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Coarse root biomass and architecture of hybrid aspen 'Crandon' (*Populus alba* L. × *P. grandidenta* Michx.) grown in an agroforestry system in central Iowa, USA

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ABSTRACT

In this study, we evaluated 'Crandon' coarse root biomass and architecture grown at different topographic positions and fertilizer rates. Complete excavations were conducted on a subset of trees after the first growing season and showed that root biomass was strongly related to stem biomass (R^2 = 0.93), but not topographic position or fertilizer rate. After the third growing season, subsamples of roots were collected from another subset of trees and showed coarse root architecture variables to be strongly related to several metrics of the tree and root size (R^2 = 0.61 to 0.82), while also differing by topographic position. Equations relating root biomass to stem biomass were derived from both methodologies (complete excavation v. subsampling for architecture measurements), and comparison of the equations indicated no difference in slopes (p = 0.59) or intercepts (p = 0.90), although the subsampling approach had a weaker model fit. Our results suggest 'Crandon' roots (i) adhere to strong allometric relationships with stem biomass, (ii) alter their architecture within the constraints of this allometric relationship according to site conditions, and (iii) can be subsampled to estimate root biomass from root architecture parameters with similar accuracy (but less precision) compared to complete excavations.

KEYWORDS
Biomass allocation; fertilizer; pipe theory; root geometry; topography

Introduction

The importance of adventitious rooting within the genus *Populus* has been thoroughly described (Friend, Scarascia-Mugnozza, Isebrands, & Heilman, 1991; Haissig & Davis, 1994; Heilman, Ekuan, & Fogle, 1994; Pregitzer & Friend, 1996), especially with regard to the influence of rooting on the commercial deployment of unrooted and rooted planting stock (Zalesny, Jr., Riemenschneider, & Hall, 2005) for traditional forest products as well as providing ecosystem services (Zalesny et al., 2016). In addition, several studies have endeavored to quantify poplar root biomass production (Coleman, Friend, & Kern, 2004; Coyle & Coleman, 2005; DesRochers & Lieffers, 2001; Douglas, McIvor, & Lloyd-West, 2016) and distribution in the soil profile (Douglas, McIvor, Potter, & Foote, 2010; Fortier, Truax, Gagnon, & Lambert, 2013; McIvor, Douglas, & Benavides, 2009) with different planting

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materials and growing conditions, while fewer seem to have evaluated how growing conditions influence poplar root architecture and associated ecosystems services such as soil stabilization and erosion control (e.g., Reubens, Poesen, Danjon, Geudens, & Muys, 2007). While hybrid poplars and hybrid aspens have been primarily deployed on relatively flat or minimally sloping terrain in the Midwestern United States, interest in their use as components of agroforestry systems in the region has led to testing of their establishment potential on landscapes with higher slopes (Headlee, Hall, & Zalesny, 2013a; Thelemann et al., 2010). Thus, the importance of understanding root system development (e.g., biomass accumulation and architecture) in response to site conditions such as slope and nutrient availability is vital for increasing the success of these systems.

As a result, greater knowledge about the effects of topographic position and fertilizer on coarse root biomass and architecture is needed. Previous research has indicated that changes in woody biomass allocation for poplars (*Populus* spp.) are often primarily a function of tree size and genotype (Coleman et al., 2004; Coyle & Coleman, 2005; King, Pregitzer, & Zak, 1999; Wullschleger et al., 2005) rather than resource availability; in other words, greater resource availability simply pushes woody biomass allocation farther along a species-specific, size-based (allometric) curve. These findings may be related to the pipe theory of plant water transport (Coutts, 1987; Shinozaki, Yoda, Hozumi, & Kira, 1964), which describes the cumulative cross-sectional area of downstream components (e.g., stems) as being directly proportional to the cumulative cross-sectional area of upstream components (e.g., coarse roots).

Within the constraints of an allometric biomass relationship and the pipe theory, however, responses to resource availability may still occur through changes in root architecture parameters such as the number, diameter, and/or length of roots (Curt, Coll, Prévosto, Balandier, & Kunstler, 2005; Di Iorio, Lassere, Scippa, & Chiatante, 2005; Mou, Mitchell, & Jones, 1997). Approaches for evaluating root architecture range from empirical modeling (Nygren, Lu, & Ozier-Lafontaine, 2009) to highly-detailed 3D reconstruction of woody root systems (Danjon & Reubens, 2008; Danjon, Sinoquet, Godin, Colin, & Drexhage, 1999). Previous research suggests that root architecture parameters are relatively consistent within a root system (Salas, Ozier-Lafontaine, & Nygren, 2004) and that root biomass may be estimable from relatively few measurements of stem and root sizes (Drexhage, Chauvière, Colin, & Nielsen, 1999). Simple empirical models based on such measurements would then help to alleviate the need for complete excavation of the root system, which exhibits inherent difficulties in data collection, as well as increased time and labor requirements of sampling procedures relative to aboveground harvesting (Wiese, Riemenschneider, & Zalesny, 2005).

In this paper, we describe research evaluating coarse root biomass and architecture for the hybrid aspen clone ‘Crandon’ (*Populus alba* L. × *P. grandidenta* Michx.) (Gatherum, Gordon, & Broerman, 1967; Goerndt & Mize, 2008) grown at different topographic positions and supplied with different rates of fertilizer in central Iowa, USA. Research on aboveground biomass at the site demonstrated that topographic position and fertilization had significant impacts on tree growth over the first three growing seasons; for example, the highest fertilizer rate tested was associated with approximately twice as much aboveground biomass as the unfertilized control (Headlee et al., 2013a). To evaluate belowground biomass, we conducted a complete excavation of root systems for a subset of trees following the first growing season. After the third growing season, we subsampled the roots systems of another subset of trees to evaluate root architecture parameters that could be used to estimate root system biomass. Thus, we were able to evaluate the root biomass production (first year) and architecture (third year) of 'Crandon' as functions of allometric relationships and/or resource availability (i.e., topographic position and fertilizer rate), and also compare root biomass equations derived from these first-year versus third-year measurements.

Materials & methods

Site description

The study site is located adjacent to Big Creek approximately 20 km southwest of Ames, IA, and lies on an east-facing hillside which ranges in elevation from approximately 305 to 325 m above sea level. Plots measuring 18.3 × 24.5 m were established at each of five topographic positions (floodplain, toe slope, back slope, shoulder slope, and summit), with three plots placed along the north-south axis of each topographic position, for a total of 15 plots. In the spring of 2009, two sets of trees were planted: 48 trees per plot spaced at 3.0 × 3.65 m for long-term evaluation of growth and environmental impacts relative to other perennial and annual biomass cropping systems, and 24 trees per plot at half-spacing (1.5 × 3.65 m) for short-term evaluation of the effects of topographic position and fertilizer rate during establishment (i.e., first 3 years). The short-term trees were randomly assigned to one of four fertilizer rates (0, 10, 20, or 40 g tree^{-1} of 20–10-5 NPK tablets; Henry Field's Seed and Nursery Co., Aurora, IN, USA) with six trees per fertilizer rate per plot. Of these, two trees per fertilizer rate per plot were randomly selected for root excavation, one after the first growing season and one after the third growing season, and are the subject of this paper (5 topographic positions × 3 plots × 4 fertilizer rates × 2 trees = 120 total trees). The soils, layout, site preparation, and planting of the tree plots have been described in detail elsewhere (Headlee, 2012; Headlee et al., 2013a). Similarly, the other biomass cropping systems evaluated in the study have been described in detail by Ontl, Hofmockel, Cambardella, Schulte, and Kolka (2013) and Wilson et al. (2014).

Stem and root measurements

During the dormant season, stem biomass (bole and branches) of the sample trees was harvested, diameter was measured at the base of each cut stem (10 cm aboveground), the material was oven-dried at 100°C until stable weights were observed, and dry biomass was measured to the nearest 0.1 g for each tree. The ranges of observed dry weights are summarized in Table 1, by topographic position and fertilizer rate. For the roots, a complete excavation was conducted with shovels for the trees harvested after the first year (n = 60). The excavated roots were washed, placed in paper bags, and dried and weighed in the same manner as described above for the stems. For the trees harvested after the third year (n = 60), a 60 cm tree spade (Caretree Systems, Inc., Columbus OH) was used to excavate all roots within a radius of approximately 30 cm of the tree stump, and three randomly-selected coarse roots (≥ 2 mm diameter) per tree were also excavated using shovels (starting at their origin from the hole created by the tree spade and extending outward through the plots). Each set of roots was placed in a plastic bag,

Table 1. Summary of stem biomass and root dry biomass data for 1-year-old and 3-year-old trees, by topographic position and fertilizer rate. The data for 3-year-old root biomass are estimates based on root architecture; all other data are measured values.

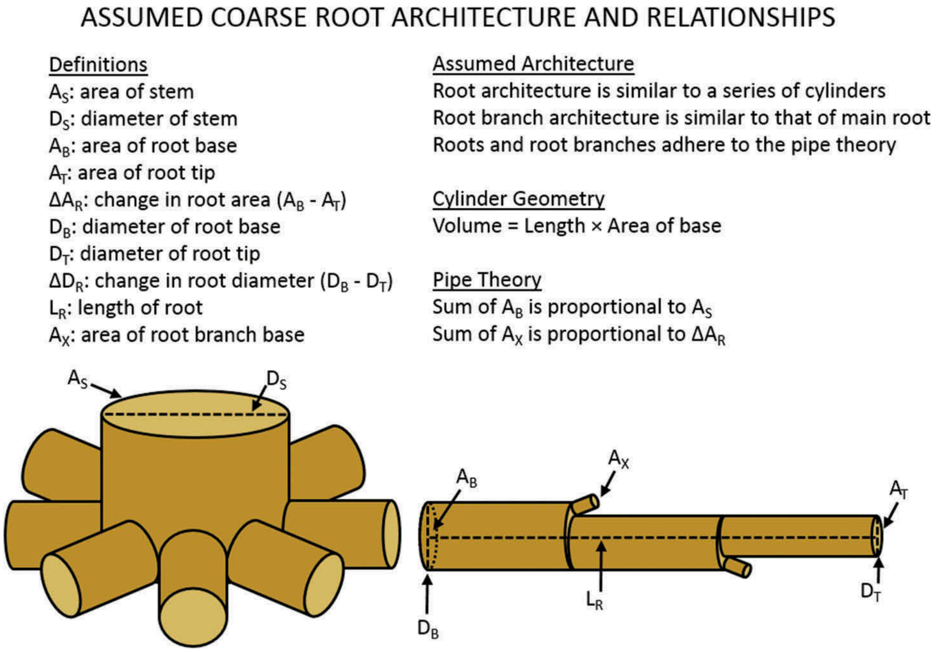
	1-Year-Old Trees		3-Year-Old Trees	
	Stem Biomass (g)	*Root Biomass (g)*	*Stem Biomass (g)*	*Root Biomass (g)*
Topographic Position				
Summit	7.7–69.4	7.9–52.2	170–1972	61–730
Shoulder	1.6–75.8	3.8–60.0	613–1994	362–963
Backslope	3.9–98.7	7.5–59.2	472–3490	157–1748
Toeslope	1.6–99.7	4.0–81.9	184–2746	182–1216
Floodplain	2.1–55.6	4.6–65.1	447–3958	55–1799
Fertilizer (g tree^{-1})				
0	2.1–45.5	4.6–48.9	472–1910	79–601
10	3.6–74.9	7.5–81.9	205–2024	55–809
20	1.6–90.9	3.8–64.7	170–2624	143–863
40	1.6–99.7	4.0–65.1	428–3958	157–1799

transported to the greenhouse, and kept in cold storage (5°C) until root architecture measurements were taken.

The coarse roots connected to the stump (n = 1,802) were individually measured at the end severed by the tree spade for diameter (to the nearest 0.01 cm) using digital calipers, and the subsample of coarse roots originating from the hole created by the tree spade (n = 180) were measured for length (to the nearest cm) as well as the following diameters: root base (where the root was severed by the tree spade), root tip (where the root was severed during shovel excavation), and base of each root branch arising from the root. Individual cross-sectional area (CSA) of each root and root branch was then calculated from these diameter measurements; total root CSA per tree and total root branch CSA area per root were also calculated, to facilitate simultaneous testing of the pipe theory along with root architecture. Change in root diameter and CSA (base minus tip) were also calculated for each subsampled root, for use as a metric of root size that accounts for differences in the completeness of the harvest of the root. The root branches were then removed and the subsampled roots were washed, placed in paper bags, and dried and weighed as previously described. The dry mass data were subsequently used to determine root mass per unit volume, where volumes were estimated from the root length and diameter measurements as described by Bolte et al. (2004). The mean ± standard error for root specific gravity was 0.345 ± 0.008 dry g cm^{-3}, which is slightly lower than the value of around 0.39 g cm^{-3} typically observed for the stem wood of ‘Crandon’ trees (Hall, Hart, & Peszlen, 2001; Headlee et al., 2013b).

Estimation of total coarse root biomass

Root architecture variables were used to estimate total coarse root biomass with the assumptions that 1) the architecture of coarse roots can be reasonably described as a series of connected cylinders which reduce in size each time a root branch diverges from the root, 2) root branches, and any subordinate branches arising from them, will have similar architecture as the roots and root branches from which they arise, and 3) coarse roots and their root branches adhere to the pipe theory of plant water transport. A schematic of the simplified root structure associated with these assumptions is shown in Figure 1. The first assumption allows



ASSUMED COARSE ROOT ARCHITECTURE AND RELATIONSHIPS

Definitions

A_S: area of stem
D_S: diameter of stem
A_B: area of root base
A_T: area of root tip
ΔA_R: change in root area (A_B - A_T)
D_B: diameter of root base
D_T: diameter of root tip
ΔD_R: change in root diameter (D_B - D_T)
L_R: length of root
A_X: area of root branch base

Assumed Architecture

Root architecture is similar to a series of cylinders
Root branch architecture is similar to that of main root
Roots and root branches adhere to the pipe theory

Cylinder Geometry

Volume = Length × Area of base

Pipe Theory

Sum of A_B is proportional to A_S
Sum of A_X is proportional to ΔA_R

Figure 1. Diagram of the simplified architecture of 'Crandon' roots assumed for the current study. Roots and root branches are assumed to be analogous to a series of cylinders and adhere to the pipe theory of plant water transport.

the volume of each root to be calculated from the lengths and CSA of its cylinders. The second assumption is based on previous research with fractal root models suggesting that root architecture parameters are similar across root orders (Salas et al., 2004) and allows root architecture parameters to be estimated from primary roots alone, which greatly reduces the time and labor required for excavation and measurements. The third assumption allows the size and number of roots to be estimated from the size of the stem, and the size and number of root branches to be estimated from the size of the root. Together, these assumptions allow for estimation of the volume of a root as

$$V_R = \sum (A_i \times L_i) = A_0 \times 30 + A_1 \times L_1 + A_2 \times L_2 \tag{1}$$

where V_R is estimated volume of the root (cm^3), A_i is the area of the *i*th cylinder along the root (cm^2), and L_i is the length of the *i*th cylinder along the root (cm). A length of 30 cm is shown here for the length of the root segment attached to the stump (A_0) as this was the radius of the hole excavated by the tree spade, because root branches lack these segments attached to the stump, this term is excluded when estimating root branch volumes.

Previous research indicates that the use of the means of root architecture parameters produce similar results as more detailed methods while allowing much simpler calculations (Nygren et al., 2009); thus, root volumes in this study were estimated using mean CSA (e.g., total CSA of roots divided by number of roots) rather than estimated for each individual root or root branch. Similarly, the length between each root branch and the reduction in cylinder area at each root branch were also treated as equal to their means (e.g., the total change in root CSA divided by the number of root branches). Based on these volume estimates, total root biomass was then estimated as

$$B_R = 0.345 \times N_R(V_R + N_{X1}(V_{X1} + N_{X2}(V_{X2}\ldots))) \tag{2}$$

where B_R is estimated total root biomass (g), 0.345 is the observed mean root weight per unit volume (g cm^{-3}; described in the preceding section), N_R is the number of roots, V_R is the volume per root (cm^3), N_{Xi} is the number of *i*th-order root branches, and V_{Xi} is the volume per branch of *i*th-order root branches (cm^3). Because the estimated number of third-order root branches per second-order root branch was found to be less than one (mean = 0.8 branches), we concluded our calculations of total root volume and biomass with second-order root branches.

Data analysis

The root biomass data for the 1-year-old trees were analyzed as a two-way factorial of topographic position and fertilizer rate (fixed effects, completely randomized design), with stem biomass used as the covariate and random effects of plot and tree within the plot. Both root biomass and stem biomass were log-transformed to homogenize variance. For the 3-year-old trees, the root architecture variables of total CSA of the roots (covariate = CSA of the stem) and number of roots (covariate = stem diameter) were also analyzed as a completely randomized, fixed effects, two-way factorial of topographic position and fertilizer rate, with random effects of plot and tree within plot. The sub-sampling of roots allowed the remaining variables (root length, total CSA of root branches, and number of root branches) to be analyzed with the additional random effect of root within the tree, and the variability among trees to be tested against the variability among roots within trees. Number of root branches was used as the covariate for root length, change in CSA of the root was used as the covariate for total CSA of the root branches, and change in diameter of the root was used as the covariate for the number of root branches. Analysis of variance (ANOVA) was conducted using PROC MIXED in SAS® (SAS Institute Inc., Cary, NC, USA), with denominator degrees of freedom determined via the Kenward-Rogers method (Littell, Stroup, & Freund, 2002). Based on the results of the ANOVAs, model factors determined to be statistically significant ($p < 0.05$) were used to develop regression equations with PROC GLM in SAS®. Statistical contrasts in PROC GLM were also used to evaluate the biomass equations developed from the first-year and third-year measurements for differences in slopes and intercepts (Littell et al., 2002).

Results

For trees harvested after the first growing season, root biomass was found to be significantly influenced by stem biomass ($p < 0.0001$), but not by topographic position or fertilizer rate (Table 2). Additionally, the relationship between root biomass and stem biomass was found to be very strong ($R^2 = 0.93$). Thus, coarse root biomass appeared to be primarily a function of stem size, rather than resource availability as influenced by topographic position and fertilizer rate.

For trees harvested after the third growing season, the analyses indicated that the total CSA of roots was significantly influenced by ($p < 0.0001$) and strongly related to

Table 2. Results of ANOVA and regression analyses for 'Crandon' root biomass (B_R; g), sum of root cross-sectional area (ΣA_R; cm^2), number of coarse roots (N_R), root length (L_R; cm), sum of root branch cross-sectional area (ΣA_X; cm^2), and number of root branches (N_X). Covariates for each parameter are shown in the Equation column and include stem biomass (B_S; g), cross-sectional area of the stem (A_S; cm^2), diameter of the stem (D_S; cm), number of root branches (N_X), change in root cross-sectional area (ΔA_R; cm^2), and change in root diameter (ΔD_R; cm). Intercepts were suppressed when not significantly different from zero (ΣA_R, N_R, ΣA_X, and N_X), and treatment-specific coefficients (*a*, *b*, and *c*) were fit when significant treatment effects were observed (N_R, L_R, and N_X).

	ANOVA (*p*-values)					Regression	
Parameter	*Covariate*	*Position*	*Fertilizer*	*Pos. × Fert.*	*Tree*	*Equation*	R^2
1-Year-Old Trees							
B_R	**< 0.0001**	0.77	0.42	0.39	n/a	$= 2.66B_S^{0.68}$	0.93
3-Year-Old Trees							
ΣA_R	**< 0.0001**	0.42	0.16	0.26	n/a	$= 1.35A_S$	0.82
N_R	**< 0.0001**	**0.04**	0.66	0.60	n/a	$= aD_S$	0.73
L_R	**< 0.0001**	**0.01**	0.83	0.94	**0.03**	$= b + 13.36N_X$	0.61
ΣA_X	**< 0.0001**	0.72	0.22	0.46	0.23	$= 0.475\Delta A_R$	0.70
N_X	**< 0.0001**	0.87	0.86	0.82	**0.03**	$= c\Delta D_R$	0.81

(R^2 = 0.82) the CSA of the stem but was not significantly influenced by topographic position or fertilizer rate. This indicates that the roots adhered to the pipe theory, regardless of resource availability. The number of roots was significantly related to stem diameter ($p < 0.0001$) as well as topographic position ($p = 0.04$), indicating that 'Crandon' does alter this aspect of its rooting in response to site conditions. Comparison of means by topographic position (Figure 2(a)) showed that the floodplain had significantly more roots per unit stem diameter (14.5 roots per cm) than the other topographic positions (8.9 to 11.3 roots per cm). Linear regression using these

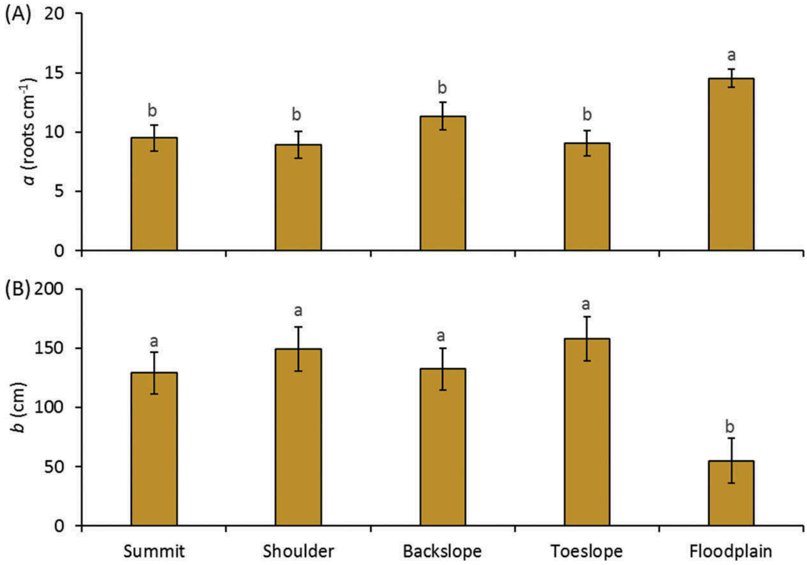


Figure 2. Comparisons among topographic positions for coefficients *a* (number of roots per unit stem diameter; panel A) and *b* (root length intercept; panel B). Statistically significant differences ($p < 0.05$) are indicated by different letters above the standard error bars.

position-specific coefficients demonstrated that the number of roots had a relatively strong relationship with stem diameter ($R^2 = 0.73$).

Root length was found to be significantly related to the number of root branches ($p < 0.0001$) and topographic position ($p = 0.01$), and also differed significantly between trees within treatments ($p = 0.03$); this suggests that 'Crandon' alters its root length in response to both broad scale (topographic) and micro-site (between-tree) conditions. Comparison of means by topographic position (Figure 2(b)) showed that the floodplain had a significantly lower root length intercept (55 cm) than the other topographic positions (129 to 158 cm). Because the tree effects also capture topographic position effects, tree-specific coefficients were fit in the regression analysis and resulted in a moderately strong relationship between root length and the number of root branches ($R^2 = 0.61$). Similar to the total CSA of the roots, the total CSA of the root branches was significantly related to only the covariate (change in CSA of the root; $p < 0.0001$), with a relatively strong model fit ($R^2 = 0.70$). This indicates that the root branches also adhered to the pipe theory, regardless of resource availability. The number of root branches was significantly related to the change in diameter of the root ($p < 0.0001$) and also differed significantly between trees within treatments ($p = 0.03$), indicating that 'Crandon' alters its root branching in response to micro-site conditions. The regression analysis included tree-specific coefficients for the number of root branches per unit change in root diameter and showed a strong model fit ($R^2 = 0.81$).

The estimates of 3-year-old coarse root biomass derived from the root architecture parameters and dry weight data had a moderately strong relationship with stem biomass ($R^2 = 0.56$) and produced a similar equation as was observed for the 1-year-old trees (Figure 3). Statistical contrasts revealed no significant differences between the two equations in terms of slope ($p = 0.59$) or intercept ($p = 0.90$). Thus, subsampling of 3-year-old trees produced a similar equation for predicting root biomass from stem biomass (albeit with a weaker fit) compared to complete excavations of 1-year-old trees.

Discussion

In terms of our underlying assumptions, the similarity between the root biomass equation developed from complete excavations and that developed from our root architecture model suggests that 'Crandon' root architecture can be reasonably approximated as a series of connected cylinders (first assumption). The ANOVA results also suggest a certain amount of consistency in root architecture within a given root system (second assumption), in that the variability among trees was typically greater than the variability among roots within trees. We also specifically tested and confirmed that the roots and their root branches conform to the pipe theory (third assumption), as the sum of the CSA of the roots and root branches were strongly related to the CSA of the stem and change in CSA of the root, respectively. Thus, the underlying assumptions of the model appear to be reasonable, to the extent that we were able to test them in the current study. While our root biomass estimates for the 3-year-old trees included extrapolation to second-order root branches, it should be noted that the contribution of these roots to the estimates of total root biomass was relatively small (mean = 11% of total estimated root biomass) compared to roots (mean = 67%) and first-order root branches (mean = 22%).

While research on stem biomass at our site showed significant differences due to topographic position and fertilization, the same research also showed that allocation of

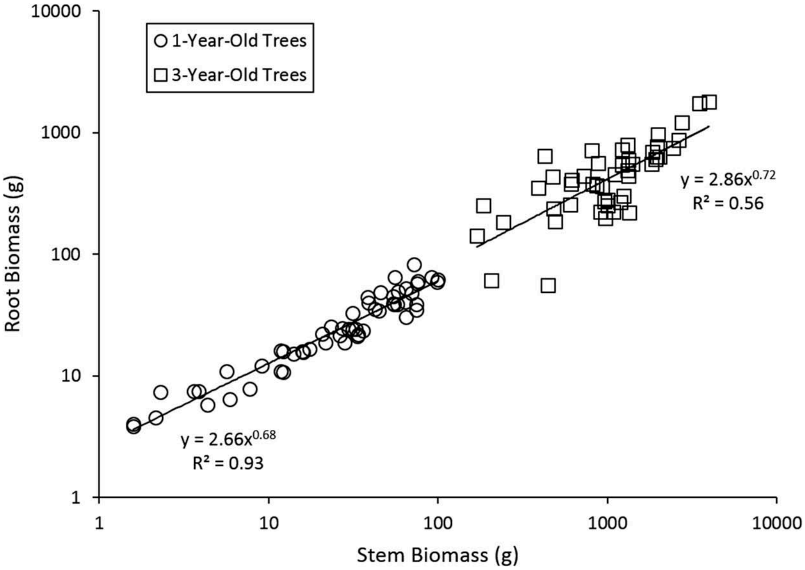


Figure 3. Root biomass measured after the first growing season (circles) and estimated via root architecture parameters after the third growing season (squares) for 'Crandon' trees, in relation to measured stem biomass.

stem biomass between boles and branches was primarily a function of tree size (Headlee et al., 2013a). Similarly, the current study shows that root biomass was primarily a function of tree size for 1-year-old trees, and root biomass estimates derived from root architecture relationships suggest a comparable (albeit weaker) trend for 3-year-old trees. This weaker model fit may be at least partially attributable to the use of estimated mean root architecture parameters as well as the nested structure of Equation 2, which amplifies the errors associated with the estimated means. Of particular interest is the potential error introduced through the root length parameter, as it had the weakest model fit (R^2 = 0.61) of those evaluated in the current study. Thus, additional work aimed at improving root length estimates appears warranted. For example, greater predictive power may be attainable by subsampling a larger number of roots per tree, as our sub-sampling of 3 roots per tree was on average only about 10% of the total number of roots.

Similarly, model fit for root length and possibly other root architecture variables might also be improved by accounting for differences in growing conditions at the micro-site level. Previous research has shown that variability among (and even within) individual tree roots can occur when heterogeneous soil conditions such as patches of nutrient-rich soil are present, particularly with hardwood trees (Mou et al., 1997). Our results suggest that the majority of the variation in 'Crandon' root architecture was explained by allometric relationships along with topographic and between-tree differences, as indicated by the R^2 values in Table 2. However, it is possible that much of the remaining variability represents individual roots (and/or root segments) deviating about the means of the root architecture parameters. Thus, information on micro-site conditions may be useful for improving model fit, although incorporation of such information would likely require considerably more complex models.

Regarding topographic effects on root architecture, the floodplain was observed to have more roots per unit stem diameter and less length per root compared to the other topographic positions (Figure 2). Because the sum of the CSA of the roots is proportional to stem area, as dictated by the pipe theory, the greater number of roots per unit stem size also implies a smaller average CSA per root. Thus, for a given stem diameter, the floodplain trees had a larger number of roots that were shorter in length and smaller in diameter than the other topographic positions. Brassard et al. (2009) describe a tendency for trees to produce more numerous roots concentrated closer to the tree under conditions of higher resource availability; this may explain the changes in root architecture for the floodplain in the current study, as research with the other cropping systems at our site showed the floodplain to have higher levels of soil nitrogen, organic matter, and water conductivity than the other topographic positions (Ontl et al., 2013). The inverse nature of the relationship between the number and size of roots also indicates a trade-off in root architecture that allows the trees to respond to site conditions while maintaining an allometric relationship between aboveground and belowground biomass.

While the results of the current study are encouraging, we recommend broader testing of these methods using additional genotypes and/or sites, especially given the potential for plasticity in rooting across variable environments (Pregitzer & Friend., 1996). Detailed information about the orientation of the roots (e.g., uphill v. downhill direction, horizontal v. diagonal v. vertical angles) could also be included in future studies to produce spatial models of root systems. We collected basic spatial orientation data on a subset of our 3-year-old trees, and the results (though insufficiently replicated for statistical analyses) indicate that the percentages of roots oriented uphill and downhill were approximately 50% each, and the percentages oriented horizontally (roughly < 30 degrees relative to ground level), diagonally (roughly 30 to 60 degrees), and vertically (roughly > 60 degrees) were approximately 65%, 20%, and 15%, respectively. These observations were similar across topographic positions, which is in contrast to reports of differences in spatial distribution of roots associated with topography (Di Iorio et al., 2005; McIvor et al., 2009); however the aforementioned studies involved older trees (i.e., ≥ 10 years old), and therefore may reflect an age effect that had yet to develop at our site. The observation that the majority of roots were oriented horizontally is consistent with previous descriptions of poplar roots being mostly horizontal and near the soil surface (Pregitzer & Friend, 1996).

Conclusion

In summary, our results demonstrate that coarse root biomass in hybrid aspen 'Crandon' is primarily a function of tree size, that 'Crandon' alters its root architecture in response to site conditions within the constraints of this allometric relationship, and that a comparable root biomass equation may be developed using root architecture information from subsampling techniques versus complete excavations of root systems. Among the root architecture variables evaluated here, root length appears especially deserving of additional study aimed at improving model fit. We thus encourage wider testing with 'Crandon' as well as other clones and species, particularly to determine the extent to which greater subsampling and/or other adjustments may improve the fit of biomass equations derived from root architecture measurements.

Acknowledgments

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References

Bolte, A., Rahmann, T., Kuhr, M., Pogoda, P., Murach, D., & Gadow, K. V. (2004). Relationships between tree dimension and coarse root biomass in mixed stands of European beech (*Fagus sylvatica* L.) and Norway spruce (*Picea abies* [L.] Karst.). *Plant and Soil*, *264*, 1–11. doi:10.1023/B:PLSO.0000047777.23344.a3

Brassard, B. W., Chen, H. Y. H., & Bergeron, Y. (2009). Influence of environmental variability on root dynamics in northern forests. *Critical Reviews in Plant Science*, *28*, 179–197. doi:10.1080/07352680902776572

Coleman, M. D., Friend, A. L., & Kern, C. C. (2004). Carbon allocation and nitrogen acquisition in a developing *Populus deltoides* plantation. *Tree Physiology*, *24*, 1347–1357.

Coutts, M. P. (1987). Developmental processes in tree root systems. *Canadian Journal of Forest Research*, *17*, 761–767. doi:10.1139/x87-122

Coyle, D. R., & Coleman, M. D. (2005). Forest production responses to irrigation and fertilization are not explained by shifts in allocation. *Forest Ecology and Management*, *208*, 137–152. doi:10.1016/j.foreco.2004.11.022

Curt, T., Coll, L., Prévosto, B., Balandier, P., & Kunstler, G. (2005). Plasticity in growth, biomass allocation and root morphology in beech seedlings as induced by irradiance and herbaceous competition. *Annals of Forest Science*, *62*, 51–60. doi:10.1051/forest:2004092

Danjon, F., & Reubens, B. (2008). Assessing and analyzing 3D architecture of woody root systems, a review of methods and applications in tree and soil stability, resource acquisition and allocation. *Plant and Soil*, *303*, 1–34. doi:10.1007/s11104-007-9470-7

Danjon, F., Sinoquet, H., Godin, C., Colin, F., & Drexhage, M. (1999). Characterisation of structural tree root architecture using 3D digitizing and AMAPmod software. *Plant and Soil*, *211*, 241–258. doi:10.1023/A:1004680824612

DesRochers, A., & Lieffers, V. J. (2001). Root biomass of regenerating aspen (*Populus tremuloides*) stands of different densities in Alberta. *Canadian Journal of Forest Research*, *31*, 1012–1018. doi:10.1139/x01-037

Di Iorio, A., Lassere, B., Scippa, G. S., & Chiatante, D. (2005). Root system architecture of *Quercus pubescens* trees growing on different sloping conditions. *Annals of Botany*, *95*, 351–361. doi:10.1093/aob/mci033

Douglas, G. B., McIvor, I. R., & Lloyd-West, C. M. (2016). Early root development of field-grown poplar: Effects of planting material and genotype. *New Zealand Journal of Forestry Science*, *46*, 1. doi:10.1186/s40490-015-0057-4

Douglas, G. B., McIvor, I. R., Potter, J. F., & Foote, L. G. (2010). Root distribution of poplar at varying densities on pastoral hill country. *Plant and Soil*, *333*, 147–161. doi:10.1007/s11104-010-0331-4

Drexhage, M., Chauvière, M., Colin, F., & Nielsen, C. N. N. (1999). Development of structural root architecture and allometry of *Quercus petraea*. *Canadian Journal of Forest Research*, *29*, 600–608. doi:10.1139/x99-027

Fortier, J., Truax, B., Gagnon, D., & Lambert, F. (2013). Root biomass and soil carbon distribution in hybrid poplar riparian buffers, herbaceous riparian buffers and natural riparian woodlots on farmland. *SpringerPlus*, *2*, 539. doi:10.1186/2193-1801-2-539

Friend, A. L., Scarascia-Mugnozza, G., Isebrands, J. G., & Heilman, P. E. (1991). Quantification of two-year-old hybrid poplar root systems: Morphology, biomass, and ^{14}C distribution. *Tree Physiology*, *8*, 109–119.

Gatherum, G. E., Gordon, J. C., & Broerman, B. F. S. (1967). Effects of clone and light intensity on photosynthesis, respiration and growth of aspen-poplar hybrids. *Silvae Genetica*, *16*(4), 128–132.

Goerndt, M. E., & Mize, C. (2008). Short-rotation woody biomass as a crop on marginal lands in Iowa. *Northern Journal of Applied Forestry*, *25*, 82–86.

Haissig, B. E., & Davis, T. D. (1994). A historical evaluation of adventitious rooting research to 1993. In T. D. Davis & B. E. Haissig (Eds.), *Biology of adventitious root formation* (pp. 275–331). New York, NY: Plenum Press.

Hall, R. B., Hart, E. R., & Peszlen, I. (2001). Modifying *Populus* environmental responses: Impacts on wood quantity and quality. In N. Morohoshi & A. Komamine (Eds.), *Molecular breeding of woody plants* (pp. 229–238). Amsterdam, the Netherlands: Elsevier Science B.V.

Headlee, W. L. (2012). *Productivity and biometry of hybrid poplars with respect to establishment, regeneration, regional modeling, and utilization of bio-energy byproducts*. PhD dissertation, Iowa State University, Ames, IA. 140 p.

Headlee, W. L., Hall, R. B., & Zalesny, R. S. (2013a). Establishment of alleycropped hybrid aspen "Crandon" in central Iowa, USA: Effects of topographic position and fertilizer rate on aboveground biomass production and allocation. *Sustainability*, *5*, 2874–2886. doi:10.3390/su5072874

Headlee, W. L., Zalesny, R. S., Hall, R. B., Bauer, E. O., Bender, B., Birr, B. A., … Wiese, A. H. (2013b). Specific gravity of hybrid poplars in the north-central region, USA: Within-tree variability and site × genotype effects. *Forests*, *4*, 251–269. doi:10.3390/f4020251

Heilman, P. E., Ekuan, G., & Fogle, D. B. (1994). First-order root development from cuttings of *Populus trichocarpa* × *Populus deltoides* hybrids. *Tree Physiology*, *14*, 911–920.

King, J. S., Pregitzer, K. S., & Zak, D. R. (1999). Clonal variation in above- and below-ground growth responses of *Populus tremuloides* Michaux: Influence of soil warming and nutrient availability. *Plant and Soil*, *217*, 119–130. doi:10.1023/A:1004560311563

Littell, R. C., Stroup, W. W., & Freund, R. J. (2002). *SAS® for linear models* (4th ed., pp. 466). Cary, NC: SAS Institute Inc.

McIvor, I. R., Douglas, G. B., & Benavides, R. (2009). Coarse root growth of Veronese poplar trees varies with position on an erodible slope in New Zealand. *Agroforestry Systems*, *76*, 251–264. doi:10.1007/s10457-009-9209-y

Mou, P., Mitchell, R. J., & Jones, R. H. (1997). Root distribution of two tree species under a heterogeneous nutrient environment. *Journal of Applied Ecology*, *34*, 645–656. doi:10.2307/2404913

Nygren, P., Lu, M., & Ozier-Lafontaine, H. (2009). Effects of turnover and internal variability of tree root systems on modeling coarse root architecture: Comparing simulations for young *Populus deltoides* with field data. *Canadian Journal of Forest Research*, *39*, 97–108. doi:10.1139/X08-158

Ontl, T. A., Hofmockel, K. S., Cambardella, C. A., Schulte, L. A., & Kolka, R. K. (2013). Topographic and soil influences on root productivity of three bioenergy cropping systems. *New Phytologist*, *199*, 727–737. doi:10.1111/nph.12302

Pregitzer, K. S., & Friend, A. L. (1996). The structure and function of *Populus* root systems. In R. F. Stettler, H. D. Bradshaw Jr., P. E. Heilman, & T. M. Hinckley (Eds.), *Biology of populus and its implications for management and conservation* (pp. 331–354). Ottawa, Canada: NRC Res Press.

Reubens, B., Poesen, J., Danjon, F., Geudens, G., & Muys, B. (2007). The role of fine and coarse roots in shallow slope stability and soil erosion control with a focus on root system architecture: A review. *Trees*, *21*, 385–402. doi:10.1007/s00468-007-0132-4

Salas, E., Ozier-Lafontaine, H., & Nygren, P. (2004). A fractal root model applied for estimating the root biomass and architecture in two tropical legume tree species. *Annals of Forest Science*, *61*, 337–345. doi:10.1051/forest:2004027

Shinozaki, K., Yoda, K., Hozumi, K., & Kira, T. (1964). A quantitative analysis of plant form – The pipe model theory. I. Basic analysis. *Japanese Journal of Ecology*, *14*, 97–105.

Thelemann, R., Johnson, G., Sheaffer, C., Banerjee, S., Cai, H., & Wyse, D. (2010). The effect of landscape position on biomass crop yield. *Agronomy Journal*, *102*, 513–522. doi:10.2134/agronj2009.0058

Wiese, A. H., Riemenschneider, D. E., & Zalesny, R. S., Jr. (2005). An inexpensive rhizotron design for two-dimensional, horizontal root growth measurements. *Tree Planters' Notes*, *51*, 40–46.

Wilson, D. M., Heaton, E. A., Schulte, L. A., Gunther, T. P., Hall, R. B., Headlee, W. L., … Boersma, N. N. (2014). Establishment and short-term productivity of annual and perennial bioenergy crops across a landscape gradient. *BioEnergy Research*, *7*, 885–898. doi:10.1007/s12155-014-9409-9

Wullschleger, S. D., Yin, T. M., DiFazio, S. P., Tschaplinski, T. J., Gunter, L. E., Davis, M. F., & Tuskan, G. A. (2005). Phenotypic variation in growth and biomass distribution for two advanced-generation pedigrees of hybrid poplar. *Canadian Journal of Forest Research*, *35*, 1779–1789. doi:10.1139/x05-101

Zalesny, R. S., Jr., Stanturf, J. A., Gardiner, E. S., Perdue, J. H., Young, T. M., Coyle, D. R., … Hass, A. (2016). Ecosystem services of woody crop production systems. *BioEnergy Research*, *9*(2), 465–491. doi:10.1007/s12155-016-9737-z

Zalesny, R. S., Jr., Riemenschneider, D. E., & Hall, R. B. (2005). Early rooting of dormant hardwood cuttings of *Populus*: Analysis of quantitative genetics and genotype × environment interactions. *Canadian Journal of Forest Research*, *35*, 918–929. doi:10.1139/x05-018