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Shoot Position Affects Root Initiation and Growth of Dormant Unrooted Cuttings of *Populus*

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Abstract

Rooting of dormant unrooted cuttings is crucial to the commercial deployment of intensively cultured poplar (*Populus* spp.) plantations because it is the first biological prerequisite to stand establishment. Rooting can be genetically controlled and subject to selection. Thus, our objective was to test for differences in rooting ability among cuttings from three positions on cutting orchard plants of five genomic groups ([Bartr. ex Marsh × *P. trichocarpa* Torr. & Gray *P. deltoides*] × *P. deltoides* 'BC', *P. deltoides* 'D', *P. deltoides* × *P. maximowiczii* A. Henry 'DM', *P. deltoides* × *P. nigra* L. 'DN', *P. nigra* × *P. maximowiczii* 'NM'). Cuttings, 20 cm long, were randomly planted at 1.2- x 2.4-m spacing across three planting dates during 2001 and 2002 at Ames, Iowa, USA (42.0°N, 93.6°W); Waseca, Minnesota, USA (44.1°N, 93.5°W); and Westport, Minnesota, USA (45.7°N, 95.2°W). We measured root dry weight, number of roots, and total root length from harvested cuttings after 14 d of growth. Rooting traits varied relative to stem position but interactions of genomic groups and positions and genotype x environment interactions existed on multiple-year and single-year bases. Position accounted for the second highest amount of variation (≥ 5%) for all rooting traits. Cuttings from the basal third of the shoot system of the stool plant exhibited nearly two times more rooting as those from middle and apical regions, whereas middle cuttings exhibited similar rooting trends as apical cuttings, for all rooting traits. The percentage of cuttings rooted across years was greatest with basal cuttings for the BC, D, DM, and DN genomic groups (> 50%). Middle cuttings of the NM group survived at a greater rate (88%) than did basal (80%) and apical (72%) cuttings. Single-year analyses of interactions of genomic groups and positions showed rooting was greatest with basal cuttings for BC, D, and DN genotypes. Basal cuttings of the DM and NM genomic groups did not clearly outperform middle and apical cuttings, and differences among all cutting positions were site- and year-dependent.

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Key words: Poplar, lateral rooting, adventitious rooting, rooting ability, short rotation intensive culture.

Introduction

There is a predicted shortage of *P. tremuloides* Michx. (quaking aspen) and *P. grandidentata* Michx. (bigtooth aspen) in the North Central United States within 10 to 20 years due to a lack of suitable aspen stumpage within harvestable diameter classes (PIVA, 2003). Thus, recent attention in the North Central United States focuses on increasing production from intensively managed plantations because production from such plantations reduces pressure on native forests (GLADSTONE and LEDIG, 1990). Selected poplar clones (*Populus* spp.) are suited to intensive culture because they are fast-growing, relatively easy to propagate vegetatively, and require shorter harvest cycles than aspen (DICKMANN, 2001; HEILMAN, 1999). Poplar plantations can provide fiber, energy (liquid fuels and biomass for electricity), phytoremediation benefits, raw material for engineered lumber products, cordwood (firewood), riparian stabilization, agroforestry opportunities, wildlife habitat, and aesthetic values (HEILMAN, 1999; JOSLIN and SCHOENHOLTZ, 1997). Four *Populus* species commonly used in North American breeding programs are *P. deltoides* (eastern cottonwood), *P. trichocarpa* (western black cottonwood), *P. nigra* (European black poplar), and *P. maximowiczii* (Japanese poplar).

The ability of poplars to form lateral and adventitious roots from dormant unrooted cuttings is crucial to the commercial deployment of intensively cultured poplar plantations because rooting is the first biological prerequisite to stand establishment. Information is lacking about the genetics and physiology underlying the ability of stem cuttings to root (HAISSIG and DAVIS, 1994; HAISSIG et al., 1992), and an increased knowledge of genetic and environmental covariances between root and shoot developmental systems is desirable (RIEMENSCHNEIDER et al., 1996). Breeding for enhanced rooting ability is a key component of poplar clonal development (RIEMENSCHNEIDER and BAUER, 1997).

Propagation conditions used with unrooted hardwood poplar cuttings are critical to rooting (HANSEN et al., 1983; PHIPPS et al., 1977). Poor care and handling of hardwood cuttings can limit root development in otherwise suitable clones, whereas careful propagation methods can support root growth in recalcitrant clones. Stem cuttings are produced in orchards commonly called stool beds. Cutting position on the stool plant is an important consideration. There is a general trend of improved survival and root growth when cuttings originate close to the base of the stool plant (HARTMANN et al., 1997). BLOOMBERG (1959) tested for differences in number of roots and root length among hardwood cuttings from four regions of stool plants of *P. trichocarpa* (the upper quarter of the shoot, middle two quarters of the shoot, and the lower quarter of the shoot), and he reported greater values for cuttings from the lower quarter than from the other areas. He recommended using cuttings made from the lower quarter of the parental shoot to improve rooting success. Similarly, FEGE and BROWN (1984) evaluated dormant hardwood poplar cuttings and reported the highest carbohydrate content in cuttings from basal portions of stool plants. Carbohydrates often are associated with increased survival and root system production (NGUYEN et al., 1990).

Comparatively high rooting success with increased distance from the apex of the parental shoot also has been reported for other species (HANSEN, 1986; 1989). O'ROURKE (1944) used four cuttings from each parent shoot of *Vaccinium corymbosum* L. (highbush blueberry) to test whether parental shoot position affected rooting ability of cuttings. He concluded the percentage of rooting was highest for cuttings made close to the base of the parental shoot. The rooting percentages ranged from 6% (apex) to 66% (base).

Our preliminary studies of selected poplar genomic groups showed better rooting with cuttings made close to the apex of the parental shoot than more basal cuttings, which is a reversal of the common rooting advantage of basal cuttings. The objective of our current study was to test for differences in rooting ability among dormant unrooted poplar cuttings from three positions on stool plants of five poplar genomic groups.

Table 1. – Genomic groups, clones, and their origin in an experiment testing for differences in rooting ability among cuttings of poplar (*Populus* spp.) based on their positions on the shoot system of the parental stool plant.

Genomic group	Clone	Origin
(*P. trichocarpa* × *P. deltoides*)	NC13563	D. Riemenschneider, U.S. Forest Service
× *P. deltoides*	NC13570	"
(BC)	NC13624	"
	NC13649	"
	NC13686	"
	NC14042	"
P. deltoides	D110	C. Mohn, Univ. of Minnesota
(D)	D105	"
	D117	"
	D133	"
P. deltoides × *P. maximowiczii*	25	V. Steenacker, Belgium
(DM)	DM105	C. Mohn, Univ. of Minnesota and
	NC14103	D. Riemenschneider, U.S. Forest Service
	NC14105	"
	NC14106	"
P. deltoides × *P. nigra*	DN17	France, a.k.a. 'Robusta'
(DN)[a]	DN34	Europe, a.k.a. 'Eugenei'
	DN5	Netherlands, a.k.a. 'Gelrica'
	DN70	Europe
P. nigra × *P. maximowiczii*	NM2	Germany
(NM)	NM6	Germany

[a] Euramerican hybrids with the common designations of *P.* × *euramericana* Guin. and *P.* × *canadensis* Moench.

Materials and Methods

Clone and site selection

Twenty-one clones (*Table 1*) were sampled at random from five *Populus* genomic groups in December 2000 based on their growth potential and anticipated range of rooting abilities. Shoots were collected from stool beds established at the Hugo Sauer Nursery in Rhinelander, Wisconsin, USA (45.6°N, 89.4°W). Cuttings, 20 cm long, were prepared during December and January of 2001 and 2002 and separated according to stem position of the cutting on the stool plant. Cuttings made from the upper third, middle third, and lower third of the parental stool plant are hereafter referred to as apical, middle, and basal positions, respectively. Cuttings were sealed in polyethylene bags and stored at 5°C until each planting date during spring 2001 and 2002. Cuttings were soaked in water for 3 d before planting. There were three planting dates for each combination of year and site. Test plots were established at Ames, Iowa, USA (42.0°N, 93.6°W); Waseca, Minnesota, USA (44.1°N, 93.5°W); and Westport, Minnesota, USA (45.7°N, 95.2°W). Sites were chosen because of their inclusion in a *Populus* Regional Testing Program (RIEMENSCHNEIDER et al., 2001), and because they represented a latitudinal gradient from central Iowa to central Minnesota and a range of soil types typical of poplar plantations (Ames – Hanlon fine sandy loam, Waseca – Clarion loam / Webster clay loam, and Westport – Estherville sandy loam). A completely random experimental design was used for each planting date with four ramets per combination of clone and parental shoot position and a spacing of 1.2 x 2.4 m between cuttings. Two border rows of clones DN34 and NM2 were established at all plantings, except at Westport in 2002, where only one border row of clone NM2 was planted due to spatial constraints.

Data analysis

Individual trees were harvested two weeks after planting. A soil mass up to 65 cm in diameter and 40 cm deep around the developing cutting was excavated. Roots were isolated by washing and photographed with a computerized image-capturing system. Images were stored on high-resolution (High 8 format) video tape and subsequently converted to 8-bit grayscale Tagged Image File Format (TIFF) digital images. The TIFF images were analyzed with the Optimas™ 6.2 image analysis software (Optimas Corporation, Bothell, Washington) to determine dimensions and numbers of leaves and roots. Leaves, stems, lateral roots, callus, and callus roots were dissected from each cutting, bagged, and dried at 70°C for dry weight determination.

Clones were pooled by genomic group for analysis. Data on root dry weight, number of roots, and total root length were subjected to analysis of variance according to the Statistical Analysis System (SAS®) (PROC GLM; SAS INSTITUTE, INC., 2000) on multiple-year (Model I) and single-year (Model II)

Table 2. – Analysis of variance mean squares and expected mean squares in an experiment testing five poplar genomic groups during 2001 and 2002 at Ames, Iowa; Waseca, Minnesota; and Westport, Minnesota, for differences in root dry weight, number of roots, and total root length among stem cuttings based on their positions on the shoot system of the parental stool plant. Probabilities associated with F variance-ratios are listed within parentheses, with significant ($\alpha \leq 0.05$) ratios in bold.

Source	df	Mean squares			Expected mean squares[a, b]
		Root dry weight (mg)	Number of roots	Total root length (cm)	
Year	1	0.15	651.20	685.44	$\sigma^2 + 657.38\sigma^2_{YP} + 394.43\sigma^2_{YG} + 657.38\sigma^2_{YD} + 657.38\sigma^2_{YS} + 1972.10\sigma^2_{Y}$
		(0.8269)	(0.4042)	(0.6439)	
Site	2	3.20	422.97	6109.96	$\sigma^2 + 438.61\sigma^2_{SP} + 263.17\sigma^2_{SG} + 438.61\sigma^2_{SD} + 657.92\sigma^2_{YS} + 1315.80\sigma^2_{S}$
		(0.4370)	(0.6042)	(0.3135)	
Year*Site	2	2.37	636.14	2326.43	$\sigma^2 + 755.37\sigma^2_{YS}$
		(<0.0001)	**(<0.0001)**	**(<0.0001)**	
Date	2	4.06	1927.33	9386.32	$\sigma^2 + 438.53\sigma^2_{DP} + 263.12\sigma^2_{DG} + 438.53\sigma^2_{SD} + 657.79\sigma^2_{YD} + 1315.60\sigma^2_{D}$
		(0.1285)	**(0.0230)**	(0.0967)	
Year*Date	2	0.20	94.59	129.29	$\sigma^2 + 755.20\sigma^2_{YD}$
		(0.0375)	**(0.0014)**	(0.2633)	
Site*Date	4	1.11	184.41	2216.12	$\sigma^2 + 503.61\sigma^2_{SD}$
		(<0.0001)	**(<0.0001)**	**(<0.0001)**	
Genomic group[c]	4	1.85	738.92	3773.89	$\sigma^2 + 294.54\sigma^2_{GP} + 294.54\sigma^2_{DG} + 294.54\sigma^2_{SG} + 441.81\sigma^2_{YG} + 883.62\sigma^2_{G}$
		(0.2858)	(0.0712)	(0.2289)	
Year*Genomic group	4	0.45	82.37	563.52	$\sigma^2 + 441.81\sigma^2_{YG}$
		(<0.0001)	**(0.0001)**	**(0.0001)**	
Site*Genomic group	8	0.18	28.89	404.18	$\sigma^2 + 294.65\sigma^2_{SG}$
		(0.0035)	**(0.0422)**	**(<0.0001)**	
Date*Genomic group	8	0.42	106.54	817.69	$\sigma^2 + 294.60\sigma^2_{DG}$
		(<0.0001)	**(<0.0001)**	**(<0.0001)**	
Position	2	6.76	1497.13	10574.00	$\sigma^2 + 262.71\sigma^2_{GP} + 437.85\sigma^2_{DP} + 437.85\sigma^2_{SP} + 656.77\sigma^2_{YP} + 1313.50\sigma^2_{P}$
		(0.0062)	**(0.0039)**	**(0.0113)**	
Year*Position	2	0.25	32.36	301.36	$\sigma^2 + 754.33\sigma^2_{YP}$
		(0.0186)	(0.1061)	**(0.0446)**	
Site*Position	4	0.15	10.37	320.97	$\sigma^2 + 503.07\sigma^2_{SP}$
		(0.0481)	(0.5788)	**(0.0102)**	
Date*Position	4	0.22	56.30	565.37	$\sigma^2 + 502.94\sigma^2_{DP}$
		(0.0063)	**(0.0036)**	**(0.0001)**	
Genomic group*Position	8	0.49	107.00	957.21	$\sigma^2 + 294.27\sigma^2_{GP}$
		(<0.0001)	**(<0.0001)**	**(<0.0001)**	
Error	4478	0.06	14.42	96.85	σ^2
Total	4535				

[a] Type III expected mean squares and appropriate F-tests generated by using the "RANDOM" statement in "PROC GLM" of the Statistical Analysis System (SAS INSTITUTE, INC., 2000).

[b] σ^2_ = variance attributed to term in the model: σ^2_Y = year, σ^2_S = site, σ^2_{YS} = year*site, σ^2_D = date, σ^2_{YD} = year*date, σ^2_{SD} = site*date, σ^2_G = genomic group, σ^2_{YG} = year*genomic group, σ^2_{SG} = site*genomic group, σ^2_{DG} = date*genomic group, σ^2_P = position, σ^2_{YP} = year*position, σ^2_{SP} = site*position, σ^2_{DP} = date*position, σ^2_{GP} = genomic group*position, σ^2 = error.

[c] Genomic groups are: BC = (*P. trichocarpa* × *P. deltoides*) × *P. deltoides*, D = *P. deltoides*, DM = *P. deltoides* × *P. maximowiczii*, DN = *P. deltoides* × *P. nigra*, NM = *P. nigra* × *P. maximowiczii*.

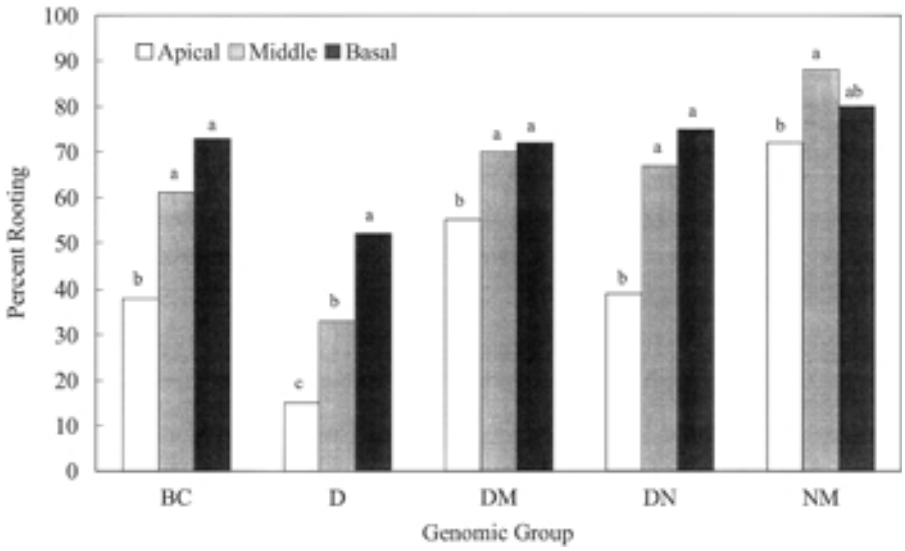

Figure 1. – Percent rooting of stem cuttings across three sites during 2001 and 2002 of five genomic groups of poplar in an experiment testing for differences in rooting ability among cuttings based on their positions on the shoot system of the parental stool plant. Genomic groups are: BC = (*P. trichocarpa* × *P. deltoides*) × *P. deltoides* (n = 432 per position), D = *P. deltoides* (n = 288 per position), DM = *P. deltoides* × *P. maximowiczii* (n = 360 per position), DN = *P. deltoides* × *P. nigra* (n = 288 per position), NM = *P. nigra* × *P. maximowiczii* (n = 144 per position). Cuttings made from the upper third, middle third, and lower third of the parental stool plant are designated apical, middle, and basal, respectively. Positions with the same letter above bars within each genomic group are not different according to Fisher's protected least significant difference (LSD) (α = 0.05, LSD for genomic groups as follows: BC = 14, D = 14, DM = 13, DN = 18, NM = 13).

bases assuming all random effects. Nonsignificant ($\alpha > 0.25$) interaction terms from the original all-effects model were pooled with the residual error term to increase precision of F-tests. Variance components were determined for both models by using restricted maximum likelihood (REML) estimation in PROC VARCOMP of SAS® (SAS INSTITUTE, INC., 2000). Variance estimates were then used to estimate broad-sense heritability (H), the percentage of phenotypic variation among genomic groups due to combined genetic effects, on an individual-tree basis (WILCOX and FARMER, 1968). Cutting diameter at the time of harvest ranged from 0.3 to 1.6 cm and was used as a covariate in the analyses. Final means were adjusted to account for this variation in cutting size. Fisher's least significant difference (LSD) was used to compare adjusted means of main effects for both models.

Results

Multiple-year analysis

Analyses of variance indicated stem position effects across years (*Table 2*). Within plot error contributed to the greatest amount of variation for all growth parameters (74 to 79%), while position accounted for the second greatest amount of variation (≈ 6%). Position also accounted for much more variation when its contributions to significant interactions were included. Genomic groups accounted for less variation (< 3%), which also was expressed by low broad-sense heritability estimates (≤ 0.04). Mean root dry weight adjusted for cutting diameter was 5.4, 3.9, and 3.5 mg for basal, middle, and apical cuttings, respectively (LSD = 0.6, standard error = 0.2, α = 0.05, n = 1512 for each position). Mean number of roots adjusted for cutting diameter was 3.5, 2.8, and 2.4 for basal, middle, and apical cuttings, respectively (LSD = 0.3, standard error = 0.1, α = 0.05, n = 1512 for each position). Mean total root length adjusted for cutting diameter was 7.2, 5.2, and 4.3 cm for basal, middle, and apical cuttings, respectively (LSD = 0.7, standard error = 0.3, α = 0.05, n = 1512 for each position). Year, site, date, and genomic group main effects generally were negligible for each growth parameter. However, there were many significant interactions (*Table 2*).

Table 3. – Root dry weight, number of roots, and total root length (adjusted for cutting diameter) in an experiment testing for differences in rooting ability among stem cuttings of five genomic groups[a] of poplar based on their positions on the shoot system of the parental stool plant. Values for positions with the same letter in each column are not different according to Fisher's protected least significant difference (LSD) (α = 0.05). n = 756 cuttings for each combination of year and position. Standard errors are in parentheses.

	Root dry weight (mg)		Number of roots		Total root length (cm)	
Position[b]	2001	2002	2001	2002	2001	2002
Apical	3.6 b	3.4 c	2.1 b	2.7 c	4.4 b	4.3 c
	(0.3)	(0.3)	(0.1)	(0.2)	(0.4)	(0.4)
Middle	3.4 b	4.5 b	2.2 b	3.3 b	4.3 b	6.1 b
	(0.3)	(0.3)	(0.1)	(0.2)	(0.4)	(0.4)
Basal	5.4 a	5.4 a	3.2 a	3.8 a	7.0 a	7.5 a
	(0.3)	(0.3)	(0.1)	(0.2)	(0.4)	(0.4)

[a] Genomic groups are: BC = (*P. trichocarpa* × *P. deltoides*) × *P. deltoides*, D = *P. deltoides*, DM = *P. deltoides* × *P. maximowiczii*, DN = *P. deltoides* × *P. nigra*, NM = *P. nigra* × *P. maximowiczii*.

[b] Cuttings made from the upper third, middle third, and lower third of the parental stool plant are designated apical, middle, and basal, respectively.

The interaction of genomic group and position was highly significant for all parameters ($P < 0.0001$). Rooting percentages for middle and basal cuttings were not different (α = 0.05) for BC, DM, and DN genomic groups, whereas all positions were different for the D genomic group (α = 0.05) (*Figure 1*). Within the NM genomic group, rooting percentage of basal cuttings was not different from that of apical or middle cuttings, but

Table 4. – Root dry weight, number of roots, and total root length (adjusted for cutting diameter) for each combination of genomic group and position by year and site in an experiment testing for differences in rooting ability among stem cuttings of poplar based on their positions on the shoot system of the parental stool plant. Standard errors for each combination of site and position (DM = 60 cuttings, NM = 24 cuttings) are in parentheses.

	2001				2002				
Genomic group[a]-position[b]	Ames	Waseca	Westport	Over all sites	Ames	Waseca	Westport	Over all sites	Over all years & sites
	Root dry weight (mg)								
DM									
Apical	2.9 (1.0)	3.8 (1.3)	2.1 (0.7)	3.0 (0.6)	6.8 (1.4)	1.6 (0.8)	1.3 (0.6)	3.2 (0.6)	3.1 (0.4)
Middle	4.3 (1.0)	1.7 (1.3)	2.3 (0.7)	2.7 (0.6)	5.6 (1.4)	0.8 (0.8)	1.4 (0.6)	2.7 (0.6)	2.7 (0.4)
Basal	5.1 (1.0)	4.4 (1.3)	1.5 (0.7)	3.7 (0.6)	1.6 (1.4)	0.4 (0.8)	0.8 (0.6)	0.9 (0.6)	2.3 (0.4)
NM									
Apical	4.4 (1.5)	6.3 (2.0)	3.8 (1.1)	5.0 (0.9)	6.7 (2.2)	3.7 (1.2)	1.6 (0.9)	4.0 (0.9)	4.4 (0.7)
Middle	5.5 (1.5)	5.3 (2.0)	2.5 (1.1)	4.3 (0.9)	9.6 (2.2)	5.4 (1.2)	2.2 (0.9)	5.8 (0.9)	5.0 (0.7)
Basal	4.1 (1.5)	4.3 (2.0)	5.1 (1.1)	4.4 (0.9)	4.0 (2.2)	7.2 (1.2)	2.6 (0.9)	4.7 (0.9)	4.4 (0.7)
	Number of roots								
DM									
Apical	1.7 (0.4)	2.2 (0.5)	1.9 (0.4)	2.0 (0.3)	4.1 (0.6)	1.9 (0.5)	1.7 (0.4)	2.6 (0.3)	2.3 (0.2)
Middle	2.4 (0.4)	1.6 (0.5)	1.8 (0.4)	1.9 (0.3)	4.3 (0.6)	1.4 (0.5)	1.8 (0.4)	2.5 (0.3)	2.2 (0.2)
Basal	3.7 (0.4)	2.5 (0.5)	2.0 (0.4)	2.7 (0.3)	2.2 (0.6)	1.3 (0.5)	1.8 (0.4)	1.8 (0.3)	2.2 (0.2)
NM									
Apical	2.6 (0.7)	3.8 (0.7)	3.2 (0.6)	3.3 (0.4)	5.6 (1.0)	4.8 (0.8)	2.3 (0.7)	4.2 (0.5)	3.7 (0.3)
Middle	3.7 (0.7)	3.0 (0.7)	3.0 (0.6)	3.2 (0.4)	6.2 (1.0)	4.4 (0.8)	3.1 (0.7)	4.5 (0.5)	3.9 (0.3)
Basal	2.3 (0.7)	2.8 (0.7)	4.3 (0.6)	3.1 (0.4)	3.5 (1.0)	4.9 (0.8)	2.6 (0.7)	3.7 (0.5)	3.3 (0.3)
	Total root length (cm)								
DM									
Apical	3.5 (1.4)	4.4 (1.2)	2.6 (0.8)	3.7 (0.7)	8.8 (1.8)	2.3 (1.0)	1.9 (1.0)	4.3 (0.7)	4.0 (0.5)
Middle	5.7 (1.4)	2.3 (1.2)	2.6 (0.8)	3.4 (0.7)	8.8 (1.8)	1.1 (1.0)	2.2 (1.0)	4.1 (0.7)	3.8 (0.5)
Basal	6.9 (1.4)	4.1 (1.2)	2.0 (0.8)	4.3 (0.7)	3.1 (1.8)	0.8 (1.0)	1.3 (1.0)	1.7 (0.7)	3.0 (0.5)
NM									
Apical	7.3 (2.2)	6.9 (2.0)	4.7 (1.3)	6.4 (1.1)	9.1 (2.8)	6.0 (1.6)	2.3 (1.5)	5.8 (1.2)	6.0 (0.8)
Middle	7.9 (2.2)	5.5 (2.0)	3.0 (1.3)	5.3 (1.1)	13.5 (2.8)	7.4 (1.6)	4.2 (1.5)	8.4 (1.2)	6.8 (0.8)
Basal	7.7 (2.2)	5.2 (2.0)	7.2 (1.3)	6.6 (1.1)	6.7 (2.8)	9.7 (1.6)	5.4 (1.5)	7.4 (1.2)	6.9 (0.8)

[a] Genomic groups are: DM = *P. deltoides* × *P. maximowiczii* and NM = *P. nigra* × *P. maximowiczii*.

[b] Cuttings made from the upper third, middle third, and lower third of the parental stool plant are designated apical, middle, and basal, respectively.

middle cuttings rooted at a greater percentage than apical cuttings ($\alpha = 0.05$). Similar interaction trends existed for root dry weight, number of roots, and total root length.

Single-year analysis

Analyses of variance indicated parental shoot position effects in 2001 (root dry weight, $P < 0.0001$; number of roots, $P = 0.0040$; and total root length, $P = 0.0055$) and 2002 (root dry weight, $P = 0.0368$; number of roots, $P = 0.0169$; and total root length, $P = 0.0348$). Position main effects accounted for greater amounts of variation in 2001 (≈ 7%) than 2002 (≈ 5%). Genomic groups did not account for much variation (< 5%), and broad-sense heritabilities were low (0.01 to 0.06). During 2001, basal cuttings were different from apical and middle cuttings, but cuttings of apical and middle positions were not different from each other (*Table 3*). In addition, all positions were different during 2002 for all rooting traits, with basal and apical cuttings exhibiting the greatest and poorest rooting, respectively.

The interaction of genomic group and position was significant for all rooting parameters in both years ($P < 0.0001$). These data were similar to those of the multiple-year analysis, with the greatest rooting success from basal cuttings for the BC, D, and DN genomic groups. However, DM and NM cuttings exhibited different trends. Basal cuttings did not clearly outperform apical and middle cuttings, and differences among all cutting positions were site- and year-dependent (*Table 4*).

Discussion

Within plot error accounted for 74 to 79% of the variation in rooting. We attribute this high error to microsite differences that were magnified by the short time of the study. We believe loss of roots during excavation was minimal given the small, unbranched structure of the root systems studied.

Our results show the stem position on the stool plant accounted for ≈ 6% of the variation in rooting of these poplar genomic groups. Across genomic groups and years, cuttings from the basal third of the shoot system on the parental stool plant outperformed those from middle and apical regions, and middle cuttings generally outperformed apical cuttings. Genomic groups varied in their ability to produce roots from cuttings made from different regions of the parental shoot. The BC, D, and DN genomic groups exhibited greater rooting on cuttings from the basal section of the parental shoot. In contrast, using basal cuttings was not clearly superior for the DM and NM genomic groups; differences between middle and basal cuttings from plants of these two groups were site- and year-dependent. Specific information for the DM and NM genomic groups has not been reported previously, and attempts to plant cuttings based on such genotypic classifications are lacking. Therefore, we recommend locally based pilot studies testing the rooting ability of DM and NM genomic groups before broad-scale use.

Our results found a much greater role for interactions than those of previous studies. Plausible mechanisms for better rooting with cuttings made from basal stem positions than those made from apical positions are similar to previous studies and include cutting diameter (mass) differences potentially associated with increased carbohydrate storage, initiation of preformed root primordia, and differences in organogenic activity along the stem of the parental shoot.

Rooting may improve as diameter and associated mass of the cuttings increases. The diameter of the cuttings at the time of harvest increased toward the base of the stool plant (0.3 to 1.6 cm). Cutting diameter was a significant covariate ($P < 0.0001$) for all rooting traits. Thus, we report genotype-specific responses to differences in cutting diameter, with cuttings of large diameter (≥ 0.6 cm) exhibiting greater survival than those with diameter < 0.6 cm. Robison and Raffa (1998) also cited above-average growth with cutting diameter ≥ 0.6 cm. Dickmann et al. (1980) tested the influence of cutting diameter on early survival and growth of *Populus* clones by using cuttings that were from < 0.6 to 1.9 cm in diameter. The "best" survival and shoot growth occurred in cuttings at least 0.6 cm in diameter, with shoot height of smaller-diameter (< 0.6 cm) cuttings only 30 to 67% of larger-diameter (0.6 to 1.9 cm) cuttings. Hansen et al. (1983) reported improved rooting as diameter increased with cuttings ranging in diameter from 0.95 to 2.54 cm. Furthermore, we believe the improved rooting from basal cuttings of our BC, D, and DN genomic groups may have been the result of increased carbohydrate reserves stored in the large-diameter cuttings. A positive relationship between carbohydrate content and cutting diameter has been reported (Fege and Brown, 1984), along with evidence citing increased total carbohydrate content in basal portions of cuttings directly before root initiation (Okoro and Grace, 1976). In addition to storage and availability, carbohydrate metabolism may have differed among our genomic groups, which has been reported elsewhere for DN hybrids and hybrids between *P. balsamifera* L. (balsam poplar) and *P. deltoides* (Tschaplinski and Blake, 1989).

The existence of preformed root primordia also may increase rooting success of basal cuttings compared with apical cuttings. We hypothesize that variation in number of roots may be the result of differences among clones in their ability to develop preformed primordia throughout the parental shoot during the preceding growing season. Number of primordia may be directly related to the position on the stem of the parental shoot. Clones that root well may have an abundance of preformed primordia, whereas clones that root less prominently may have relatively fewer preformed primordia. Smith and Wareing (1974) concluded fewer preformed root primordia existed in smaller-diameter cuttings made from apical regions of the parental shoot than among larger-diameter, basal cuttings. Differences in root distribution along cuttings within positions also have been reported. Ying and Bagley (1977) discovered apical cuttings rooted primarily from the bottom of cuttings, whereas roots differentiated throughout basal cuttings from top to bottom. In addition, better rooting with basal cuttings of large diameter may be the result of more available auxin than in apical cuttings of small diameter. Translocation of auxin to the root system may have contributed to higher auxin content in basal sections of the stool shoots. Auxin increases the rooting success of cuttings (Haissig, 1972; Farmer, 1966), perhaps due to its influence on the initiation of preformed root primordia. Future studies could be designed to test for the existence of preformed root primordia and to test for correlations between extent and distribution of preformed root primordia and number of roots.

Organogenic activity may have decreased toward the apex of the parental shoot. This decrease of activity may have been present in our BC, D, and DN genomic groups. However, we speculate that organogenic activity did not decrease in shoots of the DM and NM genomic groups, which may have led to better rooting on apical cuttings compared with those from the other groups. Bloomberg (1963) tested cuttings of *P.* × *canadensis* Moench (Euramerican poplar) hybrids and *P. trichocarpa* for differences in initial rooting based on parental shoot position. He reported basal cuttings exhibited greater root length than did apical cuttings for all genotypes. In addition, number of roots and root length on *P. trichocarpa* cuttings increased towards the base of the shoot, suggesting that organogenic activity decreased towards the apex (Bloomberg,

1959). Furthermore, HANSEN and TOLSTED (1981) cited greater survival for cuttings made from basal stem positions than those made from apical positions in a hybrid of *P. alba* L. (white poplar) and *P. grandidentata*. YING and BAGLEY (1977) reported more roots on *P. deltoides* cuttings made from the base of the parental stool shoot than cuttings made from the middle and apical sections of the shoot.

Conclusion

Cuttings from the basal third of the shoot system of the stool plant exhibited nearly two times more rooting as those from middle and apical regions, whereas middle cuttings exhibited similar rooting trends as apical cuttings. Single-year analyses of interactions of genomic groups and positions showed rooting was greatest with basal cuttings for BC, D, and DN genotypes. Basal cuttings of the DM and NM genomic groups did not clearly outperform middle and apical cuttings, and differences among all cutting positions were site- and year-dependent.

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