

## ROOTING OF STEM CUTTINGS FROM 12-14-YEAR OLD SCOTCH PINE ORTETS

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ABSTRACT .--Hardwood stem cuttings were collected from upper and lower crown positions of 24 Scotch pine (*Pinus sylvestris* L.) ortets. The ortets were 12-years-old at the beginning of the three year experiment and had started strobili production at ages 10 to 12. Percent rooting and number of ortets having at least one cutting that rooted declined each year of the study. Percent rooting was generally higher in cuttings collected from the lower crown position than the upper; however, in some ortets percent rooting was greater from the upper crown position. Percent rooting ranged from 0 to 79%, and averaged 10% over all ortets, years, and crown positions. The relationship between sexual maturity and rooting potential is discussed.

### INTRODUCTION

Vegetative propagation has received increasing interest in tree improvement programs as a means of maximizing genetic gains, as it captures both additive and non-additive genetic variation. There are many technical problems to overcome before asexual propagation can be used operationally. One major problem has been the inability to propagate proven genotypes, either through tissue culture or traditional macro-propagation techniques.

Typically, juvenile-mature correlations for volume growth are not available for coniferous species so selection of superior individuals has to be delayed until approximately one-third to one-half rotation age. Coniferous rotations (excluding those for Christmas tree production) in the northeastern United States are greater than 40 years. Selection, therefore, has to be delayed until 20 years, by which time the

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genotypes are said to be "physiologically mature".<sup>1</sup> Stem cuttings taken from these mature individuals no longer exhibit high root regeneration potential.

All meristems of a plant are not of the same physiological maturity. A cone of juvenility exists where meristems in the interior and lower crown positions are more juvenile than meristems in exterior and upper crown positions (16).

A meristem's chronological age is not an indicator of physiological maturity. Physiological maturity is related to the number of cell divisions the meristematic region has undergone and to the degree to which those cells elongate (2,6,10,15). Also, there are degrees of sexual maturity in conifers; female strobili are produced two or more years before male strobili (3,9).

One cause of the loss of high regeneration potential has been attributed to the onset of sexual maturity. Some observations indicate that loss of rooting potential and onset of sexual maturity occur concomitantly (1), while others have observed high rooting potential in ortets that are sexually mature (14 and Struve, unpublished data). The relationship between loss of adventitious rooting potential in stem cuttings and onset of sexual maturity needs clarification.

Scotch pine was chosen for study because it is an important reforestation species in Europe and the northeastern United States. Further, some varieties are precocious, with individuals producing strobili as early as three years from seed germination (4).

To study the relationship between sexual maturity and adventitious root regeneration potential of stem cuttings, hardwood stem cuttings were collected for propagation from upper and lower (putative physiologically mature and juvenile, respectively) crown positions of 24 Scotch pine ortets for three consecutive years, at ages 12 to 14.

#### MATERIALS AND METHODS

Twenty-four phenotypically superior Scotch pine ortets were selected in 1976 for desirable crown form, dark blue-green winter foliage color, and moderate-to-fast growth. The ortets were F1 progeny from predominantly intraprovenance (Guadarrama Mts., Spain) full-sib crosses using 13

<sup>1</sup>There are at least two definitions of physiological maturity. One is based on the ability to form reproductive structures. A physiologically mature meristem has the ability to initiate and develop reproductive structures. A physiologically juvenile one does not. Another definition is based on the ability to form adventitious roots. A meristem is physiologically mature when it exhibits low adventitious root regeneration potential, juvenile when it exhibits high potential.

parents. The ortets were field planted near University Park, PA as two-year-old seedlings in 1972. The plantation was thinned twice to minimize crown competition. In 1982 the ortets ranged in height from 4 to 6 m. Beginning in 1982 and again in 1983 and 1984, 25 hardwood stem cuttings were collected from each of two crown positions; lower (0-2 m height) and upper (greater than 2 m height) on each ortet. Current season's terminal shoots from lateral branches were collected the last week in January and shipped to Columbus, Ohio for propagation. Cuttings were stored no longer than one week at 3°C before sticking. Cuttings were trimmed to 15 cm length, conelets were removed, basal ends were dipped in 3,000 ppm K-IBA (potassium salt of indole-3-butyric acid) for 5 seconds, and cuttings were stuck in a 1:1 (v:v) peat:perlite propagation medium. Cuttings were rooted under intermittent mist, 6 seconds mist every 6 minutes, from dawn to dusk. Greenhouse temperature was programmed at 25° day/18° C night. Cuttings were propagated under natural photoperiod. During the 5 month propagation period the cuttings were observed for presence of strobili. In each year, a randomized complete block design with 2 replications of 12 cuttings from each crown position of each ortet was used.

Rooting response was evaluated the first week in June. Any cutting having one or more roots greater than 2 mm in length was classified as rooted, and ortet rooting percentages were calculated. All rooted cuttings, hereafter referred to as ramets, were potted in 3:1 (v:v) pine bark:peat medium in 3.8 liter containers, hardened off and grown in a greenhouse. Greenhouse conditions were 25° day/18° night temperature. Natural light was supplemented as needed with low pressure sodium lamps to give a 20 hour light period of at least 450 foot candles at plant height. All ramets were fertilized weekly with a complete liquid fertilizer.

In 1982 ramets (those resulting from cuttings rooted in 1982) were kept under greenhouse conditions until August 1983. At that time one-half the ramets were field planted at University Park, PA. The remaining ramets were placed in a container area and overwintered in a double layer inflated polyethylene house. In February, 1984 the containerized ramets were brought into a greenhouse and exposed to conditions as outlined above. As the buds broke, sex of strobili was noted.

Only treatment means are presented, as data contained many zeros and were not normally distributed.

## RESULTS

The average rooting percentages declined each year of the study (Table 1). Average rooting response was greater in cuttings collected from the lower crown position than the upper in 1982 and 1983, with no difference between crown positions in 1984. Similarly the number of ortets having at least one rooted cutting declined each year of the study (Table 2).

Individual ortet rooting response in some cases differed greatly from the averages presented in Table 1. Cuttings of Ortet 5 collected in 1982 from the upper crown position were the only ones that formed adventitious roots in any of the three years (Table 3). Other examples of deviations from typical response patterns were cuttings collected from the upper crown position for Ortets 13 and 14 in 1984, Ortet 19 in 1983, Ortet 20 in 1983 and 1984 and Ortet 25 in 1983. Rooting response varied greatly among years for individual ortets and crown positions within ortets.

Dormant buds elongated during propagation, but little needle elongation occurred. Shoot and needle elongation in those that elongated during the propagation phase was slight after the ramets were potted and placed under the greenhouse growing conditions. However, the growing conditions did induce resting buds formed during this period to elongate in a normal manner without exposure to chilling temperatures. Resting buds were formed by fall when the ramets were transferred to outdoor conditions in August, 1983.

The 1982 ramets all broke bud when brought into a heated greenhouse in February, 1984. Every ramet produced strobili; those ramets originating from cuttings collected from the upper crown position produced female strobili and those ramets from cuttings collected from lower crown positions produced male strobili.

#### DISCUSSION

The yearly decreases in average rooting percentages support the idea that rooting response decreases with increasing chronological age, an observation made in numerous studies and reviews (5,10). Others have observed that strobili production increases with increasing chronological age (3,9). These correlations of decreasing rooting potential with increasing chronological age and strobili production have led to the implication that attainment of sexual maturity results in loss of adventitious root regeneration potential (7,11).

However, chronological age is not the same as physiological maturity, defined here as ability to form reproductive structures. The chronologically oldest portion of a tree (the juncture of stem and root system) is physiologically the most juvenile (7,8). Confusion exists as to development of which mature characteristics are the result of epigenetic (ontogenetic changes), and which are the result of physiological (environmental or complexity factors) changes (7,11). It is not known whether loss of rooting potential in stem cuttings is an epigenetic change (and thus a relatively stable change) or physiological change (and thus reversible). If loss of rooting potential results from physiological change

<sup>1</sup> Many characteristics other than strobili production and rooting potential have been used as indicators of physiological juvenility or maturity; leaf shape and size, growth rate and duration of growth, thorniness, and wood quality.

then proven genotypes can be successfully propagated in high percentages once the physiological cause(s) has been overcome. Data in Table 3 suggest that loss of rooting potential in stem cuttings is not an epigenetic event. At times rooting response was greater in stem cuttings collected from upper crown positions, and presumably more physiologically mature, than cuttings collected from lower crown positions. Rooting potential of truly juvenile Scotch pine cuttings was not studied, as the youngest chronologically aged material was 12-years-old and sexually mature. For comparison, hardwood stem cuttings from 9-month-old non-strobili producing Scotch pine seedlings averaged 53%, with a range of 23 to 73% (Struve, unpublished data). Several sexually mature 12 and 13-year-old ortets had rooting responses equal to or better than the 9-month-old seedling average. Similar high rooting potential has been observed in sexually mature eastern white pine (Struve, unpublished data) and Douglas-fir (13) stem cuttings. The results indicate that the onset of sexual maturity does not prevent rooting in Scotch pine stem cuttings. The loss of rooting potential might be due to a physiological age effect (complexity factor). Therefore, physiological maturity would best be defined in terms of ability to produce strobili.

The large drop in rooting response between 1983 and 1984 might be more attributed to adverse weather conditions than advancing maturity or increased complexity. Stem cuttings from 35 different younger ortets (plantation age 9-years-old) rooted in 1984 under similar conditions to the 24 ortets in this study averaged 4%. The 1983 growing season was characterized by hot, dry conditions and the 1983-1984 winter was colder than normal prior to cutting collection. Pre-severance environmental conditions can greatly affect rooting potential (12,14). Other explanations, e.g. unknown pathogenic attack, are possible.

Adventitious root regeneration potential is under genetic control. No rooted cuttings were obtained during the three year study from four ortets, while percent rooting of cuttings from lower crowns of five ortets exceeded 40% in two of three years. Because of the great variation in rooting response among years, a precise estimation of ortet rooting potential cannot be determined from one rooting trial.

Table 1. Annual rooting of Scotch pine hardwood stem cuttings collected from two crown positions on each of 24 ortets.

Crown Position	AVERAGE PERCENT ROOTING <sup>1</sup>		
	1982	1983	1984
Upper	12	8	2
Lower	18	16	3
Yearly Average	15.0	12.0	2.5

<sup>1</sup> Average of 576 cuttings for each crown position.

Table 2. Number of Scotch pine ortets (by year) in which at least one of 24 cuttings rooted. Cuttings were collected from upper and lower crown positions.

NUMBER OF ORTETS HAVING AT LEAST ONE ROOTED CUTTING

Crown Position	1982	1983	1984	1982-1984
Upper	12	10	4	15
Lower	17	13	5	18
Upper or Lower	19	15	6	20

Table 3. Rooting response of hardwood stem cuttings taken from two crown positions of 24 Scotch pine ortets for three years.

ORTET	CROWN POSITION	PERCENT ROOTING <sup>1</sup>		
		1982	1983	1984
76-HG-4	Upper	17	46	0
	Lower	58	63	0
5	Upper	13	0	-
	Lower	0	0	-
6	Upper	38	0	4
	Lower	38	4	4
7	Upper	0	0	0
	Lower	13	4	0
8	Upper	0	0	0
	Lower	21	8	0
9	Upper	4	0	0
	Lower	0	0	0
10	Upper	4	0	0
	Lower	13	0	0
11	Upper	21	29	0
	Lower	21	42	4
12	Upper	0	0	0
	Lower	0	0	0
13	Upper	30	17	21
	Lower	46	58	4
14	Upper	46	21	17
	Lower	79	42	0
15	Upper	8	0	0
	Lower	8	0	0
16	Upper	0	0	0
	Lower	0	0	0
17	Upper	0	0	0
	Lower	0	0	0
18	Upper	0	0	0
	Lower	0	0	0
19	Upper	0	13	0
	Lower	0	4	0
20	Upper	0	8	4
	Lower	21	0	0
21	Upper	0	0	0
	Lower	4	0	0
22	Upper	0	0	0
	Lower	4	0	0
23	Upper	0	0	0
	Lower	25	25	8
25	Upper	0	4	0
	Lower	4	0	0
28	Upper	21	4	0
	Lower	33	17	0

Table 3 continued.

ORTET	CROWN POSITION	PERCENT ROOTING <sup>1</sup>		
		1982	1983	1984
76-HG-29	Upper	25	21	0
	Lower	54	42	42
30	Upper	35	38	0
	Lower	38	67	0

<sup>1</sup> Based on two replications of 12 cuttings in each year.

#### LITERATURE CITED

1. Black, D. K. 1972. The influence of shoot origin on the rooting of Douglas-fir stem cuttings. Comb. Proc. Int. Plant Prop. Soc. 22:142-157.
2. Borchert, R. 1976. The concept of juvenility in woody plants. Acta. Horticulturae 56, Juvenility in woody Perennials. 21-36.
3. Dorman, K. 1976. The Genetics and Breeding of Southern Pines. U.S.D.A. For. Sci. Agr. Handbook. 471.
4. Gerhold, H. D. 1966. Selection for precocious flowering in Scotch pine. U.S. For. Ser. Res. Paper NC-6:4-7.
5. Girouard, R. M. 1974. Propagation of spruce by stem cuttings. N.Z. J. For. Sci. 4(2):140-149.
6. Greenwood, M. S. 1984. Phase change in loblolly pine: shoot development as a function of age. Phys. Plant. (In press).
7. Hackett, W. P. 1983. Phase change and intracclone variability. Hort-Science. 18:840-844.
8. Hartmann, H. T. and D. E. Kester. 1983. Plant Propagation, Principles and Practices. 4th ed. Prentice-Hall, Englewood Cliffs, NJ, USA. 206.
9. Koski, V. 1975. Natural pollination in seed orchards with special reference to pines. In. Seed Orchards, Faulkner ed., Forest Commission Bulletin. No. 54. London. p. 83-91.
10. Nienstaedt, H., F. C. Cech, F. Mergen, C. Wang and B. Zalz. 1958. Vegetative propagation for forest genetics research and practice. J. For. 56(11):826-839.

11. Olesen, P. O. 1978. On cyclophysics and topophysics. *Silvae Gen.* 27(5):173-178.
12. Roberts, A. N. and L. H. Fuchigami. 1973. Seasonal changes in auxin effects on rooting of Douglas fir stem cuttings as related to bud activity. *Physiologia Plantarum* 28:215-221.
13. Roberts, A. N., B. J. Tomasovic, and L. H. Fuchigami. 1974. Synchronicity of bud break dormancy in Douglas fir and its relation to scale removal and rooting ability. *Physiologia Plantarum* 31:211-216.
14. Roberts, A. N. and F. W. Moeller. 1978. Phasic development and physiological conditioning in the rooting of Douglas fir shoots. *Combined Proceedings of the International Plant Propagators' Society* 28:32-39.
15. Schaffalitz de Muchaldell, M. 1959. Investigation on aging of apical meristems in woody plants and its importance in silviculture. *Forest. Forsgsv. Danm.* 25:310-455.