Deep Soil: Quantification, Modeling, and Significance of Subsurface Carbon and Nitrogen

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Abstract

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Soil is the primary sink for C in forest ecosystems, but is often overlooked in ecosystem C budgets. Efforts to quantify C pools often sample soils to a depth of 0.2 m despite observations that deep soil C is neither scarce nor entirely stable. This study examined the systematic sampling depth for ecosystem C analyses in the Pacific Northwest, and compared best-fit models of C in deep soil layers with laboratory measurements. Forest floor samples and mineral soil bulk density samples were collected at regular intervals from the soil surface to depths of 2.5 m from 22 sites across the coastal Pacific Northwest Douglas-fir zone. Soil samples were screened to 4.7 mm and analyzed for C content. We found that systematic soil sampling shallower than 1.5 m significantly underestimated total soil C. On average, sampling to 2.5 m compared to 0.5 m increased total C by 156% (85.3 Mg ha^{-1} to 132.7 Mg ha^{-1}), and 21% of total C within the depth range sampled was below 1.0 m. A nonlinear mixed model using an inverse polynomial curve form and predicting total C to 2.5 m given only data to 1.0 m was reliable for 20 of 22 sites; the sites that could not be accurately modeled carried the greatest C at depth and contained noncrystalline minerals. Shallow soil sampling at best provides a biased estimate and at worst leads to misleading conclusions regarding soil C. Researchers seeking to quantify soil C or measure change over time should sample deep soil to create a more complete picture of soil pools and fluxes.

Nitrogen (N) is one of the primary limiting nutrients in Pacific Northwest forests, as well as many other ecosystems in the United States. Efforts to quantify total soil N and to monitor N cycling have often sampled soils to a depth of 0.2 m, occasionally to 1.0 m depth, or the bottom of the B horizon. However, tree roots often extend many meters into the soil and can produce hydraulic redistribution during seasonal drought that brings soil water (and dissolved nutrients) from deep layers up to surface horizons. This study examined the systematic sampling depth for ecosystem N analyses in the Pacific Northwest, and compared best-fit models of N in deep soil layers with observed quantities. At 22 sites across the Pacific Northwest Douglas-fir zone, forest floor and mineral soil bulk density samples were collected at depths of 0.1 m, 0.5 m, 1.0 m, 1.5 m, 2.0 m, and 2.5 m. Mineral soil was screened to 4.75 mm and analyzed for total N content. Systematic sampling shallower than 2.0 m produced significantly smaller estimates of total N. On average, only 3% of total soil N was in the litter layer, and 31% was below 1.0 m depth (almost 2700 kg N ha^{-1}). Over 45% of soil N was below 1.0 m at three sites. A nonlinear mixed effect model using the Langmuir equation predicted total N to 2.5 m with -12.4% mean error given data to 1.0 m, and -7.6% mean error with data to 1.5 m. Shallow sampling of soil N in studies of biogeochemical cycling, forest management impacts, or ecosystem monitoring at best provides a biased estimate and at worst produces misleading conclusions. Research and monitoring efforts seeking to quantify soil N or measure fluxes should sample deep soil to create a more complete picture of soil pools and changes over time.

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CHAPTER I – DEEP SOIL CARBON [§]

1. INTRODUCTION

Soil represents the most important long-term carbon (C) reservoir in terrestrial ecosystems because it contains more C than plant biomass and the atmosphere combined (Jobbagy and Jackson, 2000). Despite this fact, soil remains the most under-reported element in ecosystem analyses in the primary literature (Harrison et al., 2011). In particular, detailed mechanistic explanations for soil C flux have not yet been proposed in subsurface soil, defined here as below 0.5 m (Fang et al., 2005; Fontaine et al., 2007).

Emissions of CO_2 and CH_4 in the United States from fossil fuel combustion and other sources totaled 5,691.8 Tg CO_2-C in 2011, and greenhouse gas emissions have increased, on average, by 0.4 percent per year since 1990 (US Environmental Protection Agency, 2013). Consequently, areas where C compounds may be removed from the climate system are particularly critical to the interests of protecting human health and preserving the ecosystem services upon which we rely (Lorenz et al., 2011). Among terrestrial ecosystems, forests have been identified as important potential sinks for C in the face of climate change (Birdsey et al., 2006). Within the contiguous United States, Pacific Northwest forests represent the single most important ecosystem for C sequestration – around 39% of total storage in all US forests, despite representing a much smaller proportion of total forested area (Birdsey, 1992). Homann (2005) found that current C storage in the Pacific Northwest is less than half of the potential total ecosystem storage, indicating that substantial C sequestration is theoretically possible in the

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future, depending on harvest-age trends, the occurrence of wildfires, and the rate of forest conversion.

Our ability to investigate total ecosystem C or change in C over time and across management treatments relies upon accurate measurement of the total soil C stock because it is the single largest C sink in forest ecosystems (Dixon et al., 1994; Harrison et al., 2003; Lal, 2005; Zabowski et al., 2011). A substantial portion of total soil C can be found in subsurface layers (Diochon and Kellman, 2009; Harrison et al., 2011; Kaiser et al., 2002; Zabowski et al., 2011). Jobbagy and Jackson (2000) projected that 56% of soil C globally can be found below a depth of 1m. Nevertheless, the major National Programs for forest C estimates in the US – the Forest Inventory and Analysis (FIA) and Forest Health Monitoring (FHM) programs of the United States Department of Agriculture Forest Service – sample soils only to 0.2 m (O'Neill et al., 2005). This represents a major systematic underestimation of total soil C, an error that propagates to any model or budget that relies upon this data (Hamburg, 2000).

Defense of shallow forest soil sampling has relied upon studies that identify residence times ranging from hundreds to thousands of years for C in subsurface horizons (Chabbi et al., 2009; Fang and Moncrieff, 2005; Kaiser et al., 2002). While residence times for resistant C compounds in deep soil horizons may be quite long, the assumption that this C will remain non-reactive may be less accurate than previously thought, particularly in soil with deep rooting zones. Recent research has given considerable support to the hypothesis that deep soil C may become reactive under certain conditions. Studies show that the stability of C in deep soil layers is controlled by the supply of fresh, energy rich organic matter (Fontaine et al., 2007); that labile and resistant C pools respond similarly to changes in soil temperature (Fang et al., 2005); that nitrogen fertilization affects soil C both in surficial and subsurface horizons (Adams et al., 2005; Canary

et al., 2000); and that retention of organic matter postharvest may increase the recalcitrant soil organic pool in intensively managed timber stands (Strahm et al., 2009). The resistant C in deep soil was formed under a particular set of environmental conditions that, by every account, will be altered by continuing climate change and human influences (Fang et al., 2005; Richter, 2007; Richter and Yaalon, 2012). Natural and man-made changes in factors and processes that affect soil genesis can spur rapid movement of C into and loss of C from soil material that has remained stable for long periods of time (Fontaine et al., 2007; Harrison et al., 2011). Moreover, increases in the recalcitrant soil organic C pool are an important potential long-term sink for atmospheric C (Lorenz et al., 2007). Ignoring subsurface soil horizons effectively eliminates our ability to accurately assess C sequestration in the more recalcitrant pool.

This study, conducted in coastal Pacific Northwest Douglas-fir forests, addresses three objectives: (1) to determine the effect of systematic sampling to variable depths on estimates of forest soil C; (2) to evaluate the ability of mathematical models to accurately predict total soil C to 2.5 m based upon 1.0 m sampling (a depth usually reachable with a non-powered soil auger); and (3) to assess which soils are most important to sample more deeply.

2. MATERIALS AND METHODS

Twenty-two sites were selected from the Stand Management Cooperative (SMC) Type V Long-Term Site Productivity plots to cover a range of soils across the parent materials and climatic conditions of the coastal Pacific Northwest region (Table 1). All study sites were intensively managed, 20-to-25 year old Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco) plantations within the region bounded by the Pacific Ocean and the Cascade Range, and thus under a maritime-influenced climate which is generally characterized by mild, wet winters and

warm, dry summers (Figure 1). The long-term mean annual temperature across the twenty-two sites ranged from 6.1°C at the Vailton site in the Cascade foothills to 11.6°C at the Jory site in the Willamette Valley (mean annual temp across all sites was 9.0°C). The lowest mean January temperature was 0.5°C at the Tokul site in the northern end of the Puget Trough, and the highest mean July temperature was 19.8°C at the Honeygrove site in the Willamette Valley. Mean annual precipitation ranged from 813 mm at the Olete site on the east side of the Olympic Range to 3302 mm at the Hoko site on the west side of the Olympic Range (mean annual precipitation across all sites was 1780 mm).

Soil pits were dug using an excavator to 2.5 or 3.0 m and classified using data from the US Department of Agriculture Natural Resources Conservation Service soil surveys (Soil Survey Staff, 2013). Bulk density samples were taken using a soil corer of a known volume in the middle of succeeding soil layers bounded by the mineral soil surface and depth intervals of 0.1, 0.5, 1.0, 1.5, 2.0, 2.5 and 3.0 m (when excavation was not impeded by bedrock or hardpan). Major horizons were identified and recorded, along with horizon thickness and profile depth (Figure 2). In all cases, soil pits extended into the C horizon, often extensively. Additionally, one forest floor sample per site was gathered from randomly placed 0.3 m x 0.3 m quadrats. Samples were sealed in plastic bags and immediately returned to the lab, where they were refrigerated at 3°C until analysis.

Hamburg (2000) showed how inadequate soil sampling gives rise to inaccurate conclusions in analyses of ecosystem processes, and we followed his recommendation that measurements of soil bulk density and carbon concentration come from the same samples. The bulk density mineral soil samples were weighed, dried in an oven at 60°C to constant weight, and then re-weighed. Holub (2011) demonstrated that sieving soil to <2 mm unnecessarily discards

significant amounts of soil C in the 2 mm to 4.75 mm fraction. Consequently, our samples were separated into coarse and fine soil fractions using a 4.75 mm sieve. All obvious roots remaining in the sieve were discarded. The >4.75 mm fraction was weighed to determine mass and percent content of coarse and fine material at each depth interval.

Subsamples of the <4.75 mm fraction were ground with a mortar and pestle for elemental analysis. Organic horizons likewise were weighed, dried at 60°C to constant weight, and reweighed. Samples were ground to <0.5 mm using a Wiley Mill. Total C analysis was performed using a PerkinElmer 2400 CHN analyzer.

The total carbon (C_t) for each sample layer was calculated from: the layer height (H) (cm) between the sample depth and the depth of the sample immediately above it or the surface; the bulk density (D_b) (g cm^{-3}) of the <4.75 mm fraction; and the C concentration (C_{con}), using the equation

$$C_t = H\, D_b\, C_{con} \qquad (1)$$

The result of this calculation for each sample was then converted from g cm^{-2} to Mg C ha^{-1}. Equation (1) assumes that C_{con} is constant throughout a layer of height H. Thus, C_{tot} calculated for any given layer will likely underestimate C from the top portion of the layer and overestimate C from the bottom portion of the layer. These differences should approximately cancel, giving a reliable estimate for C_{tot} in a given layer. However, due to the high rate of change in C_{con} near the soil surface, this method will slightly underestimate C_{tot} at the surface.

To address the question of appropriate soil sampling depth, total cumulative C was calculated for interval layers 0-0.1 m, 0-0.5 m, 0-1.0 m, 0-1.5 m, 0-2.0 m, 0-2.5 m, and 0-3.0 m. Due to the unpredictable variability of organic horizon C across sites, only mineral soil was included in these analyses. The cumulative C in each interval was calculated by addition of C from the

deepest measured layer in the interval to the total of the more shallow depth intervals. For example, the 0-3.0 m interval includes all C in the measured layers from the surface to 3.0 m.

To test differences between soil C estimates for different soil sampling depths, mean total C across the cumulative depth intervals was compared using a linear mixed effects model in R (R Core Team, 2013). Using the *nlme* package (Pinheiro et al., 2013), depth intervals were compared as a fixed effect factor. The individual soil profiles were treated as a random block effect to account for the expected differences between different soils and to isolate the effect of sampling depth on the cumulative soil C estimates. Because samples taken from the same profile are spatially autocorrelated and thus violate the assumption of independent measures, the model incorporated a Gaussian spatial autocorrelation structure between sampling depths for the particular soil from which the samples were taken. Model-fitting with mean total C values showed strong departure from normality in the tails of residuals upon examination of the Normal Quantile-Quantile plot; a log transformation was performed on the data, and examination of the Normal Q-Q plot for the transformed data showed satisfactory agreement with the assumption of normally distributed residuals. The mean cumulative C for the 0-3.0 m layer was not included in the modeling or statistical analysis due to an inadequate number of soil profiles that could be sampled to that depth ($n = 7$ compared to $n = 20$-22 for other layers). ANOVA of the linear mixed-effects model was used to test for differences between means, and Tukey's Honestly Significant Difference (Tukey's HSD) was used to calculate multiple comparisons between the sampling depth intervals using the *multcomp* package in R (Hothorn, 2008). All statistical tests were performed at $\alpha = 0.01$ because the F-statistic is an approximation in ANOVA analysis of linear mixed models, and thus provides a greater degree of statistical confidence. Denominator degrees of freedom for the ANOVA F-test were calculated using the Satterthwaite method.

A series of functions were tested using Proc NLMIXED (SAS Institute Inc. 2008) to determine whether a single equation form could be fit to the soil C profiles (to a 2.5 m depth) at all 22 sites. In these functions, soil depth (i.e., the midpoint of each sampling interval) was used as the predictor of soil C content, and site was treated as a random effect. The group of functions initially tested were: logarithmic, first-degree inverse polynomial (hyperbola), type III exponential, the power function (Bennema, 1974), Schumacher's equation (Schumacher, 1939), the log-log equation of Jobbagy and Jackson (2000), the Langmuir equation, the negative exponential function (Minasny et al., 2006), the Chapman-Richards function, and the organic matter function used by Russell and Moore (1968). Where applicable, functions were fit to the C concentration profiles (concentration decreasing with depth) and to the cumulative soil C content (total content typically increasing toward an asymptote with depth). Four of the tested functions fit the data relatively well across all sites: these models always converged, were significant ($P < 0.05$), and had satisfactory distribution of residuals upon visual assessment. All four of the best-fitting functions fit cumulative soil C content rather than the C concentration profile. These functions were:

the first-degree inverse polynomial function:

$$C_t = D / (a + b\,D) \qquad \text{(Sit and Poulin-Costello, 1994)}$$

the Langmuir equation:

$$C_t = C_{max}\,a\,D / (1 + a\,D) \qquad \text{(Johnson et al., 2011)}$$

a logarithmic function:

$$C_t = a + b \ln(D) \qquad \text{(Johnson et al., 2011)}$$

and a type III exponential function:

$$C_t = a\,e^{b/D} \qquad \text{(Sit and Poulin-Costello, 1994)}$$

where C_t is total carbon content (Mg ha^{-1}) between the soil surface and depth D (cm), C_{max} is the maximum C content of the soil profile (Mg ha^{-1}), and a and b are fitted constants for the functions.

To test the capacity of these functions to predict deep soil C without sampling beyond 1.0 m, each function was then used to predict each soil's total C to a depth of 2.5 m, after fitting the function only with data from sample intervals of 0-0.1, 0.1-0.5, 0.5-1.0 m.

3. RESULTS AND DISCUSSION

3.1 SOIL CARBON AND SAMPLING DEPTH

The total C in the profile for each soil series (0-2.5 m depth) ranged from 61 to 321 Mg ha^{-1} (Figure 3). By comparison, mean organic-horizon C was 7.3 Mg ha^{-1} (with a range from 2.3 to 14.1 Mg ha^{-1}). Total C in organic horizons did not correlate with total C in the mineral soil (R^2 = 0.04) and thus subsequent analysis focused solely on mineral soil. ANOVA of total C at increasingly large intervals of depth showed that some mean estimates of C were significantly different than others (F = 188.51, P< .001). The F-test and p-value was calculated with 101 denominator degrees of freedom from the Satterthwaite approximation. Tukey's HSD revealed 4 groups of means that were significantly different from each other: the total soil C estimates for the 0-0.1 m and 0-0.5 m sampling depths were different from both each other and all other mean estimates; the 0-1.0 m, 0-1.5 m, and 0-2.0 m sampling depth means form a third group; and finally the 0-1.5 m, 0-2.0 m, and 0-2.5 m sampling depths form the final group of means (Figure 4; P<0.005). Controlling for spatial autocorrelation between samples and differences between sites, sampling to 1.0 m will give significantly less total soil C than sampling the whole profile (to a depth of 2.5 m).

On average, the greatest contribution to total soil C came from the 0.1-0.5 m layer (Figure 5). Though mean C concentration in this layer was less than half that of the surface to 0.1 m layer, increased bulk density and layer thickness resulted in more total C in the layer. Mineral soil C concentration decreased regularly with depth with averages between 0.32% and 0.15% in layers below 1.0 m. By comparison, average litter layer C concentration was 33%. Despite very low C concentrations, average soil C content in the deep layers never decreased below total C in the litter layer due to much higher bulk density. Deep soil layers contribute substantially to total soil C, even when C concentrations are low.

Given the need to establish a standard soil sampling depth for national C accounting, our data suggest that, for the soils in this region, sampling to at least 1.0 m would substantially increase the accuracy of total soil C estimates. However, an average of 21% of total C was located below 1.0 m, with a range from 6% in the Ovall series to 57% in the Vailton series. Jobbagy and Jackson (2000) projected that 56% of soil C globally across forest, grassland, shrubland, and other ecosystems can be found between 1.0 m and 3.0 m depth. For temperate coniferous forest soils their estimate of soil C below 1.0 m was 41% compared to our estimate of 21% (to 2.5 m; this increases to 27% if we include the 2.5-3.0 m estimate from 7 soils). The difference between these estimates can be explained by differences in methods; Jobbagy and Jackson (2000) calculated C below 1.0 m by extrapolating from surface to 1.0 m data using a log-log equation that does not become asymptotic with depth, whereas our observed soil C profiles are best fit by functions that predict a sharper decline in soil C in deep parts of the soil profile. In general, methods that sample as deeply as possible in the soil profile will better approximate total C. Since accuracy of these estimates is critical in appropriate calculation of the total soil C reservoir (Hamburg, 2000; Zabowski et al., 2004), in measuring C flux (Lorenz and Lal, 2005), and in

properly modeling C dynamics over landscapes or ecosystems (Lal, 2005; Schrumpf et al., 2011), I recommend sampling soils as deeply as necessary to test defined hypotheses or meet objectives while considering budget and labor constraints.

3.2 MODELING SOIL CARBON PROFILES

Four soil C profiles selected to represent the range of C profiles among the 22 soils in this study are shown in Figure 6. The C profile of the Shelton soil series is typical of lowland glacial soils of the Puget Trough physiographic region. This soil is formed in young (~14,000 years) glacial materials and has a high coarse fragment content, with volcanic ash in surface horizons; cumulative C levels off significantly below the 0.1-0.5-m depth interval. Among the four mathematical functions fit to this profile, the fit of the logarithmic function was somewhat poorer than the other three functions at intermediate depths, as it did not capture the sharp decline in the rate of C accumulation below 0.5 m.

The Melbourne soil series is a well-drained loam formed in sedimentary residuum, found in the foothills. Although total C to 2.5 m is similar to the Shelton series, distribution of C in the profile is more even, producing a more gradual accrual of C with depth. In contrast with the Shelton series, the exponential function fit the C profile best, whereas the other three functions became asymptotic too rapidly and thus underestimated total C to 2.5 m.

The Honeygrove series is a very deep, well-drained Ultisol, with approximately twice the C content of the Shelton and Melbourne series, and an intermediate C profile shape relative to those series. The poorest fit was the Langmuir equation, which became asymptotic too early as a result of the value for the 0.5-1.0 m interval.

The Hoko series (308 Mg C ha^{-1}) is representative of the two sampled soils with very high C content; Vailton (269 Mg C ha^{-1}) is the other. Again the Langmuir equation became asymptotic too rapidly and did not account for the continuing rate of accrual lower in the soil profile.

Across all 22 soils, the inverse polynomial function had the most uniform distribution of residuals across soil depth (Figure 7; Table 2). The Langmuir equation consistently underestimated cumulative soil C at greater depths, whereas the logarithmic function tended to underestimate cumulative C near the surface and then overestimate it at a depth of 2.5 m. The exponential function overestimated cumulative C at intermediate depths but underestimated it at 2.5 m. Measured vs. predicted cumulative C to 2.5 m (i.e., curve fit at 2.5 m) appears in Figure 8. The inverse polynomial function again showed the most consistent fit across soils series. The greatest error in model fit occurred for the two soils highest in soil C. For the inverse polynomial, Langmuir, logarithmic, and type III exponential functions, the mean percentage error in predicting cumulative C at 2.5 m for the 22 soils was -2.0, -6.4, 2.7, and -5.0, respectively. Excluding the two soils highest in C, the mean percentage error for the remaining 20 soils was -1.7, -5.5, 4.0, and -4.5 for the same equations, respectively.

To determine the feasibility of accurately predicting cumulative soil C to a 2.5 m depth using data only to a 1.0 m depth, the four selected functions were then fit with cumulative C data from depth intervals of 0-0.1, 0.1-0.5, and 0.5-1.0 m. Using these fitted equations for each soil series, cumulative C was predicted to 2.5 m. Error increased for all four functions (Figure 9) relative to the fit with the full 0-2.5-m dataset (Figure 8). The greatest error occurred for the logarithmic and type III exponential functions (Table 2); conversely, the Langmuir equation predicted cumulative C to 2.5 m nearly as well with data from only 0-1.0 m as it did with data from 0-2.5

m. Mean percentage error in prediction across the 22 soils, and for the 20-soil sample excluding the high-C profiles, was lowest for the inverse polynomial function (Table 2).

While the predictions of the best-fit models were generally adequate for total soil C to 2.5 m, several exceptions should be noted. First, the Hoko and Vailton series (both Andisols) were not well modeled, despite the importance of these C-rich soils for C accounting. Second, considerably more soil C is located below 2.5 m in these soils, and estimates produced by these models should not be construed as the same as sampling to bedrock. Our sampling only reached bedrock at two sites, the Kinney and O'brien series soils. Finally, none of the soils modeled in this study were Spodosols; as noted by Johnson et al. (2011) Spodosols resist standard C modeling techniques due to the irregular increase of C concentration in subsurface, spodic horizons.

3.3 SOIL PARENT MATERIAL AND CARBON CONTENT

Several of the soils sampled in this study provide useful examples of the broad variations in factors that influence soil C profiles of the coastal Pacific Northwest region. These factors include variations in parent materials (volcanic, glacial, and sedimentary), soil age, and climatic gradients. Here we discuss typical glacial and sedimentary soils, as well as volcanic soils with the highest total soil C.

The recent glacial history of the Puget Trough was accompanied by active volcanism in the nearby Cascade Range, which influenced the formation of many soils in the region. All sites with glacial parent materials selected for this study, except the Scamman series, have been impacted by volcanic activity to some extent. The Scamman series consists of deep, poorly drained soils that formed in mixed material weathered from glacial sources and can be found on high terraces

and footslopes of adjacent hills. It had the least total soil C of all sites (58 Mg ha^{-1}), of which 14% was found below 1.0 m.

Tokul series soils are formed in similar, poorly drained conditions on glacial till, but with the addition of loess and volcanic ash. These soils contain amorphic mineralogy and andic materials near the surface. Both the Scamman and Tokul series occur along the western slopes of the North Cascade Range. The C profile of the Tokul site excavated in this study contained 128 Mg C ha^{-1}, considerably more than the Scamman site despite being considerably younger, and having similar climatic conditions (~1400 mm precipitation per year, and mean annual temperature of 10°C).

The Honeygrove and Jory series are typical of soils developed from colluvium and residuum derived from sedimentary and basic igneous rocks. Both are very deep, well drained Ultisols, with the highest mean annual temperatures of all 22 sites (11.7°C at the Jory site; 10.6°C at the Honeygrove site). Despite similar conditions and relatively close proximity (Figure 1), the Honeygrove site contained substantially more soil C than the Jory site (158 Mg ha^{-1} to 94 Mg ha^{-1}, respectively), and the C accrued more gradually down the Honeygrove profile than the Jory profile. The differences in C distribution could be accounted for by differences in topographical position between the two sites: the Honegrove site is on a toe slope in the Oregon Coast Range, whereas the Jory site is near a peak in the hills south of the Willamette Valley. Climate also differs: the Jory site receives only 1143 mm per year, while the Honeygrove site receives 1905 mm precipitation per year, which could lead to increased leaching of organic matter to deeper soil layers.

The two series with the highest total C and greatest C below 1.0 m were the Hoko (308 Mg ha^{-1} to 2.5 m, 116 Mg ha^{-1} below 1.0 m) and Vailton series (269 Mg ha^{-1} to 2.5 m, 133 Mg ha^{-1}

below 1.0 m). Despite similar soil C profiles, the Hoko and Vailton series soils differ in many key environmental factors. The Hoko series is a young (~14,000 years), moderately deep, moderately well drained soil formed in glacial till mixed with volcanic ash. The excavated site is located on the west side of the Olympic peninsula at low elevation (61 m) and has the highest annual precipitation of all 22 sites (3302 mm). The Vailton series is considerably older (~40,000,000 years) and consists of deep, well-drained soils formed in volcanic ash and colluvium and residuum from siltstone and shale. These soils are found exclusively at somewhat higher elevations; the excavated site is 634 m above sea level in the Cascade foothills. Annual precipitation for Vailton series soils is slightly above average (2032 mm compared to mean 1780 mm across 22 sites), and the annual average temperature is the lowest from all 22 sites (6.1°C). The primary similarity between these soils is the presence of large quantities of noncrystalline minerals, including allophane, imogolite, and ferrihydrite (Soil Survey Staff, 2013). These amorphous or nanocrystalline minerals exhibit high adsorption of humic material, particularly humic and fulvic acids (Kennedy et al., 2002; Oades, 1988; Tisdall and Oades, 1982), and can preferentially preserve certain organic compounds from biological degradation for long periods of time (Baldock and Skjemstad, 2000; Calabi-Floody et al., 2011; Marschner et al., 2008).

The models tested in this study lacked the ability to adequately account for continuing, non-asymptotic accrual of C in deep layers, and thus poorly predicted total soil C for the Hoko and Vailton series (Table 2). In such cases, fully sampling the deep soil (> 1.0 m) remains the only satisfactory means for estimating total soil C. We suggest a simple diagnostic to evaluate where such deep (and expensive) sampling should take place: in soils of the Andisol order, particularly those with a histic epipedon (such as the Aquand Great Group), or those with mineralogy classes that are characterized by amorphous minerals or Al and Fe oxides. Examples include the

halloysitic, amorphic, and ferrihydritic mineralogy classes (Soil Survey Staff, 1999). This diagnostic is by no means flawless; while it would successfully identify the Hoko and Vailton series (ferrihydritic and amorphic classes, respectively), it would also identify the Tokul (amorphic, Aquic Vitrixerand) and Hoquiam series (ferrihydritic, Typic Fulvudand) as important to sample more deeply. Both the Tokul and Hoquiam sites carried 8-14% of total soil C below 1.0 m, and were well modeled by the inverse polynomial equation. However, for the objective of strategically sampling deep soil C at a landscape scale, some false positives (i.e., sites wrongly suspected of high deep-soil C content) are preferable to randomly located deep sampling and overlooking soils with large quantities of deep soil C.

Sampling of these volcanically influenced soils for C should endeavor to be as deep as possible, or until a restrictive layer is reached. Though the analyses here are limited to the top 2.5 m of the soil profile, our soil pit at the Vailton site reached 3.5 m. Total soil C for this site was 317 Mg ha^{-1} to 3.5 m, and 15% of this total was found below 2.5 m. If substantial adsorption of organic matter to amorphous or clay minerals has occurred in a soil, sampling to bedrock will provide the most accurate assessment of total soil C.

Deep soil sampling is both expensive and labor intensive (Harrison et al., 2003). However, the difficulty or expense of a method is no justification for inadequate alternatives (Harrison, et al., 2011). Sampling protocols that call for many shallow soil pits could be improved by, at minimum, sampling more deeply in a targeted subset of soils that are more likely to have substantial C in subsurface (>0.5 m) layers. While we believe sampling to 1.0 m – and preferably to 2.5 m – provides a superior alternative to conventionally shallow soil sampling, we also recognize that it fails to capture a truly complete picture of total soil C. The development of

methods to sample soil accurately down to bedrock would be a boon to biogeochemistry and soil science generally.

4. CONCLUSIONS

1. Rocky forest soils of the coastal Pacific Northwest have substantial total soil C below 0.5 m. When sampling depths were increased from 0.5 m to 1 m and 2.5 m, average total soil C increased by 21 and 48 Mg ha^{-1}, respectively.

2. A nonlinear mixed effect model using an inverse first-degree polynomial was developed that adequately predicted total soil C to 2.5 m on a soil-specific basis given sampling to 1.0 m. Reasonable predictions were made for forested soils of this region ranging from Entisols to Ultisols and across glacial, sedimentary, and volcanic parent materials; the exceptions were deep volcanic soils with noncrystalline minerals throughout the profile.

3. Andisols that feature noncrystalline mineralogy may contain more soil C than other soil types particularly at depths below 1 m.

4.1 SOIL SAMPLING CONSIDERATIONS

Soil sampling protocols in the coastal Pacific Northwest need to take these factors into account. How deep to sample soil depends on the objectives of the investigation and the soils in question. Shallow soil sampling (< 1m depth) will underestimate soil C stocks and will provide incomplete information on the magnitude and direction of soil C flux. Sampling protocols that call for a large number of shallow samples can be improved by adding a sub-set of deep soil samples targeted to soils where substantial C is likely to be stored in subsurface horizons. Sampling

deeper in the soil profile is more expensive and time consuming but it will provide better estimates of ecosystem C budgets and fluxes, particularly for soils with andic properties.

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CHAPTER II – DEEP SOIL NITROGEN

5. INTRODUCTION

Nitrogen (N) is the primary limiting nutrient in many forest ecosystems in the Pacific Northwest (Van Miegroet and Cole, 1984, 1985; Walker and Gessel, 1991; Cole, 1995). Consequently, the pools, fluxes and transformations of N have been studied in great detail to decipher the complete N cycle and to understand the fate of anthropogenic N inputs. Standard practice for N stock and flow budgets account for N either in the top 0.2 m of soil (Amelung *et al.*, 1998; Hart and Sollins, 1998), in the top 1.0 m of soil (Adams *et al.*, 2005), or to the bottom of the B horizon (Van Miegroet and Cole, 1984; Huntington *et al.*, 1988); leaching loss of NO_3 past this point is considered lost to the system (Van Miegroet and Cole, 1984, 1985). At the landscape scale, the USDA Forest Inventory and Analysis Program monitors soils to a depth of 0.2 m (O'Neill *et al.*, 2005), despite evidence that substantial amounts of carbon (C) and N across all soil orders are stored in subsurface layers, even below 1.0 m (Whitney and Zabowski, 2004; Zabowski *et al.*, 2011; James *et al.*, in press).

Mobile forms of N should not be assumed to move strictly downward in soil, particularly when tree roots provide passive movement of water toward dry regions of soil whether they are downward, lateral, or even upward from wetted soil. The Mediterranean climate of the coastal Pacific Northwest leads to surface soil drying during summer, precisely when trees are most in need of water and nutrients. The high demand for water coupled with drought can induce hydraulic redistribution of soil water from deep soil layers to surface soil through plant roots, providing a mechanism for upward transport of dissolved organic N (DON) and NO_3, as well as P and base cations derived from weathering (Quijano *et al.*, 2013). Hydraulic redistribution replenished 28% and 40% of water depleted from the top 2 m of soil in a Douglas-fir stand at

Wind River during summer drought (Brooks *et al.*, 2002; Brooks *et al.*, 2006). In a review of temperate to semi-arid environments, Sardans (2014) noted that hydraulic redistribution was widespread across species and ecosystems, and contributed 17-81% of total transpired water. This upward flux of the soil solution is a predominately nocturnal process that occurs when tree transpiration is reduced and surface soil water potential is high (Brooks *et al.*, 2006). The summer dry season is also marked by little NO_3 leaching and low soil solution concentrations (Strahm *et al.*, 2005). This can drive mineral surface adsorption equilibria toward release of NO_3 and DON in soils with substantial anion exchange capacity, precisely during periods of hydraulic redistribution. There is some evidence that plants performing hydraulic redistribution have higher N uptake and nutrient status, as well as higher ecosystem N turnover, though this has not been specifically tested in Douglas-fir ecosystems (Armas *et al.*, 2012).

Furthermore, tree roots have been documented to reach many meters in depth across ecosystems and plant species. Stone and Kalisz (1991) documented Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco) rooting to greater than 3 meters depth in numerous studies and noted that across over 200 tree species roots deeper than 1.0 m were often found in young stands (1-4 years). Root interactions with the soil are not limited to the upper soil horizons; sampling only surface layers (<0.5 m) ignores all N in deeper layers that is biologically available, and can substantially bias N pool estimates (Whitney and Zabowski, 2004). Considering the substantial land area occupied by soils deeper than 1.0 m in the Pacific Northwest and globally, accurately quantifying subsurface N is important to allow a comparison of traditional soil sampling depths with depths compatible with the maximum extent of tree rooting zones. This study addresses three objectives within the coastal Douglas-fir forests of the Pacific Northwest: (1) to determine the effect of sampling depth on estimates of soil N; (2) to evaluate the ability of mathematical

models to accurately predict total N to 2.5 m based upon shallower sampling to 1.0 m or 1.5 m; and (3) to assess which soils are most important to sample deeply for N.

6. MATERIALS AND METHODS

Twenty-two sites were selected from University of Washington Stand Management Cooperative Type V research plots. All sites were located within same-age (20-24 years) intensively managed Douglas-fir plantations within the region bounded by the Pacific Ocean and the Cascade Range (Figure 1). The selection of sites under similar ecological regimes allowed us to isolate the effects of parent materials, climate, and topography on soil development. Douglas-fir is the dominant overstory tree species in the region, and consequently a wide range of soil types can be found within this ecoregion (Table 1). The Pacific Northwest is a maritime-influenced climate generally characterized by warm, dry summers and mild, wet winters. Mean annual temperature across all sites was 9.0°C, ranging from 6.1°C at the Vailton site in the Cascade foothills to 11.6°C at the Jory site on the margins of the Willamette Valley. Generally, lower temperatures were found at the northerly and high elevation sites, while higher temperatures were found in southerly and lower elevation sites. The lowest mean January temperature was 0.5°C at the Tokul site in the northern end of the Puget Trough, and the highest mean July temperature was 19.8°C at the Honeygrove site in the Willamette Valley. Mean annual precipitation across all sites was 1780 mm, ranging from 813 mm at the Olete site on the east side of the Olympic Range to 3302 mm at the Hoko site near sea level on the western edge of the Olympic Range.

Using an excavator, soil pits were dug to 2.5 or 3.0 m. Profiles were classified by comparison between samples and NRCS Web Soil Survey data (Soil Survey Staff, 2013). Bulk density

samples were taken in the middle of succeeding soil layers bounded by the mineral soil surface and depth intervals of 0.1, 0.5, 1.0, 1.5, 2.0, 2.5 and 3.0 m using a soil corer of known volume. Excavation was halted at 1.0 m bedrock at the Kinney site and at 2.0 m hardpan at the O'Brien site. Soil pits extended into the C horizon at all sites (Figure 2). A forest floor sample was taken at each site using a randomly placed 0.3 m x 0.3 m quadrat. Both bulk density samples and forest floor samples were sealed in plastic bags and immediately returned to the lab, where they were stored at 3.0°C until analysis.

Litter layer samples were weighed, dried at 60°C to a constant weight, and reweighed. The samples were ground to <0.5 mm using a Wiley Mill. Soil subsamples used in elemental analysis were taken from the bulk density samples to avoid potential biases as recommended by Hamburg (2000). The bulk density soil samples were weighed, dried in an oven at 60°C to a constant weight, then reweighed. Because sieving samples to <2mm unnecessarily discards meaningful quantities of soil C and N (Holub, 2011), samples were sieved to <4.75 mm, and rock weight was recorded. All large roots were discarded. All calculations of bulk density and total soil N were based upon the total weight of samples minus the >4.75 mm fraction. Whitney and Zabowski (2004) showed that substantial proportions of total soil N may be found in the >2mm coarse fraction. In our analysis, N found in the >4.75 mm fraction was considered null, and consequently may be a source of underestimation. For elemental analysis, subsamples of the <4.75 mm fraction were ground with a mortar and pestle to a fine powder. N concentration was obtained using a PerkinElmer 2400 CHN Analyzer.

Total nitrogen (N_t) for each sample layer was calculated from: the bulk density (D_b) (g cm^{-3}) of the <4.75 mm fraction; the nitrogen concentration (N_{con}), and the layer height (H) (cm) using the equation

$$N_t = D_b\, N_{con}\, H \qquad (1)$$

and converted to kg N ha^{-1}. Bulk density samples were taken from the middle of each sample layer. Because equation (1) assumes that N_{con} from the point sample is constant throughout the layer height (H), the estimated N_t is likely an underestimate of the actual N in the top portion of the layer and an overestimate of the N in the bottom portion of the layer. These errors should approximately cancel, giving a reliable estimate for N_t in the calculated layer.

To address the question of appropriate sampling depth, total N was calculated for interval layers corresponding to soil pit depths of: 0- to 0.1-m, 0- to 0.5-m, 0- to 1.0-m, and so on to 0- to 3.0-m. Organic horizon N was excluded from these analyses. Total N for each of these intervals was calculated as the sum of all measured layers ($N_{0\text{-}0.1} + N_{0.1\text{-}0.5} + \ldots$) within the interval.

Because N values at each sample point are spatially autocorrelated and violate the assumption of independent measures necessary for traditional linear modeling and ANOVA tests, mean N estimates were compared using a linear mixed effects model in R (R Core Team, 2012). Using the *nlme* package (Pinheiro et al., 2013), mean total N at increasing depth intervals was compared as a function of depth as a fixed effect factor and site as a random block effect (accounting for the expected total N differences between soils). Fitting the model with a range of correlation structures did not significantly improve the model fit (AIC or BIC), and consequently no explicit structure was used in the final model. Examination of the Normal Q-Q plot for model residuals showed large departures from normality in the tails. The Hoko site, which contained by far the most N of all 22 soils (as well as 12369 kg ha^{-1} below 1.0 m, or 46.5% of total), was excluded as an outlier; subsequent examination of the residual Normal Q-Q plot showed better agreement with the assumption of normally distributed errors, and an Anderson-Darling test showed no significant departure from normality (A=0.553, P=0.1509). The mean cumulative N

for the 0- to 3.0-m layer was excluded due to an inadequate number of soil profiles sampled to that depth (n=7 compared to n=20-22 for shallower pits to 2.5 m).

ANOVA of the linear mixed effects model was used to test for differences between means, followed by Tukey's HSD to calculate multiple comparisons between sampling depth intervals using the *multcomp* package in R (Hothorn, 2008). Denominator degrees of freedom for the ANOVA F-test were calculated using the Satterthwaite method. The conservative value $\alpha = 0.01$ was used for all statistical tests because the F-statistic and subsequent p-value are approximations in ANOVA of linear mixed effects models, and thus a greater degree of statistical confidence is desirable.

A series of nonlinear mixed effect model functions were tested to determine whether a single equation could be fit to the soil N profiles to a depth of 2.5 m at all 22 sites using the nlme package in R (Pinheiro et al., 2008). The group of functions initially tested to determine best fit were chosen based upon previous use in the literature to estimate total soil C or N values. These functions included: logarithmic, first-degree inverse polynomial (Sit and Poulin-Costello, 1994), the power function (Bennema, 1974), type III exponential function, the Langmuir equation (Johnson *et al.*, 2011), the negative exponential function (Minasny *et al.*, 2006), the Chapman Richards function, the log-log equation of Jobbagy and Jackson (2000), the Gompertz function, and the organic matter function of Russel and Moore (1968). Because the equations would subsequently be used to estimate soil N to 2.5 m based upon shallower sampling procedures, equations that contained more than 2 coefficients were excluded (such as the Gompertz and Chapman Richards functions). Four of the tested functions fit the data relatively well across all sites: these models always converged, were significant ($P < 0.05$), and had satisfactory

distribution of residuals upon visual assessment. These best-fitting functions fit cumulative soil N rather than N concentration profiles. The functions were:

the log-log function:

$$\ln(N_t) = a \ln(D) + b \qquad \text{(Jobbagy and Jackson, 2000)}$$

the Langmuir equation:

$$N_t = N_{max}\, a\, D / (1 + a\, D) \qquad \text{(Johnson } et\ al.\text{, 2011)}$$

the logarithmic function:

$$N_t = a + b \ln(D) \qquad \text{(Johnson } et\ al.\text{, 2011)}$$

And the type III exponential function:

$$N_t = a\, e^{b/D} \qquad \text{(Sit and Poulin-Costello, 1994)}$$

where N_t is total nitrogen content (kg ha^{-1}) between the soil surface and depth D (m), and N_{max}, a, and b are fitted constants.

Due to time, budget, safety, or logistical limitations, it is often not possible to dig sample pits as deep as 2.5 m. Consequently, it is of much practical use to estimate total soil N given shallower sampling. To test the capacity of these functions to make such predictions, each function was fitted using a shallower subset of data (0- to 1.0-m or 0- to 1.5-m) and extrapolated to 2.5 m. The functions were fit twice with 1.0 m and 1.5 m maximum depth data to investigate the effect of small increases in sampling depth on prediction error.

7. RESULTS AND DISCUSSION

7.1 SOIL NITROGEN AND SAMPLING DEPTH

Excluding the litter layer, total soil N in the profiles (0 to 2.5 m depth) ranged from 2660 kg ha^{-1} in the Olete series to 26600 kg ha^{-1} in the Hoko series, with a mean of 8660 kg ha^{-1} (Figure

10). On average, only 273 kg ha^{-1} or just over 3% of total N was found in the litter layer. The organic horizon N was not correlated with total mineral soil N ($R^2 = 0.045$) and was excluded from subsequent statistical analysis.

ANOVA of total N at increasingly large intervals of depth showed that some mean estimates were significantly different than others ($F = 78.55$, $P < 0.001$). The Satterthwaite method calculated 101 denominator degrees of freedom for the F-test and subsequent p-value. Tukey's HSD showed 5 groups of means that were significantly different from each other: the 0- to 0.1-m and 0- to 0.5-m means were significantly different from both each other and all other means; the 0- to 1.0-m and 0- to 1.5-m means form a third group; the 0- to 1.5-m and 0- to 2.0-m means form the fourth group; and the 0- to 2.0-m and 0- to 2.5-m means form the fifth and final group ($P < 0.005$; Figure 11). This shows that, controlling for spatial autocorrelation between samples and differences between sites, sampling shallower than 2.0 m will give a significantly different estimate of total soil N than sampling the whole profile (to a depth of 2.5 m).

The greatest contribution to total soil N, on average, comes from the 0.1- to 0.5-m depth interval (Figure 12). Across all profiles, N concentration decreases steadily with depth. Average soil N concentration varies between 0.025 and 0.018% between 1.0 m and 2.5 m with measured concentrations ranging from 0.062% at 2.5 m in the Hoko soil to 0.002% at 2.5 m in the Jory soil. Despite low N concentrations, between 8 and 9% of total soil N was found in each of the 1.0- to 1.5-m, 1.5- to 2.0-m, and 2.0- to 2.5-m intervals, on average. Excluding the Kinney site where excavation was stopped at 1.0 m bedrock, an average of 2691 kg N ha^{-1} (or 31% of total) was found below 1.0 m. Over 45% of total N was found beneath 1.0 m at three sites (Barneston 2, Hoko, and Vailton), while only the Astoria and Jory sites carried less than 10% of total N below 1.0 m. These values largely coincide with the 7-35% underestimation of soil N based upon

1.0 m sampling found by Whitney and Zabowski (2004). Sub-1.0 m soil layers can account for substantial portions of total N; ignoring these deep layers in nutrient pool analyses could lead to large differences in estimated pools and fluxes and, consequently, to inaccurate conclusions.

Only 50% of root biomass can be found above the top 0.3 m of soil in temperate coniferous forests such as those in the Pacific Northwest (Jackson *et al.*, 1996). In a comprehensive review covering over 200 species, Stone and Kalisz (1991) recorded Douglas-fir maximum rooting depths of 3.7 m in a 4 year old stand, and between 3 and 10 m depth in mature to old growth stands. In this study, roots were seen beneath 1.0 m in at least 14 of the 22 sites, some as deep as 2.3 m. The assumption that N leached deeper than 1.0 m or past the B-horizon is completely lost to the system may be deeply flawed, especially in soils that have a significant anion exchange capacity. Upward mobility of NO_3 and DON through hydraulic redistribution coupled with root growth toward wetted portions of soil during the primary growing season provides a clear mechanistic hypothesis for the transport and potential uptake of N stored in deep soils of the Pacific Northwest.

7.2 MODELING SOIL NITROGEN PROFILES

Four cumulative soil N profiles that bracket the range of total soil N and profile distributions are shown in Figure 13, along with the fit of all four model functions. The Honeygrove soil is a very deep, well-drained Ultisol derived from colluvium and residuum that contains above-average soil N for the soils in this study. The N profile is intermediate between more asymptotic soils (like Cloquallum), and those that are near-linear (Vailton & Hoko). The profile is fit well by the Langmuir, logarithmic, and exponential functions, but is over-estimated by the log-log function due to the large decline in soil N deep in the profile.

The Blachly series is a coarse Inceptisol formed from residual and colluvial igneous and sedimentary rocks that contains approximately average soil N for the soils in this study. The profile N distribution in this soil is more gradual than the Honeygrove series, though not as near to linear as the Hoko series. Also unlike the Honeygrove soil, the Blachly series was fit best by the log-log function, and is most severely underestimated by the exponential function.

The Cloquallum series is a somewhat poorly drained Inceptisol formed in silty lacustrine deposits around the southern tip of Puget Sound. It contains some of the least soil N of all 22 sites, and is close to asymptotic at depth. Consequently, the log-log and logarithmic function overestimate N at depth, while the exponential and Langmuir functions fit more closely.

Finally, the Hoko series is a moderately well drained Andisol that carries by far the most soil N of all sites, over three times the average for all 22 soils. The distribution of N is nearly linear with depth, leading to large underestimation of total N using the logarithmic and exponential functions. The log-log function fits this soil very well, and the Langmuir equation underestimates total N only slightly.

In general, the log-log function best fit soils with gradual accrual of N with depth. The Langmuir function showed the greatest flexibility in fitting soils with both near linear and more asymptotic profile distributions. The exponential function fit soils with an asymptotic distribution of soil N with depth best. Likewise, the logarithmic function best fit profiles with greater curvature, but was slightly more flexible than the exponential function in fitting soils with gradual accrual of N with depth.

Across all 22 sites, the residuals of the Langmuir equation are the smallest of all four functions, and show no consistent pattern above or below 0 (Figure 14). The log-log function has the second smallest residuals, but consistently overestimates N at 0.1 m and underestimates N at

1.0 m. This lack of consistent fit throughout the profile suggests that larger errors will develop when soil N is extrapolated to depth using this function. The logarithmic function has several large residuals both at 0.5 m and at 2.5 m, and consistently underestimates many soil N profiles. The exponential function likewise underestimates soil N at 2.5 m, and consistent patterns are visible in the residuals. Both the logarithmic and exponential functions show less consistency in curve fitting, suggesting that extrapolation to depth could result in significant departure from measured values. Measured vs. predicted cumulative N to 2.5 m appears in Figure 15. The Langmuir equation again shows the most consistent fit across soils. The log-log function is a close second, though soil N is consistently overestimated at 2.5 m. In soils with high total N content (greater than ~10,000 kg ha^{-1}), both the logarithmic and exponential functions substantially underestimate N at 2.5 m.

The lack of deep soil research in the literature bespeaks the difficulty of excavating and measuring soil beneath 1.0 m. For landscape scale research, ecosystem monitoring efforts, and other research efforts where many soil samples across a landscape are desired, deep soil sampling at every point is unlikely due to time and budget limitations. Consequently, it would be of much practical value to be able to estimate deep soil N across a landscape based upon a subset of deeply sampled points and extrapolation at shallower sample points. To determine the feasibility of accurately predicting cumulative soil N to 2.5 m using data to only 1.0 m or 1.5 m (depths typically reachable by a non-powered soil auger), the four selected functions were fit with cumulative N data from the depth intervals of 0- to 0.1-m, 0.1- to 0.5-m, and 0.5- to 1.0-m (the increment from 1.0- to 1.5-m was included when extrapolating based upon 1.5 m sampling). Error increased for all four functions relative to the fit with the full 0 to 2.5 m dataset, especially when extrapolating using data to 1.0 m (Figure 16). By contrast, extrapolating based upon 1.5 m

sampling substantially improved cumulative N estimates (Figure 17). Based upon extrapolation from 1.0 m data, error decreased from the exponential function, to the log-log function, the Langmuir equation, and finally to the logarithmic function (Table 3). The additional data point to 1.5 m decreased the mean percent error of all four function's estimates of cumulative N by between 2 and 11 percent. Whereas the Langmuir, logarithmic, and exponential functions had negative error, the log-log function had positive error; for practical purposes, using both the Langmuir equation (which has the least error based upon 1.5 m sampling) and the log-log function to extrapolate to depth would provide a means to reasonably bracket the likely cumulative soil N values in individual soils.

While predictions for the best-fit models were generally adequate for total soil N to 2.5 m based upon 1.5 m sampling, there are exceptions. All except the log-log function underestimate cumulative soil N in the Hoko series based upon shallow sampling; the log-log function provided a very accurate estimate of the Hoko series, but overestimated N in almost all other soils. All four equations substantially underestimated cumulative soil N for both the Barneston 2 and Vailton series. Both of these soils carry >50% of their total soil N below 1.0 m. All three of these series (Hoko, Barneston, and Vailton) are Andisols that have developed from deposits of volcanic material that must be sampled deeply to account for continuing accrual of N in deep layers.

7.3 SOIL MORPHOLOGY AND NITROGEN CONTENT

Several of the soils sampled in this study provide useful examples of the broad variations in factors that influence soil N cycling in the coastal Pacific Northwest region. These factors include climatic gradients, variation in parent materials, soil age, texture, and topography.

However, detailed disturbance histories were not available for these sites. The frequency of stand disturbance is important for N cycling in the coastal Pacific Northwest primarily due to colonization of N-fixing red alder (*Alnus rubra*) in early succession, which can result in inputs of 100-300 kg N ha^{-1} yr^{-1} (Van Miegroet and Cole, 1984; Binkley *et al.*, 1994). Depending on the density of red alder, the duration of their dominance in a stand, and frequency of disturbance, substantial amounts of N can build up in the soil, leading to high soil N (Perakis and Sinkhorn, 2011).

There are substantial differences between soil orders in this study, both in terms of total N to 2.5 m as well as the proportion of N stored in deep layers. Andisols carry 11100 kg N ha^{-1} on average (across 9 sites), of which 55% is below 0.5 m and 36% is below 1.0 m. Ultisols contain nearly as much N on average, 10600 kg ha^{-1} (across 3 sites), while Inceptisols (8 sites) and Alfisols (2 sites) hold 5950 and 5450 kg N ha^{-1}, respectively; the relative allocation of N with depth is relatively consistent across these three soil orders, with 41-44% of N below 0.5 m and 21-25% below 1.0 m. These differences suggest that pedogenic processes play an important role in controlling the trajectory of N cycling.

Higher than average retention of N in Andisols both in the total soil and at depth is likely related to the diagnostic mineralogical properties associated with this order. The Hoko and Vailton soil series are good examples from this study - the Hoko series is classified as a ferrihydritic soil, while the Vailton series is classified as amorphic (Soil Survey Staff, 1999). Noncrystalline minerals, including ferrihydrite, allophane and imogolite, possess variable charge surfaces that can develop both substantial cation and anion exchange capacities, especially in deeper horizons (Strahm and Harrison, 2007). This variable surface charge allows retention of NO_3^-, NH_4, amino acids, and other organic materials through adsorption to mineral surfaces,

particularly in deeper horizons where soil pH tends to be higher than in the surface soil (Strahm *et al.*, 2005). The Hoko series contains by far the most soil N of the sites in this study (26600 kg ha^{-1}), of which 46% is located below 1.0 m. Likewise, the Vailton series carries just over 50% of its 12500 kg N ha^{-1} below 1.0 m. Besides mineralogical qualities, the Hoko and Vailton sites do not share common site characteristics. The Hoko series is a young (~14,000 years), moderately deep, moderately well drained soil formed in glacial till mixed with volcanic ash. The excavated site is located at low elevation (61 m) on the western Olympic peninsula with the highest annual precipitation of all 22 sites (3302 mm). On the other hand, the Vailton series is considerably older (~4,000,000 years) and consists of deep, well-drained soils formed in volcanic ash as well as colluvium and residuum from siltstone and shale. The site is located at relatively high elevation (634 m) in the foothills of the Cascade Range with precipitation just above average across the 22 sites in this study (2032 mm compared to mean 1780 mm). The diagnostic nature of mineralogy in classifying Andisols, together with retention of N controlled by this mineralogy, suggests that ecosystem monitoring efforts and landscape-scale research should carefully consider targeted deep soil sampling of Andisols, particularly those with mineralogy classifications that are characterized by noncrystalline minerals or Al and Fe oxides.

The large difference in total soil N between Ultisols and other soil orders (Inceptisols and Alfisols) makes some intuitive sense. High historical inputs of N that reach saturation levels in soil favor nitrification of ammonia into nitrate, which can leach through the soil and remove base cations such as Ca, Mg, K, and Na, resulting in the characteristic low base saturation of Ultisols. The Honeygrove and Jory series are typical Ultisols developed from colluvium and residuum derived from sedimentary and igneous rocks. As two of the lowest latitude sites in this study, they have the highest mean annual temperatures, 11.7°C at the Jory site and 10.6°C at the

Honeygrove site. However, the Jory soil contains considerably less N (7990 compared to 12200 kg ha^{-1} in the Honeygrove series), as well as only 9% of total N below 1.0 m compared to 24% at the Honeygrove site. Topography likely plays a role in explaining these differences: the Jory site is located along a ridge top in the hills south of the Willamette Valley, while the Honeygrove site is on a toe slope in the Oregon Coast Range. Accumulation of N from soil deposition and downslope water transport is likely at the Honeygrove site, whereas erosion and downslope groundwater movement carries N away from the Jory site. Precipitation also differs between sites (1143 mm at the Jory site; 1905 mm at the Honeygrove site) leading to differential leaching rates of N to deep soil layers.

On average, Inceptisols in this study have only 53% of the total soil N observed in Andisols. The relatively young nature of soil development that is typical of Inceptisols could explain much of this difference. The Olete series, for example, is a well drained, very gravelly silt loam; the site carries the least soil N of all 22 soils in this study. 29% of total N was below 1.0 m, but N levels were low throughout this soil profile, suggesting that the large proportion of N at depth is due to relatively low surface soil N. These soils are typically found along strongly sloping to steep uplands, leading to erosion and exaggerated drainage of N away from the site. The Ragnar series likewise carries a substantial proportion of N deeper than 1.0 m (38%), though total N was nearly as low as in the Olete soil. Ragnar soils are young (~14,000 years) sandy loams formed on glacial outwash. Coarse texture distinguishes both the Olete and Ragnar series from other soils in this study. The formation of clays and secondary minerals associated with soil development plays a key role in adsorbing and storing DON (such as amino acids) and inorganic forms of N (NO_3^- and NH_4^+). Consequently, soils with relatively little development and coarse texture lack the mineralogical storage mechanisms necessary to hold on to N.

8. CONCLUSIONS

1. Rocky forest soils of the Pacific Northwest carry substantial soil N below 0.5 m. When sampling depths were increased to 1.0 m and 2.5 m, average total soil N increased by 1700 and 4250 kg ha^{-1} respectively.

2. A nonlinear mixed effects model using the Langmuir equation can predict total soil N to 2.5 m on a soil specific basis with 12.4% mean error given sampling to 1.0 m and 7.6% mean error given sampling to 1.5 m. Reasonable predictions were made for soils from Inceptisol, Ultisol, Alfisol, and Andisol orders and across glacial, sedimentary, and volcanic parent materials. A log-log curve form better modeled deep volcanic soils with noncrystalline minerals throughout the profile.

3. Soils of the Andisol order contained more soil N relative to Inceptisols, Alfisols, and (to lesser extent) Ultisols, as well as a greater proportion of N in deep layers, due to the development of variable charge on mineral surfaces that can store both organic and inorganic forms of N.

8.1 SOIL SAMPLING CONSIDERATIONS

Soil sampling protocols in the coastal Pacific Northwest need to take into account the vertical distribution of N in soil. Shallow soil sampling (< 1.0 m depth) will underestimate soil N stocks, which could bias both the magnitude and direction of N flux. Long-term monitoring and landscape scale research efforts that must establish systematic sampling depths – and that typically emphasize a greater number of shallow samples – should consider adding a subset of deep soil samples that target particular deep soils where substantial stores of N are likely to be found. While sampling deep soil is more expensive and time consuming, it will provide substantially better estimates of soil N that is biologically accessible and ecologically important.

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Table 1. Soil series, taxonomic class, texture, range in percentage coarse fragments over all horizons, and parent material for 22 forest soils in western Washington and Oregon. Data from Soil Survey Staff (2013).

Soil Series	Taxonomic Class	Texture	Coarse Fraction (%)	Parent material
Astoria (A)	Andic Humudept	Silt loam	6-88	Colluvium and residuum derived from sandstone and siltstone
Barneston 1 (Bn1)	Typic Vitrixerand	Gravelly coarse sandy loam	12-81	Volcanic ash and glacial outwash
Barneston 2 (Bn2)	Typic Vitrixerand	Gravelly loam	25-59	Volcanic ash and loess over glacial outwash
Blachly (Bl)	Humic Dystrudept	Silty clay loam	31-70	Residuum and colluvium derived from basic igneous and sedimentary rock
Cloquallum (C)	Aquandic Dystrudept	Silt loam	15-44	Silty lacustrine deposits with volcanic ash
Hoko (Hk)	Aquic Durudand	Gravelly silt loam	12-53	Alpine Basal till with volcanic ash
Honeygrove (Hg)	Typic Palehumult	Gravelly clay loam	0-20	Colluvium and residuum derived from sandstone, siltstone, and volcanic rock
Hoquiam (Ho)	Typic Fulvudand	Silt loam	0-50	Old alluvium over glacial drift
Jory (J)	Xeric Palehumult	Silty clay loam	0-4	Colluvium and residuum derived from basalt
Kinney (K)	Andic Humudept	Silt loam	49-80	Residuum weathered from igneous rock with a mantle of volcanic ash
Lemolo (L)	Typic Humaquept	Silt loam	7-65	Oceola mudflow deposits
Melbourne (M)	Ultic Palexeralf	Loam	0	Residuum from siltstone
O'Brien (Ob)	Typic Fulvudand	Silt loam	6-58	Glacial outwash
Olete (Ol)	Vitrandic Haploxerept	Very gravelly silt loam	16-79	Colluvium and residuum from basalt
Ovall (Ov)	Andic Humixerept	Gravelly loam	0-28	Glacial drift over residuum with volcanic ash near surface
Ragnar (R)	Vitrandic Dystroxerept	Fine sandy loam	0-16	Glacial outwash
Scamman (Sm)	Aquic Palexeralf	Silt loam	0-49	Mixed glacial and sedimentary materials
Shelton (Sh)	Typic Haploxerand	Gravelly sandy loam	36-76	Basal till with volcanic ash
Tokul (Tk)	Aquic Vitrizerand	Gravelly loam	19-77	Volcanic ash and loess over glacial drift
Tolovana (Tv)	Typic Fulvudand	Silt loam	0-74	Colluvium and residuum derived from tuffaceous sedimentary rock
Vailton (V)	Alic Hapludand	Silt loam	0-33	Residuum and colluvium from siltstone and shale mixed with volcanic ash
Windygap (W)	Xeric Haplohumult	Silt loam	0-7	Colluvium and residuum derived sedimentary rock

Table 2. Prediction error (Mg C ha^{-1}) for four functions used to extrapolate total soil C content to 2.5 m based on samples collected between 0 to 1.0 m for 22 forest soils in western Washington and Oregon.

		Function			
Soil series	**Subgroup**	**Inv. poly.**	**Lang-muir**	**Log.**	**Exp.**
		Prediction error (Mg C ha^{-1})			
Astoria (A)	Andic Humudept	-18.7	-9.3	6.7	-28.4
Barneston 1	Typic Vitrixerand	-16.7	-6.1	-1.2	-20.8
Barneston 2	Typic Vitrixerand	-1.6	-15.6	-9.8	-18.8
Blachly (Bl)	Humic Dystrudept	-6.8	-5.9	17.1	-12.9
Cloquallum (C)	Aquandic Dystrudept	2.6	-4.4	15.6	-5.2
Hoko (Hk)	Aquic Durudand	-66.1	-50.1	-62.4	-115.4
Honeygrove (Hg)	Typic Palehumult	-16.0	-14.6	1.1	-33.8
Hoquiam (Ho)	Typic Fulvudand	1.2	-5.3	26.2	-7.2
Jory (J)	Xeric Palehumult	-4.0	-4.6	10.7	-11.6
Kinney (K)	Andic Humudept	9.3	-2.0	27.4	5.7
Lemolo (L)	Typic Humaquept	-2.3	-14.5	-6.5	-28.8
Melbourne (M)	Ultic Palexeralf	-11.4	-2.9	2.0	-13.4
O'Brien (Ob)	Typic Fulvudand	-4.6	-7.9	23.6	-17.4
Olete (Ol)	Vitrandic Haploxerept	3.8	-3.8	21.8	-1.7
Ovall (Ov)	Andic Humixerept	21.0	-8.5	49.3	0.6
Ragnar (R)	Vitrandic Dystroxerept	-14.1	-5.2	0.8	-17.9
Scamman (Sm)	Aquic Palexeralf	0.3	-3.0	10.8	-3.9
Shelton (Sh)	Typic Haploxerand	-1.6	-3.1	14.3	-5.5
Tokul (Tk)	Aquic Vitrizerand	-0.4	-7.2	21.0	-11.3
Tolovana (Tv)	Typic Fulvudand	3.6	-7.5	31.4	-8.1
Vailton (V)	Alic Hapludand	-106.2	-56.8	-89.2	-128.8
Windygap (W)	Xeric Haplohumult	-11.7	-5.2	3.9	-16.7
	Mean error (Mg C ha^{-}	-10.9	-11.1	5.2	-22.8
	Mean error (Mg C ha^{-}	-3.4	-6.8	13.3	-12.8
	Mean error (%)	-5.6	-7.0	8.0	-13.9
	Mean error (%)*	-3.1	-5.8	11.5	-11.0

* Mean calculated without Hoko and Vailton series, the two series not well predicted by the models.

Table 3. Prediction error (kg N ha^{-1}) for four functions used to extrapolate total soil N content to 2.5 m based on samples collected between 0 to 1.0 m and 0 to 1.5 m for 22 forest soils in western Washington and Oregon. Mean error was calculated as absolute values of prediction errors.

		Extrapolation from 0 to 1.0 m				Extrapolation from 0 to 1.5 m			
Soil Series	**Subgroup**	**Log-Log**	**Lang-muir**	**Log**	**Exp.**	**Log-Log**	**Lang-muir**	**Log**	**Exp.**
		Prediction Error (kg N ha^{-1})				*Prediction Error (kg N ha^{-1})*			
Astoria (A)	Andic Humudept	6313	1346	1688	-633	3368	1036	1545	181
Barneston 1 (Bn1)	Typic Vitrixerand	1275	7	-5	-1025	508	-464	-112	-905
Barneston 2 (Bn2)	Typic Vitrixerand	-2881	-5680	-5596	-6882	-1562	-1051	-4348	-5394
Blachly (Bl)	Humic Dystrudept	523	-1609	-1514	-2888	-127	-1626	-1320	-2355
Cloquallum (C)	Aquandic Dystrudept	1607	404	383	-515	915	6	269	-436
Hoko (Hk)	Aquic Durudand	49	-10555	-9925	-12762	217	-3211	-7479	-7568
Honeygrove (Hg)	Typic Palehumult	4231	-807	-447	-2644	2749	297	83	-1203
Hoquiam (Ho)	Typic Fulvudand	2508	526	634	-875	1322	-83	534	-566
Jory (J)	Xeric Palehumult	4227	1148	1339	-496	2412	624	1210	-31
Kinney (K)	Andic Humudept	2719	1531	1515	508	2308	1011	1471	454
Lemolo (L)	Typic Humaquept	-745	-1749	-1820	-2550	-813	-1677	-1581	-2255
Melbourne (M)	Ultic Palexeralf	3000	640	748	-881	1270	-141	467	-675
O'Brien (Ob)	Typic Fulvudand	4302	-527	-149	-2604	2121	-468	53	-1341
Olete (Ol)	Vitrandic Haploxerept	634	74	-88	-446	201	-305	-238	-543
Ovall (Ov)	Andic Humixerept	3907	299	530	-1313	1798	-89	361	-848
Ragnar (R)	Vitrandic Dystroxerept	195	-369	-534	-865	-143	-671	-605	-915
Scamman (Sm)	Aquic Palexeralf	67	-579	-703	-1149	-166	-785	-667	-1104
Shelton (Sh)	Typic Haploxerand	1093	-663	-620	-1826	272	-965	-629	-1546
Tokul (Tk)	Aquic Vitrizerand	1819	-199	-113	-1430	627	-682	-270	-1223
Tolovana (Tv)	Typic Fulvudand	3812	200	475	-1462	1501	-467	202	-1052
Vailton (V)	Alic Hapludand	-2124	-5078	-4929	-6406	-1951	-2916	-4159	-5255
Windygap (W)	Xeric Haplohumult	1798	-1849	-1604	-3542	178	-1873	-1489	-2764
	Mean Error (kg ha^{-1})	1742	-1068	-943	-2395	773	-659	-759	-1698
	Mean Error (%)	20.2	-12.4	-10.9	-27.7	8.9	-7.6	-8.8	-19.6
	Absolute Mean Error (kg ha^{-1})	2265	1629	1607	2441	1206	930	1322	1755

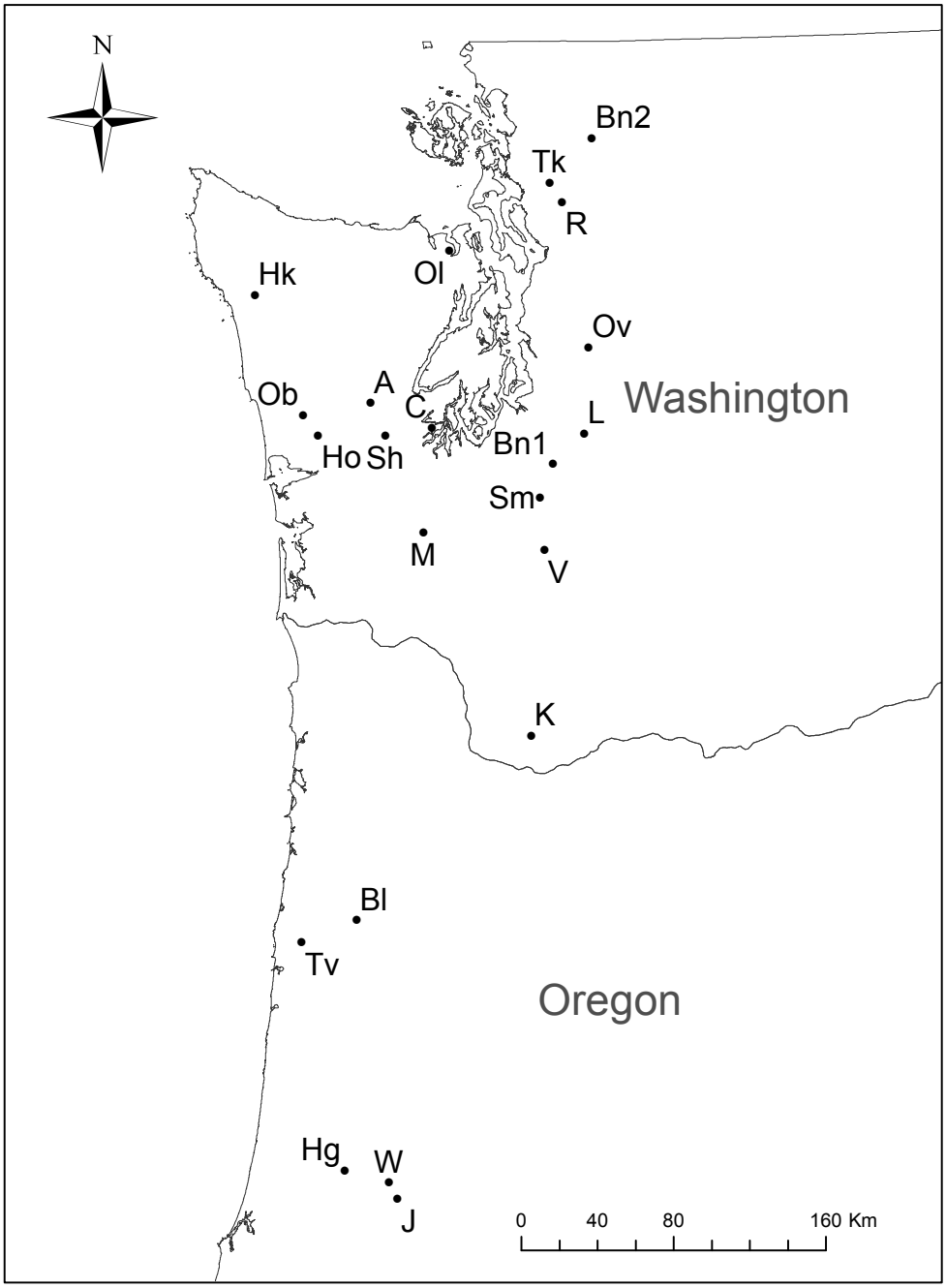

Figure 1. Locations of 22 soil series sampled in the Pacific Northwest Douglas-fir Zone of Washington and Oregon. One soil profile was excavated at each site. Map units for each series are listed in Table 1.

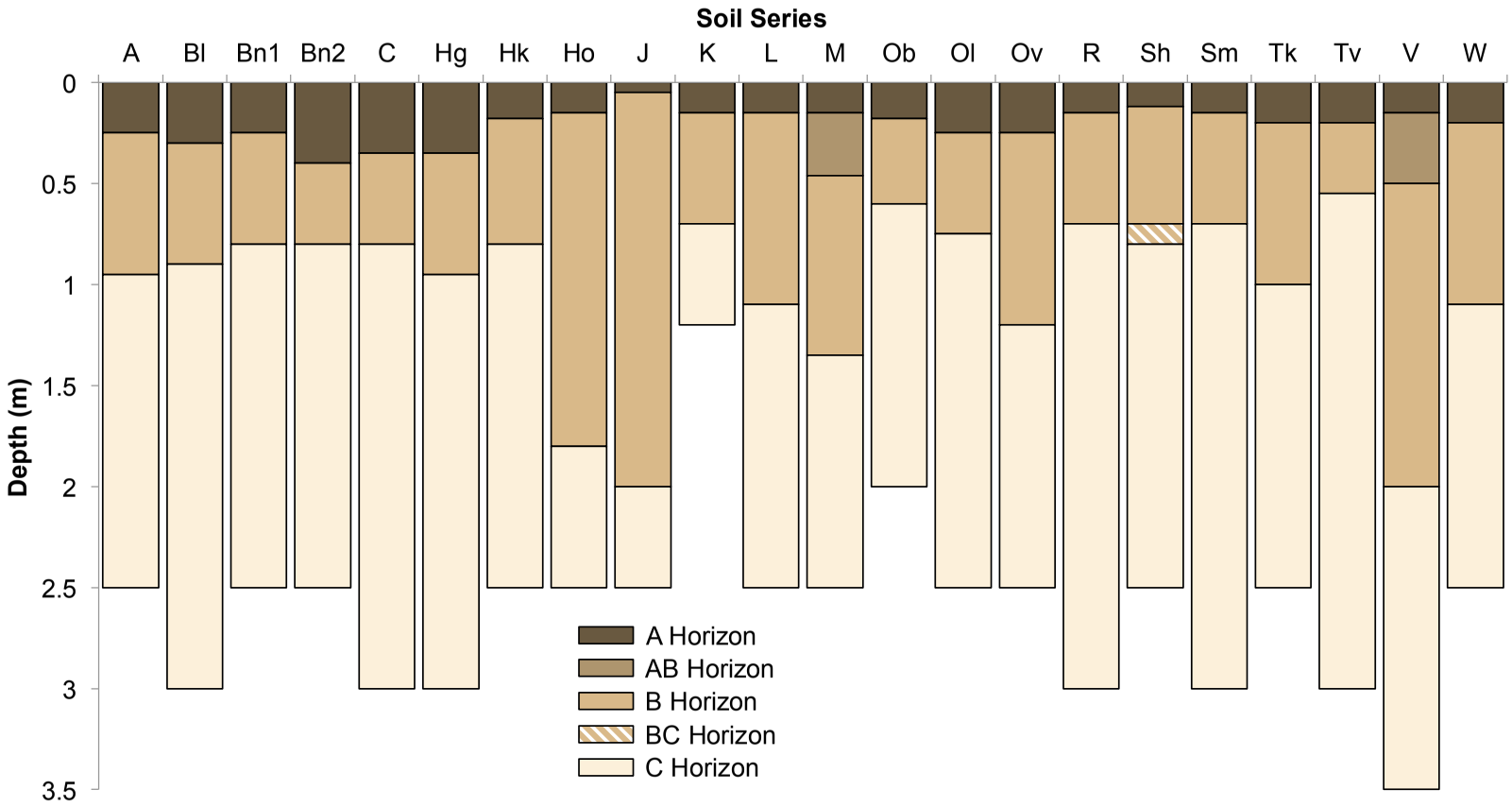

Figure 2. Major genetic horizons and profile depth in 22 soil series in Washington and Oregon. Profiles were sampled to at least 2.5 m except where bedrock or compacted material impeded further excavation. See Table 1 for key to forest soil series indicated on x-axis.

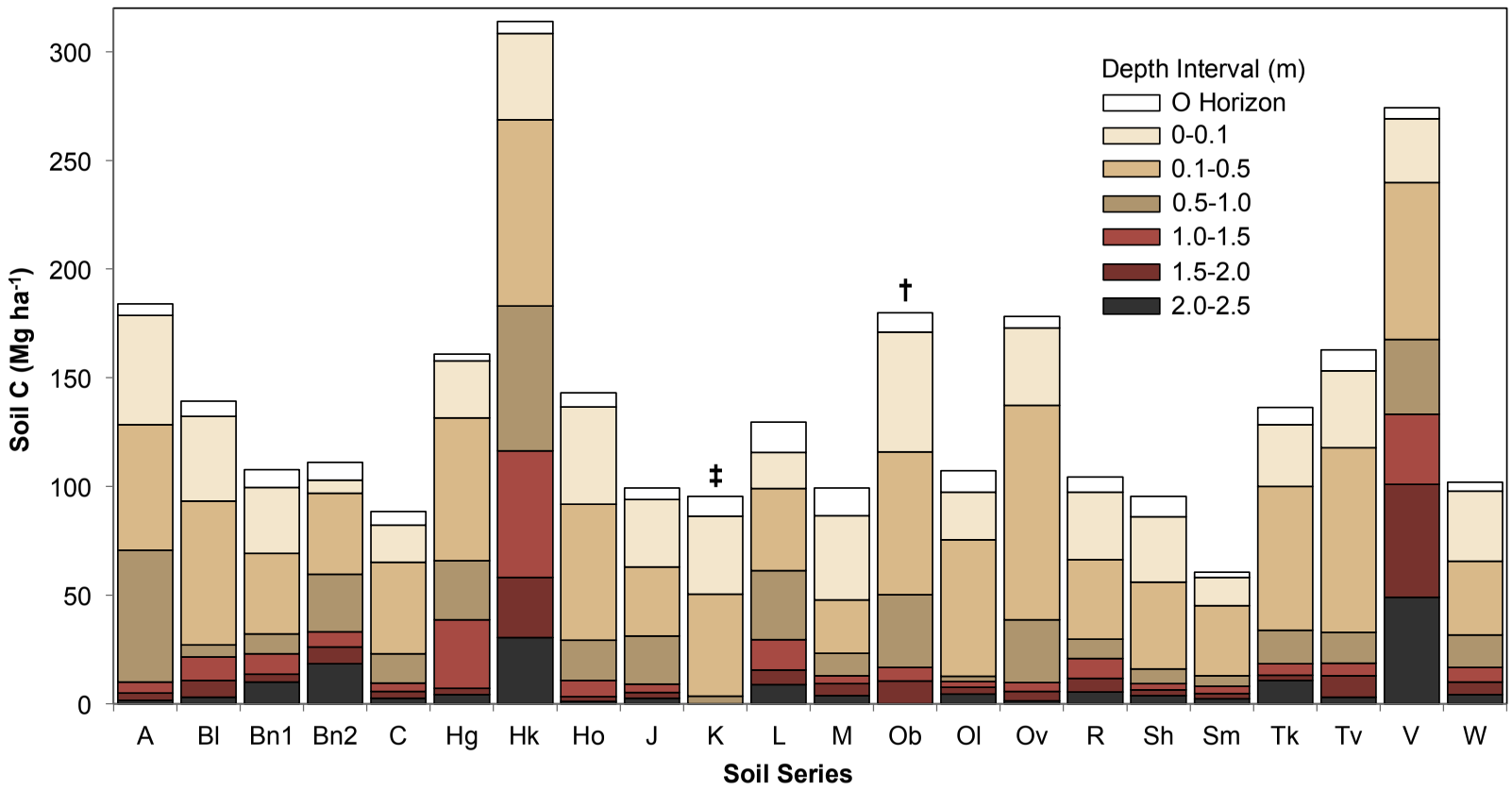

Figure 3. Soil C (Mg ha^{-1}) within each sampled depth interval for 22 forest soils in western Washington and Oregon. † Soil pit excavation impeded by compacted glacial till at 2.0 m. ‡ Soil pit reached igneous bedrock at 1.0 m. See Table 1 for key to forest soil series indicated on x-axis.

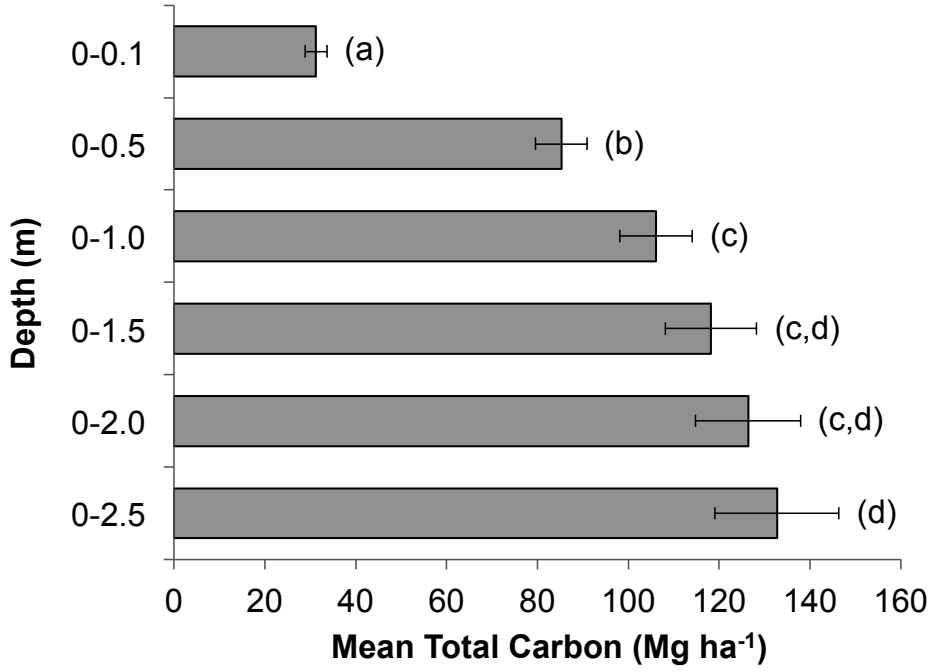

Figure 4. Mean total carbon at increasing depth intervals. Error bars represent ±1 SE. Means accompanied by the same letter are not significantly different (Tukey's HSD, $P < .01$).

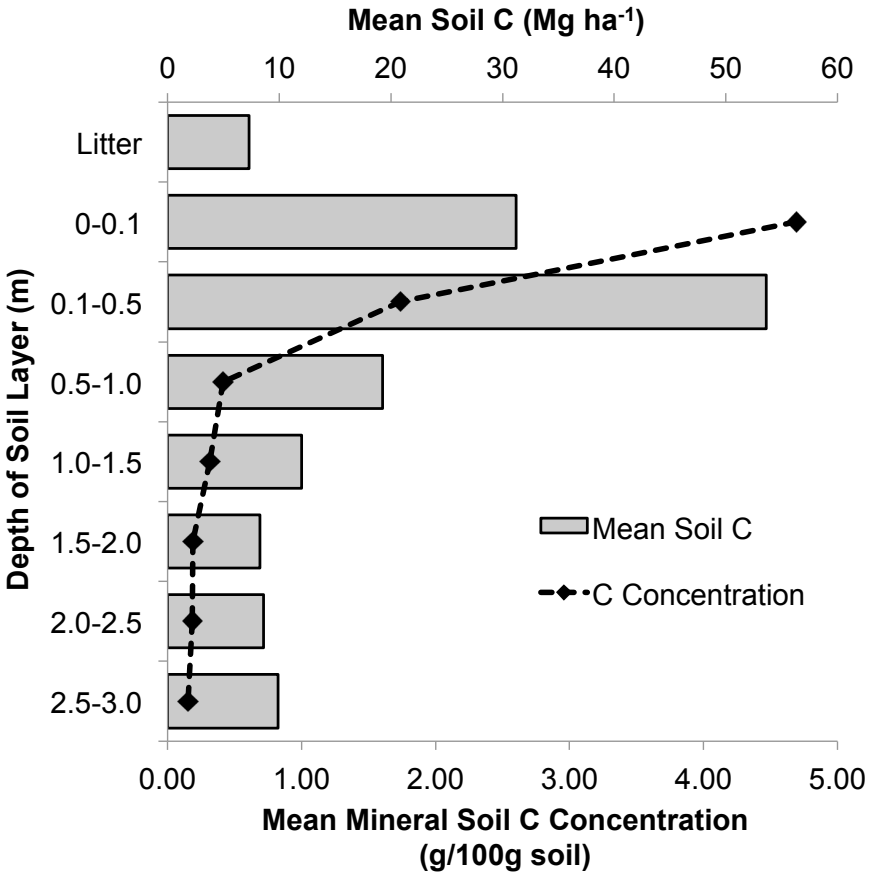

Figure 5. Mean soil C and mineral soil C concentration in succeeding soil layers from 22 soil profiles in western Washington and Oregon.

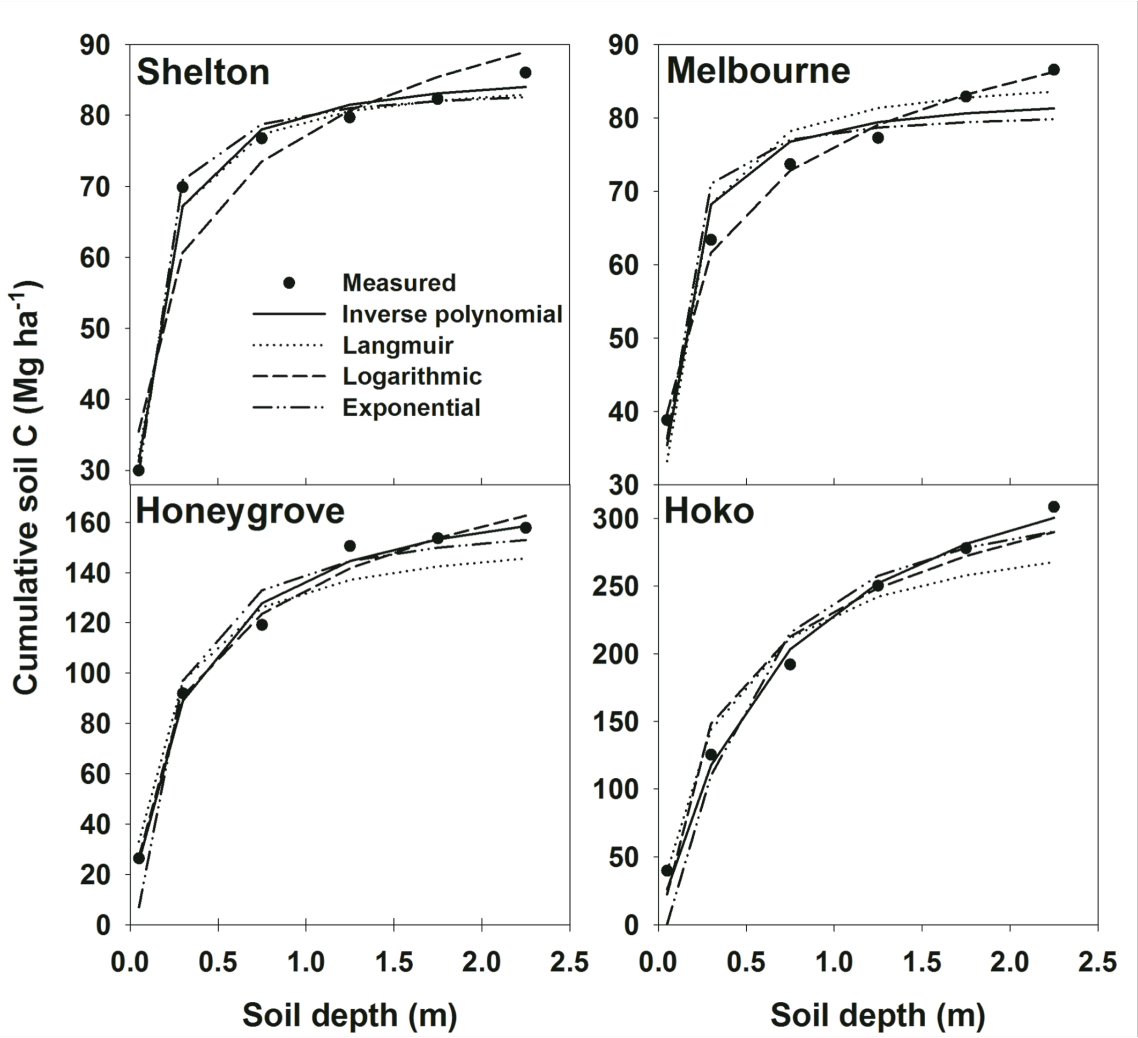

Figure 6. Cumulative soil C profiles (sample interval midpoints are plotted) for four soil series, representative of the range of profiles from 22 forest soils in western Washington and Oregon. Four functions are fit to the data for each soil series.

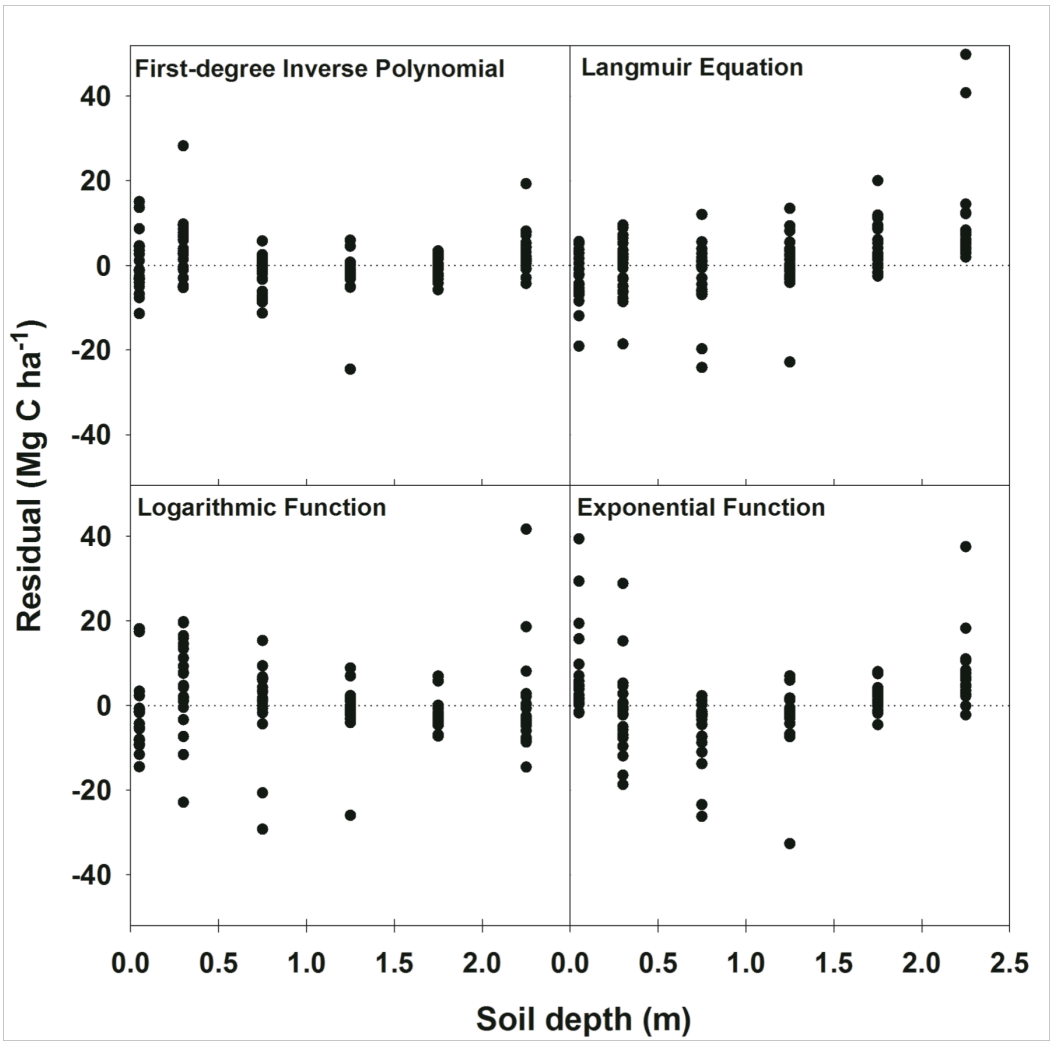

Figure 7. Residuals produced by fitting four different functions to 22 cumulative soil C profiles (0-2.5-m depth) of forest soils in western Washington and Oregon.

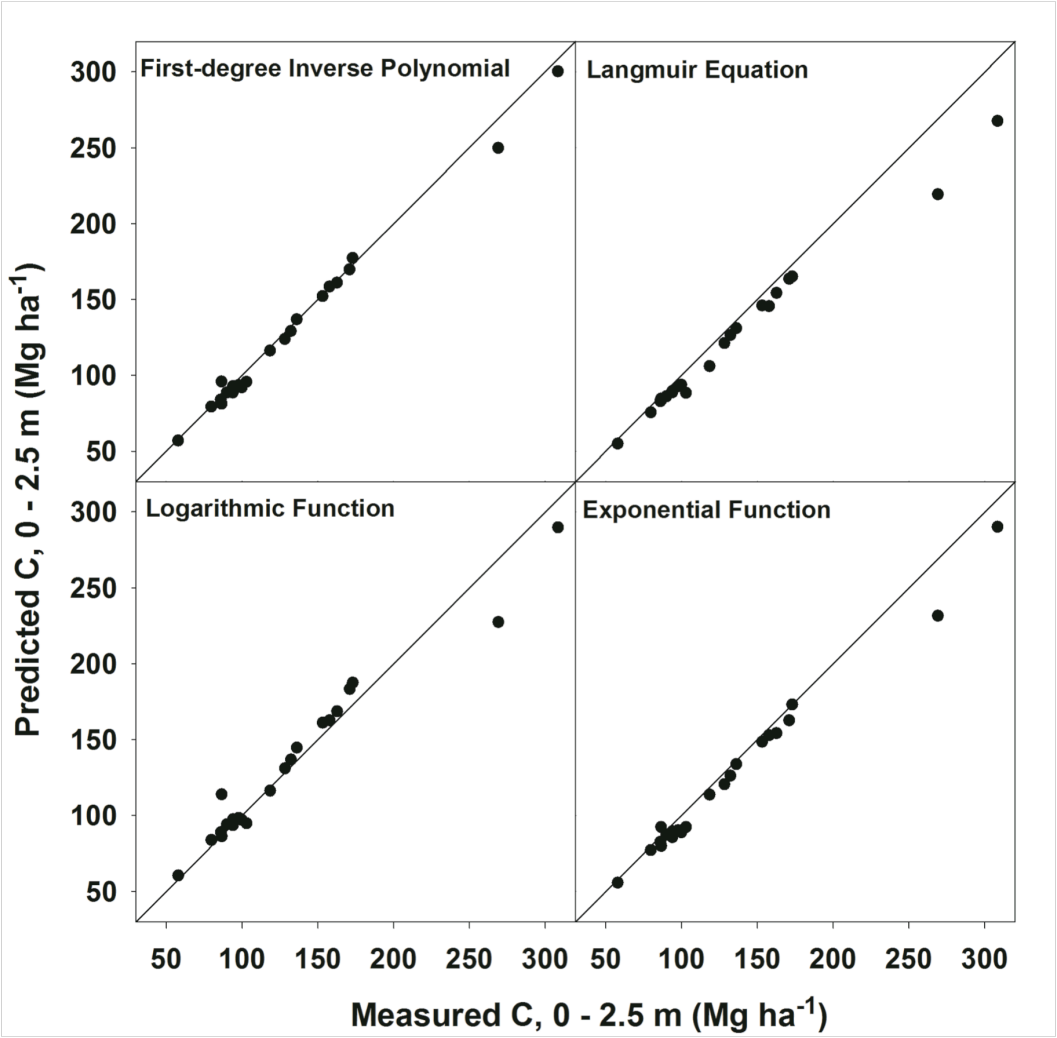

Figure 8. Predicted versus measured soil C (Mg ha^{-1} from 0-2.5 m) for 22 forest soils in western Washington and Oregon, based on four functions fit to cumulative soil C profiles (0-2.5-m depth).

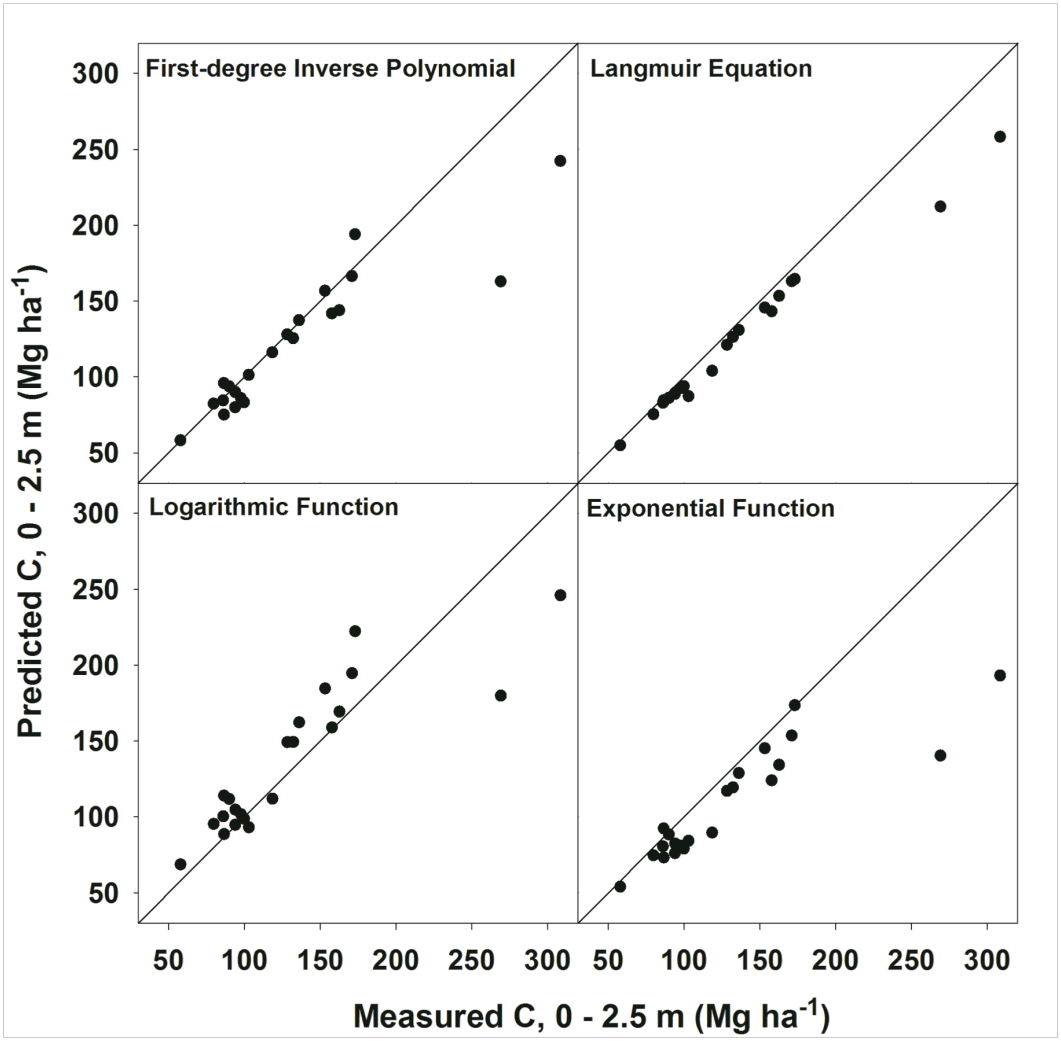

Figure 9. Predicted versus measured soil C (Mg ha^{-1} from 0-2.5 m) for 22 forest soils in western Washington and Oregon, based on four functions fit to cumulative soil C profiles to a 1.0-m depth that were subsequently used to predict C to a 2.5-m depth.

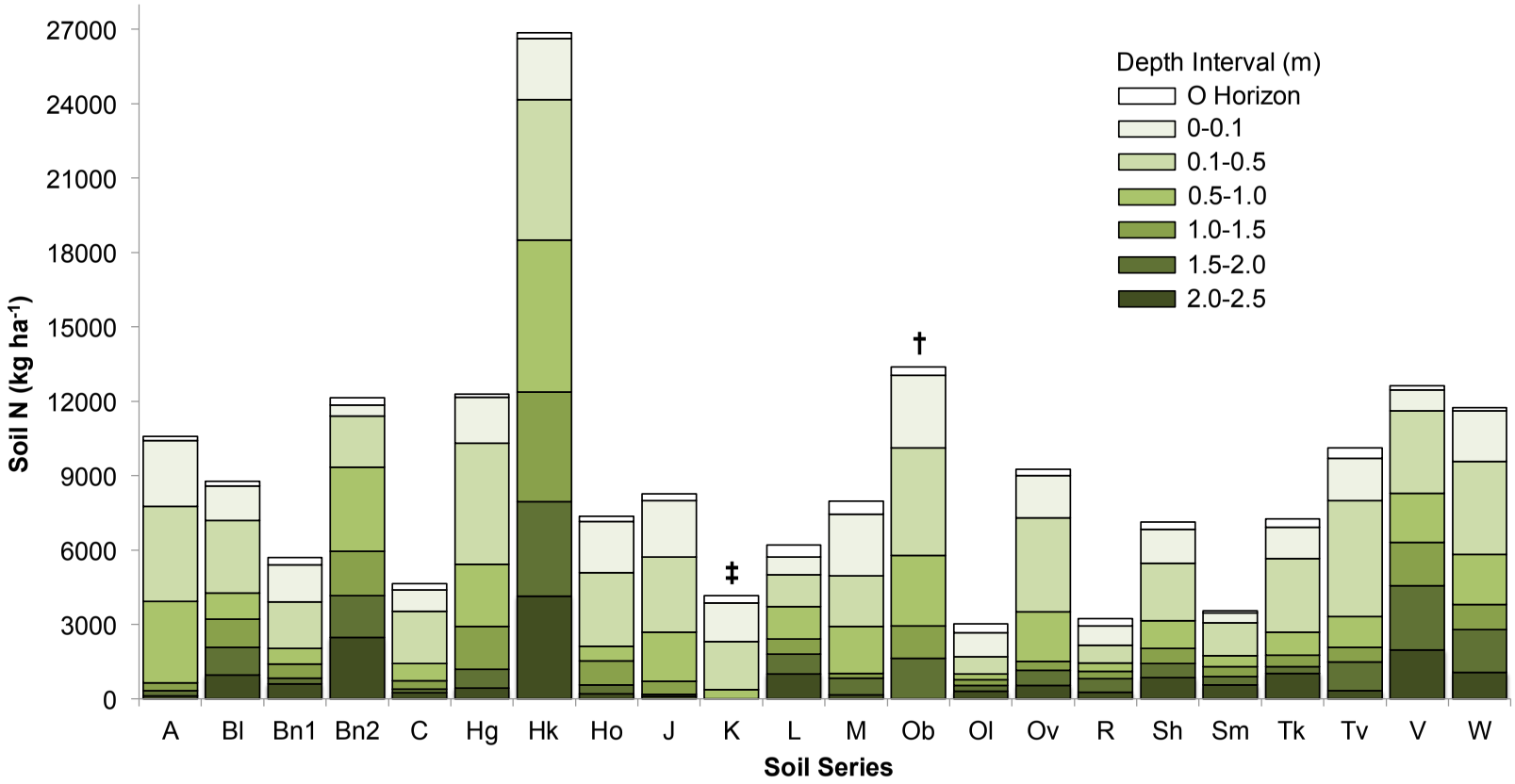

Figure 10. Soil N (kg ha^{-1}) within each sampled depth interval for 22 forest soils in western Washington and Oregon. † Soil pit excavation impeded by compacted glacial till at 2.0 m. ‡ Soil pit reached igneous bedrock at 1.0 m. See Table 1 for key to forest soil series indicated on x-axis.

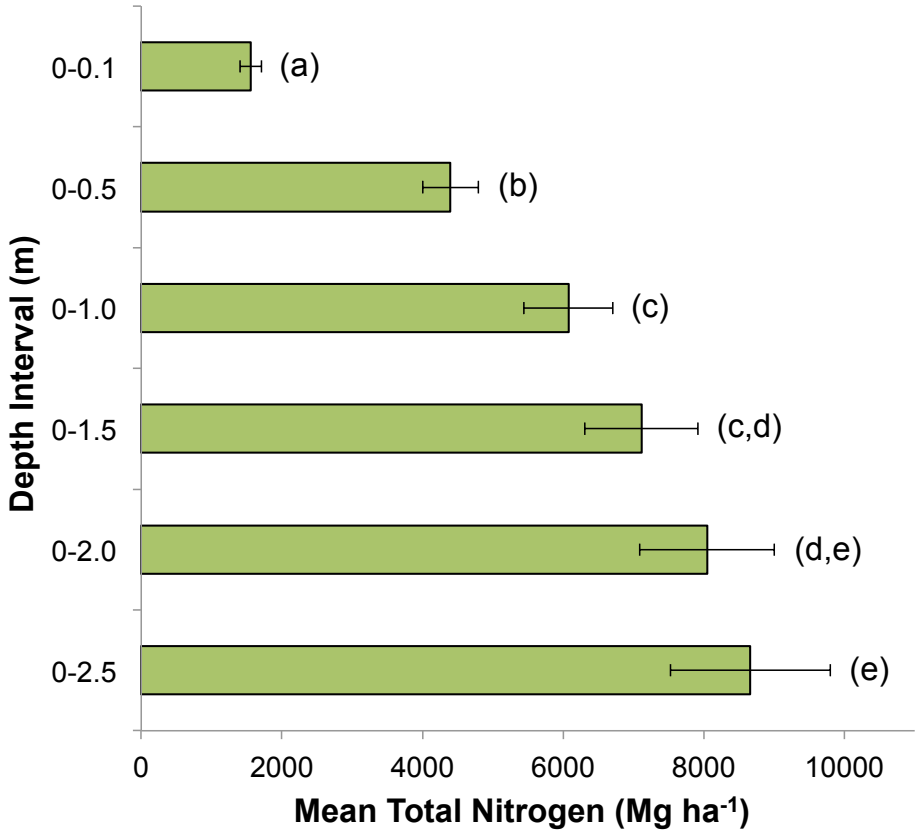

Figure 11. Mean total N at intervals of increasing depth. Error bars represent ±1 SE. Means accompanied by the same letter are not significantly different (Tukey's Honestly Significant Difference, $P < 0.01$).

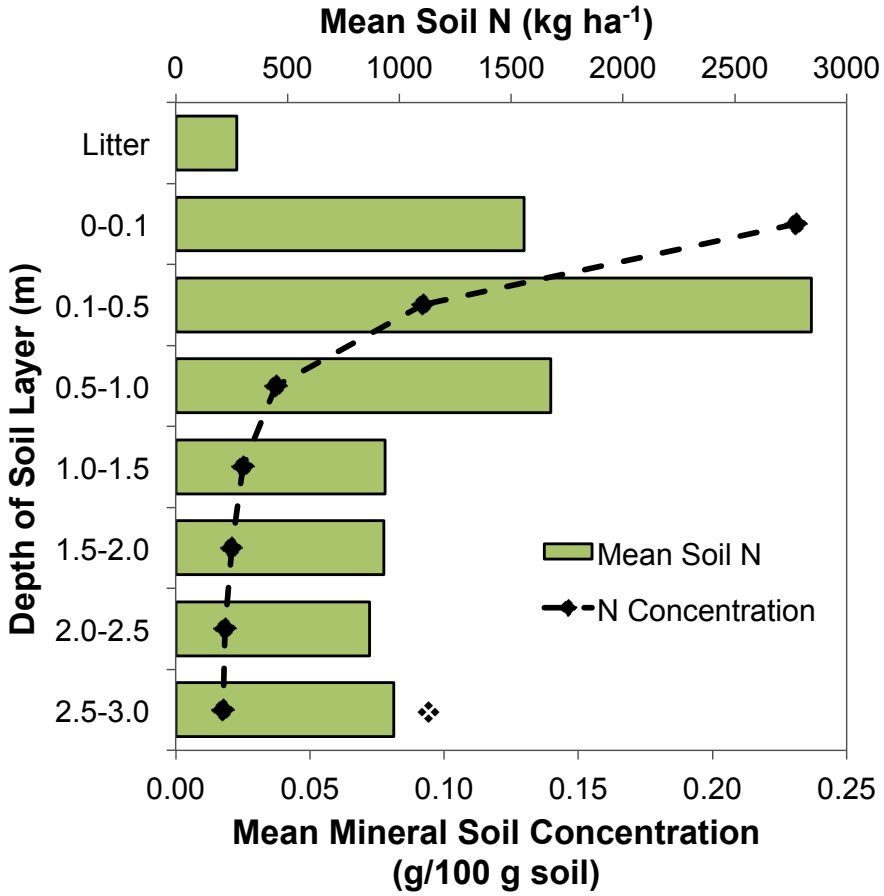

Figure 12. Mean soil N and mineral soil N concentration in succeeding soil layers from 22 soil profiles in western Washington and Oregon.

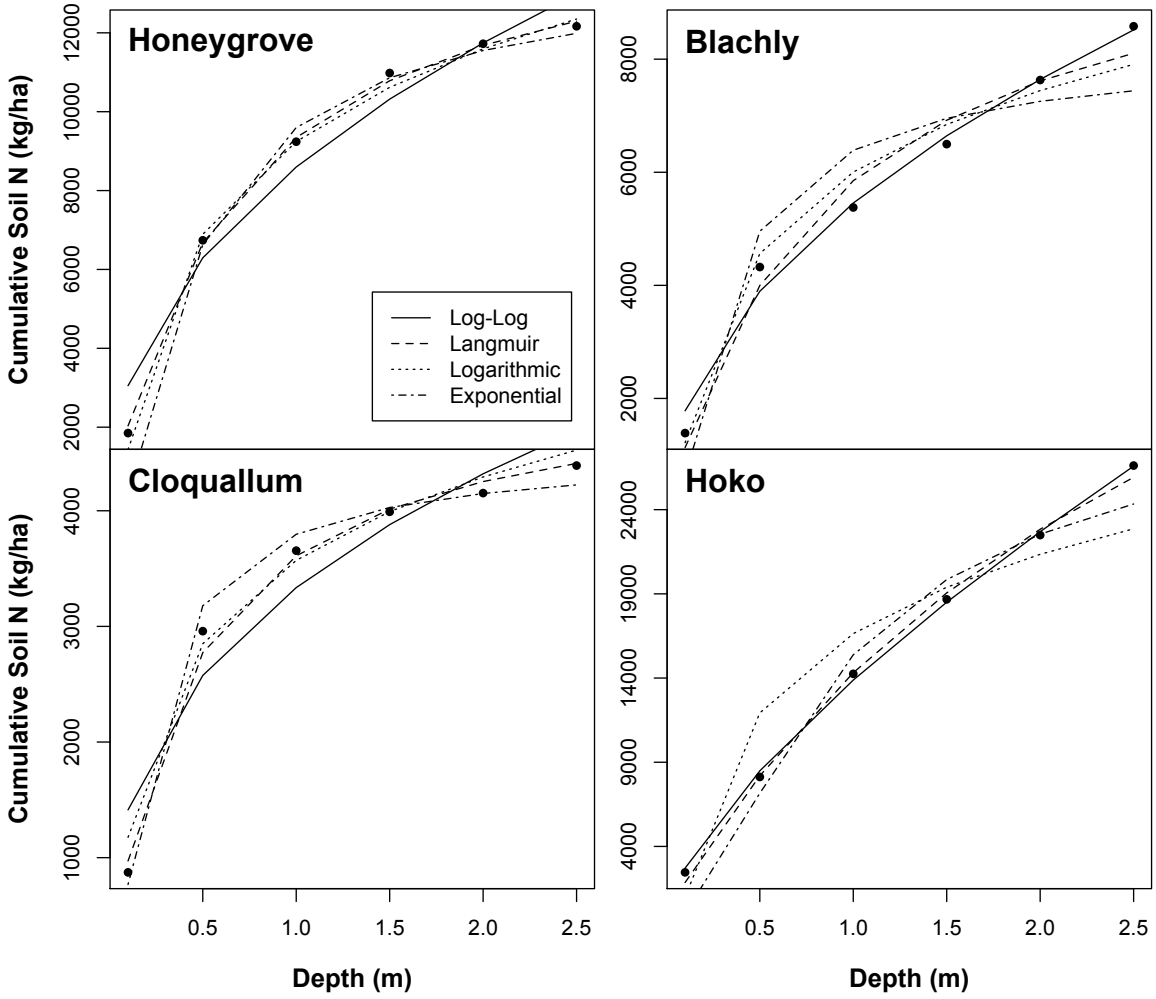

Figure 13. Cumulative soil N profiles for four soil series representative of the range of profiles from 22 forest soils in western Washington and Oregon. Four functions are fit to the data for each soil series.

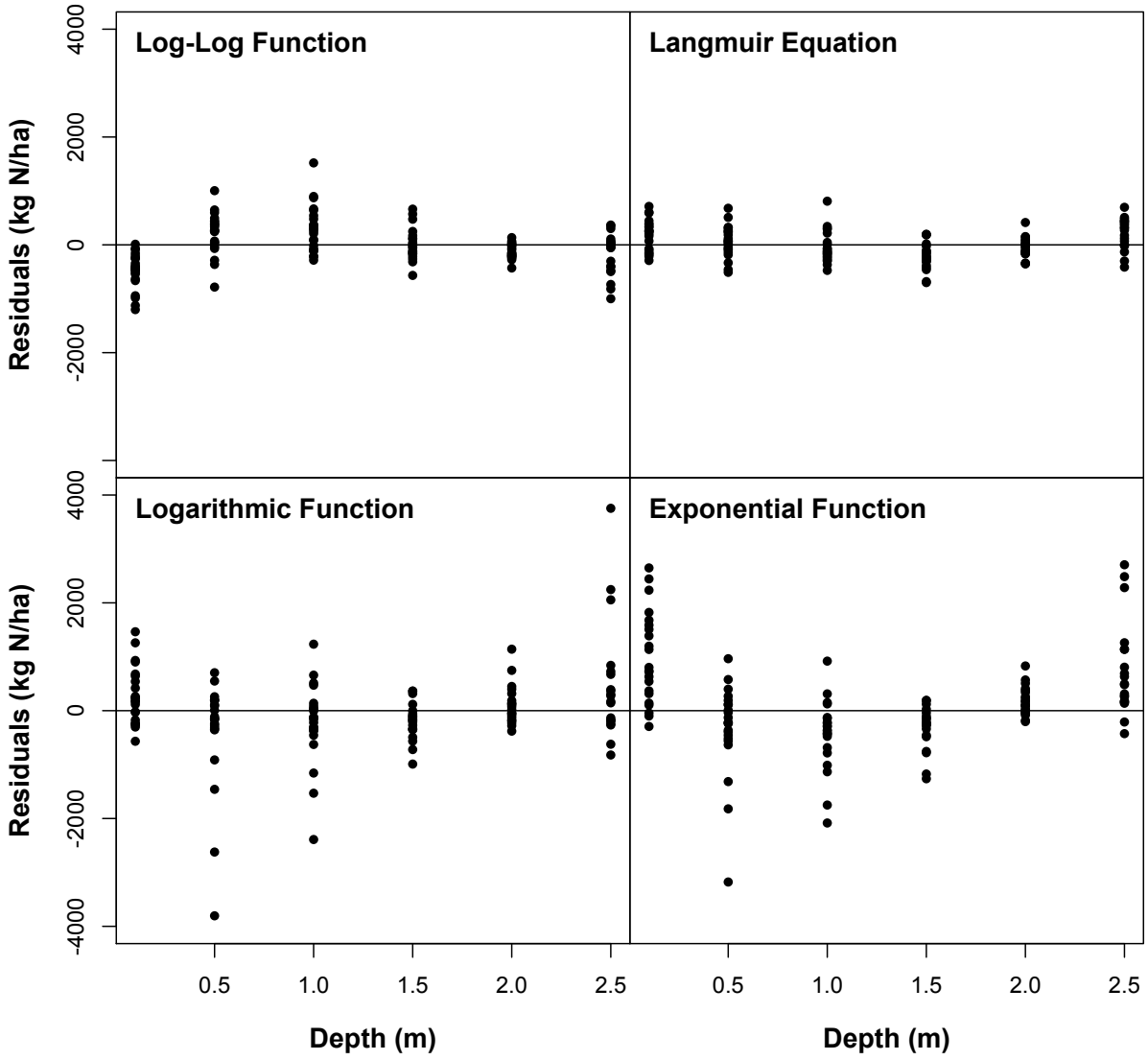

Figure 14. Residuals produced by fitting four different functions to 22 cumulative soil N profiles (0- to 2.5-m depth) of forest soils in western Washington and Oregon.

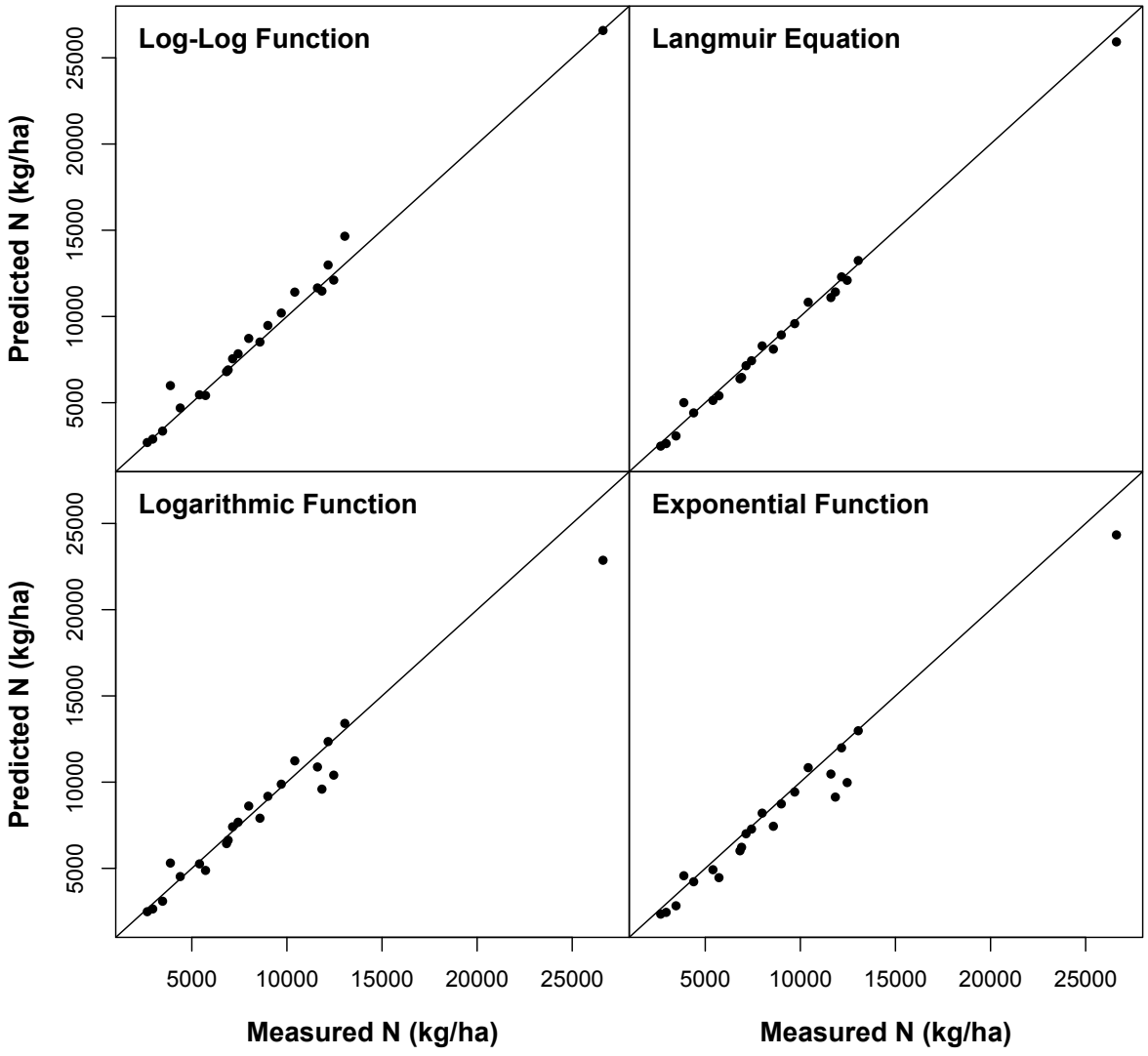

Figure 15. Predicted versus measured soil N (kg ha^{-1}, from 0- to 2.5-m) for 22 forest soils in western Washington and Oregon, based upon four functions fit to cumulative soil N profiles from 0 to 2.5 m depth.

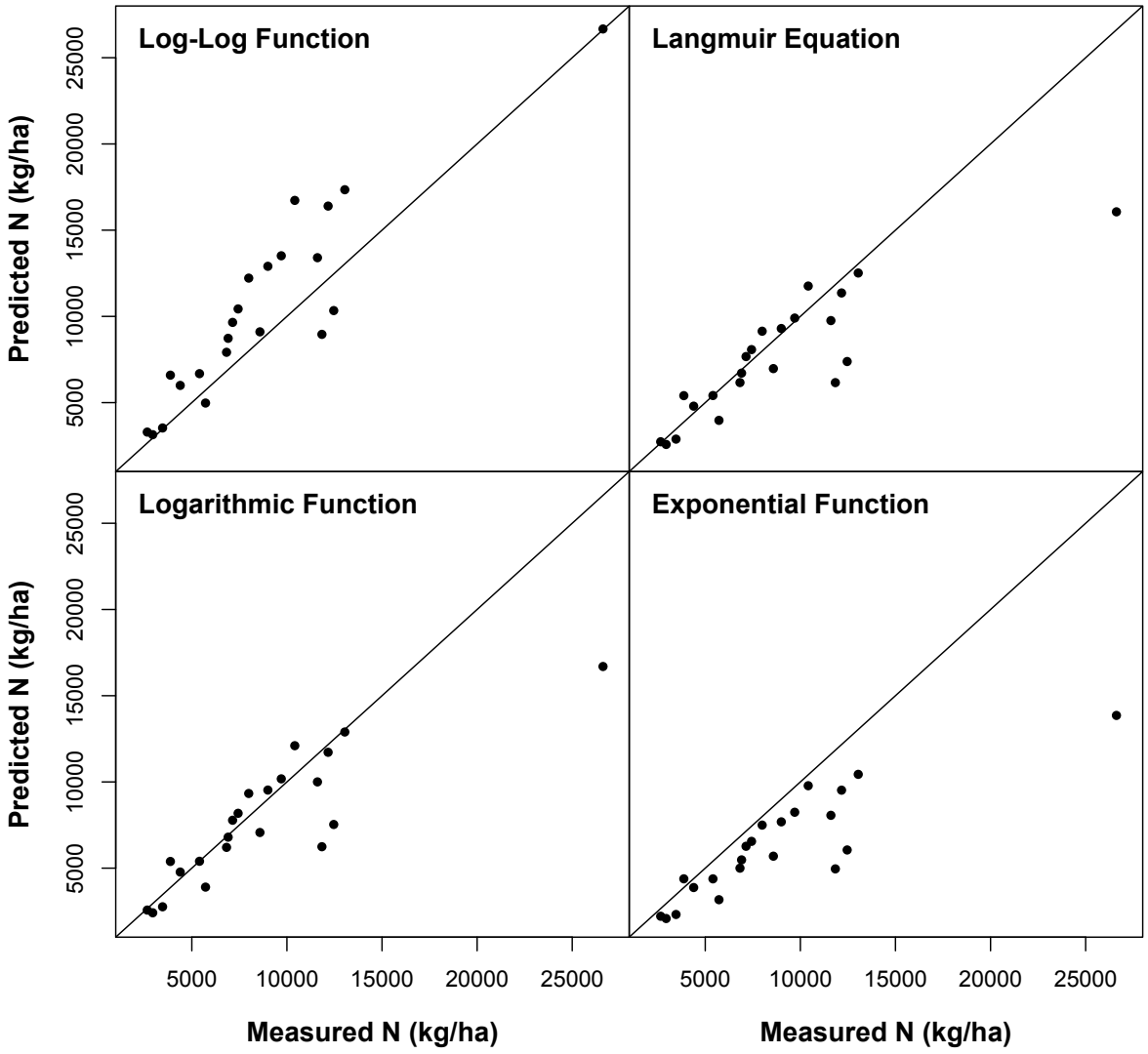

Figure 16. Predicted versus measured soil N (kg ha^{-1}, from 0- to 2.5-m) for 22 forest soils in western Washington and Oregon, based upon four functions fit to cumulative soil N profiles truncated at 1.0 m depth that were subsequently used to predict N to 2.5 m depth.

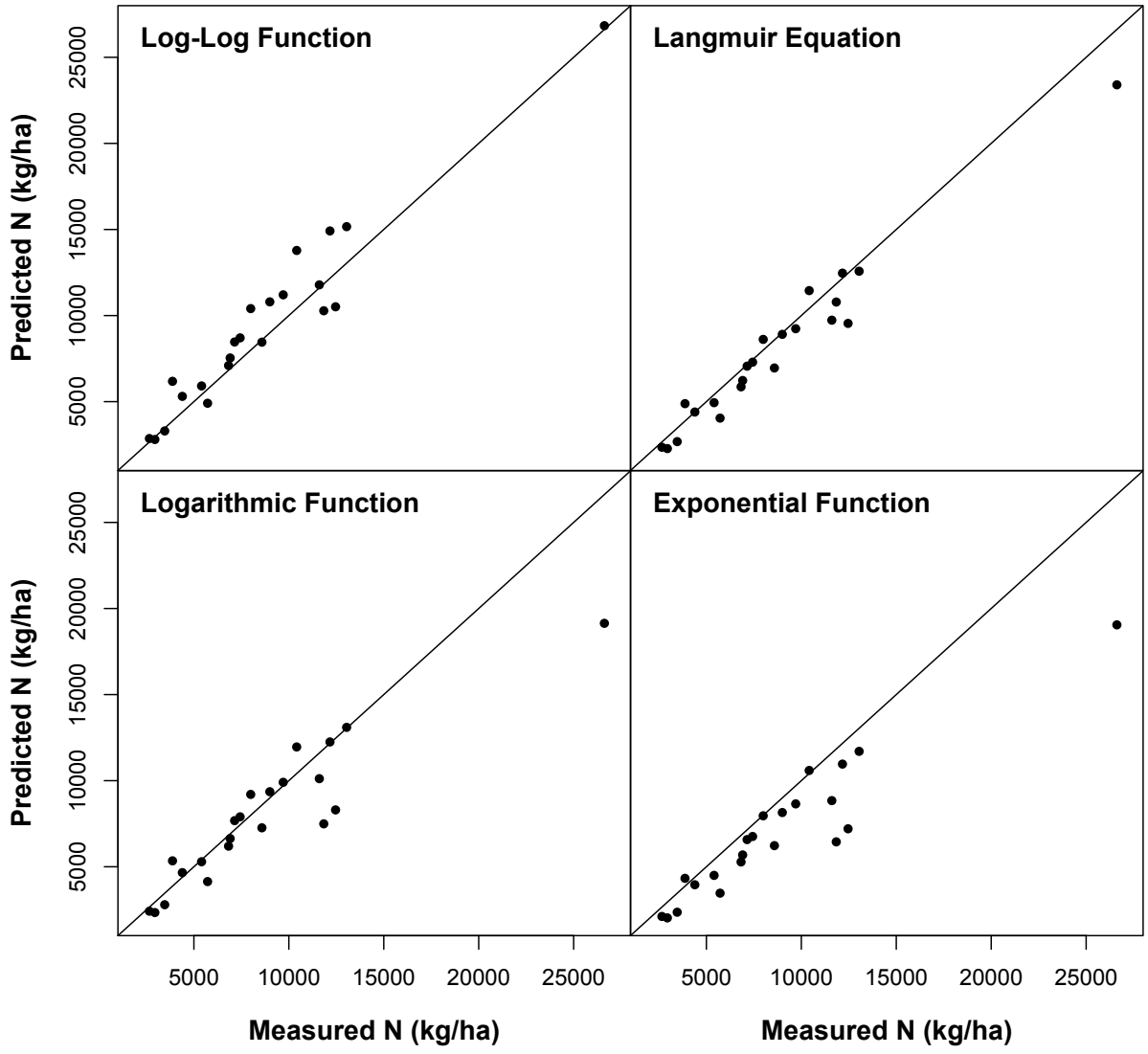

Figure 17. Predicted versus measured soil N (kg ha^{-1}, from 0- to 2.5-m) for 22 forest soils in western Washington and Oregon, based upon four functions fit to cumulative soil N profiles truncated at 1.5 m depth that were subsequently used to predict N to 2.5 m depth.