Cutting Type and Auxin Treatment Affect Rooting of *Cupressus cashmeriana*[1]

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Abstract

Cupressus cashmeriana is an ornamental evergreen conifer with little published information on vegetative propagation. Two experiments were conducted to determine the effects of cutting type and auxin concentration and form 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 salt of indole-3-butyric acid (K-IBA). Experiment 2 evaluated the same cutting types as experiment 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*.

Index words: Kashmir cypress, adventitious rooting, rooting hormone, *Cupressus tortulosa* (syn).

Species used in this study: Kashmir cypress (*Cupressus cashmeriana* Carrière).

Significance to the Nursery Industry

Cupressus cashmeriana is a highly attractive ornamental tree that makes an excellent landscape or containerized plant. The tree can be successfully grown outdoors in the southern United States, yet is sold sparingly across the country. This may be due to the lack of understanding and the minimal information published on the proper propagation techniques. Additionally, this tree is considered critically endangered by the International Union for the Conservation of Nature (IUCN). Results from this study indicate 1–2 year old growth from relatively young trees is ideal for vegetative propagation and rooting hormone may not stimulate adventitious root formation.

Introduction

Kashmir cypress (*Cupressus cashmeriana* Carrière) is an evergreen conifer, native to Kashmir and Tibet. This ornamental tree can reach up to 60 ft (18 m) and has a broadly conical habit with pendulous flat blue-green branches (2, 7). Trees can bear viable seed, but this is of irregular occurrence so vegetative propagation is preferred (6). There is little published information on propagation of this species.

Studies have indicated that cutting type can affect rooting success of other *Cupressus* L. (cypress) species (1, 9). Rooting percentages of *C. arizonica* (Arizona cypress) were highest during the hardwood stage, followed by semi-hardwood while softwood cuttings had the lowest rooting percentage (9). Vakouftis et al., (10) indicates *C. macrocarpa* Hartw. ‘Goldcress’ (Monterey cypress) cuttings rooted in higher percentages when taken in the winter (hardwood cuttings) compared to all other seasons.

Another factor that is important for successful cutting propagation of hardwood species is auxin type and concentration (5). Rooting of hardwood and semi-hardwood cuttings of *C. arizonica* (9) increased linearly when auxin concentrations increased from 0 to 16,000 ppm. However, auxin treatments ranging from 0 to 9000 ppm did not affect the number or length of roots formed on *C. macrocarpa* cuttings (10). This study was conducted to determine the effects auxin form and concentration as well as cutting type on adventitious rooting of *C. cashmeriana*.

Materials and Methods

Experiment 1. On January 28, 2009, 216 cuttings were taken from 2–3 m (6–10 ft) tall *C. cashmeriana* trees that were growing in a glass covered greenhouse located in Kennett Square, PA, that was heated at 13C (55F) and vented at 18C (65F). Trees were growing in square 38 liter (10 gal) containers with a peat based substrate and were regularly irrigated with a complete 20-10-20 (N-P2O5-K2O) fertilizer at 250 ppm N. There were three cutting treatments (softwood, hardwood, and mallet) and four auxin treatments consisting of 0, 2500, 5000, and 10,000 ppm potassium salt of indole-3-butyric acid (K-IBA). All cuttings were approximately 10–15 cm (4–6 in) in length. Softwood cuttings were absent of woody tissue were collected from the terminal end of the branches; hardwood cuttings consisted of sub-terminal woody material of the previous year’s growth; finally, mallet cuttings consisted of a hardwood cutting with a small mallet section of adjoining older wood that was 2–3 cm (0.8–1.2 in) in length. Foliage was removed from the basal 3–4 cm (1.2–1.6 in) portion of the cuttings. Hardwood cuttings were wounded on opposite sides, and mallet cuttings were wounded on the side opposite of the shoot. Wounds were 2–3 cm (0.8–1.2 in) in length, approximately 0.5 cm (0.2 in) in width and exposed both the cambium and xylem. K-IBA treatments were created by dissolving the K-IBA in deionized water, and 4 cm (2 in) of the basal ends of cuttings were dipped into treatment solutions for 10 seconds following wounding. After the cuttings were dipped in the K-IBA solutions, they were immediately planted into a 72-cell plug tray with square cells that measured 3.5 × 3.5 × 12.5 cm (1.4 × 1.4 × 4.9 in). Cells were filled with Sunshine Mix #4 (Conrad Fafard Inc. Agawam, MA). Trays were then placed in a

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Table 1. **Effect of cutting type and auxin type on rooting weights and rooting percentage of *Cupressus cashmeriana* 20 weeks after taking cuttings for experiment 2.**

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

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

glass-covered greenhouse that was heated at 21C (70F) and vented at 24C (75F) and shaded with white wash to reduce light levels by approximately 50%. Cuttings were given 8 weeks to root, during which, they were automatically misted for 10 seconds 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 data collected. Data included the formation of callus or roots. Any cuttings that formed one or more roots were removed from the experimental flat. Unrooted cuttings were returned to the experimental flat.

Experiment 2. On March 1, 2011, 72 cuttings were taken from 2–3 m (6–10 ft) *C. cashmeriana* trees different from those used in experiment 1. However, they were growing under the same greenhouse conditions. The same cutting types were taken as Experiment 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. Auxin treatments were created by dissolving the corresponding auxin treatments in deionized water. All other experimental parameters were the same as Experiment 1. After 140 days 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 60C (140F) for 48 hours and weighed.

Data analysis. Both experiments were a randomized complete block design with a factorial arrangement of three cutting treatments and four auxin treatments. Each cutting was considered an experimental unit and treatments were replicated 18 and 6 times for experiments 1 and 2, respectively. At harvest dates during experiment 1, each cutting was assigned a value of 1 or 0 when being evaluated for callus and root formation. All data were statistically analyzed with ANOVA and means presented in Table 1 were separated by LSD test at p = 0.05 (SAS Institute, 2002, Cary, NC 27513). Interactions were considered significant at $p \leq 0.05$. Error bars in figures 1 and 2 were determined using the standard error function of Sigmaplot and data in figure 2 was analyzed with the regression analysis function (SPSS Inc., Chicago, IL).

Results and Discussion

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 between date both and cutting treatments (Fig. 1). At each date the cutting treatment by auxin treatment interaction was nonsignificant. At 8 weeks, over 60% of the softwood and hardwood cuttings formed callus, which was 3 fold greater than mallet cuttings. The percentage of softwood and hardwood cuttings with callus decreased significantly over time which 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 percent rooting was 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 the main effects of cutting type and auxin rate for the percent of cuttings forming callus and rooting percentage were significant. Furthermore, when analyzing each cutting type there were no significant interaction with date, therefor data was pooled across all dates. Pooled data for each cutting type were analyzed with regression analysis to determine linear and quadratic relationships between auxin rate and dependent factors. For softwood cuttings, the relationship between auxin treatment and callus formation and rooting percentage were not significant (data not shown). For mallet cuttings, there was a decreasing linear relationship between auxin treatment and rooting percentage (P = 0.02). However, due to the overall low rooting percentage of mallet cuttings (5.6%), this relationship has minimal

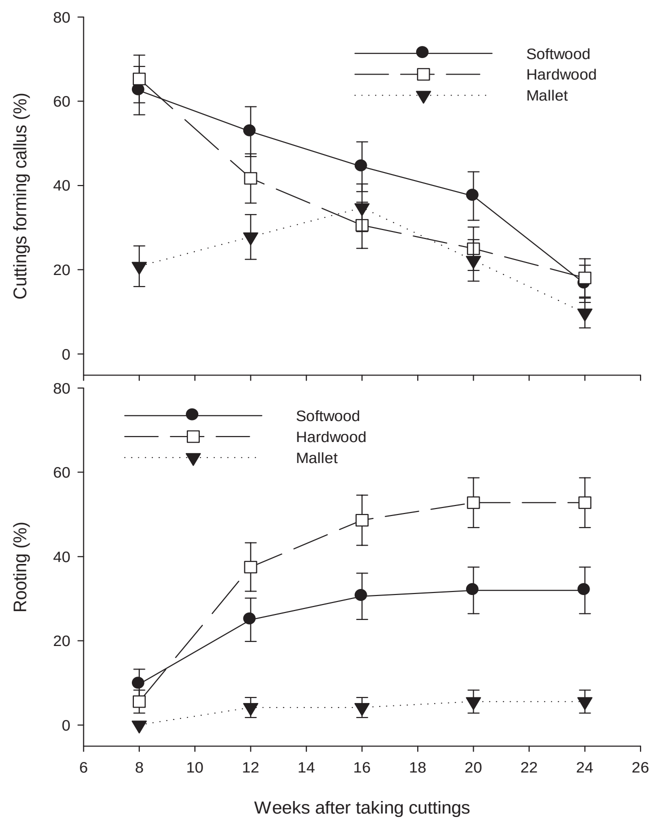

Fig. 1. **Effect of cutting type of *Cupressus cashmeriana* on the percentage of cuttings that formed callus and rooting percentage every 4 weeks from 8–24 weeks after taking cuttings for experiment 1. Means are averaged over K-IBA treatments and error bars represent standard error.**

importance (data not shown). For hardwood cuttings, there was a positive linear relationship between auxin treatment and percent of cuttings forming callus (P = 0.03) and a negative linear relationship between auxin treatment and rooting percentage (P < 0.001) (Fig. 2). Although these linear relationships were significant, R-squared values indicate these relationships are poorly correlated. The quadratic response was also significant; however P-values were greater than linear response P-values.

Experiment 2. The results of experiment 2 were similar to experiment 1 in that softwood and hardwood cuttings had higher rooting percentages compared to mallet cuttings. Indeed, hardwood and softwood cuttings also had significantly higher root weights than mallet cuttings.

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 experiment 1, there was a significant positive linear relationship between K-IBA rate and the percentage of cuttings forming callus, yet the reverse occurred for rooting percentage. Saini (8) indicated rooting percentages decreased by more than 50% when rooting hormones were applied to tip cuttings of *C. cashmeriana*. With *C. glabra* 'Blue Pyramid' (Arizona smooth bark cypress) and *C. sempervirens* 'Glauca' (Mediterranean cypress), rooting percentage of semi-hardwood cuttings increased by 49 and 15%, respectively when IBA dip concentrations increased from 3000 and 8000 ppm (1). Conversely, in the same experiment with *C. macrocarpa* 'Donard Gold' (Monterey cypress), rooting decreased 57% when IBA dip concentrations increased from 3000 to 8000 ppm. Another study with *C. sempervirens* indicated there was no significant difference in the number of roots per cutting or root length when IBA was applied as talc, dissolved in water or dissolved in alcohol (3). However, when cuttings were treated with auxin compared to the untreated control there was an increase in root number 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 experiment one, rooting of hardwood cuttings was over 20% greater than softwood cuttings. Although this difference was not significant in experiment two, successful rooting of soft and hardwood cuttings was much greater than mallet cuttings in both experiments. A study by Lahiri (6) indicated *C. cashmeriana* rooting percentage was greatest from cuttings taken from seedings (95%), while 5% or less of soft and hardwood cuttings rooted when taken from mature trees. Dirr and Heuser (4) specify that for successful rooting of both *C. arizonica* (Arizona cypress) 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.

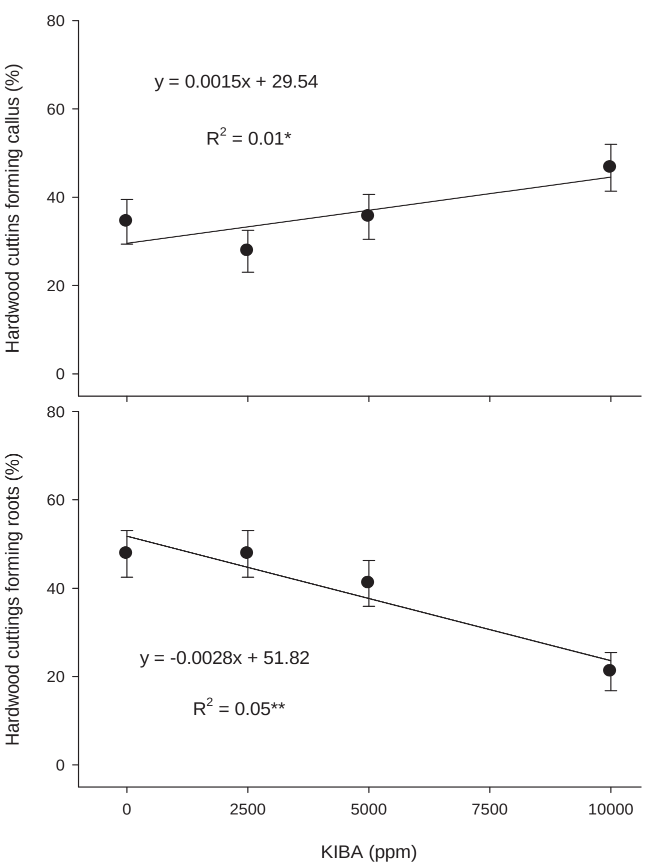

Fig. 2. Relationships between K-IBA concentration and percent of *Cupressus cashmeriana* hardwood cuttings forming callus and rooting percentage for experiment 1. Each symbol represents the mean of 18 cuttings at each evaluation date and error bars represent standard error. * and ** indicate significant at P < 0.05 and 0.001, respectively.

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