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## The Distribution of Tree Roots in Douglas-fir Forests in the Pacific Northwest in Relation to Depth, Space, Coarse Organic Matter and Mineral Fragments

### Abstract

This study evaluated relationships between site or tree characteristics and below-ground materials in Douglas-fir forests of the Pacific Northwest. We core-sampled living roots, dead organic matter, and mineral fragments at three soil depths on a 300-sample grid at nine forested sites in western Washington and Oregon resulting in approximately 7200 samples. We explored relationships between materials across depths and at three scales—point, block and site—and examined the data to quantify the degree of spatial clustering. Mass of roots and organic matter declined with depth; there were no consistent patterns with depth for mineral fragments. Correlations between values at the same point but at different depths were low for roots and organic matter at all sites (mean  $r$  values  $< 0.3$ ). Total soil carbon per site was negatively correlated ( $r = -0.85$ ) with total root mass; correlations between other site or tree variables and below-ground materials were much lower ( $r = -0.21$  to  $0.34$ ). There were no detectable spatial patterns in root mass. The number of samples needed to estimate mean root mass was calculated by site and for several desired precision levels; this power analysis will aid others working at similar sites in determining necessary sample sizes *a priori*, or conversely, in estimating level of precision given a particular sample size. Precision of  $\pm 1.5 \text{ kg m}^{-3}$  could be achieved with less than 100 samples in the upper layer at most sites.

**Keywords:** Spatial variability; forest soils; soil C; tree roots; sample precision.

### Introduction

Below-ground ecology of forests is poorly understood compared to our vast knowledge of above-ground ecology of forest ecosystems. Most information on root systems comes from herbaceous plants, especially those of crop species for which water and nutrient uptake may limit plant productivity (Torrey and Clarkson 1975, Coutts 1987, Kramer and Boyer 1995, Lynch 1995, Eshel and Beeckman 2013) and detailed information is lacking for many larger forest plants (Hermann 1977, Sutton 1969). To better understand and

manage processes in below-ground systems, forest managers and scientists need to understand where roots are distributed within the soil, variability of root density and how roots may impact soil development (Zinke 1962). Such information is key to understanding how forests function below ground, predicting above- and below-ground carbon storage, and designing sampling approaches and hypotheses for future experiments.

A better understanding of root architecture, including the spatial distribution of roots, can lead to a better understanding of plant productivity (Lynch 1995). Clustering of roots occurs for three main reasons: 1) they cluster in space because daughter roots are necessarily concentrated close to their mother root (Logsdon and Allmaras 1991), 2) they cluster by depth depending on availability of water

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and nutrients with soil layer, and 3) they cluster based on changes in the local environment of the root such as cracks in compacted soil or patches of nutrients or residues from previous plants (Bengough et al. 2000) or water availability. The spatial distribution of roots in three dimensions differs by plant species and with the spacing and arrangement of individual plants (van Noordwijk et al. 1985). Tree roots move around rocks (Coutts and Phillipson 1976) and often follow former root channels (cf. Carlson et al. 1988), especially in compacted soils or soils with naturally high strength or strong structure. Tree root distribution has also been reported to correspond to soil type (Curt et al. 2001). Since coarse organic materials in soil are primarily decomposed roots, it could be speculated that if roots are clustered, then organic materials might also be clustered. On the other hand, if roots are not clustered and exhibit rapid turnover, we might expect organic matter distribution in soils to become more uniform over time as roots grow, die and then colonize new locations. Coarse mineral fragments, particularly gravel and small rocks, could be clustered as a result of topography and water movement. For example, if outwash processes resulted in areas with similar size and distribution of coarse fragments, then point sampling on such soils might be expected to result in spatial correlation of our samples.

Tree root systems are notoriously difficult and expensive to sample, and thus, to quantify. To efficiently study root systems and related below-ground characteristics we need to understand not only the degree of clustering but also the variability in root distribution with depth and across spatial scales. In this study, we use a new data set of more than 7000 soil samples from nine forested sites in western Washington and Oregon to quantify living roots, coarse organic material, and mineral fragments with depth and at three spatial scales. We asked first, does the scale of sampling affect how we describe and quantify tree roots? Next, we know that roots and other below-ground characteristics become more difficult to sample with depth so we asked how effectively we could predict root mass or coarse organic materials in a deeper layer from mass in a shallower layer? We also asked if there were spatial patterns in the

data that might provide ecological insight or that would suggest alternative sampling approaches for future projects. At the site level, we asked if roots or coarse organic components are related to stand or climatic factors. We also used this data set to examine how sample size affects the precision of the estimate of mean root mass and to estimate the sample sizes needed to estimate mean root mass with a desired precision level.

## Materials

### Site Selection and Sample Collection

Nine forested sites were selected in western Washington and Oregon (Figure 1, Table 1). These sites had been previously harvested, regenerated and all except two, WA1 and OR1, had been thinned in the 1970s, 1980s or 1990s (Holub 2011). Site selection criteria included: trees large enough for commercial harvesting in a future phase of the study, stands dominated by Douglas-fir, forest stands large enough to install a 300-point sampling grid with reasonably uniform conditions (slope, aspect, landform, soils, and site conditions), and soils not mapped as rocky as the sampling method was not suitable for use on sites with large rocks. Each study site was delineated to include only one stand of trees (with similar age, species composition, and stand structure). Stand ages ranged from 35 to 50 years at the Washington sites and from 39 to 62 years in Oregon: site index (height of dominant and codominant trees at age 50) ranged from 36 to 42 m in Washington and from 36 to 44 m in Oregon. The stand at one site (WA4) was 49% (by basal area) Douglas-fir (*Pseudotsuga menziesii*) and 50% western hemlock (*Tsuga heterophylla*), stands at two sites (WA1 and OR4) were about 66% Douglas-fir and 30% western hemlock, and stands at the remaining six sites were at least 80% Douglas-fir. Other tree species present (all less than 2.5% of the basal area) were western redcedar (*Thuja plicata*), Sitka spruce (*Picea sitchensis*), noble fir (*Abies procera*), grand fir (*Abies grandis*), red alder (*Alnus rubra*) and big-leaf maple (*Acer macrophyllum*). All sites were on land owned by Weyerhaeuser Company. Additional detail on the sites and sampling procedures is available (Holub 2011).

TABLE 1. Description of sites sampled for below-ground materials > 4.75 mm. All soils were classified as well drained except OR2 which was classified as somewhat poorly drained.

| Region                                        | WA Coast |                     |                        |                       | WA Cascades           |                      |                      |                       | OR Coast             |                      |  | OR Cascades |  |
|-----------------------------------------------|----------|---------------------|------------------------|-----------------------|-----------------------|----------------------|----------------------|-----------------------|----------------------|----------------------|--|-------------|--|
|                                               | Site ID  | WA1                 | WA2                    | WA3                   | WA4                   | OR2                  | OR3                  | OR5                   | OR1                  | OR4                  |  |             |  |
| Elevation (m)                                 |          | 143                 | 466                    | 167                   | 833                   | 282                  | 751                  | 551                   | 691                  | 628                  |  |             |  |
| Slope (%)                                     |          | 18                  | 21                     | 19                    | 6                     | 8                    | 18                   | 21                    | 23                   | 33                   |  |             |  |
| Aspect                                        |          | NNW                 | SE                     | W                     | WNW                   | WNW                  | W                    | SE                    | NNW                  | WSW                  |  |             |  |
| Annual precipitation <sup>a</sup><br>(mm)     |          | 2468                | 2111                   | 1263                  | 1941                  | 1323                 | 1592                 | 2103                  | 2306                 | 1796                 |  |             |  |
| Mean annual<br>temperature <sup>b</sup> (°C)  |          | 11.9                | 11.0                   | 13.0                  | 9.5                   | 12.1                 | 11.2                 | 10.9                  | 11.3                 | 12.6                 |  |             |  |
| NRCS soil series <sup>c</sup>                 |          | Ilwaco/Narel        | Cathlamet              | Cathcart              | Reichel               | Mayger               | Orford               | Olyic                 | Blachly              | McCully              |  |             |  |
| NRCS soil series depth                        |          | Very deep           | Very deep              | Deep                  | Deep                  | Deep                 | Very deep            | Deep                  | Very deep            | Deep                 |  |             |  |
| Soil Subgroup                                 |          | Typic<br>Fulvudands | Andic<br>Dystroxerepts | Typic<br>Haploxerands | Typic<br>Fulvivryands | Aquic<br>Palehumults | Typic<br>Palehumults | Typic<br>Haplohumults | Typic<br>Dystrodepts | Humic<br>Dystrodepts |  |             |  |
| Stand age (yr)                                |          | 39                  | 39                     | 35                    | 50                    | 39                   | 56                   | 51                    | 46                   | 62                   |  |             |  |
| Site index <sup>d</sup> (m)                   |          | 41                  | 42                     | 41                    | 36                    | 44                   | 36                   | 39                    | 41                   | 38                   |  |             |  |
| Basal area (m <sup>2</sup> ha <sup>-1</sup> ) |          | 53                  | 44                     | 50                    | 54                    | 47                   | 57                   | 51                    | 65                   | 41                   |  |             |  |
| Sample area (ha)                              |          | 4.6                 | 3.3                    | 8.1                   | 3.5                   | 3.9                  | 5.9                  | 3.6                   | 9.1                  | 5.5                  |  |             |  |

<sup>a</sup>Mean annual precipitation based on PRISM predictions (Daly et al. 2008) 1971–2000: <http://www.prism.oregonstate.edu/> (accessed 01 July 2010).  
<sup>b</sup>Mean annual temperature based on DayMet predictions 1980–2003 (Thornton et al 2012). Oak Ridge National Laboratory Distributed Active Archive Center, Oak Ridge TN. Available online at: <http://daymet.ornl.gov> (accessed 14 February 2013).  
<sup>c</sup>USDA Natural Resource Conservation Service classifications. Soil series in Oregon based on mapped polygons; series in WA based on descriptions from soil pits (USDA, NRCS 2013).  
<sup>d</sup>Site index, tree height at age 50 calculated using King 1966.

At each site, a fixed-distance grid of 300 sample points was laid out with a random starting point and a random azimuth. The 300 sampling points were divided into 25 blocks with 12 points in each block. The blocks were set up for the purposes of compositing samples for chemical analyses in a separate substudy; they provide a reasonable way to combine observations at an intermediate scale for this study. There were 12 samples per block for the upper two soil layers and eight samples per block for the deepest layer (see explanation for reduced sample size below).

The distance between sample points was fixed within a site but varied between sites from 9 to 15 m as needed to fill the whole uniform area identified at each site. Areas of roads or other non-typical characteristics were not included in the sampling area: aside from those non-uniform areas, the grid was rigorously adhered to except when it was physically impossible to sample at the specified point (e.g., tree boles, large-diameter roots, or large rocks). In those instances, the point was moved in 0.5 m intervals perpendicular to the obstacle or in a spiral fashion around smaller obstacles until a suitable sampling point was encountered.

At each point, samples of forest floor were removed and then the mineral soil was sampled in three depth increments: 0–15 cm, 15–30 cm, and 30–100 cm (30–60 cm was used at WA3 and WA4 as rockiness below 60 cm limited our ability to collect deeper samples using an auger on those sites). At each point a custom-made push corer (7.62 cm inside diameter) was driven into the soil using a sledge hammer to collect the samples at the 0–15 cm and 15–30 cm layers. Two-thirds of the points were selected for sampling the deepest layer (30–100 cm or 30–60 cm); this layer was sampled using a commercially available clay auger with an effective inside diameter of 6.22 cm. Due

to the loss of a few samples, there were 794 to 800 samples per site.

Soil samples were processed by first discarding any obvious living plants (e.g., moss or small vascular plants) or material that was clearly part of the forest floor (e.g., twigs or needles) and not part of the mineral soil. We then sieved each sample to collect the material > 4.75 mm (material retained on a #4 sieve). This sieve size allowed soil and very fine organic materials to pass but retained live roots, small dead organic material and coarse mineral fragments. Smaller sieve sizes would not allow the organic material to pass that contained a lot of carbon and thus, should be counted as part of the soil carbon pool. The material > 4.75 mm was separated into living roots, non-living coarse organic material, and coarse mineral fragments. The root material was placed into a dish of water, gently stirred, and then removed and patted

dry on paper toweling; more aggressive cleaning was not done to avoid loss of root material during cleaning. Living roots were determined based on root appearance—primarily plumpness of small roots, texture, and presence of breaks in the bark. Roots were not separated by sizes for this study but a subset of composited root samples were separated into three diameter size classes to provide an indication of the sizes of roots sampled in the augers. The size classes used for this characterization were: < 2 mm (typically called fine roots), 2 mm to 1 cm (small roots) and > 1 cm (medium roots). The percentage of roots in the size categories varied by depth and site with the root weight in the < 2 mm diameter category ranging from approximately 44% of the total in the 0–15 cm layer down to 13% of the total in the 30–100 cm layer. Roots > 1 cm in diameter (and generally < 2 cm) ranged from 17% of the total in the surface layer to 36% in the deepest layer. A few slivers of larger roots were sometimes included in the auger samples, these ranged from 0 (most com-

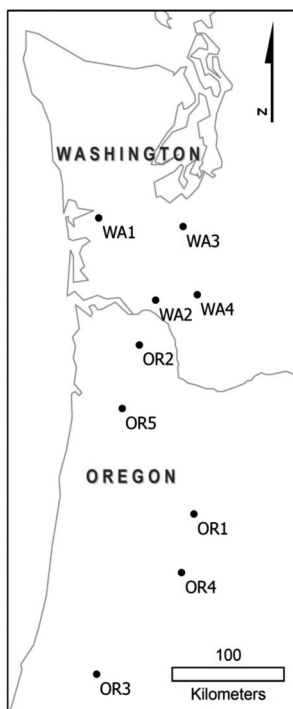


Figure 1. Location of the nine forested stands sampled for soil components.

mon) to a maximum of 3% of the sample total by weight. The non-living organic material such as dead roots, buried branches or boles, and charcoal was separated from the samples by hand; obvious clumps of soil were gently removed but the samples were not washed. Each subsample was placed in an individual bag, dried to constant mass at 65 °C and weighed. Coarse mineral fragments included partially decomposed rock fragments, gravel, small rocks, pumice paragravel, concretions, and peds with strong structure. If we were not able to crush the material between our fingers we considered it to be a coarse fragment; this ambiguity in classification (i.e., soil versus coarse mineral fragment) only occurred in a few samples, all of which had low values for this characteristic. Hereafter, we refer to this component as mineral or mineral fragments.

Soil C was calculated for each depth and site. Soil samples were sieved to remove non-soil components, dry mass determined, and total C determined by LECO combustion (Holub et al. 2011). For the correlation analyses of carbon, roots, and organic matter with tree-level characteristics, e.g., basal area and trees per acre, we adjusted the values for roots and organic matter for the deepest soil layers at WA3 and WA4 in an attempt to partially compensate for the fact that these layers were only sampled to 60 cm instead of 100 cm. The deepest layers at WA3 and WA4 were 30 cm thick, while the deepest layers for the other seven sites were 70 cm thick. If we had assumed that roots and organic material were evenly distributed in the 30–100 cm layer, we could have multiplied the values in the 30–60 layer by 2.33 (i.e., 70/30) to obtain an estimate for the value in the 30–100 layer that accounted for the 40 cm thick layer that was not sampled. However, in the absence of specific information, we assumed that root mass would have declined with depth by at least 1/3 and reduced the multiplier to 1.5.

## Data Analysis

Root, coarse organic material, and mineral data were converted from g per sample to kg per m<sup>3</sup> (based on the volume of each auger sample) to facilitate comparisons across depths and with

values from other studies. Root data were not normally distributed (highly skewed to the right) and describe mass or volume, so were cube-root transformed for most analyses. Root, organic material, and mineral data were summarized at three scales: point, block, and site. The point scale is the scale at which each observation represents one sampled point in space. To summarize point data at the block scale, we took the mean of all point observations within each block as defined above. At the site scale, we simply used the mean of all observations at each of the nine study sites. For spatial analysis, latitude and longitude were converted to planar coordinates (UTM Zone 10N, NAD 83).

*How Do Component Values Vary With Depth and Across Scales?*—Component values of mass were summarized (mean, maximum value, and standard deviation) by site and depth. Boxplots of roots, organic matter, and coarse mineral fragments were used to compare the distribution of each material, lumped together across sites, at each of the three depths, and across each of the three scales (R Development Core Team 2013).

*Are There Correlations Between Materials Collected at Different Soil Depths?*—To explore correlations across materials at the same depth or for the same material across depths, we created scatterplots of a selection of potential relationships (e.g., root mass at 0–15 cm versus root mass at 15–30 cm) within the data from all sites together at each of the three scales. To summarize findings across sites, we calculated correlation coefficients between the same materials at two depths for each site independently.

*Are There Spatial Patterns in the Sieved Components? If So, Do They Differ by Scale?*—Values for each point were plotted over space to explore the data and screened for spatial pattern. We also looked for spatial pattern by plotting data values against north-south and east-west coordinates to look for evidence of any systematic trends in the data. We looked for spatial trends at both the point and the block scale.



We used the robust semivariogram option in R to investigate spatial autocorrelation (Ribeiro and Diggle 2001). The variogram calculates the empirical variability in the data at multiple distances and describes the degree to which observations located more closely in space are more similar to each other than observations located further apart (Diggle and Ribeiro 2007). Variograms were calculated and plotted for each component, depth and site for both the point and the block scale data.

*Are There Correlations Between Soil Carbon and Root Mass or Other Site-Scale Characteristics (e.g., Stand or Climatic Factors)?*—We calculated correlation coefficients between roots, organic material, and mineral materials at the site-scale and between these components and site characteristics including stand age, basal area, elevation, mean annual temperature and precipitation, and soil carbon. Soil carbon values and site characteristics were taken from Holub et al. 2011.

*Effect of Sample Size on Precision*—Sampling is often done to estimate a mean value of a variable, e.g., root volume, across all potential observations within a study site. The precision of that estimate of the mean is a function of sample size. We used a Monte Carlo approach ( $n = 1000$  iterations) to estimate precision as a function of sample size from our observed data. For each of seven potential sample sizes (10, 30, 50, 100, 150, 300, 500), we selected observations with replacement and calculated both the standard error of the sample mean and the resultant width of the 95% confidence interval. To assess site-to-site differences in potential precision, analysis was completed independently for all nine study sites. To provide the best possible estimate of precision that could be expected at a new site that is similar to our study sites, we also analyzed the data pooled across all sites. Because root mass and variability of root mass differed for roots at different depths, the analysis was completed independently for the top layer and for roots at the deepest layer. Precision was estimated as the width of the 95% confidence interval ( $2 \times 1.96 \times \text{SE of estimated mean}$ ).

## Results

### Mass by Component, Layer, and Site

Mass of all three components varied substantially by depth (Table 2, Figure 2) and was not normally distributed at any of the three scales. All values for mass (roots, organic materials or mineral fragments) at all depths on all sites had distributions which were skewed to the right (low number of large values) and for some components on some sites, there were substantial numbers of zero or very low values. Over all sites and depths, the percentage of samples with zero values was 2.6% for roots, 14.1% for organic matter, and 9.5% for mineral fragments. Although many samples had low root mass, the percentage with no root mass (i.e., values of 0) was only 0.3% in the surface layer and increased with depth to 6.5% at the deepest layer. The percentage of samples with zero mass followed a similar pattern of increase with depth for organic matter; however, there was no pattern with depth for mineral fragments. Median values were always less than the mean values; in some cases the high values had such an influence that the median value was less than 10% of the mean (e.g., organic matter at the deepest layer for OR3 and OR5) at the point level.

Oven dry mass of live roots at individual sample points ranged from 0 to 86 kg m<sup>-3</sup> with a mean of 2.2 kg m<sup>-3</sup> (median of 1.2 kg m<sup>-3</sup>). Mean root mass ranged from 0.4 kg m<sup>-3</sup> to 4.3 kg m<sup>-3</sup> (median 0.1 kg m<sup>-3</sup> to 3.4 kg m<sup>-3</sup> (Table 2). For all sites, mean root mass was highest in the top layer and least in the bottom layer. The ratio of root mass between the top and middle layers ranged from 1.0 to 1.8; thus, although root mass, on average, was lower in the 15–30 cm than the 0–15 cm layer, the differences were not great. The ratio of roots from the middle layer to the bottom layer ranged from 1.7 to 3.9, reflecting the more substantial drop off in mass to the deepest layer. The ratio of root mass between layers did not appear to be affected by rooting depth as both the soil with the lowest ratio between layers and the one with the highest ratio were from sites with rooting depth classified as very deep ( $> 152$  cm to a layer that retards root development; USDA, NRCS 2013).

TABLE 2. Summary statistics (mean, median, maximum and standard deviation) for living roots, organic matter, and mineral fragments by site and soil depth. WCst = Washington Coast, WCas = Washington Cascades, OCst = Oregon Coast, OCas = Oregon Cascades. Minimum values were almost always close to zero so were not shown to reduce the number of columns.

| Region/Site | Depth (cm) | n   | Living Roots (kg m <sup>-3</sup> ) |        |     | Organic Matter (kg m <sup>-3</sup> ) |      |        | Mineral Fragments (kg m <sup>-3</sup> ) |      |      |
|-------------|------------|-----|------------------------------------|--------|-----|--------------------------------------|------|--------|-----------------------------------------|------|------|
|             |            |     | Mean                               | Median | Max | SD                                   | Mean | Median | Max                                     | SD   | SD   |
| WCst/WA1    | 0-15       | 297 | 1.2                                | 0.9    | 7   | 1.1                                  | 4.8  | 2.7    | 59                                      | 7.0  | 283  |
|             | 15-30      | 300 | 1.0                                | 0.6    | 11  | 1.1                                  | 2.6  | 0.8    | 84                                      | 7.6  | 406  |
|             | 30-100     | 197 | 0.6                                | 0.1    | 15  | 1.6                                  | 0.7  | 0.4    | 11                                      | 1.3  | 353  |
| WCst/WA2    | 0-15       | 300 | 1.3                                | 0.8    | 18  | 2.0                                  | 3.7  | 1.7    | 63                                      | 7.1  | 366  |
|             | 15-30      | 299 | 1.1                                | 0.5    | 17  | 2.0                                  | 3.0  | 1.0    | 56                                      | 6.4  | 481  |
|             | 30-100     | 199 | 0.4                                | 0.1    | 6   | 0.8                                  | 1.6  | 0.3    | 44                                      | 4.6  | 576  |
| WCas/WA3    | 0-15       | 299 | 3.0                                | 2.2    | 20  | 2.9                                  | 5.0  | 2.2    | 65                                      | 8.5  | 1049 |
|             | 15-30      | 300 | 2.7                                | 1.2    | 41  | 2.9                                  | 2.6  | 0.7    | 61                                      | 6.3  | 1110 |
|             | 30-60      | 200 | 2.0                                | 0.3    | 21  | 3.3                                  | 1.2  | 0.1    | 42                                      | 4.0  | 455  |
| WCas/WA4    | 0-15       | 300 | 3.4                                | 2.5    | 25  | 3.4                                  | 6.7  | 2.0    | 93                                      | 13.3 | 264  |
|             | 15-30      | 300 | 2.2                                | 1.2    | 30  | 3.4                                  | 3.6  | 1.0    | 69                                      | 9.2  | 584  |
|             | 30-60      | 200 | 1.2                                | 0.3    | 15  | 2.0                                  | 1.3  | 0.3    | 22                                      | 3.0  | 188  |
| OCst/OR2    | 0-15       | 300 | 2.9                                | 2.1    | 27  | 3.2                                  | 2.0  | 0.5    | 39                                      | 5.4  | 657  |
|             | 15-30      | 300 | 2.9                                | 1.2    | 86  | 3.2                                  | 2.7  | 0.1    | 116                                     | 10.6 | 376  |
|             | 30-100     | 200 | 1.7                                | 0.8    | 15  | 2.3                                  | 0.5  | 0      | 18                                      | 1.8  | 19   |
| OCst/OR3    | 0-15       | 299 | 3.1                                | 2.5    | 20  | 2.7                                  | 4.9  | 1.6    | 88                                      | 9.9  | 984  |
|             | 15-30      | 299 | 3.0                                | 1.4    | 53  | 2.7                                  | 5.0  | 0.6    | 232                                     | 20.9 | 641  |
|             | 30-100     | 200 | 1.9                                | 0.6    | 27  | 3.4                                  | 1.3  | 0.1    | 47                                      | 5.5  | 232  |
| OCst/OR5    | 0-15       | 300 | 4.3                                | 3.4    | 26  | 3.6                                  | 2.5  | 1.0    | 64                                      | 6.5  | 618  |
|             | 15-30      | 300 | 3.1                                | 2.0    | 30  | 3.6                                  | 1.9  | 0.3    | 96                                      | 8.7  | 634  |
|             | 30-100     | 200 | 2.3                                | 1.4    | 20  | 2.8                                  | 1.6  | 0.2    | 21                                      | 3.4  | 601  |
| OCas/OR1    | 0-15       | 300 | 2.6                                | 1.7    | 47  | 3.9                                  | 3.6  | 0.7    | 194                                     | 9.1  | 1269 |
|             | 15-30      | 300 | 1.8                                | 0.9    | 42  | 3.9                                  | 3.4  | 0.4    | 14                                      | 15.3 | 434  |
|             | 30-100     | 200 | 1.6                                | 0.6    | 24  | 3.1                                  | 0.8  | 0.2    | 39                                      | 1.7  | 657  |
| OCas/OR4    | 0-15       | 299 | 4.0                                | 2.5    | 45  | 5.0                                  | 2.8  | 1.0    | 47                                      | 5.3  | 805  |
|             | 15-30      | 299 | 2.2                                | 1.3    | 41  | 5.0                                  | 1.9  | 0.6    | 60                                      | 4.8  | 469  |
|             | 30-100     | 200 | 1.6                                | 0.6    | 32  | 3.3                                  | 1.0  | 0.2    | 28                                      | 2.9  | 247  |



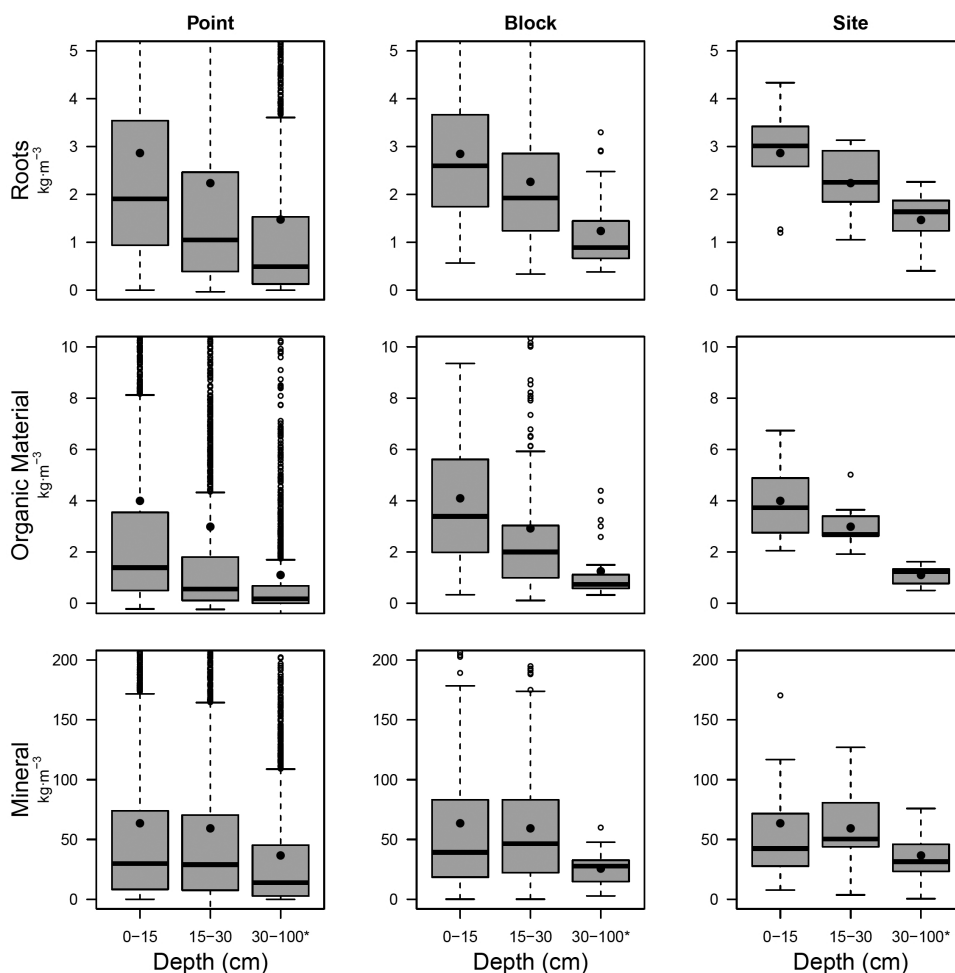


Figure 2. Mass (g) of roots, organic matter, and coarse mineral fragments retained on a 4.75 mm sieve per site and sampling layer summarized at three scales across all sites combined. Values for the bottom layer were calculated from the 30–60 or 30–100 cm sample depending on site (see text for details). For each component, the middle line shows the median, the box the interquartile range, and whiskers extend to the most extreme data point (or no more than 1.5 times the interquartile range). Mean values are displayed with a solid black dot. Y-axes were kept the same across scales; this resulted in excluding some extreme observations from the plots. Maximum values are shown in Table 2.

Variability in root mass from point to point tended to increase with depth. Root mass of the deepest soil layer was generally greater in the Oregon sites than the Washington ones.

Oven dry mass of coarse organic material at individual sample points ranged from 0 to 232 kg m<sup>-3</sup> (Table 2, Figure 2) with a mean of 2.7 kg m<sup>-3</sup> (median of 0.8 kg m<sup>-3</sup>). Generally, mass of coarse organics was highest in the surface layer and decreased with depth. There was more organic matter in the top layer than in the middle layer

for all sites except OR2. The deepest layer always had less organic material than the top or middle layers. Standardized mass of organic materials for the deepest layer did not exhibit the same trend of higher values in Oregon than in Washington that was observed for roots.

Mass of mineral fragments at individual sample points ranged from 0 to 1269 kg m<sup>-3</sup> (Table 2) with a mean of 53.3 kg m<sup>-3</sup> (median of 31.8 kg m<sup>-3</sup>). OR2 had very low amounts of mineral fragments at most sample points but the maximum value for

coarse fragments in the shallowest layer at that site was higher than for several other sites, thus, providing another indication of the variability found within the sites. The amount of mineral fragments decreased with depth for most of the sites but the pattern was not strong (Figure 2). Mass of mineral fragments at the block or site levels were underestimated for sites such as WA3 and WA4 which had large rocks that necessitated moving the sample points.

### Correlations Between Contents at Different Depths

Correlations between values at different depths in each core were evaluated to determine if values at one layer could predict values at another layer. Correlations between roots in the top and middle depths were quite low (Figure 3) and correlations between non-living organic materials across these same two depths were only slightly higher. For both roots and coarse organic materials, correlations between components at different depths were generally less than 0.4 with median values for roots < 0.2. Correlations between mineral fragments across layers were somewhat higher than those for roots or organic material but the values were also quite variable with some sites exhibiting very low or very high values. Correlations across components—roots versus organic matter or mineral fragments—were also very low (data not shown).

Relationships between mass of roots in different layers are difficult to see at the point scale (Figure 4). Visual examination of plots of root mass at the block level showed weak but positive relationships (Figure 4) as did plots at the site scale. Mass of roots at 0–15 cm were positively correlated with roots at each of the 2 deeper layers when evaluated at the site scale. There were no obvious patterns in the relationship between mass of roots and mass of organic matter or between mass of roots and mineral fragments at any of

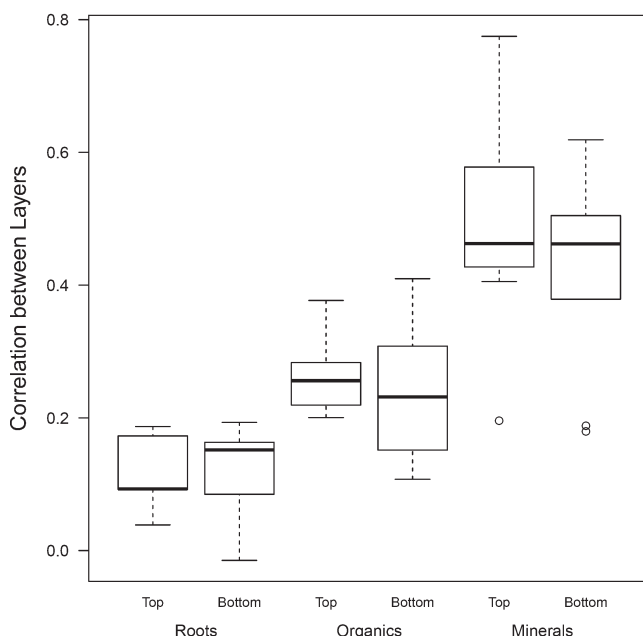


Figure 3. Correlations between layers of each material across sites. “Top” correlations describe the relationship between the values for a particular material (roots, organic materials, minerals) in the top layer (0–15 cm) and the values for that same material in the middle (15–30 cm) layer. “Bottom” correlations describe the relationship between the values for a particular material (roots, organic materials, minerals) in the middle (15–30 cm) layer and the values for that same material in the bottom layer (30–60 or 30–100 cm, see text for details). Correlations were calculated using cube-root transformed data; missing values were excluded.

the three scales; thus, scaling up did not always improve the relationship.

### Correlations Between Site Variables and Below-ground Components

By far the strongest correlations between the site variables and the below-ground components involved soil carbon. Soil carbon was negatively correlated with total root mass ( $r = -0.85$ , Figure 5); the negative relationship was present at individual layers and ranged from  $r = -0.60$  for the deepest layer to  $r = -0.93$  for the middle layer. Stand age was positively correlated with total mass of sampled roots ( $r = 0.50$ ) or root mass in the top or bottom layers ( $r = 0.56$ ) but more weakly correlated with root mass in the middle

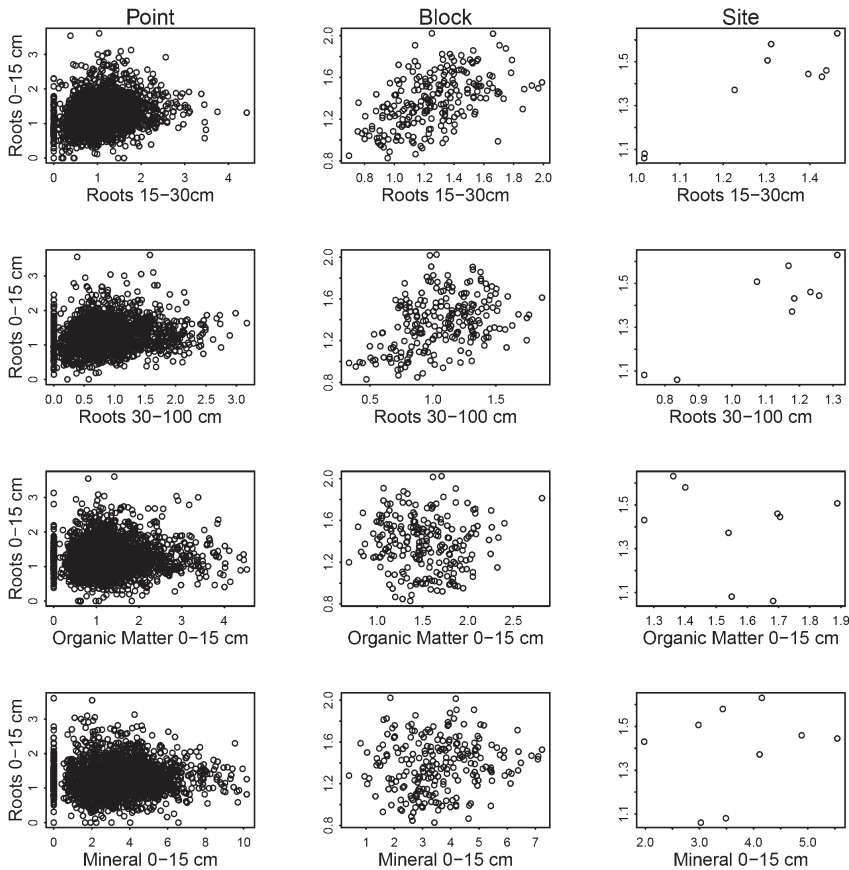


Figure 4. Effect of scale on relationships between roots in the top layer of soil and other factors. Mass of root material in the upper layer (0–15 cm) of soil as a function of root mass from 15–30 cm depth (first row); root mass in the deepest layer (second row); organic matter in the upper layer of soil (third row); and coarse mineral fragments in the upper layer of soil (bottom row). Data describing mass of roots, organic material, and minerals are in kg/m<sup>3</sup> and were cube-root transformed. Each column describes an analysis at a different spatial scale. The left column displays point data; the middle column displays data at the block scale; and the right column displays average values for each of the nine sites.

layer ( $r = 0.29$ ). Total root mass was positively but weakly correlated with basal area of all trees ( $r = 0.14$ ) or basal area of Douglas-fir trees ( $r = 0.32$ ). Total mass of roots was only weakly correlated with organic matter ( $r = -0.21$ ) or mean annual temperature ( $r = 0.07$ ).

### Spatial Variation

Plots of component values over space revealed surprisingly few spatial patterns. Visual examination of the values for each soil depth for individual points did not reveal any obvious spatial pattern in roots or organic material (cf., Figure 6 for three

sites; sites not shown revealed no patterns); however, for mineral fragments there were indications of clustering of higher values in some locations, for example OR3 in Figure 6). Bubble plots at the block level indicated no clustering as what few patterns appeared to be present at the point level were lost during block averaging.

Plotting component values against geographic coordinates also did not reveal spatial trends with respect to east-west or north-south gradients (Figure 7). There were a few exceptions in the larger data set. For example, at OR5, the root and organic matter values from the deepest layer

tended to increase then level off from east to west and to decrease from north to south. In general, coarse mineral fragment mass at OR3 tended to increase from east to west and to increase and then level off from north to south.

Variograms for the root and organic matter point data were generally flat (Figure 7 and data not shown). A few exceptions were: root mass at the deepest layer for OR5 and possibly OR3, and organic matter at the middle depth for OR4 (data not shown). On the other hand, the variograms for the point data for mineral fragments indicated strong autocorrelations for some, but not all, of the sites. The site with the strongest autocorrelation was OR3 where the variograms were similar for all three depths. Some sites exhibited autocorrelation for mineral fragments at one or more depths (cf., WA1 or OR5, strong at one or more of the surface layers but much weaker at the deepest layer).

## Precision

We used our sample information to calculate the precision, i.e., width of the 95% confidence interval, with which we can expect to estimate mean root mass as a function of sample size, for each site and for all sites combined at the top and bottom sampling layers (Figure 8). As expected, the precision at which we can estimate the mean increases substantially with sample size. In other words, the width of the 95% confidence interval decreases with sample size.

We estimate that for data from a new site to achieve a width of the 95% CI of no greater than  $0.8 \text{ kg m}^{-3}$  for the upper soil layer would require a sample size of approximately 300 samples. However, in some sites, for example WA1, far fewer samples would be needed. A desired precision of  $\pm 1.5 \text{ kg m}^{-3}$  could be achieved with less than 100 samples in the upper layer at most sites (Figure 8).

## Discussion

Detailed data on below-ground forest ecology can improve our vision of how roots are distributed in space, both within sites, across sites, and with depth. We observed that root mass generally decreased with depth and that root mass at any one

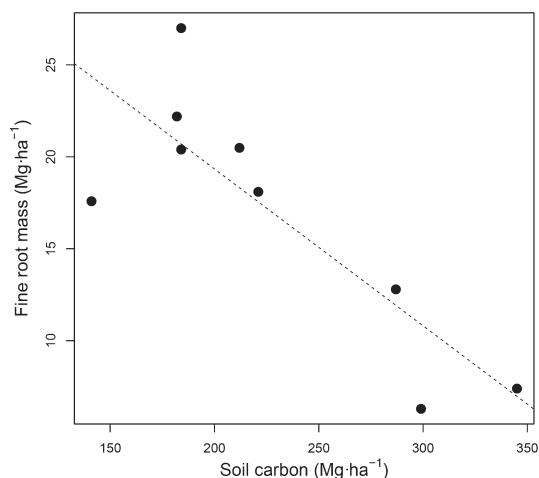


Figure 5. Relationship between root mass (cube-root transformed) and soil carbon for the nine sites in the study ( $r = -0.85$ ).

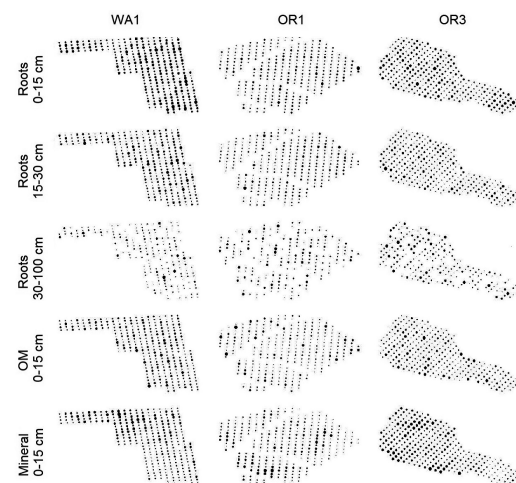


Figure 6. Spatial configuration of data for three example sites. Data were cube-root transformed before plotting. The spatial arrangement of points corresponds to the location of points in the field (each site had a different outline) with the blank linear areas in OR1 corresponding to the location of current or former roads. The size of each point represents the value of that point relative to the other values at that site. Relative values are presented to enable comparisons of spatial pattern within a site. Note that because the overall magnitude of values differed by material and site, comparisons of point size across materials or sites are not possible.

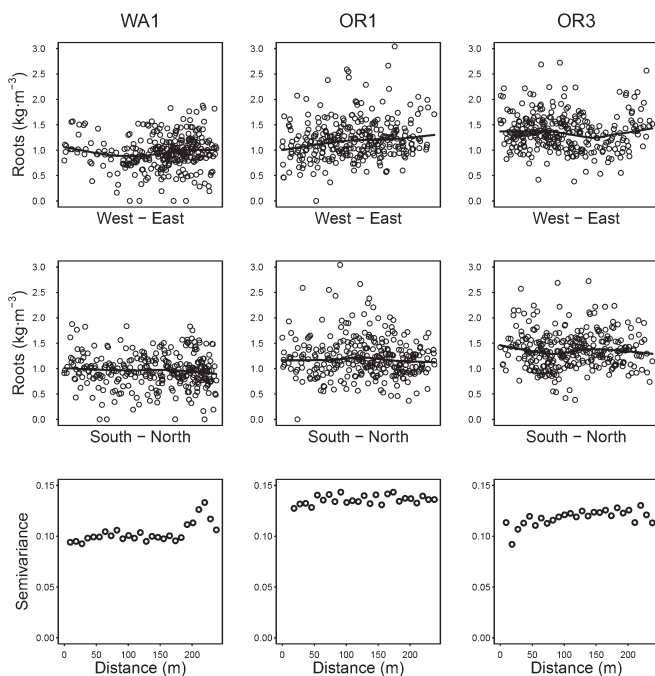


Figure 7. Spatial trends in root data (kg m<sup>-3</sup>) from the 0–15 cm layer across three example sites. Spatial configuration of relative values for the same sites shown in Figure 6. The first row of data describes the trend in values moving from west to east. The second row of data displays the trend in values moving from south to north. Robust semi-variograms are presented in the final row of plots.

location is relatively poorly correlated with root mass at nearby locations either with respect to depth or horizontal distance. The only site-scale driver of root density was soil carbon; sites with less soil carbon had higher root mass. Forest soil characteristics have been described for many sites but most reports are based on relatively few samples or only sample at one depth. In fact, some reports estimate root mass based on above-ground characteristics (e.g., Smithwick et al. 2002) due to the expense and difficulty in obtaining field values. Root mass in this study was similar to values for Douglas-fir fine roots reported by some researchers (Vogt et al. 1987, Ares et al. 2007, Dumm et al. 2008) but substantially lower than values reported by others (Santantonio and Hermann 1985, Curt et al. 2001). Values in the literature differ in the methods used in sampling, including variation in the depth sampled, and in

how the results were reported with some authors reporting on an area basis and others using volume.

### Below-ground Biomass as a Function of Soil Depth

The decline of root mass and coarse organic materials with depth is not universal but has been commonly observed. It has been attributed to greater nutrient availability in surface layers, moisture availability during summer rainfall events, or to poorer aeration and stronger soil structure with depth (Coile 1937, Eis 1974, Sutton 1991). The actual factors associated with rooting patterns in forest soils are complex and not completely known, however, soil texture, the presence of organic pockets or a hardpan (Eis 1974) and the effect of nutrition on primary and secondary root growth (Coutts and Phillipson 1976) have been studied and the biological pattern of root initials and growth described for several species (Coutts 1989). Profiles of Douglas-fir root biomass were documented in 10 cm layers for seven

different soil types in France (Curt et al. 2001). Although all their profile types exhibited declines in root biomass below 30 or 40 cm, biomass in the surface three 10 cm layers exhibited different patterns by soil type ranging from fairly uniform, rapid decline, or slight increase with depth, thus, demonstrating how root mass can be quite variable in surface horizons.

The patterns with depth (Figure 2) for roots and organic materials are consistent with our general expectation that roots are most common close to the surface and decline with depth; however, the data do not appear to support any generalizations on root or organic matter mass with site characteristics. It would be interesting to develop and test specific hypotheses as to why root mass declined more rapidly with depth at some sites than others with similar soil drainage and texture classifications. Curt et al. (2001) suggest hypotheses related to

nutrition (e.g., cations in A or illuvial horizons) and physical characteristics that could be explored.

### Lack of Spatial Pattern in Root Systems

We usually assume that near things are more related than those farther away and this relationship has been invoked as Tobler's First Law of geography (Tobler 1970). However, empirical "laws" in science may merely represent compact descriptions of patterns and are not always true (Miller 2004). First-order lateral roots grow away from the plant, and this phenomenon has been called extropy (Noll 1894 as cited in Coutts 1989). Extropy results in roots exploring the available soil volume and also (for major roots) provides physical stability. Plants are programmed with potential architecture (the vertical and horizontal spacing and orientation of root initials) but this potential architecture can be radically modified by local soil conditions (Coutts 1989). Even in agricultural fields with uniform spacing of one type of plant, it is common to observe non-uniform root patterns (Hamblin 1985).

The almost complete lack of spatial correlation of root mass on our sites suggests that the relatively uniform above-ground stand conditions were not indicative of strong patterning below ground. In addition, given the size of the core (7.62 cm diameter auger) in relation of the size of the grid (points spaced 9 to 15 m apart), we sampled a very small percentage of the soil volume in relation to the distance between points. The range of spatial dependence can be much shorter than the distances used in most grid sampling (Kweon 2012). Root systems are sparsely and irregularly distributed in soils and it may take very large numbers of samples or much larger samples to detect spatial patterns. Our analyses at the block scale did result in large increases in spatial pattern, indicating that larger samples may be appropriate to detect patterns. Although 300 points per site represents a large sampling effort, at the closest spacing between grid points, if we had sampled 15 to 20 times as many points as did, we would still have only sampled 0.1% of the surface area. For example, Kravchenko et al. 1999 used 1752 points to evaluate soil spatial variability in an agricultural field in Illinois, an

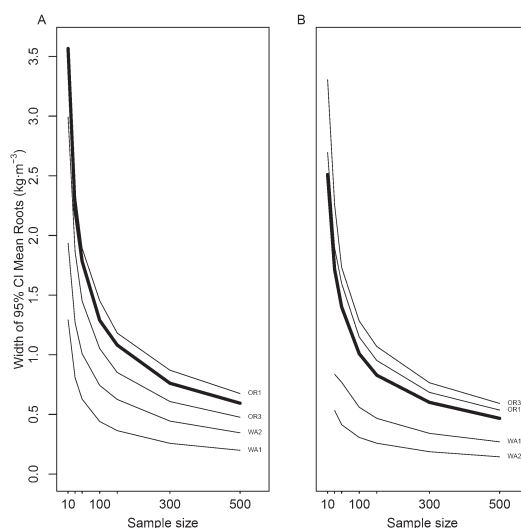


Figure 8. Full width of the 95% confidence interval on estimated mean root mass per volume as a function of sample size. Because root masses and variability of root mass differed for roots by level, the analysis was completed independently for: A) the top layer (0–15 cm) and B) the deepest layer (30–100 cm for all sites except WA3 and WA4, see text for details). In each case, precision was estimated for each site independently. The dark line represents precision analysis for data mixed together from all sites which would be our best estimate of the situation at a new site. Lines for only four of the nine sites are shown to facilitate labeling; the three sites used in other figures are shown plus WA2 to display the upper- and lower-most line for the two depths. Tick marks on the x-axis describe sample sizes for which precision was calculated to fit the curve. The width of the CI was calculated as  $2 \times 1.96 \times \text{SE of estimated mean}$ .

environment likely to be much more uniform than a forested site in the Pacific Northwest. Patterns may also have been somewhat obscured by our need to shift samples away from a tree bole, log, or large rock and spatial correlation may vary over time. For example, very young stands could have higher spatial correlations as root systems have not yet extended far beyond their starting location. Future research could investigate the relationship between root mass and distance from or the size of the closest tree.

Coarse organic matter in the soil is partially a legacy of formerly living roots, particularly for



samples not collected close to the soil surface. The mass of roots and mass of coarse organic matter were generally not related. This could be a function of differences in past stand histories. For example, stands having a relatively recent history of large-diameter conifer trees might have a higher probability of large-diameter roots still present in the soil. On the other hand, a stand with a recent history of smaller-diameter shrubs or hardwood trees might have a greater percentage of its former root mass decomposed and now sample-able as part of the soil carbon layer as opposed to remaining as dead and decaying but still intact organic materials.

While spatial correlations were very low for both root and organic matter mass for most sites and layers, auto-correlations were higher for mineral fragments at least on some sites. This makes sense from a soil development standpoint as points close together will often be similar in micro-topography and have often experienced the same soil formation factors. Root system formation, however, is a very plastic process and can be influenced by plant location as well as moisture, nutrition, and soil physical properties (Coutts 1987, 1989).

#### Effect of Scale on Our Results

At the point level, there was no effective correlation between values obtained at different levels; that is, values for roots, organic matter, and coarse fragments in the surface layer were not correlated with values at deeper layers. This was still true at the block scale for organic matter, but root mass was better correlated at block level than point level. We observed a much better correlation at the block level than the point level for coarse mineral fragments. The correlations at the site level are improved, in part, because of the much wider range in values when all sites are evaluated together rather than individually. For example, the linear or curvilinear relationship between root mass at 0–15 cm and mass at 15–30 cm would have been much poorer without the inclusion of WA1 and WA2 with their lower root system mass (upper right hand panel in Figure 4).

#### Variation in Below Ground Biomass Across Douglas-fir Forests in the Pacific Northwest

The four-fold range in root mass we observed may seem large high given the narrow range in stand age and basal area we sampled but is consistent with previous reports. Curt et al. (2001) reported a similar range in fine root biomass across their seven soil types, however, if the two soil types with rooting less than 60 cm are excluded, the range was substantially less. Vogt et al. (1987) also reported a four to five fold range in fine root mass for forested sites but their sites included a wider range in age, productivity and tree species composition than in our study. Our exploratory analyses with site-level characteristics did not suggest any particular relationships between root mass and basal area or other above-ground characteristics. However, the range in site and stand characteristics combined with the small number of sites resulted in inter-correlations between factors (e.g., cooler sites tended to be wetter, Holub 2011).

There is no standard method of root sampling in forest stands (e.g., Atkinson 2000) nor one which will give unbiased results under all conditions (e.g., Harrison et al 2003, Moir et al 1969). For example, site characterization might be efficiently done with a grid-based system, while following the development of individual roots or manipulative studies might be warranted to explore physiologically-based hypotheses. In addition, sampling schemes may differ if there is a temporal component to the study or one which focuses on different depths. Our sampling procedure resulted in mostly fine and small-diameter roots; 65% to 82% of our roots were  $\leq 1$  cm in diameter but the percentage by size class varied by site and depth with the greatest percentage of roots  $< 2$  mm in the 0–15 cm layer and the lowest percentage in the deepest layer. Curt et al. (2001), who sampled a large number of sites, demonstrated that fine root mass was correlated with some edaphic variables, but these correlations were not significant when they looked at mass of medium or coarse roots.

Soil carbon was strongly but negatively correlated with total mass of our sampled roots ( $r = -0.85$ ). We sampled both fine and small-diameter roots (generally up to 1 or occasionally 2 cm in

diameter); existing literature focuses primarily on fine roots (roots < 2 mm in diameter as those roots are the fraction which turns over most quickly). Tree roots, especially fine roots, are an important part of carbon cycling (Santantonio and Hermann 1985) but relatively little detail is known about the factors important in the below-ground aspects of the C cycle (Johnson et al. 2004). The mineral soil carbon (to 1 m depth) on these sites was in the range of 150–300 Mg C ha<sup>-1</sup> (Holub 2011); this was much larger than what was in the root or organic matter components we sampled (combined root and organic matter totals for all three layers ranged from 11 to 19 Mg C ha<sup>-1</sup>). While it could be very useful to be able to predict various components of the C cycle from each other, it is unlikely that such relationships would be strong if a wide range of soil types, and stand conditions were evaluated.

The longevity of fine roots is an important factor in C cycling calculations and many factors may influence longevity. For example, in a 140-year-old Norway spruce stand in Germany, mean fine root age based on radiocarbon values ranged from five years in the organic layer to 13 years in the 40–60 cm mineral soil (Gaul et al. 2009). Mean C/N ratio increased with depth in their study and the authors speculated that rates of root herbivory may have been reduced when C/N ratios were low and this may have contributed to greater longevity. Decomposition rates of soil organic carbon were higher in the rhizosphere of ponderosa pine (*Pinus ponderosa*) and Fremont cottonwood (*Populus fremontii*) in pot studies using three soil types and this rhizosphere priming effect was long lasting (Dijkstra and Cheng 2007); the authors suggested that increased C input into soil does not necessarily lead to increased carbon storage, but may actually enhance soil organic decomposition and result in a significant net soil carbon loss. Tree roots in the field would be much more sparsely distributed than those in pots, and thus, would cover a wider range in root size and woodiness. It is not known how far this rhizosphere effect in a pot extends beyond the roots or if this effect would be present under field conditions.

Since roots are a major source of the carbon which is incorporated into soil, it is important to understand root development, root longevity, the pathways by which roots decompose and are incorporated into soil C, and the factors which influence the decomposition of soil carbon. In addition, there may be feedback mechanisms between root development or longevity and soil carbon; that is, less carbon may be allocated to root development in trees growing in soils with high nutrient availability or water holding capacity. Small differences in soil carbon could be very important in modeling carbon pools, but are difficult to estimate due to their spatial and temporal variability (Vance 2003). Soil texture and mineralogy can affect nutrient adsorption and these factors can also influence organic carbon content in soil (Strahm and Harrison 2007). In addition, species may differ in their sensitivity to climate or nutrient variables (Vogt et al 1996). Understanding why there is a strong negative relationship between root mass and soil C as was observed in this study, and if this relationship can be altered, should help foresters make better management decisions in relation to carbon sequestration.

Above- and below-ground plant biomass are strongly coupled over a wide range of plant sizes and species (Enquist and Niklas 2002), however, living root biomass in cores and basal area of tree stems in our study were poorly correlated ( $r = 0.14$ ). Otto and Fischer (2009) also reported poor relationships between root mass and number of trees or distance to trees. These poor correlations do not imply that mass in above- and below-ground plant parts are not related but rather that the variables we used were not good surrogates for either total above- or below-ground biomass. In addition, we had a relatively narrow range in values of plant size compared to Enquist and Niklas (2002) who included plant sizes that differ by orders of magnitude.

### Sampling Designs for Quantifying Roots

For sparsely distributed plant parts such as roots, substantial effort is required to collect samples and if sampling is done without knowledge of what is below the sampling point, many samples

will be zeros or very low values. A large number of sampling points will be required to determine the mean with any precision.

The overarching study from which these samples were taken was designed to assess soil carbon over time and not specifically to sample roots or coarse organic matter. A study designed specifically to sample all root system components would not have moved a sample point if it fell on a large root as was done in this study. However, aside from this caveat, the basic design was reasonable for estimating mean mass of small-diameter living roots and examining the relationships across points, blocks, and sites as well as between layers. Future investigations of spatial pattern might best be approached with a random selection of sampling locations, possibly augmented with some additional points to fill in obvious areas with no samples, rather than using a grid. For example, Ping et al. (2010) reported more reliable estimates of root biomass in grasslands using stratified random sampling rather than sampling fixed positions.

Our analysis of precision versus sample size indicated that by 150 samples we have gained

most precision for estimating the sample mean; however, there were clear differences in sample precision obtained across the sites, even with a fairly narrow range in species composition and tree age. The required sampling intensity for a given precision will also vary with soil depth. In addition, our study sites were selected for within-site uniformity in mapped soil series, landform, slope, and aspect; thus, sites with less uniformity would presumably require higher sample sizes to achieve the same precision. Future sampling on these or other sites would need to be focused on specific questions to be efficient. Thus, sampling on more diverse sites might help in answering questions related to the effects of climate or stand conditions while more intensive sampling or manipulative studies would be appropriate to generate or test hypotheses on root distribution.

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