 % of the earth's reactive C. Even though it has been estimated that over 30 % of global NPP is used for the production of fine roots, this highly dynamic pool remains largely an unknown variable in C budget models (*e.g.*, CBM-CFS3). Our lack of understanding of fine root dynamics in northern temperate forest systems continues to hamper efforts to accurately parameterize these C budget models.

Although the direct methods, particularly the use of the minirhizotron technology, appear to provide better species- and site-specific estimates of fine root biomass pools, the results from the current study do suggest the need for species- and diameter class-specific standard root lengths (*i.e.*, factor for converting measured root lengths to biomass when using minirhizotron technologies). It was determined that there was a significant difference in fine root standard root length (SRL) between northern temperate tree species and diameter classes. Angiosperms tended to had significantly higher average SRLs compared to gymnosperms. Within these species groupings, tolerant, late successional species had higher SRLs compared to their intolerant, early successional counterparts. Standard root lengths dropped significantly (>200 %) as diameter class increased.

The results from the comparison of the indirect and direct methods to estimate fine root biomass, productivity, and turnover in the northern temperate forest types used in this study accentuate the potential challenges associated with incorporating site specific fine root research into broader generalizations applied to large land masses. These constructs inevitably incorporate inherent errors associated with aboveground biomass estimates, conversions factors to belowground estimates (indirect methods), high within-site variability associated with direct measurements, and all the numerical accounting methods and assumptions needed to arrive at fine root estimates. A simple example using Ontario's land class area data for the boreal region compared the fine root biomass estimates from the CBM-CFS3 model to those generated using the aboveground/fine root biomass ratios created in the current study using the minirhizotron estimates. These results indicated that the CBM-CFS3 model would underestimate fine root carbon stores in Ontario's Boreal forest by over 70 M Tonnes.

Future research efforts will require a more thorough examination of fine root dynamics in order to ensure wider scientific acceptance of broad-based models and their ability to predict the impacts of forest management activities or climate change.

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APPENDIX 1:

ANOVA results examining factors influencing Standard Root lengths (SRL)

4-factor ANOVA model (Groundhog R. site only):

Source	df	MS	F ratio	Pr > F
Date (D)	4	56.8	1.64	0.162
Species (S)	1	6.8	0.20	0.657
Dia. Class (C)	5	2823.5	81.62	**<0.001**
Soil Layer (L)	2	86.9	2.51	0.082
D * S	4	16.2	0.47	0.760
D * C	20	10.9	0.31	0.998
D * L	8	21.6	0.62	0.758
S * C	5	63.6	1.84	0.103
S * L	2	17.8	0.51	0.599
C * L	10	43.2	1.25	0.256
D * S * C	20	10.1	0.29	0.999
D * C * L	40	13.2	0.38	0.999
S * C * L	10	19.0	0.55	0.855
D * S * C * L	37	13.4	0.39	0.999
Error	704	34.6		
	872			

2-factor ANOVA model (Nimitz and Groundhog R. sites):

Source	df	MS	F ratio	Pr > F
Species (S)	2	167.6	5.50	**0.004**
Dia. Class (C)	5	2868.2	94.13	**<0.001**
S * C	10	38.8	1.27	0.240
Error	904	30.5		
	921			

1-way ANOVA model (all sites):

Source	df	MS	F ratio	Pr > F
Species (S)	3	138.2	34.31	**<0.001**
Error	47	4.0		
	50			