

Portland State University

PDXScholar

---

Environmental Science and Management  
Faculty Publications and Presentations

Environmental Science and Management

---

2007

# Estimating Nutrient Uptake by Mature Tree Roots Under Field Conditions: Challenges and Opportunities

Melissa S. Lucash

*Portland State University, lucash@pdx.edu*

Dave M. Eissenstat

*Pennsylvania State University*

J. Devereux Joslin

*Belowground Forest Research*

Karis J. McFarlane

*Oregon State University*

Ruth D. Yanai

*SUNY College of Environmental Science and Forestry*

Follow this and additional works at: [https://pdxscholar.library.pdx.edu/esm\\_fac](https://pdxscholar.library.pdx.edu/esm_fac)



Part of the [Natural Resources Management and Policy Commons](#), and the [Other Environmental Sciences Commons](#)

Let us know how access to this document benefits you.

---

## Citation Details

Lucash, Melissa S.; Eissenstat, Dave M.; Joslin, J. Devereux; McFarlane, Karis J.; and Yanai, Ruth D., "Estimating Nutrient Uptake by Mature Tree Roots Under Field Conditions: Challenges and Opportunities" (2007). *Environmental Science and Management Faculty Publications and Presentations*. 111. [https://pdxscholar.library.pdx.edu/esm\\_fac/111](https://pdxscholar.library.pdx.edu/esm_fac/111)

This Post-Print is brought to you for free and open access. It has been accepted for inclusion in Environmental Science and Management Faculty Publications and Presentations by an authorized administrator of PDXScholar. Please contact us if we can make this document more accessible: [pdxscholar@pdx.edu](mailto:pdxscholar@pdx.edu).

1 Estimating nutrient uptake by tree roots under field conditions:  
2 challenges and opportunities

3  
4 Melissa S. Lucash<sup>1</sup>, Dave M. Eissenstat<sup>2</sup>, J. Devereux Joslin<sup>3</sup>, Karis J. McFarlane<sup>4</sup>  
5 and Ruth D. Yanai<sup>1\*</sup>

6 <sup>1</sup>Department of Forest and Natural Resources, State University of New York  
7 College of Environmental Science and Forestry, Syracuse, NY 13210, USA;  
8 <sup>2</sup>Department of Horticulture, Pennsylvania State University, 218 Tyson Building,  
9 University Park, PA 16802, USA; <sup>3</sup> Belowground Forest Research, Apartado 104-  
10 5655, Santa Elena de Monteverde, Puntarenas, Costa Rica and <sup>4</sup>Department of  
11 Forest Engineering, 204 Peavy Hall, Oregon State University, Corvallis, OR 97331  
12 USA

13 Author for correspondence:

14 \*Ruth D. Yanai

15 <sup>1</sup>Department of Forest and Natural Resources Management  
16 State University of New York College of Environmental Science and Forestry  
17 Syracuse, NY 13210  
18 Phone: 315-470-3955  
19 FAX: 315-470-6954  
20 Email: [rdyanai@mailbox.syr.edu](mailto:rdyanai@mailbox.syr.edu)

21

22 Running head: Estimating nutrient uptake by trees

23

## 1 **Summary**

2 Nutrient uptake by tree roots is difficult to measure accurately under field  
3 conditions using existing methods. In this review, we discuss current techniques  
4 for measuring uptake at the root surface including excised root, depletion,  
5 lysimeter and isotopic tracer methods. Although many insights have been gained  
6 using these methods, each has drawbacks. Estimates of uptake are affected by  
7 the sampling scheme, experimental conditions, whether roots are excised or not,  
8 concentrations of ions, and the rate of efflux of ions. Two recently developed  
9 approaches, SUM column method and digital autoradiography, hold considerable  
10 promise for improving our estimates of uptake by tree roots. A greater focus on  
11 methods development is critical to more accurately measuring uptake under  
12 conditions representative of those in the field.

13

14

15

16

17

18

19

20

21 **Key words:** autoradiography, efflux, excision, ion uptake, nutrient concentration,  
22 sand culture

## 1 **Introduction**

2 Surprisingly little is known about rates of nutrient uptake by tree roots in the field,  
3 despite their importance for the growth and survival of trees. Root systems of  
4 trees are extensive, spatially variable, morphologically and physiologically  
5 heterogeneous, and often intertwined with those of other plants. These  
6 characteristics make it difficult to accurately measure uptake under conditions  
7 representative of those in the field. The most common approach to estimating  
8 nutrient uptake by trees has been to construct nutrient budgets; however, this  
9 technique does not provide any information on processes at the root scale.  
10 Measurements of specific root uptake or uptake capacity are required. In this  
11 study, we define specific root uptake as the rate of uptake of nutrients per unit root  
12 mass. We define uptake capacity as specific root uptake at non-limiting  
13 concentrations. Current techniques for measuring specific root uptake, such as  
14 excised roots, depletion, lysimeters and isotopic tracers, have provided valuable  
15 information, but still fall short of providing realistic and accurate estimates of rates  
16 of nutrient uptake by trees in the field.

17 In the past, specific root uptake has been measured primarily using the fine  
18 roots of agricultural crops or tree seedlings in solution culture. Results of these  
19 studies are difficult to extrapolate to the field because roots in solution differ in  
20 age, morphology and physiology from those grown in solid media or the field  
21 (Skene et al. 1998). Furthermore, the nature of the rhizosphere and the  
22 movement of nutrients to the root surface are different in soils than in solution

1 culture (Barber 1995). Finally, roots of seedlings in solution culture are seldom  
2 mycorrhizal (Van der Driessche 1971; Ingestad and Lund 1979; Bledsoe and  
3 Rains 1981), whereas many tree roots depend on mycorrhizas to supplement  
4 nutrient uptake (Smith and Read 1997).

5         Despite the drawbacks, scientists still commonly use the solution culture  
6 method because rates of uptake can be readily measured. In contrast, current  
7 techniques for use with mature trees, the focus of this review, are subject to  
8 numerous methodological problems. For example, roots are often excised or  
9 disturbed prior to measurements, which may artificially increase the loss of  
10 nutrients from roots and thereby reduce net uptake (Bloom and Caldwell 1988). In  
11 addition, estimates of uptake vary widely, depending on the timing of sampling and  
12 experimental conditions such as nutrient concentrations and experiment duration.  
13 Continued development of methods is critical to a better understanding of nutrient  
14 uptake by mature trees, and how it varies among species, through the growing  
15 season, at different stages of plant development, and under different soil  
16 conditions.

17         More realistic estimates of uptake would also be useful in the development  
18 of models of nutrient uptake by mature tree roots. Current models are often  
19 unable to even accurately predict uptake by seedlings (Kelly et al. 1992; Kelly et  
20 al. 1994; Van Rees et al. 1999; Kelly et al. 2001); yet the parameters used in these  
21 models, based upon data from experiments on seedlings, are used to simulate  
22 uptake by mature trees. There is little reason to expect that parameters measured

1 on seedlings will accurately reflect uptake by mature trees, especially since trees  
2 change physiologically as they age (Espeleta and Eissenstat 1998; Law et al.  
3 2001). Measuring uptake using roots of mature trees under conditions  
4 representative of the field would be advantageous to modelers seeking to  
5 understand plant-soil interactions.

6 In this review, we will first describe methods currently used to measure  
7 specific uptake rates of roots of mature trees. We classify these methods into two  
8 categories: (1) methods in which roots are excavated before measurement and (2)  
9 methods in which undisturbed roots are measured in soil. In the second section,  
10 we focus on the methodological challenges to obtaining more realistic estimates of  
11 uptake. In the third section, we describe recent technological advances that hold  
12 promise for the future. Finally, we describe opportunities for future research,  
13 which ultimately may lead to technological improvements and a better  
14 understanding of uptake by tree roots in the field.

15

## 16 **Methods used to measure uptake by trees**

### 17 ***Uptake by disturbed roots***

#### 18 *Excised root method*

19 The excised root technique (Epstein et al. 1963a) is commonly used for measuring  
20 specific uptake by seedlings and trees in the laboratory and in the field. In this  
21 method, roots are excavated from the soil, excised, and sealed inside cheesecloth  
22 “teabags”. The bags are placed in an aerated solution containing a radioactive or

1 stable tracer of the nutrient of interest. After periods ranging from 10 min to 2 h,  
2 the rate of uptake is determined by analyzing tracer accumulation in the root.

3 This method has been used extensively since the 1960's and has proven  
4 extremely valuable for characterizing the nutritional status of trees (Bowen 1970;  
5 Jones et al. 1994; Hogberg et al. 1998) and measuring ion uptake by roots  
6 (Epstein et al. 1963b; Huang et al. 1992). Studies with excised roots were  
7 considered superior to studies with intact roots, since excision eliminates  
8 interactions between the root and shoot, which could complicate interpretation of  
9 the results (Hoagland and Broyer 1936).

10 More recently, however, some scientists have expressed reservations  
11 about using excised roots, arguing that excavation and excision can alter root  
12 respiration and uptake (Saglio and Pradet 1980; Bloom and Caldwell 1988).  
13 Moreover, the excised root method estimates only gross influx, defined as the rate  
14 of entry of the ion into the root. To estimate net uptake, defined as the difference  
15 between influx and efflux, two tracers (e.g.  $^{32}\text{P}$  and  $^{33}\text{P}$ ) must be used (Elliott et al.  
16 1984), or the tracer method must be combined with the depletion method (Clark et  
17 al. 2000), which is described below. Therefore, rates obtained using excised roots  
18 may not provide realistic estimates of *in situ* uptake. Rather this method is most  
19 appropriate for use in comparative studies to determine which factors affect gross  
20 nutrient uptake (e.g. nutrient concentration, temperature, plant age), assuming  
21 excision does not have an interactive effect on the factor of interest. The use of

1 this method is also limited by the special handling and disposal procedures for  
2 radioactive tracers and the relatively high analytical costs of stable isotopes.

3

#### 4 *Depletion method*

5 The depletion method offers a possible improvement over the excised root method  
6 in that isotopes are not required and the roots remain intact (Gessler et al. 1998;  
7 BassiriRad et al. 1999; Lucash et al. 2005). In this method, roots of mature trees  
8 (Gessler et al. 1998; Gessler et al. 2002; Lucash et al. 2005) or seedlings (Bhat  
9 1982; Marschner et al. 1991; BassiriRad et al. 1997; Gessler et al. 1998;  
10 BassiriRad et al. 1999) are excavated without detaching them from the tree, and  
11 placed in aerated nutrient solutions. Alternatively, roots are pruned and allowed to  
12 re-grow for several months in plastic trays containing soil (Escamilla and  
13 Comerford 1998) or bags containing a sand-soil mixture (McFarlane and Yanai  
14 2006) before they are placed in nutrient solution. In both techniques, the depletion  
15 of nutrients from solution is measured by periodically sampling the solution to  
16 compute the net uptake rate.

17 The most significant advantage of this technique is that the roots are still  
18 attached to the tree and can continue to transport carbon, water, and nutrients bi-  
19 directionally. Carbohydrate supply may be particularly important for ions such as  
20 nitrate and ammonium that require substantial energy for uptake (Bloom et al.  
21 1989; Bloom et al. 1992). Another advantage of this technique is that both



1 analytical techniques and supplies can be inexpensive, and there are fewer  
2 handling restrictions than with techniques requiring radioisotopes.

3       There are several disadvantages to the depletion method. This method  
4 involves excavating roots, which can affect rates of uptake. For example, efflux is  
5 greater than influx in some field studies, although net uptake is clearly not negative  
6 over the lifetime of the plant. In the Methodological Challenges section, these  
7 problems are discussed in more detail.

8

### 9 ***Uptake by undisturbed roots in porous media***

10 Both the excised root and the intact depletion methods characterize uptake by  
11 roots that have been removed from the soil. Excavation may damage the roots  
12 and sever the extramatrical hyphae of their fungal associates, causing a reduction  
13 in uptake. To eliminate the effects of disturbance on uptake, alternative  
14 approaches are needed.

15

### 16 *Lysimeter method*

17 In the lysimeter method, seedlings or small trees are grown in porous media in  
18 containers ranging in size from small pots (Colpaert et al. 1999) to large tanks  
19 (Weinbaum et al. 1994; Syvertsen and Smith 1996). The root systems may be  
20 inoculated with mycorrhizal fungi (Colpaert et al. 1999). Nutrient solution is added  
21 to the medium and removed for sampling using a peristaltic, vacuum or sump  
22 pump. The leachate is weighed and analyzed for nutrient concentration; the

1 differences in nutrient content between the initial solution and the leachate are  
2 used to compute uptake.

3         One of the main advantages of this technique is that it can be used to  
4 assess the relative contribution of mycorrhizas to nutrient uptake without disturbing  
5 the root system (Colpaert et al. 1999). This technique has been used to quantify  
6 nitrogen uptake of small citrus (Syvertsen and Smith 1996) and plum trees  
7 (Weinbaum et al. 1994) in large lysimeters in the field but uptake was expressed at  
8 the tree level (kg N/ tree). One of the main drawbacks to this method is that it  
9 cannot be used for large trees, since the entire root system is limited by the size of  
10 the container.

11

### 12 *Isotopic tracer method*

13 To measure uptake by intact roots using isotopic tracers, nutrient solution  
14 containing tracers is applied to pots containing sand (Cui and Caldwell 1997; Proe  
15 et al. 2000; Yoder and Caldwell 2002) or peat (Ohlund and Nasholm 2004). In the  
16 field, isotopes can be applied with fertilizer (Weinbaum and Van Kessel 1998;  
17 Dinkelmeyer et al. 2003) or injected into the soil (Caldwell et al. 1985). In both  
18 techniques, uptake is calculated by analyzing the amount of tracer in the root and  
19 shoot tissue. In some studies, uptake has been calculated at the root scale (Cui  
20 and Caldwell 1997; Yoder and Caldwell 2002; Ohlund and Nasholm 2004), while  
21 in others uptake has been expressed at the whole-plant level (Proe et al. 2000;  
22 Dinkelmeyer et al. 2003).

1           The tracer method is useful for measuring gross uptake by undisturbed  
2 mycorrhizal roots (Ohlund and Nasholm 2004) and distinguishing between uptake  
3 and remobilization of nutrients, such as nitrogen (Weinbaum and Van Kessel  
4 1998; Proe et al. 2000) and potassium (Proe et al. 2000). The relative  
5 competitiveness of different species (Caldwell et al. 1985; Yoder and Caldwell  
6 2002) and seasonal trends in uptake (Nambiar and Bowen 1986) can also be  
7 quantified using this method. This technique is difficult to apply to large trees in  
8 the field (Dinkelmeier et al. 2003; McKane et al. 2003) due to problems of isotope  
9 dilution, sampling large plants and determining the concentration at the root  
10 surface.

11

## 12 **Methodological Challenges**

13 In this section, we describe the problems with existing methods to draw attention  
14 to the limits of our current techniques. These challenges need to be overcome to  
15 obtain more realistic estimates of uptake by trees.

16

### 17 ***Bias introduced by root sampling approaches***

18 Selecting which roots to sample, when to sample and how long to sample are all  
19 critical decisions in uptake studies. Since these factors affect measured uptake  
20 rates, the method of selecting roots and the timing of sampling should be  
21 considered before implementing a study.

1           Most researchers measure uptake by young, fine diameter roots because  
2 they are considered to be most active in nutrient uptake. Older, thicker roots,  
3 however, may also take up nutrients and are a significant proportion of root  
4 biomass in mature trees. Depletion of  $\text{NH}_4^+$  and magnesium was observed in the  
5 rhizosphere of both young and old roots of Norway spruce, indicating that uptake  
6 occurred in both ages of root (Dieffenbach et al. 1997). In cherry roots,  
7 phosphorus uptake was not statistically different between young (white) and old  
8 (woody) roots (Atkinson and Wilson 1979).

9           Diurnal variation in uptake should be considered when measuring uptake  
10 rates. Many studies with agricultural crops report diurnal rhythms of uptake of  
11  $\text{NO}_3^-$  (Hansen 1980; Pearson et al. 1981; Scaife and Schloemer 1994; Delhon et  
12 al. 1996) and  $\text{NH}_4^+$  (Ourry et al. 1996; Macduff et al. 1997). Only one study to  
13 date has examined diurnal patterns of uptake by trees (Gessler et al. 2002). In  
14 that study, the diurnal patterns of  $\text{NH}_4^+$  uptake by mature trees were species-  
15 specific; spruce exhibited little diurnal fluctuations in rates, while beech had higher  
16  $\text{NH}_4^+$  uptake during the day than the night.

17           Uptake also varies seasonally, but selecting the best time to conduct  
18 experiments is problematic because the timing of uptake differs among species.  
19 Ammonium uptake by mature loblolly pine roots was higher in April than July  
20 (Lucash et al. 2005). In another study in which the depletion method was used,  
21 uptake of  $\text{NH}_4^+$  by mature subalpine spruce and beech was higher in summer than  
22 spring (Gessler et al. 1998).

1           Seasonal patterns in uptake also vary from year to year. In a two-year  
2 study, uptake of phosphate by excised roots of balsam fir in April was two times  
3 higher in the first than the second year (Langlois and Fortin 1984). In another  
4 study, uptake of  $\text{NH}_4^+$  by intact roots of spruce was similar in the first and second  
5 years, but uptake by intact beech roots was significantly higher in July and Sept of  
6 the second year (Gessler et al. 1998).

7           The duration of the experiment can have dramatic effects on estimates of  
8 uptake. In experiments with crop plants in which concentrations were kept  
9 constant, uptake varied significantly over time. Nitrate uptake dropped by 50%  
10 after the first 14 h, and potassium uptake decreased by 27% after 36 h (Glass et  
11 al. 1987). Therefore, the length of the experiment can affect uptake rates, and  
12 rates measured over different durations may not be comparable. Sampling  
13 intervals that have been used in experiments with seedlings and mature trees  
14 have ranged from 10 min (Lajtha 1994) to 30 min (Bhat 1982) for P, 15 min  
15 (Rothstein et al. 2000) to 1 day (Eltrop and Marschner 1996) for  $\text{NO}_3^-$  and  $\text{NH}_4^+$ ,  
16 and 2 h (Lucash et al. 2005) to 1-2 days for  $\text{K}^+$ , calcium and  $\text{Mg}^{2+}$  (Bledsoe and  
17 Rains 1981). In experiments with long durations, hypoxic conditions may develop  
18 if the roots are not adequately aerated and uptake rates may be suppressed  
19 (Escamilla and Comerford 1998)

20

1 ***Effects of excision and disturbance on uptake are variable***

2 Although the effects of excision on uptake have never been studied in tree  
3 seedlings or mature trees, the effects of excision on uptake have been shown to  
4 be ion-specific in agricultural crops. Excision has been shown to decrease  $\text{NO}_3^-$   
5 (Bloom and Caldwell 1988; Aslam et al. 1996) and  $\text{NH}_4^+$  uptake (Bloom and  
6 Caldwell 1988), root respiration (Saglio and Pradet 1980; Bloom and Caldwell  
7 1988; but see Lipp and Andersen 2003) and carbohydrate supply (Clarkson et al.  
8 1974; Saglio and Pradet 1980). In contrast, excision had no effect on P uptake in  
9 two studies (Gronewald and Hanson 1980; Gronewald and Hanson 1982). The  
10 effects of excision on  $\text{K}^+$  and  $\text{Ca}^{+2}$  uptake are not well established. Excision of  
11 barley roots significantly decreased  $\text{K}^+$  uptake in two studies (Glass 1978; Bloom  
12 and Caldwell 1988), but had no effect on  $\text{K}^+$  uptake in another (Huang et al. 1992).  
13 Excision reduced Ca influx in corn (Rincon and Hanson 1986) but had no effect on  
14 uptake by barley (Clarkson et al. 1974).

15 Treatment differences within studies can be misinterpreted if excision has  
16 an interactive effect on the treatment of interest. For example, excision had a  
17 greater effect on uptake by barley at low than at high temperatures (Clarkson et al.  
18 1974); thus, the effects of temperature on uptake may be difficult to evaluate using  
19 this method. The effects of excision are species-specific (Huang et al. 1992),  
20 which complicates the task of comparing treatment differences among species.  
21 The effects of excision on uptake have never been studied in tree species, as far  
22 as we know, but we can expect them to be equally complex.

1           Comparing results across studies using the excised root method is difficult  
2 because there is no standard protocol. For example, root segment length varies,  
3 even though influx rates per unit mass increase with segment length (Gronewald  
4 and Hanson 1980; Huang et al. 1992). In addition, the length of time the excised  
5 roots are kept in solution (also known as the “aging” effect) affects uptake rates  
6 (Glass 1978; Huang et al. 1992). For example,  $K^+$  influx rates of excised barley  
7 roots increased by 50% between 1 h and 2 h (Glass 1978). Establishing  
8 methodological guidelines for the excised root method would make it easier to  
9 compare results across studies.

10           In both the excised root and the depletion method, roots of seedlings and  
11 mature trees are excavated from the soil before uptake is measured. In one  
12 study, roots of four tree species were exposed to pretreatments to reduce damage  
13 to the roots or allow roots to recover from the disturbance associated with  
14 excavating intact roots (McFarlane and Yanai 2006). Unexpectedly, roots given  
15 pretreatments did not have consistently higher uptake rates than roots recently  
16 disturbed, indicating that pretreatments designed to minimize disturbance may not  
17 be effective.

18           In another study, the effects of disturbance on uptake were studied by  
19 examining how uptake was affected by the transfer of seedlings to solution culture.  
20 In that study,  $NO_3^-$  uptake was similar between undisturbed mycorrhizal loblolly  
21 pine seedlings grown in sand-filled lysimeters and seedlings transferred to solution  
22 culture (Lucash et al. in review). This result may indicate that mycorrhizal hyphae

1 are not important for uptake of mobile nutrients such as  $\text{NO}_3^-$  (Eltrop and  
2 Marschner 1996). Alternatively, the negative effect on uptake of severing the  
3 extramatrical hyphae of mycorrhizas may have been masked by the positive effect  
4 of eliminating nutrient depletion zones in solution culture. More studies are  
5 necessary to distinguish the effects of root excavation from hyphal excision on  
6 nutrient uptake rates.

7

### 8 ***Underestimation of the importance of nutrient efflux***

9 As described above, the use of tracers to measure uptake detects nutrient influx  
10 but not efflux. In some situations, the rate of efflux can be a significant component  
11 of net uptake, sometimes equaling or exceeding the rate of influx. In a study of  
12 *Arabidopsis* where both influx (using the  $^{32}\text{P}$  method) and net uptake (using the  
13 depletion method) were quantified, influx of  $^{32}\text{P}$  occurred almost immediately  
14 (Clark et al. 2000). Net uptake, however, was negative for 1-3 hours, indicating  
15 that efflux exceeded influx during that period. In a set of studies using the  
16 depletion method, we found that net uptake of  $\text{NH}_4^+$ ,  $\text{Mg}^{2+}$  and  $\text{Ca}^{+2}$  over a 2 h  
17 period was positive for some tree species but not others; uptake of  $\text{K}^+$  was  
18 consistently negative (Figure 1).

19 In the excised root and depletion methods, roots are severed or excavated  
20 just prior to measurements, which may cause high rates of efflux. McFarlane and  
21 Yanai (2006) tested whether various pretreatments would reduce damage to the  
22 roots or allow roots to recover from the disturbance associated with excavating



1 intact roots. Consistent net efflux of  $\text{Ca}^{+2}$ ,  $\text{Mg}^{2+}$ ,  $\text{K}^+$ , and  $\text{Na}^+$  was observed across  
2 treatment in four tree species simultaneously with positive net uptake of  $\text{NH}_4^+$ ,  
3  $\text{NO}_3^-$ , and  $\text{PO}_4^{3-}$  and  $\text{Al}^{3+}$  (Figure 2). Therefore the disturbance associated with  
4 excavation was not responsible for nutrient efflux.

5         The fact that net nutrient efflux occurs in roots that have been excavated  
6 suggests that either uptake rates or efflux rates are not realistic under these  
7 experimental conditions. The importance of measuring nutrient efflux has not  
8 been clearly recognized, perhaps because tracer methods estimate only gross  
9 influx. The need remains for methods that minimize disturbance and produce  
10 realistic estimates of efflux and net uptake.

11

### 12 ***Influences of nutrient concentration on estimates of uptake***

13 A wide range of concentrations has been used to assess uptake rates of tree  
14 seedlings (Table 1), but the justification for the choice of concentration is seldom  
15 reported. In some studies, roots are exposed to solutions that simulate bulk soil  
16 solution concentrations (Rennenberg et al. 1996; Gessler et al. 1998), although  
17 concentrations at the root surface differ from bulk soil (Barber 1995).

18 Concentrations at the root surface can be measured, but the methods used are  
19 often destructive (Gobran and Clegg 1996; Bakker et al. 1999) and produce  
20 concentrations inconsistent with nutrient uptake models (Yanai et al. 2003). New  
21 methods that use non-destructive *in situ* sampling to characterize concentrations  
22 at the root surface (Dieffenbach et al. 1997; Dieffenbach and Matzner 2000) hold

1 promise for obtaining realistic estimates of concentrations at the root surface.  
2 These techniques may be valuable for selecting concentrations to use in  
3 laboratory experiments or improving model estimates of uptake rates. Nutrient  
4 concentrations should be carefully selected to facilitate the comparisons of results  
5 across studies and to obtain estimates of uptake using concentrations that are  
6 representative of field conditions.

7 Nutrient uptake is often measured at a range of nutrient solution  
8 concentrations and then fitted to the following Michaelis-Menten equation:

9 
$$U = \frac{V_{max} * (C_0 - C_{min})}{K_m + (C_0 - C_{min})}$$

10 where  $U$  is the uptake rate (amount per unit time per unit root),  $V_{max}$  is the  
11 maximum uptake rate (or uptake capacity) at high concentration (same units as  
12  $U$ ),  $C_0$  is the concentration at the root surface,  $K_m$  is the concentration at which  
13 uptake is  $\frac{1}{2} V_{max}$  and  $C_{min}$  is the concentration below which uptake ceases  
14 (Claasen and Barber 1974). In most studies with trees (Eltrop and Marschner  
15 1996; Rothstein et al. 1996; BassiriRad et al. 1999; Rothstein et al. 2000; Hangs et  
16 al. 2003), however, the formula is applied to data without reporting model fit or  
17 addressing whether another model might better describe the data. Depending on  
18 the concentrations used in the study, uptake may be linearly related to  
19 concentration, as observed for P uptake in red maple (Kelly and Kelly 2001) and  
20  $\text{NO}_3^-$  uptake in loblolly pine (Lucash et al. 2005).

1           In addition, internal nutrient concentrations can affect estimates of kinetic  
2 parameters. Plants have high uptake capacity following a period of nutrient  
3 deficiency (Lee and Rudge 1986; Siddiqi et al. 1989), and low uptake capacity  
4 after exposure to high concentrations, due to saturation of exchange sites at the  
5 root surface (Dean-Drummond 1982; Siddiqi et al. 1990). In one study, uptake  
6 rates of  $\text{NH}_4^+$  by Douglas-fir were reduced by high pretreatment concentrations of  
7  $\text{K}^+$ , and  $\text{K}^+$  uptake rates were reduced by high pretreatment concentrations of  $\text{NO}_3^-$   
8 (Rygiewicz and Bledsoe 1986).

9           In agricultural crops, uptake of  $\text{NO}_3^-$ ,  $\text{NH}_4^+$  and  $\text{PO}_4^{3-}$  has been observed to  
10 conform to the Michaelis-Menten model at low but not high concentrations (Siddiqi  
11 et al. 1990; Aslam et al. 1992; Wang et al. 1993; Barber 1995). This multiphasic  
12 pattern indicates that there are distinct uptake systems, which differ in their affinity  
13 for nutrients. The number of uptake systems not only varies with species (Nissen  
14 1991) and variety (Woodend et al. 1986), but also with the pretreatment  
15 concentrations (Drew et al. 1984). Future studies should determine whether  
16 uptake is multiphasic in mature trees and seedlings at concentrations normally  
17 found in the field.

18           Studies measuring uptake are often conducted by studying the uptake of  
19 one ion at a time. In nature, as well as under experimental conditions, however,  
20 roots are exposed to multiple ions simultaneously, and the presence of one ion  
21 can affect uptake of another. For example, the presence of  $\text{NH}_4^+$  inhibits  $\text{NO}_3^-$   
22 uptake by Norway spruce (Marschner et al. 1991),  $\text{K}^+$  inhibits  $\text{NH}_4^+$  uptake by

1 spruce and barley but not by rice (Wang et al. 1996), and aluminum inhibits  $\text{Ca}^{+2}$ ,  
2  $\text{NH}_4^+$ , and  $\text{K}^+$  uptake and enhances influx of  $\text{NO}_3^-$  and  $\text{PO}_4^{-3}$  by barley (Nichol et al.  
3 1993). Some depletion studies have exposed roots to solutions containing  
4 multiple ions at concentrations that attempt to simulate soil solution concentrations  
5 (Rennenberg et al. 1996; Gessler et al. 1998; Lucash et al. 2005). If the rate of  
6 uptake differs among nutrients, however, the ratios of nutrients may change over  
7 the duration of the experiment, creating artificial conditions. Studies should  
8 expose roots to multiple ions at concentrations representative of field conditions to  
9 produce realistic estimates of uptake capacity.

10

### 11 **Recent Methodological Advances**

12 Current methods for measuring specific uptake by tree roots have many  
13 drawbacks, as outlined above. Two new approaches hold considerable promise  
14 for improving our estimates of uptake by tree roots.

15

#### 16 ***Soil uptake monitoring (SUM) method***

17 The soil uptake monitoring (SUM) method is a type of lysimeter method developed  
18 to measure  $\text{NO}_3^-$  uptake of seedlings of woody plants (Scholberg et al. 2001;  
19 Lucash 2005). In this method, seedlings are grown in sand-filled PVC columns in  
20 the laboratory or in the field. To measure uptake, the valves at the base of each  
21 column are closed and nutrient solution of known volume and concentration is  
22 added to the columns. To collect the solution, the valve is opened and a vacuum

1 is applied to drain the solution into the reservoirs. The solution can be sampled at  
2 this point (Scholberg et al. 2001) or it can be repeatedly reapplied to minimize the  
3 formation of depletion zones around the roots (Lucash et al. in review). At each  
4 sampling interval, the leachate is weighed and analyzed for nutrient concentration;  
5 the differences in nutrient content between the initial solution and the leachate is  
6 used to compute uptake.

7 The SUM column method has been used to quantify uptake of  $\text{NO}_3^-$  by  
8 citrus (Scholberg et al. 2001) and uptake of  $\text{NO}_3^-$  and  $\text{NH}_4^+$  by loblolly pine (Lucash  
9 et al. in review). This method was also used to measure diurnal trends in  $\text{NO}_3^-$   
10 and  $\text{NH}_4^+$  uptake by white pine seedlings; variation between days was greater than  
11 diurnal variation (Figure 3).

12 To our knowledge, this is the first technique that allows specific uptake  
13 rates to be estimated in soil without excavating the roots. A key advantage over  
14 the solution culture method is that mycorrhizas and microbes are better able to  
15 establish in sand than solution. Most tree roots are mycorrhizal in the field (Smith  
16 and Read 1997) and the presence of mycorrhizas affects the uptake kinetics of  
17 trees (Eltrop and Marschner 1996; Constable et al. 2001).

18 Although it has been used only with seedlings to date, the SUM column  
19 method could be applied to roots of mature trees. At two field sites in Syracuse,  
20 NY, we excavated small soil pits and installed capped SUM columns vertically in  
21 the soil. We attached tubing to the cap and made sure the tubing extended  
22 slightly above the soil surface. We trained root branches of eastern hemlock and

1 sugar maple to grow into the side of the column through a small hole.  
2 Unfortunately fresh root weights in the columns averaged less than 1 g after 4  
3 months, and we were unable to detect differences in uptake between controls and  
4 plants. More studies are necessary to test methods to promote root growth in the  
5 SUM columns. The columns may need to be watered or fertilized to promote root  
6 growth, since the surrounding soil is likely to be more nutrient-rich than the  
7 medium in the columns.

8         Although the SUM column method holds promise for obtaining more  
9 realistic estimates of uptake by seedlings and mature trees, there are several  
10 drawbacks to this technique. First, as with other lysimeter methods, plants must  
11 be grown in sand rather than native soil to minimize adsorption of nutrients on the  
12 soil surface. However, uptake in sand may not be representative of uptake in  
13 native soil. Second, it is more difficult to determine the concentration of nutrients  
14 at the root surface in sand than solution culture since nutrient solution can more  
15 easily be mixed and aerated when not in a soil matrix. We homogenized the  
16 solution concentrations every ½ to 1 h by recirculating the nutrient solution, but  
17 this mixing may disturb roots, mycorrhizas and microbes in the sand. Other  
18 lysimeter studies use small containers (25 cm-long glass vials) with small  
19 seedlings (dry weights < 4 g) to maximize the availability of nutrients at the root  
20 surface (Colpaert et al. 1999). Third, differences in nitrification rates in the plant  
21 and the control SUM columns would cause estimates of  $\text{NH}_4^+$  and  $\text{NO}_3^-$  uptake to  
22 be inaccurate.

## 1 ***Spatial mapping methods***

2 A promising technique for determining which roots are actively taking up nutrients  
3 is the digital autoradiographic technique (Rubio et al. 2004). In this technique,  
4 plants are grown in sand-filled pots in the laboratory and irrigated with nutrient  
5 solution. To measure uptake, root systems are excavated but left attached to the  
6 shoot and transferred into containers with  $^{32}\text{P}$ -labelled solution. After uptake  
7 occurs, the roots are removed, excised and separated into different root classes.  
8 The root segments are scanned to measure surface area and length and placed  
9 on a phosphor screen that generates a graphical representation of the spatial  
10 distribution of  $^{32}\text{P}$ . The rate of  $^{32}\text{P}$  uptake is used to estimate specific uptake rates,  
11 as with other labeling methods.

12 This is the first technique to quantify how P uptake rates vary within a root  
13 system using intact plants. In the study, P uptake differed in basal, lateral basal,  
14 and lateral tap roots of bean plants (Rubio et al. 2004). Spatial variation in uptake  
15 along the root axis is important for understanding how plants regulate uptake and  
16 for improving uptake models, which currently do not address spatial heterogeneity  
17 in uptake within a root system (Smethurst and Comerford 1993). Like other  
18 methods that rely on uptake of tracers, this method can estimate only gross influx  
19 rates.

20

## 1 **Conclusions**

2 Measuring specific root uptake by intact roots of trees is critical to obtaining  
3 realistic rates of uptake and improving our understanding of belowground  
4 processes. The most widely used methods to measure uptake, however, rely on  
5 measurements of uptake by tree seedlings in solution culture. Methods that have  
6 measured uptake by intact roots have relied upon observations from roots that  
7 have been excised or excavated from the soil. Although these estimates of uptake  
8 may be useful for comparative purposes, more realistic estimates are necessary to  
9 describe root uptake by seedlings and mature trees in the field.

10       The task of measuring specific root uptake is limited by methodological  
11 problems which need to be addressed. Special consideration should be given to  
12 selecting the best time to sample and the concentrations to which the root will be  
13 exposed. Uptake measurements that exclude efflux should be interpreted with  
14 caution. In addition, the amount of disturbance should be carefully evaluated  
15 before selecting a method since many methods rely on observations from excised  
16 or excavated roots. Studies that compare estimates of uptake among methods are  
17 essential to determine how different experimental conditions affect estimates of  
18 uptake.

19       Recent developments using sand culture and digital autoradiography show  
20 promise, since they enable researchers to quantify uptake of intact root systems.  
21 These new methods create opportunities to more accurately measure spatial and  
22 diurnal variation in uptake rates of seedlings and mature trees in the field.



1           Although considerable progress has been made in the field of nutrient  
2 uptake by trees, many questions about how and when tree roots take up nutrients  
3 remain unanswered. How does uptake vary diurnally and seasonally for a given  
4 species or forest type? How much of that variation is controlled by nutrient  
5 availability as opposed to phenology of the tree? How much do mycorrhizas  
6 contribute to nutrient acquisition under field conditions for various tree species?  
7 Such questions not only are critical to our mechanistic understanding of nutrient  
8 acquisition, but also have practical implications for maximizing fertilizer use  
9 efficiency and forest production.

10

#### 11 **Acknowledgements**

12 We wish to thank Sarah Kulpa and Don Bickelhaupt for their technical assistance.

13 Financial support was provided by the National Science Foundation through grants

14 DEB-0087263 and 9211768.

15

## 1 **References**

- 2 Aslam, M., R.L. Travis and R.C. Huffaker. 1992. Comparative kinetics and  
3 reciprocal inhibition of nitrate and nitrite uptake in roots of uninduced and  
4 induced barley (*Hordeum vulgare* L.) seedlings. *Plant Physiol.* 99:1124-  
5 1133.
- 6 Aslam, M., R.L. Travis, D.W. Rains and R.C. Huffaker. 1996. Effect of root  
7 perturbation and excision on nitrate influx and efflux in barley (*Hordeum*  
8 *vulgare*) seedlings. *Physiol. Plant.* 97:425-432.
- 9 Atkinson, D. and S.A. Wilson 1979. The root soil interface and its significance for  
10 fruit tree roots of different ages. *In* The soil-root interface Eds. J.L. Harley  
11 and R.S. Russell. Academic Press, London, pp. 259-271.
- 12 Bakker, M.R., R. Kerisit, K. Verbist and C. Nys. 1999. Effects of liming on  
13 rhizosphere chemistry and growth of fine roots and shoots of sessile oak  
14 (*Quercus petraea*). *Plant Soil.* 217:243-255.
- 15 Barber, S.A. 1995. Soil nutrient bioavailability: a mechanistic approach. John Wiley  
16 and Sons, Inc., New York. 384 p.
- 17 BassiriRad, H., K.L. Griffin, J.F. Reynolds and B.R. Strain. 1997. Changes in root  
18  $\text{NH}_4^+$  and  $\text{NO}_3^-$  absorption rates of loblolly and ponderosa pine in response  
19 to  $\text{CO}_2$  enrichment. *Plant Soil.* 190:1-9.
- 20 BassiriRad, H.H., S.A. Prior, R.J. Norby and H.H. Rogers. 1999. A field method of  
21 determining  $\text{NH}_4^+$  and  $\text{NO}_3^-$  uptake kinetics in intact roots: effects of  $\text{CO}_2$   
22 enrichment on trees and crop species. *Plant Soil.* 217:195-204.

- 1 Bhat, K.K.S. 1982. Determination of the relationship between nutrient uptake rate  
2 and solution concentration at the root surface under field conditions:  $^{32}\text{P}$ -  
3 orthophosphate uptake by apple roots. *J. Exp. Bot.* 33:190-194.
- 4 Bledsoe, C.S. and D.W. Rains. 1981. Cation uptake by Douglas-fir seedlings  
5 grown in solution culture. *Can. J. For. Res.* 11:812-?
- 6 Bloom, A.J. and R.M. Caldwell. 1988. Root excision decreases nutrient absorption  
7 and gas fluxes. *Plant Physiol.* 87:794-796.
- 8 Bloom, A.J., R.M. Caldwell, J. Finazzo, R.L. Warner and J. Weissbart. 1989.  
9 Oxygen and carbon dioxide fluxes from barley shoots depend on nitrate  
10 assimilation. *Plant Physiol.* 91:352-356.
- 11 Bloom, A.J., S.S. Sukrapanna and R.L. Warner. 1992. Root respiration associated  
12 with ammonium and nitrate absorption and assimilation by barley. *Plant*  
13 *Physiol.* 99:1294-1301.
- 14 Bowen, G.D. 1970. Early detection of phosphate deficiency in plants. *Comm. Soil*  
15 *Sci. Plant Anal.* 1:293-298.
- 16 Caldwell, M.M., D.M. Eissenstat, J.H. Richards and M.F. Allen. 1985. Competition  
17 for phosphorus: differential uptake from dual-isotope-labeled soil  
18 interspaces between shrub and grass. *Science.* 229:384-386.
- 19 Claasen, N. and S.A. Barber. 1974. A method for characterizing the relation  
20 between nutrient concentration and flux into roots of intact plants. *Plant*  
21 *Physiol.* 54:564-568.

- 1 Clark, G.T., J. Dunlop and H.T. Phung. 2000. Phosphate absorption by  
2 *Arabidopsis thaliana*: interactions between phosphorus status and inhibition  
3 by arsenate. Australian Journal of Plant Physiology. 27:959-965.
- 4 Clarkson, D.T., M.G.T. Shone and A.V. Wood. 1974. The effect of pretreatment  
5 temperature on the exudation of xylem sap by detached barley root  
6 systems. Planta. 121:81-92.
- 7 Colpaert, J.V., K.K. Van Tichelen, J.A. Van Assche and A. Van Laere. 1999.  
8 Short-term phosphorus uptake rates in mycorrhizal and non-mycorrhizal  
9 roots of intact *Pinus sylvestris* seedlings. New Phytol. 143:589-597.
- 10 Constable, J.V.H., H. BassiriRad, J. Lussenhop and A. Zerihun. 2001. Influence of  
11 elevated CO<sub>2</sub> and mycorrhizae on nitrogen acquisition: contrasting  
12 responses in *Pinus taeda* and *Liquidambar styraciflua*. Tree Physiol. 21:83-  
13 91.
- 14 Cui, M. and M.M. Caldwell. 1997. Growth and nitrogen uptake by *Agropyron*  
15 *desertorum* and *Pseudoroegneria spicata* when exposed to nitrate pulses of  
16 different duration. Australian Journal of Plant Physiology. 24:637-642.
- 17 Dean-Drummond, C.E. 1982. Mechanisms for nitrate uptake into barley (*Hordeium*  
18 *vulgare* cv. *fergus*) seedlings grown at controlled nitrate concentrations in  
19 the nutrient medium. Plant Sci. Lett. 24:79-89.
- 20 Delhon, P., A. Gojon, P. Tillard and L. Passama. 1996. Diurnal regulation of NO<sub>3</sub><sup>-</sup>  
21 uptake in soybean plants IV. Dependence on current photosynthesis and  
22 sugar availability to the roots. J. Exp. Bot. 47:893-900.

- 1 Dieffenbach, A., A. Gottlein and E. Matzner. 1997. In-situ soil solution chemistry in  
2 an acid forest soil as influenced by growing roots of Norway spruce (*Picea*  
3 *abies* [L.] Karst.). Plant Soil. 192:57-61.
- 4 Dieffenbach, A. and E. Matzner. 2000. In situ soil solution chemistry in the  
5 rhizosphere of mature Norway spruce (*Picea abies* [L.] Karst.) trees. Plant  
6 Soil. 222:149-161.
- 7 Dinkelmeyer, H., J. Lehmann, A. Renck, L. Trujillo, J. Pereira da Silva Jr., G.  
8 Gebauer and K. Kaiser. 2003. Nitrogen uptake from <sup>15</sup>N-enriched fertilizer  
9 by four tree crops in an Amazonian agroforest. Agroforestry Systems.  
10 57:213-224.
- 11 Drew, M.C., L.R. Saker, S.A. Barber and W. Jenkins. 1984. Changes in kinetics of  
12 phosphate and potassium absorption in nutrient-deficient barley roots  
13 measured by solution-depletion technique. Planta. 160:490-499.
- 14 Elliott, G.C., J. Lynch and A. Lauchli. 1984. Influx and efflux of P in roots of intact  
15 maize plants. Plant Physiol. 76:336-341.
- 16 Eltrop, L. and H. Marschner. 1996. Growth and mineral nutrition of non-  
17 mycorrhizal and mycorrhizal Norway spruce (*Picea abies*) seedlings grown  
18 in semi-hydroponic sand culture. I. Growth and mineral nutrient uptake in  
19 plants supplied with different forms of nitrogen. New Phytol. 133:469-478.
- 20 Epstein, E., W.E. Schmid and D.W. Rains. 1963a. Significance and technique of  
21 short-term experiments on solute absorption by plant tissue. Plant Cell  
22 Physiol. 4:79-84.

- 1 Epstein, E., W.E. Schmid and D.W. Rains. 1963b. Significance and technique of  
2 short-term experiments on solute absorption by plant tissue. *Plant Cell*  
3 *Physiol.* 4:79-84.
- 4 Escamilla, J.A. and N.B. Comerford. 1998. Measuring nutrient depletion by roots  
5 of mature trees in the field. *Soil Sci. Soc. Am. J.* 62:797-804.
- 6 Espeleta, J.F. and D.M. Eissenstat. 1998. Responses of citrus fine roots to  
7 localized soil drying: a comparison of seedlings and adult fruiting trees.  
8 *Tree Physiol.* 18:113-119.
- 9 Gessler, A., J. Kreuzwieser, T. Dopatka and H. Rennenberg. 2002. Diurnal  
10 courses of ammonium net uptake by the roots of adult beech (*Fagus*  
11 *sylvatica*) and spruce (*Picea abies*) trees. *Plant Soil.* 240:23-32.
- 12 Gessler, A., S. Schneider, D. Von Sengbusch, P. Weber, U. Hanemann, C. Huber,  
13 A. Rothe, K. Kreuzer and H. Rennenberg. 1998. Field and laboratory  
14 experiments on net uptake of nitrate and ammonium by the roots of spruce  
15 (*Picea abies*) and beech (*Fagus sylvatica*) trees. *New Phytol.* 138:275-285.
- 16 Glass, A.D.M. 1978. Influence of excision and aging upon K<sup>+</sup> influx into barley  
17 roots: recovery or enhancement? *Plant Physiol.* 61:481-483.
- 18 Glass, A.D.M., M. Saccomani, G. Crookall and M.Y. Siddiqi. 1987. A  
19 microcomputer-controlled system for the automatic measurement and  
20 maintenance of ion activities in nutrient solutions during their absorption by  
21 intact plants in hydroponic facilities. *Plant Cell Env.* 10:375-381.

- 1 Gobran, G. and S. Clegg. 1996. A conceptual model for nutrient availability in the  
2 mineral soil-root system. *Can. J. Soil Sci.* 76:125-131.
- 3 Gronewald, J.W. and J.B. Hanson. 1980. Sensitivity of the proton and ion transport  
4 mechanisms of corn roots to injury. *Plant Sci. Lett.* 18:143-150.
- 5 Gronewald, J.W. and J.B. Hanson. 1982. Adenine nucleotide content of corn roots  
6 as affected by injury and subsequent washing. *Plant Physiol.* 69:1252-1256.
- 7 Hangs, R.D., J.D. Knight and K.C.J. Van Rees. 2003. Nitrogen uptake  
8 characteristics for roots of conifer seedlings and common boreal forest  
9 competitor species. *Can. J. For. Res.* 33:156-163.
- 10 Hansen, G.K. 1980. Diurnal variation of root respiration rates and nitrate uptake as  
11 influenced by nitrogen supply. *Physiol. Plant.* 48:421-427.
- 12 Hoagland, D.R. and T.C. Broyer. 1936. General nature of the process of salt  
13 accumulation by roots with description of experimental methods. *Plant*  
14 *Physiol.* 11:471-507.
- 15 Hogberg, M.N., L. Hogbom and H. Schinkel. 1998. Nitrogen-related root variables  
16 of trees along a N-deposition gradient. *Tree Physiol.* 18:823-828.
- 17 Huang, Z.Z., X. Yan, A. Jalil, J.D. Norlyn and E. Epstein. 1992. Short-term  
18 experiments on ion transport by seedlings and excised roots: technique and  
19 validity. *Plant Physiol.* 100:1914-1920.
- 20 Ingestad, T. and A.B. Lund. 1979. Nitrogen stress in birch seedlings. I. Growth  
21 technique and growth. *Physiol. Plant.* 45:137- ?

- 1 Jones, H.E., P. Högberg and H. Ohlsson. 1994. Nutritional assessment of a forest  
2 fertilisation experiment in northern Sweden by root bioassays. *For. Ecol.*  
3 *Man.* 64:59-69.
- 4 Kelly, J.M., S.A. Barber and G.S. Edwards. 1992. Modeling magnesium,  
5 phosphorus and potassium uptake by loblolly pine seedlings using a  
6 Barber-Cushman approach. *Plant Soil.* 139:209-218.
- 7 Kelly, J.M., A.H. Chappelka and B.G. Lockaby. 1994. Measured and estimated  
8 parameters for a model of nutrient uptake by trees. *N. Z. J. For. Sci.*  
9 24:213-225.
- 10 Kelly, J.M. and J.K. Kelly. 2001. Phosphorus and potassium uptake kinetics in red  
11 maple seedlings. *For. Sci.* 47:397-402.
- 12 Kelly, J.M., J.D. Scarbrough and P.A. Mays. 2001. Hardwood seedling root and  
13 nutrient parameters for a model of nutrient uptake. *Journal of Environmental*  
14 *Quality.* 30:427-439.
- 15 Lajtha, K. 1994. Nutrient uptake in eastern deciduous tree seedlings. *Plant Soil.*  
16 160:193-199.
- 17 Langlois, C.G. and J.A. Fortin. 1984. Seasonal variations in the uptake of  
18 [<sup>32</sup>P]phosphate ions by excised ectomycorrhizae and lateral roots of *Abies*  
19 *balsamea*. *Can. J. For. Res.* 14:412-415.
- 20 Law, B.E., A.H. Goldstein, P.M. Anthoni, M.H. Unsworth, J.A. Panek, M.R. Bauer,  
21 J.M. Frachebound and N. Hultman. 2001. Carbon dioxide and water vapor



- 1 exchange by young and old ponderosa pine ecosystems during a dry  
2 summer. *Tree Physiol.* 21:298-308.
- 3 Lee, R.B. and K.A. Rudge. 1986. Effects of nitrogen deficiency on the absorption  
4 of nitrate and ammonium by barley plants. *Ann. Bot.* 57:471-486.
- 5 Lipp, C.C. and C.P. Andersen. 2003. Role of carbohydrate supply in white and  
6 brown root respiration of ponderosa pine. *New Phytol.* 160:523-531.
- 7 Lucash, M.S. 2005. Methods for measuring nutrient uptake rates of intact roots of  
8 seedlings and mature trees. *In* Department of Forest and Natural  
9 Resources Management. State University of New York College of  
10 Environmental Science and Forestry, Syracuse, p. 114.
- 11 Lucash, M.S., J.D. Joslin and R.D. Yanai. 2005. Temporal variation in nutrient  
12 uptake capacity by intact roots of mature loblolly pine. *Plant Soil.* 272:253-  
13 262.
- 14 Macduff, J.H., A.K. Bakken and M.S. Dhanoo. 1997. An analysis of the  
15 physiological basis of commonality between diurnal patterns of  $\text{NH}_4^+$ ,  $\text{NO}_3^-$ ,  
16 and  $\text{K}^+$  uptake by *Phleum pratense* and *Festuca pratensis*. *J. Exp. Bot.*  
17 48:1691-1701.
- 18 Marschner, H., M. Haussling and E. George. 1991. Ammonium and nitrate uptake  
19 rates and rhizosphere pH in non-mycorrhizal roots of Norway spruce [*Picea*  
20 *abies* (L.) Karst.]. *Trees.* 5:14-21.

- 1 McFarlane, K.J. and R.D. Yanai. 2006. Measuring nitrogen and phosphorus  
2 uptake by intact roots of mature *Acer saccharum* Marsh., *Pinus resinosa*  
3 Ait., and *Picea abies* (L.) Karst. *Plant Soil*. 279:163-172.
- 4 McKane, R.B., P.T. Rygielwicz, P.A. Beedlow, C.P. Andersen, J.R. Brooks, W.E.  
5 Hogsett, M. Hynes and J.A. Laurence. 2003. Lateral root distribution of  
6 trees in an old-growth Douglas-fir forest inferred from uptake of tracer <sup>15</sup>N.  
7 *Proceedings of Ecological Society of America*
- 8 Nambiar, E.K.S. and G.D. Bowen. 1986. Uptake, distribution and retranslocation of  
9 nitrogen by *Pinus radiata* from <sup>15</sup>N-labelled fertilizer applied to podzolized  
10 sandy soil. *For. Ecol. Man.* 15:269-284.
- 11 Nichol, B.E., L.A. Oliveira, A.D.M. Glass and M.Y. Siddiqi. 1993. The effects of  
12 aluminum on the influx of calcium, potassium, ammonium, nitrate and  
13 phosphate in an aluminum-sensitive cultivar of barley (*Hordeum vulgare* L.).  
14 *Plant Physiol.* 101:1263-1266.
- 15 Nissen, P. 1991. Uptake Mechanisms. *In Plant roots: the hidden half* Eds. Y.  
16 Waisel, A. Eshel and U. Kafkafi. Marcel Dekker Inc., New York, pp. 483-  
17 502.
- 18 Ohlund, J. and T. Nasholm. 2004. Regulation of organic and inorganic nitrogen  
19 uptake in Scots pine (*Pinus sylvestris*) seedlings. *Tree Physiol.* 24:1397-  
20 1402.

- 1 Ourry, A., J.H. Macduff, M.-P. Prudhomme and J. Boucaud. 1996. Diurnal  
2 variation in the simultaneous uptake and 'sink' allocation of  $\text{NH}_4^+$  and  $\text{NO}_3^-$   
3 by *Lolium perenne* in flowing solution culture. J. Exp. Bot. 47:1853-1863.
- 4 Pearson, C.J., R.J. Volk and W.A. Jackson. 1981. Daily changes in nitrate influx,  
5 efflux and metabolism in maize and pearl millet. Planta. 152:319-324.
- 6 Proe, M.F., A.J. Midwood and J. Craig. 2000. Use of stable isotopes to quantify  
7 nitrogen, potassium and magnesium dynamics in young Scots pine (*Pinus*  
8 *sylvestris*). New Phytol.:461-469.
- 9 Rennenberg, H., S. Schneider and P. Weber. 1996. Analysis of uptake and  
10 allocation of nitrogen and sulfur compounds by trees in the field. J. Exp.  
11 Bot. 47:1491-1498.
- 12 Rincon, M. and J.B. Hanson. 1986. Controls on calcium ion fluxes in injured or  
13 shocked corn root cells: importance of proton pumping and cell membrane  
14 potential. Physiol. Plant. 67:576-583.
- 15 Rothstein, D.E., D.R. Zak and K.S. Pregitzer. 1996. Nitrate deposition in northern  
16 hardwood forests and the nitrogen metabolism of *Acer saccharum* marsh.  
17 Oecologia. 108:338-344.
- 18 Rothstein, D.E., D.R. Zak, K.S. Pregitzer and P.S. Curtis. 2000. Kinetics of  
19 nitrogen uptake by *Populus tremuloides* in relation to atmospheric  $\text{CO}_2$  and  
20 soil nitrogen availability. Tree Physiol. 20:265-270.

- 1 Rubio, G., A. Sorgona and J.P. Lynch. 2004. Spatial mapping of phosphorus influx  
2 in bean root systems using digital autoradiography. *J. Exp. Bot.* 55:2269-  
3 2280.
- 4 Rygiewicz, P.T. and C.S. Bledsoe. 1986. Effects of pretreatment conditions on  
5 ammonium and nitrate uptake by Douglas-fir seedlings. *Tree Physiol.*  
6 1:145-150.
- 7 Saglio, P.H. and A. Pradet. 1980. Soluble sugars, respiration and energy charge  
8 during aging of excised maize root tips. *Plant Physiol.* 66:516-519.
- 9 Scaife, A. and Schloemer. 1994. The diurnal pattern of nitrate uptake and  
10 reduction by spinach (*Spinacia oleraceae* L.). *Ann. Bot.* 73:337-343.
- 11 Scholberg, J.M.S., L.R. Parsons, T.A. Wheaton and K.T. Morgan. 2001.  
12 Procedures for determining the effects of environmental conditions on plant  
13 nitrogen uptake: an alternative approach. *Soil Crop Sci. Soc. Florida Proc.*  
14 60:40-49.
- 15 Siddiqi, M.Y., A.D.M. Glass, T.J. Ruth and M. Fernando. 1989. Studies of the  
16 regulation of nitrate influx by barley seedlings using  $^{13}\text{NO}_3^-$ . *Plant Physiol.*  
17 90:1426-1432.
- 18 Siddiqi, M.Y., A.D.M. Glass, T.J. Ruth and T.W. Ruffy. 1990. Studies of the uptake  
19 of nitrate in barley. I. Kinetics of  $^{13}\text{NO}_3^-$  influx. *Plant Physiol.* 90:1426-1432.
- 20 Skene, K.R., J. Raven and J.L. Sprent. 1998. Cluster root development in  
21 *Grevillea robusta* (Proteaceae). I. Xylem, pericycle, cortex and epidermis  
22 development in a determinate root. *New Phytol.* 138:725-732.

- 1 Smethurst, P.J. and N.B. Comerford. 1993. Potassium and phosphorus uptake by  
2 competing pine and grass: observations and model verification. *Soil Sci.*  
3 *Soc. Am. J.* 57:1602-1610.
- 4 Smith, S.E. and D.J. Read. 1997. *Mycorrhizal symbiosis*. Academic Press, San  
5 Diego. 605 p.
- 6 Syvertsen, J.P. and M.L. Smith. 1996. Nitrogen uptake efficiency and leaching  
7 losses from lysimeter-grown *Citrus* trees fertilized at three nitrogen rates.  
8 *Journal of American Horticultural Society.* 121:57-62.
- 9 Van der Driessche, R. 1971. Response of conifer seedlings to nitrate and  
10 ammonium sources of nitrogen. *Plant Soil.* 34:421- ?
- 11 Van Rees, K.C.J., N.B. Comerford and W.W. McFee. 1999. Modeling potassium  
12 uptake by slash pine seedlings from low-potassium-supplying soils. *Soil Sci.*  
13 *Soc. Am. J.* 54:1413-1421.
- 14 Wang, M.Y., M.Y. Siddiqi and A.D.M. Glass. 1996. Interactions between  $K^+$  and  
15  $NH_4^+$ : effects on ion uptake by rice roots. *Plant Cell Env.* 19:1037-1046.
- 16 Wang, M.Y., M.Y. Siddiqi, T.J. Ruth and A.D.M. Glass. 1993. Ammonium uptake  
17 by rice roots. II. Kinetics of  $^{13}NH_4^+$  influx across the plasmalemma. *Plant*  
18 *Physiol.* 103:1259-1267.
- 19 Weinbaum, S.A., F.J.A. Niederholzer, S. Ponchner, R.C. Rosecrance, R.M.  
20 Carlson, A.C. Whittlesey and T.T. Muraoka. 1994. Nutrient uptake by  
21 cropping and defruited field-grown 'French' prune trees. *Journal of*  
22 *American Horticultural Society.* 119:925-930.

- 1 Weinbaum, S.A. and C. Van Kessel. 1998. Quantitative estimates of uptake and  
2 internal cycling of <sup>14</sup>N-labeled fertilizer in mature walnut trees. *Tree Physiol.*  
3 18:795-801.
- 4 Woodend, J.J., A.D.M. Glass and C.O. Person. 1986. Intraspecific variation for  
5 nitrate uptake and nitrogen utilization in wheat (*T. aestivum* L.) grown under  
6 nitrogen stress. *J. Plant Nut.* 9:1213-1225.
- 7 Yanai, R.D., H. Majdi and B.B. Park. 2003. Measured and modelled differences in  
8 nutrient concentrations between rhizosphere and bulk soil in a Norway  
9 spruce stand. *Plant Soil.* 257:133-142.
- 10 Yoder, C. and M.M. Caldwell. 2002. Effects of perennial neighbors and nitrogen  
11 pulses on growth and nitrogen uptake by *Bromus tectorum*. *Plant Ecology.*  
12 158:77-84.
- 13

- 1 Table 1. Ammonium concentrations used to quantify uptake capacity in nine  
2 studies involving tree species, in order of increasing concentration.

Species	Conc. ( $\mu\text{mol L}^{-1}$ )	Technique	Author
<i>Fagus sylvatica</i> <i>Picea abies</i>	53-55	Depletion	Gessler et al. (1998)
<i>Picea abies</i>	0-150	Depletion	Marschner et al. (1991)
<i>Acer rubrum</i> <i>Acer saccharum</i>	0-200	Depletion	BassiriRad et al. (1999)
<i>Pinus ponderosa</i> <i>Pinus taeda</i>	0-500	Excised	BassiriRad et al. (1996)
<i>Populus tremuloides</i>	0-500	Excised	Rothstein et al. (2000)
<i>Picea abies</i>	800	Lysimeter	Eltrop and Marschner (1996)
<i>Acer saccharum</i>	0-1000	Excised	Rothstein et al. (1996)
<i>Betula alleghaniensis</i> <i>Carya ovata</i> <i>Fagus sylvatica</i> <i>Fraxinus americana</i> <i>Liriodendron tulipifera</i> <i>Prunus serotina</i> <i>Quercus phellos</i>	0-4000	Excised	Lajtha (1994)

3

4

## 1 **Figure Legends**

2

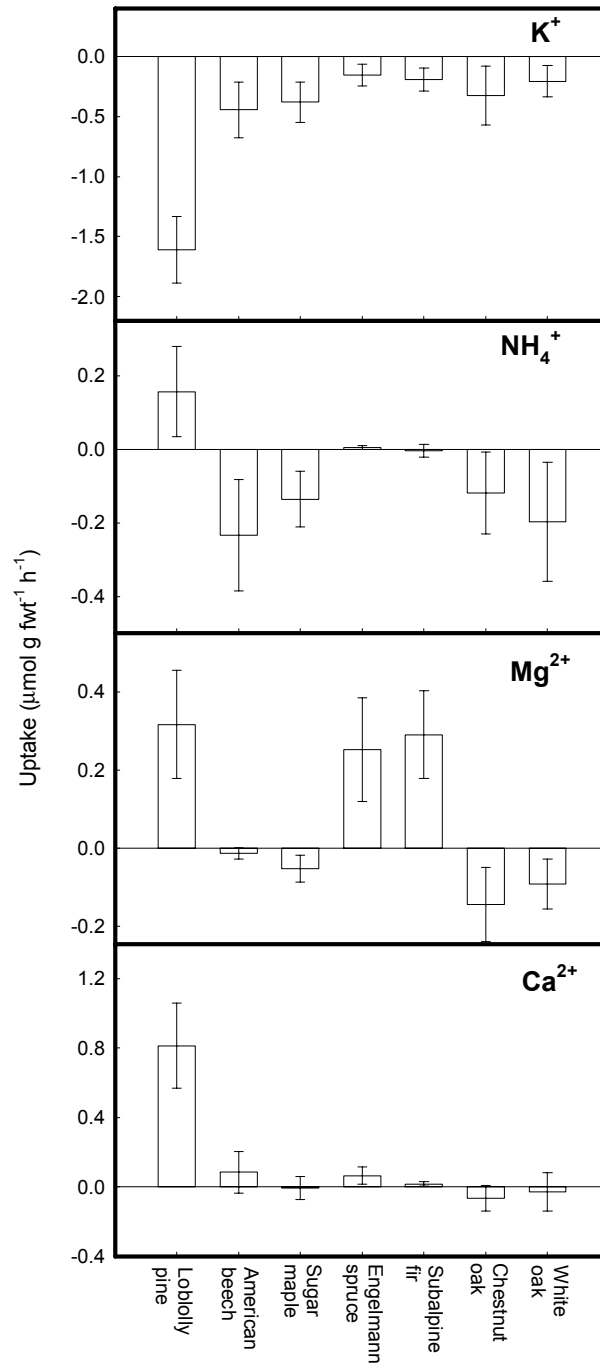
3 Figure 1. Mean net uptake (positive values) or net efflux (negative values) of by  
4 seven different tree species measured using the depletion method on intact roots.  
5 In 2000-2001, roots at Calhoun Experimental Forest, SC (loblolly pine), Hubbard  
6 Brook Experimental Forest, NH (beech and maple), Huntington Forest, NY (beech  
7 and maple), Loch Vale Experimental Forest, CO (spruce and fir), Fraser  
8 Experimental Forest, CO (spruce and fir) and Walker Branch, TN (chestnut and  
9 white oak) were excavated and placed in solutions that simulated soil  
10 concentrations of  $K^+$ ,  $NH_4^+$ ,  $Mg^{2+}$ , and  $Ca^{+2}$ . Changes in nutrient concentrations  
11 were monitored over time. Vertical bars indicate standard errors of the mean (n=  
12 4-11).

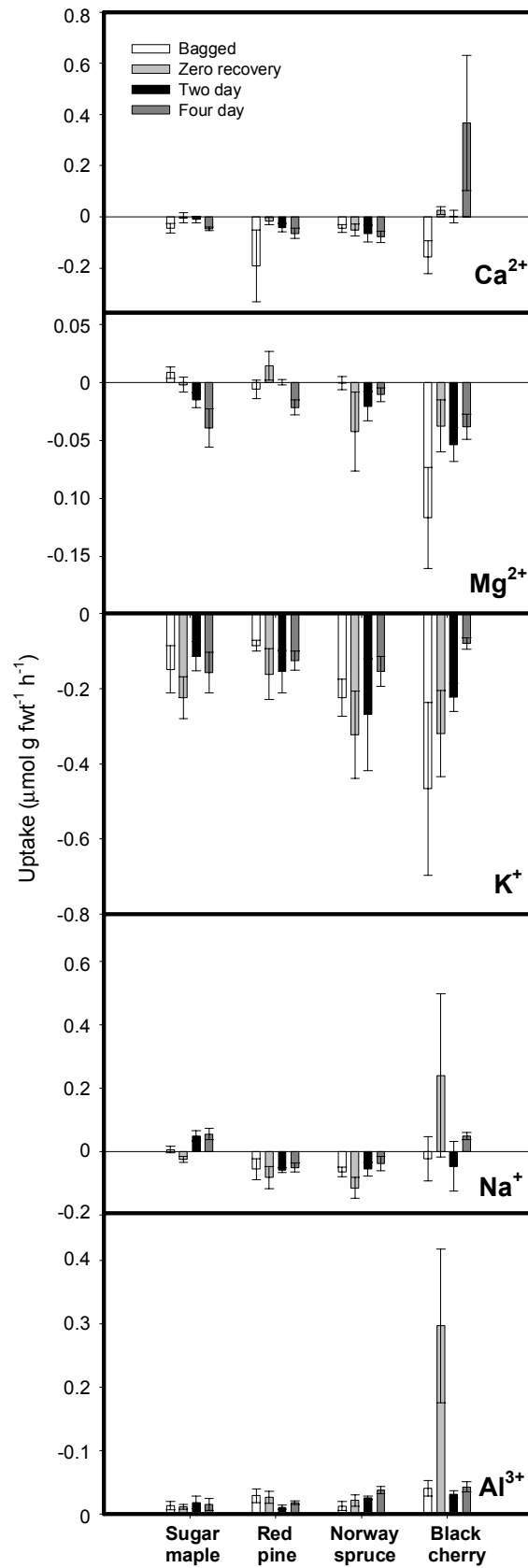
13

14 Figure 2. Mean net uptake (positive values) or net efflux (negative values) of  $Ca^{+2}$ ,  
15  $Mg^{2+}$ ,  $K^+$ ,  $Na^+$  and  $Al^{+3}$  by four tree species given pretreatments to mitigate  
16 excavation-related disturbance. "Bagged" roots were grown in bags filled with a  
17 sand-soil mixture. "Zero recovery" roots were excavated and used immediately for  
18 uptake experiments. "Two day" and "Four day recovery" roots were excavated and  
19 given two- or four-day recovery periods, prior to experiments. Vertical bars  
20 indicate standard errors of the mean (n=7-10). For a detailed description of the  
21 methodology and the effect of treatments on N and P uptake, see McFarlane and  
22 Yanai (2006).



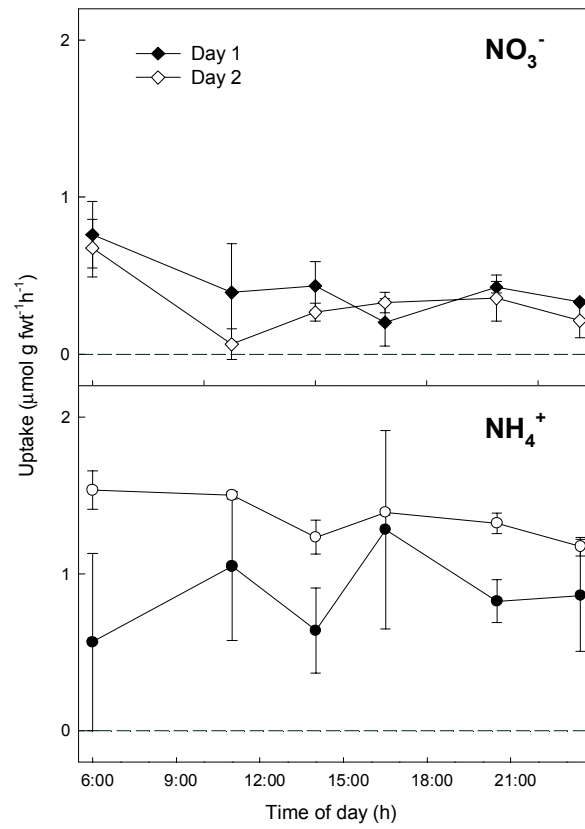
1 Figure 3. Mean net uptake of  $\text{NO}_3^-$  and  $\text{NH}_4^+$  by white pine seedlings grown in  
2 soil-uptake-monitoring (SUM) columns filled with sand. Uptake of  $\text{NO}_3^-$  and  $\text{NH}_4^+$   
3 was measured by adding  $\text{KNO}_3$  or  $\text{NH}_4\text{Cl}$  to the SUM columns. After circulating  
4 the solution in the columns for one hour, the solution was removed by a vacuum  
5 pump and analyzed for  $\text{NO}_3^-$  or  $\text{NH}_4^+$ . Uptake rates were corrected for changes in  
6 the concentration of the controls, which did not contain plants. Vertical bars  
7 indicate standard errors of the mean (n= 3).  
8





1

2



3