Rooting Walnut Hardwood Cuttings

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INTRODUCTION

The use of hardwood cuttings is often the most economical method of producing clonal rootstocks or own-rooted varieties of temperate deciduous fruit trees (Hartmann, et al., 1990). Because leaves are absent and the material is dormant, hardwood cuttings are not as vulnerable to desiccation nor are they as cumbersome to handle as leafy semihardwood cuttings. These attributes make hardwood cuttings desirable for commercial nursery production.

Since walnut is difficult to root and genotypic variation in rooting ability exists, results derived from one clone may not be applicable to other clones. Significant differences in rooting ability among *Juglans regia* (Persian or English walnut), *J. hindsii* (northern California black walnut), and their F_1 hybrid (*J. hindsii* × *J. regia*) known as Paradox have been noted (Hartmann 1978). Paradox hybrids appear to have higher rooting percentages than either parent species. In addition, F_1 hybrids differ markedly from BC_1 (Paradox × *J. regia*) hybrids in rooting ability (Sutter and McKenna, 1995). Although rooting percentages and number of roots/cutting are highest in semihardwood 'Paradox' cuttings compared to hardwood cuttings, hardwood cuttings remain an attractive method of clonal propagation for walnut because of their ease of handling.

The use of bottom heat ranging from 72 to 80F (Burger and Sutter, 1994; Lynn 1957; Sutter and McKenna, 1995) is an important factor required for optimum rooting of hardwood walnut cuttings. This requirement for bottom heat explains the poor results (only 10% to 30% rooting) obtained when one tries to root hardwood walnut cuttings directly in the nursery. When hardwood cuttings are rooted in propagation structures with bottom heat as much as 80% rooting has been achieved (Lynn, 1957; Sutter and McKenna, 1996). The difficulty in using bottom heat, however, is that it dries the lower portion of the propagation medium while the upper portion remains wet making it difficult to maintain an optimum moisture content in the medium surrounding the basal portion of the cuttings where roots form. Kester, working with several *Prunus* species determined that the optimum moisture content when using peat moss as a rooting medium is obtained by mixing peat moss and water (2 : 1, w/w) (pers. comm.). We found in preliminary experiments that this moisture content is useful for walnut cuttings as well.

The objective of our research was to determine factors other than moisture content of the rooting medium that were significant in the rooting of hardwood walnut cuttings. We investigated the effects of factors such as the method of application of auxin as well as auxin synergists on rooting of hardwood walnut cuttings.

The following is a summary of research conducted between 1993 and 1996 designed to determine the rooting potential of hardwood cuttings from a diverse collection of Paradox selections in the Walnut Improvement Program (WIP) at U.C., Davis, and an F_1 Paradox clone known for its relatively high rooting potential, Paradox Bowman Kuhn (BK).

METHODS

All plant material was collected from experimental orchards at UC Davis. Most selections, including BK, were grafted onto seedling Paradox in 1988. The trees were a diverse collection of Paradox selections obtained from the WIP at U.C., Davis. Most of these Paradox genotypes had not been screened for rooting potential. The harvest of cuttings followed a specific protocol. Shoots ranging from 2 to 5 ft were harvested in the morning from current-year shoots. They were placed upright in 5-gal pails and transported to a workroom for immediate treatment and sticking. Three node cuttings were used, 8 in. long on average, with the length largely a function of the number of nodes. Basal cuts were made 1/2 in. below a node. Stem diameters varied from 5/16 to 3/4 of an in. and terminal and subterminal cuttings were handled together. The tops of cuttings were waxed with paraffin to prevent desiccation. All cuttings were soaked for a minimum of 1 min in a 1500 ppm citric acid solution prior to auxin treatment.

Auxin was applied as an aqueous solution of the potassium salt of indole butyric acid (K-IBA) either as a quick dip or by using a KIBA-saturated toothpick. For quick dips, cuttings were soaked in 8000 ppm K-IBA to a depth of 1/2 in. for 30 sec and, allowed to dry for several min, bundled in groups of 15 cuttings with a rubber band, and placed in 5-gal pots containing peat moss moistened by using peat moss and water (2 : 1, w/w). In 1995 and 1996, cuttings were placed in individual plugs, positioned in specially designed Styrofoam® trays that allowed periodic flow-through watering. The trays were placed outdoors over bottom heat in a specially fabricated propagation structure.

Toothpick treatments consisted of soaking common round white spruce toothpicks in a 4000 ppm K-IBA solution for 18 to 24 h in an aluminum foil-wrapped vial prior to use. Pilot holes were drilled perpendicular through the entire width of each cutting in the vicinity of a node using a 3/32-in. wood drill bit and a hand-held electric drill. Toothpicks were removed from the solution using forceps, inserted into the pilot hole, and one blunt end was formed using anvil pruners. The toothpick was hammered through the pilot hole using a block of wood, and was clipped nearly flush on both sides. Cuttings were put into bundles and placed in damp peat moss or plugs as described above for quick dip treatments.

Potential auxin-synergists tested in combination with 4000 ppm K-IBA included the following: 20 ppm abscisic acid, 12 ppm o-coumaric acid, 12 ppm ferulic acid, and both 400 ppm and 2000 ppm spermine (Sigma Chemical Corp. St. Louis, Mo.). Other auxin combinations tested included 100 ppm 2-4-D + 1000 ppm NAA + 2000 K-IBA ppm; and 1500 ppm citric acid + 4000 ppm K-IBA.

RESULTS AND DISCUSSION

Rooting studies the first year resulted in low rooting percentages among the few selections that rooted. The most promising results were obtained with selections 84-121 and 84-128, but great variability was noted among replicates (Table 1). One difficulty that affected results was that the peat moss dried out in the bottom of several containers and we assumed that rooting was decreased as a result. Another important factor was the date the cuttings were collected. The greatest percentage of rooting was obtained in cuttings collected on 3 Dec 1993 (Table 1). Very little rooting was obtained in cuttings collected on 15 Dec 1993 and 11 Jan 1994 and no rooting was obtained in cuttings collected on 4 Feb 1994. In addition all selections that rooted from the Dec. 3 collection date were treated with toothpicks, not quick dips. Of the two selections in the Dec. 15 collection date that rooted, one rooted best with the quick dip and one with the toothpick application. From this data it was impossible to determine the relative effectiveness of the two methods of auxin application. It was clear, though, there was an effect of time of collection of plant material on the rooting of the cuttings. We could not verify reports by Serr (1950) who found that hardwood cuttings rooted well when collected immediately prior to bud break since, we did not collect cuttings at that time.

Table 1. Rooting percentages of hardwood cuttings of selections in the Walnut Improvement Program subjected to different methods of auxin application. Observations were taken 50 days after cuttings were treated and placed in the propagation medium[1].

	Dec.3, 1993[2]		Dec.15, 1993[2]		Jan.11, 1994[2]	
Selection	TP[3]	QD[4]	TP[3]	QD[4]	TP[3]	QD[4]
84-121-1	53[5]	0	0	0	0	0
84-121-2	15	0	0	0	0	0
84-128-1	3	0	8	0	0	0
84-128-2	15	0	0	13	0	0
85-117-19	5	0	0	0	-	-
87-050-1	-	-	0	0	4	0

[1]Note: only those selections that produced roots are included. 11 selections did not produce any roots: 85-117-20,85-117-21,87-026-2,87-027-4,87-032-1,87-112-12,87-117-16,87-117-2,Hagus-8, Sibbett, VK110-6.
[2]Date cuttings were collected
[3]Toothpick application, using 4000 ppm potassium IBA
[4]Quick dip application, using 8000 ppm potassium IBA
[5]Numbers of cuttings in each treatment ranged from 9 to 40 depending on their availability. The experiment was run one time

In order to reduce confounding of results due to genotypic variability and low numbers of available cuttings, we conducted much of the remaining research with one clone, Paradox Bowman Kuhn (BK). This 50-year-old clone, BK, is known for its relatively high rooting potential which was demonstrated in our 1994 and 1995 rooting trials. Results of rooting trials with BK in 1993-1994 indicated that application of auxin by using toothpicks was more effective than that using a quick dip (Table 2). Moisture control of the propagating medium was handled by spraying the peat moss with a hand pump sprayer with a fine spray nozzle. The peat moss became too wet in several containers resulting in rot in the bases of cuttings.

We were not able to replicate results of Lynn (1957) in which he obtained rooting percentages as high as 80% in hardwood cuttings. Perhaps one reason for the difference in results is that the methods of auxin application were different in the two studies. Lynn (1957) treated BK cuttings with a 24-h soak in 200 to 300 ppm IBA concentrations. Another reason for the differences in the two studies may have been that the BK clone was only 10 years old at the time of Lynn's studies. Our work used BK but it was a 50-year-old clone. An aging process, known to occur in clones, may have caused the reduced rooting that we obtained compared to that obtained by Lynn.

Table 2. Effect of harvest date and treatment on percent rooting of hardwood cuttings of Paradox Bowman Kuhn[1].

	Date	
Treatment	Dec. 3, 1994	Dec. 14, 1994
Quick dip	7.0%	9.0%
Toothpick application	8.0%	13.2%

[1]For each treatment 4 replicates of 15 cuttings each were used.

In the next series of experiments we applied several compounds together with K-IBA in order to improve the rooting percentages we had obtained with BK cuttings. Of the different chemicals tried, spermine, o-coumaric acid, ferulic acid, and abscisic acid all increased the percentage of rooting over that of K-IBA alone (Table 3). None of the other treatments had an effect on rooting percentages. Ortho-coumaric acid, ferulic acid, and spermine also caused a marked increase in the number of roots per rooted cutting (Table 3). The use of citric acid resulted in a greater proportion of rooted cuttings having roots originating on at least two opposite sides of the base of the cutting compared to other treatments in which a majority of rooted cuttings had roots emerging primarily from one side.

The experiment using 400 ppm spermine in addition to K-IBA was repeated in Dec. 1995 using a completely randomized complete block design with four blocks of 24 cuttings per treatment. The results confirmed the efficacy of spermine, although its effect was not as marked as in the previous year (Table 4). The addition of spermine to the rooting hormone solution using a quick dip produced rooting percentages three times as great as that obtained using the toothpick method of application. The combination of spermine and K-IBA resulted in rooting percentages three times that of K-IBA alone applied as a quick dip and 1.2 times greater than when K-IBA was applied alone using toothpicks (Table 4). There were fewer roots per rooted cutting than obtained the previous year. In addition, cuttings in which compounds were applied using toothpicks had an average of 4.6 roots per cutting compared to using a quick dip which produced an average of 2.8 roots per cutting. The use of a plug system in this experiment, which allowed more precise control of moisture in the rooting medium during the propagation period, made accurate root counts impossible.

Ultimately, successful clonal propagation of walnut by hardwood stem cuttings will depend on integrating methods of rooting cuttings and handling and transplanting newly rooted cuttings to yield high survival rates. Survival of rooted hardwood cuttings in our experiments has ranged between 21% and 27%. More research is necessary to enable production of own-rooted walnut trees to be commercially viable.

Table 3. Effect of various chemicals on the rooting of hardwood cuttings of Paradox Bowman Kuhn walnut cuttings, collected 20 Dec. 1994.

Treatment	Percent rooted/ (no. cuttings in treatment)	Mean number of roots per rooted cutting	Percentage of rooted cuttings having "2-sided" roots[2]
citric acid[1] 1500 ppm	13/(30)	4	0.7
o-coumaric acid[1] 12 ppm	61/(37)	8.2	0.4
ferulic acid[1] 12 ppm	77/(30)	10.1	0.3
spermine[1] 400 ppm	82/(28)	7.9	0.4
spermine[1] 2000 ppm	39/(28)	5.6	0.4
abscisic acid[1] 20 ppm	43/(30)	3	0.3
2,4-D 100 ppm + NAA 1000 ppm + K-IBA 2000 ppm	10/(30)	4.7	0

[1]In addition to the chemical indicated, 4000 ppm K-IBA was added. The resultant solution was applied using toothpicks soaked in the solutions.

[2]Origin of roots were scored depending on whether they originated from one side or from two opposite sides (two-sided) of the stem base.

Table 4. The effect of spermine with and without potassium indole butyric acid on rooting of hardwood Paradox Bowman Kuhn walnut cuttings. Cuttings collected 19 Dec 1995.

Chemical	Application method	Percent rooting mean	Mean number roots per cutting	Percent two-sided[3]
K-IBA 4000 ppm + spermine 400 ppm	TP[1]	75 ± 12.2	4.4 ±0.6	0.5 ±0.1
K-IBA 4000 ppm	TP	63 ± 0	4.8 ± 1.5	0.4 ± 0.1
Spermine 400 ppm	TP	1 ± 2.1	4.0 ± 8.0	0.3 ± 0.5
K-IBA 8000 ppm + spermine 800 ppm	QD[2]	54 ± 12.3	2.5 ± 0.5	0.3 ± 0.2
K-IBA 8000 ppm	QD	18 ± 8.6	3.0 ± 0.3	0.5 ± 0.4
Spermine 800 ppm	QD	0	0	-

[1] Toothpick application, using 4000 ppm potassium IBA
[2] Quick dip application, using 8000 ppm potassium IBA
[3] Origin of roots were scored depending on whether they originated from one side or from two opposite sides (two-sided) of the stem base.

Acknowledgments. We would like to thank the Walnut Marketing Board for providing the funding for this research, and all the members of the Walnut Improvement Program for providing advice and plant material.

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