The Effect of Auxin and Cutting Type on Rooting of *Cupressus cashmeriana*©

Matthew D. Taylor and Annie Raup
Longwood Gardens, 1001 Longwood Road Kennett Square, Pennsylvania 19348, USA
Email: mtaylor@longwoodgardens.org

Cupressus cashmeriana **is an ornamental evergreen conifer with little information published on vegetative propagation. Two experiments were conducted to determine the effects of auxin and cutting type on adventitious rooting. Experiment 1 evaluated three cutting treatments (softwood, hardwood, and mallet) and four auxin treatments consisting of 0, 2500, 5000, and 10000 ppm potassium indole-3-butyric acid (K-IBA). Experiment 2 evaluated the same cutting types as Expt. 1 and auxin treatments consisted of 5000 ppm K-IBA, 5000 ppm naphthalene acetic acid (NAA), 2500 ppm K-IBA in combination with 2500 ppm NAA and zero auxin applied. In both experiments, rooting of softwood and hardwood cuttings was significantly higher than mallet cuttings. Treatment of cuttings with different rates and types of auxin lead to inconclusive results, which indicate auxin may not be beneficial for rooting of *C. cashmeriana*. This research suggests that the age of plant material is one of the most significant factors for successful rooting *C. cashmeriana*.**

INTRODUCTION

Kashmir cypress, *Cupressus cashmeriana*, is an evergreen conifer, native to Kashmir and Tibet. This ornamental tree can reach up to 18 m and has a broadly conical habit with pendulous flat blue-green branches (Brickell and Cole, 2002; McDonald, 1993). Trees can bear viable seed, but this is of irregular occurrence so vegetative propagation is preferred (Lahiri, 1975). There is little published information on propagation of this species. Studies have indicated that the type of cuttings taken can affects rooting success of other *Cupressus* species (Blyth, 1989; Stubbs et al., 1997). Also Vakouftis et al., (Vakouftis, 2009) indicates *C. macrocarpa* 'Goldcrest' cuttings had higher rooting percentages when taken in the winter compared to all other seasons. Additionally, it is well known that the type of auxin and concentration can have a significant effect on rooting success of hardwood species (Hartmann, 2011). This study was conducted to determine the effects auxin and cutting type on the rooting of *C. cashmeriana*.

MATERIALS AND METHODS

Experiment 1

On 28 Jan 2009, 216 cuttings were taken from 2-3 m tall greenhouse grown *C. cashmeriana* trees. There were three cutting treatments (softwood, hardwood, and mallet) and four auxin treatments consisting of 0, 2500, 5000, and 10,000 ppm potassium indole-3-butyric acid (K-IBA) with 18 replications. All cuttings were approximately 10-15 cm in length. Softwood cuttings were absent of woody tissue, hardwood cuttings consisted of woody material of the previous year's growth, and mallet cuttings consisted of a hardwood cutting with a mallet section of older wood that was 2-3 cm in length. If foliage was present on the bottom 3-4 cm portion of a cutting it was removed. Hardwood cuttings were wounded on two opposite sides, and mallet cuttings were wounded on the side opposite of the shoot. Wounds were 2-3 cm in length. K-IBA treatments were created by dissolving the K-IBA in deionized water, and cuttings were dipped in treatment solutions for 10 sec following wounding. After the cuttings were dipped in the K-IBA solutions, they were immediately planted in a complete randomized block design into a 72-cell plug tray with square cells that measured 3.5×3.5×12.5 cm. Cells were filled with Sunshine Mix #4 (Conrad Fafard Inc. Agawam, Massachusetts) and put in a glass-covered greenhouse that was shaded with white wash to reduce light levels by approximately 50% located in Kennett Square, Pennsylvania. Cuttings were in the greenhouse for 8 weeks

and automatically misted for 10 sec when outdoor light levels accumulated to 200 $W \cdot m^{-2}$. At 8, 12, 16, 20, and 24 weeks, cuttings were removed from the rooting substrate and evaluated based on the formation of callus or roots. Any cuttings that formed one or more roots were removed from the experimental flat and repotted. If a cutting did not root, it was returned to the experimental flat for further evaluation. All rooted cuttings were not further evaluated and that experimental cutting was considered rooted for the remainder of the evaluation dates. Data were subjected to ANOVA using SAS (SAS Institute Inc., Cary, North Carolina) and interactions were considered significant at $p \leq 0.05$. Means were separated by least significant difference (LSD) and error bars in Figure 1 were determined using the standard error function of Sigmaplot (SPSS Inc., Chicago, Illinois).

Experiment 2

On 1 Mar 2011, 72 cuttings were taken from 2-3 m tall greenhouse grown *C. cashmeriana* trees. The same cutting types were taken as Expt. 1 and auxin treatments consisted of 5000 ppm K-IBA, 5000 ppm naphthalene acetic acid (NAA), 2500 ppm K-IBA in combination with 2500 ppm NAA and zero auxin applied. All other experimental parameters were the same as Expt. 1. After 140 d under mist all cuttings were removed from the substrate. If a cutting formed roots, the basal end was dipped into water to remove any substrate from the roots. Roots were then cut off, and dried at 60°C for 48 h and weighed. All experimental data in tables from both experiments was statistically analyzed as a randomized complete block design with mean separation by LSD test at $p = 0.05$ (SAS Institute, 2002, Cary, North Carolina 27513). Interactions were considered significant at $p \leq 0.05$.

RESULTS

Experiment 1

The percentage of cuttings with callus and the percentage of cuttings with roots are presented over time since there was a significant interaction with date and cutting treatment for both factors (Fig. 1). At each date the cutting treatment by auxin treatment interaction was nonsignificant. At 8 weeks, the percentage of cuttings that formed callus was over 60% for both softwood and hardwood cuttings, which were over three times greater than the percentage of mallet cuttings forming callus. The percentage of softwood and hardwood cuttings with callus decreased significantly over time and this effect was due to the increase in the percentage of cuttings that formed roots. Generally, the percentage of cuttings with roots increased from 8 to 20 weeks for all three cutting types. At 20 weeks, maximum rooting percentages were achieved for all three cutting types with hardwood cuttings having the highest rooting percentage of 52.8%. Softwood and mallet cuttings reached a rooting percentage of 31.9 and 5.6%, respectively.

The interaction between cutting type and hormone rate was significant for the percent of cuttings forming callus and the percent of cuttings forming roots. Furthermore, when analyzing each cutting type for the previous factors the auxin treatment by date interaction was nonsignificant and the data for each cutting type is presented in Table 1. Auxin treatment did not significantly affect the percentage of softwood cuttings forming callus. A higher percentage of hardwood cuttings treated formed callus when they were treated with 10,000 ppm K-IBA compared to 2500 ppm K-IBA while all other auxin treatments were intermediate. There were a higher percentage of mallet cuttings that formed callus when treated with 2500 ppm K-IBA compared to all other auxin treatments. The percentage of softwood and hardwood cuttings forming roots was lowest with the 2500 and 10,000 ppm K-IBA treatments, respectively. For mallet cuttings, treatment with 0 ppm K-IBA lead to the highest percentage of cuttings forming roots, however, this was only 6.7%

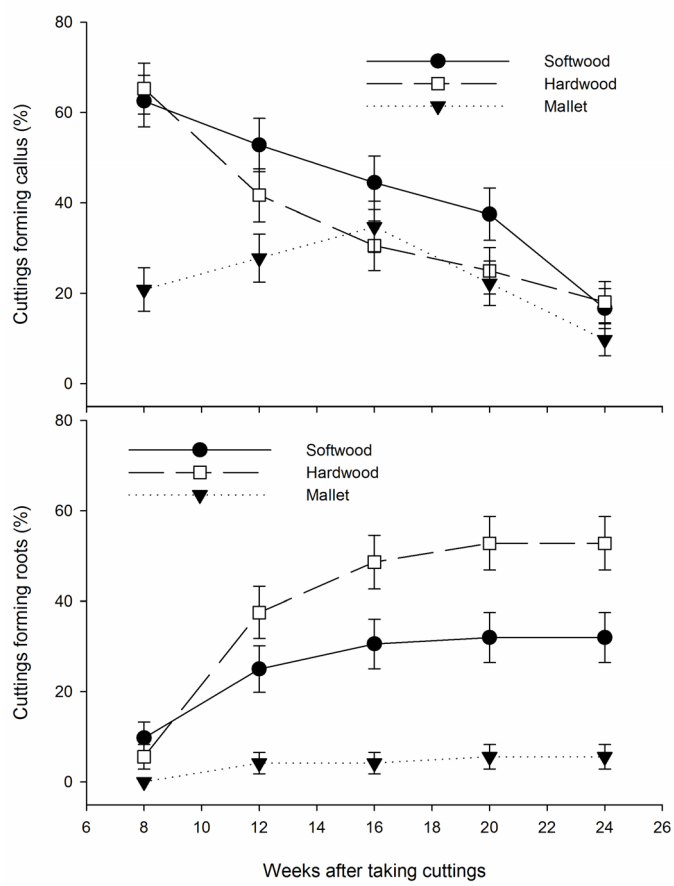

Fig. 1. Effect of cutting type of *Cupressus cashmeriana* on the percentage of cuttings that formed callus and the percentage of cuttings that formed roots every 4 weeks from 8-24 weeks after taking cuttings for Expt. 1. Means are averaged over K-IBA treatments and error bars represent standard error.

Table 1. Effect of K-IBA concentration with each cutting type on the percent of *Cupressus cashmeriana* cuttings that formed callus and the percent that formed roots averaged over dates for Expt. 1.

KIBA (ppm)	Forming callus (%)			Forming roots (%)		
	Softwood	Hardwood	Mallet	Softwood	Hardwood	Mallet
0	37.8 a[z]	34.4 ab	17.8 bc	28.9 a	47.8 a	6.7 a
2500	43.3 a	27.8 b	40.0 a	10.0 b	47.8 a	4.4 ab
5000	50.0 a	35.6 ab	10.0 c	34.4 a	41.1 a	4.4 ab
10000	40.0 a	46.7 a	24.4 b	30.0 a	21.0 b	0 b

[z]Mean separation by LSD within columns at P=0.05.

Experiment 2

The results of Expt. 2 were similar to Expt. 1 in that soft and hardwood cuttings had higher rooting percentages compared to mallet cuttings (Table 2). Indeed, hard and softwood cutting also had significantly higher root weights than mallet cuttings. Although cuttings treated with 0% auxin had a greater rooting percentage and root weights than cuttings treated with auxin, these differences were not significant.

Table 2. Effect of cutting type and auxin type on rooting weights and rooting percentage of *Cupressus cashmeriana* 20 weeks after taking cuttings for Expt. 2.

	Root weight (mg)	Rooting (%)
Cutting type		
Softwood	190 a[z]	83.3 a
Hardwood	291 a	66.7 a
Mallet	42 b	20.8 b
Auxin type		
Control (no auxin)	252 a	72.2 a
K-IBA	149 a	50.0 a
NAA	155 a	55.6 a
K-IBA + NAA	141 a	50.0 a

[z]Mean separation by LSD within columns at P=0.05.

DISCUSSION

This study indicates that auxin treatments of K-IBA at various concentrations, NAA or K-IBA and NAA in combination may not be beneficial, and could potentially have a negative effect on rooting of *C. cashmeriana*. In Expt. 1, 10,000 ppm K-IBA lead to the highest percentage of hardwood cuttings forming callus, yet lead to the lowest percentage of cutting forming roots. Saini (2001) indicated rooting percentages decreased by more than 50% when rooting hormones were applied to tip cuttings of *C. cashmeriana*. With *C. arizonica* var. *glabra* 'Blue Pyramid' and *C. sempervirens* 'Glauca', rooting percentage of semi-hardwood cuttings increased by 49 and 15%, respectively when IBA dip concentrations increased from 3000 and 8000 ppm (1989). Conversely, in the same experiment with *C. macrocarpa* 'Donard Gold', rooting decreased 57% when IBA dip concentrations increased from 3000 to 8000 ppm. Another study with *C. sempervirens* indicated there was also no significant difference in roots per cutting or root length when IBA was applied as talc, dissolved in water or dissolved in alcohol (Dirr and Heuser, 2006). However, when cuttings were treated with auxin compared to the untreated control there was an increase in roots per cutting but no increase root length. There are inconsistent rooting responses to auxin application across the *Cupressus* genus. With *C. cashmeriana*, research results indicate auxin application does not have a positive effect on adventitious root formation.

In Expt. 1, rooting of hardwood cuttings was over 20% greater than softwood cuttings. Although this difference was not significant in Expt. 2, successful rooting of soft and hardwood cuttings was much greater than mallet cuttings in both experiments. A study by Lahiri (1975) indicated *C. cashmeriana* rooting percentage was greatest from cuttings taken from seedings (95%), while 5% or less of soft and hardwood cuttings rooted. However, the soft and hardwood cuttings used in the study were taken from well-established trees. Dirr and Heuser (2006) specifie that for successful rooting of both *C. arizonica* and *C. macrocarpa*, cuttings should be taken from juvenile plants. In the current study, the ages of the trees were unknown but trees were approximately 2-3 m in height and would be of a relatively young age compared to mature established trees. This information suggests that the age of the tree may be one of the most significant factors for successful rooting. However, acquiring cuttings from seedling trees may not always be feasible. In this case, recent growth of the most juvenile trees available should be selected for successful propagation.

Literature Cited

Blyth, G. 1989. Cutting propagation of *Cupresses* and ×*Cupressocyparis*. Comb. Proc. Intl. Plant Prop. Soc. 39:154-160.

Brickell, C. and Cole, T. 2002. The American Horticultural Society Encyclopedia of

Plants and Flowers. New York: DK Publishing.
Capuana, M., Giovannelli, A. and Giannini, R. 2000. Factors influencing rooting in cutting propagation of cypress (*Cupressus sempervirens* L.) Silvae Genet. 49:277-281.
Dirr, M. and Heuser, C. 2006. The Reference Manual of Woody Plant Propagation. Varsity Press Inc. Cary, North Carolina.
Hartmann, H., Kester, D., Davies, F. and Geneve, R. 2011. Hartmann and Kester's plant propagation principles and practices. Prentice-Hall Inc., Upper Saddle River, New Jersey.
Lahiri, A.K. 1975. Propagation of *Cupressus cashmeriana* 'Royle' and *Cryptomeria japonica* 'Don' by stem cuttings. Ind. For. 101:264-268.
McDonald, E. 1993. The New Houseplant: Bringing the Garden Indoors. Macmillian Publishing Company, New York, New York.
Saini, R. 2001. Vegetative propagation in silviculture (hills) division, Darjeeling (West Bengal). Indian Forester. April. p.389-408.
Stubbs, H.L., Blazich, F.A., Ranney, T.G. and Warren, S.L. 1997. Propagation of 'Carolina Sapphire' smooth Arizona cypress by stem cuttings: Effects of growth stage, type of cutting, and IBA treatment. J. Environ. Hort. 15:61-64.
Vakouftis, G., Syros, T., Kostas, S., Economou, A.S., Tsoilpha, P., Scaltsoyiannes, A. and Metaxas, D. 2009. Effect of IBA, time of cutting collection, type of cuttings, and rooting substrate on vegetative propagation in *Cupressus macrocarpa* 'Goldcrest'. J. Prop. Ornamental Plants 9:65-70.