Effect of Light and Cutting Age on Rooting in *Quercus bicolor*, *Quercus robur*, and *Quercus macrocarpa* Cuttings©

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This study investigated the effect of light and etiolation with or without stem banding on rooting in *Quercus robur, Q. macrocarpa*, and *Q. bicolor* cuttings. Percent rooting in *Q. bicolor* and *Q. robur* cuttings, taken from greenhouse-grown stock plants averaged 84.9% and 63.8%, respectively. In a second experiment using field-grown stock plants, cuttings were taken at 3-week intervals, from four light and stem banding treatment groups. In *Q. bicolor* cuttings, rooting response was significantly increased by stock plant etiolation ($p < 0.01$) and early cutting age ($p < 0.0001$), but not stem banding ($p > 0.05$). Rooting decreased ($p < 0.0001$) as cuttings aged across treatments with an overall rooting percentage of 62.3% at 3 weeks, decreasing to 10.3% at 6 weeks and 4.7% by the 9th week. In a later field experiment, rooting response in *Q. macrocarpa* cuttings was improved by taking cuttings at 2 weeks after etiolation treatment.

INTRODUCTION

Rooting success in cutting propagation is controlled by a number of variable factors such as the physiological state of the cutting, position of the cutting on the plant, the nutritional status, and the phase of maturity of the stock plant. In a study to investigate the influence of timing and indolebutyric acid (IBA) treatments on rooting, McGuigan, et al. (1996) found, that of the four growth stages investigated (softwood, semi-hardwood, hardwood, and a transition between soft and semi-hardwood), softwood cuttings from two *Quercus phillyreoides* clones (treated with IBA) rooted best at 97.2% and 55.5%, respectively. Seasonal rooting responses have also been observed in lilacs (*Syringa*), Chinese fringe tree (*Chionanthus retusus*), and creeping fig (*Ficus pumila*) (Hartmann et al., 2002). The "narrow window" of rooting opportunity — which is very often limited to 1 or 2 weeks during the entire growing period, makes timing very important to successful propagation (Bicket and Bilderback, 1982; Chapman and Hoover, 1981). To further enhance the rooting ability of cuttings, a number of stockplant management techniques such as hedging and severe pruning have successfully been used to rejuvenate plants and improve rooting (Amissah and Bassuk, 2004; Bolstad and Libby, 1982; and Rosier, 2005).

Recently, a modified container layering technique has successfully been used to asexually propagate oaks (Amissah and Bassuk, 2004; Amissah, 2007; and Hawver and Bassuk, 2000). In a previous experiment to evaluate the effect of severely cutting back stock plants on rooting ability in *Q. bicolor* and *Q. macrocarpa* layers and cuttings, we noted a marked increase in rooting ability with the increasing severity of stock plant cutback (Amissah, 2007). In addition, pretreatments like etiolation and stem banding have been shown to enhance adventitious root formation (Bassuk and Maynard, 1987; Harrison-Murray, 1981; Maynard and Bassuk, 1987, 1992). Etiolation has been used effectively on its own or in combination with stem banding (a localized form of etiolation), to increase rooting in a range of species that are otherwise difficult to propagate vegetatively (Harrison-Murray and Howard, 1982; Maynard and Bassuk, 1985).

The goal of this study is to develop a reliable system for cutting propagation of *Quercus* species, using a combination of factors known to enhance adventitious root formation. Experiment 1 investigated the effects of light and stem banding pretreatments on rooting in *Q. bicolor* and *Q. robur* cuttings. Experiments 2 and 3 compared the effects of light and stem banding pretreatments on rooting in *Q. bicolor* and *Q. macrocarpa* cuttings as cuttings aged over a 9- and 4-week period, respectively.

MATERIALS AND METHODS

Experiment 1. The Effect of Light and Stem Banding Treatments on Rooting in *Quercus bicolor* and *Quercus robur*.

On 28 Jan. 2005, 3- to 5-year-old dormant containerized stock plants of *Q. robur* and *Q. bicolor* were brought out of the cooler (after having received 3 months of chilling at 2.2 °C/36 °F), into a warm temperature greenhouse 23.9 °C/18.3 °C day/night (75 °F/ 65 °F day/night).

The plants were immediately watered and shoots were removed leaving a 4 cm (~1.6 in.) stump above the soil surface. Thirty-three *Q. robur* and 94 *Q. bicolor* stock plants were used. The *Q. bicolor* plants were randomly assigned to one of three treatment groups (light only, light+banding, and etiolated + banding) and *Q. robur* plants were randomly assigned to one of two treatment groups (etiolated + banding and light + banding). To ensure maximum budbreak all stockplant stumps were treated with 500 ppm $GA_{(4+7)}$, repeated every 4th day until budbreak (Amissah and Bassuk, 2004).

Upon budbreak, plants were either etiolated (98% light exclusion) or grown in natural light until shoots reached a 6 cm height, which marked the start of a 3-week treatment period in which shoots were either banded or left unbanded. Stockplants to be etiolated were placed beneath black cloth tents and allowed to grow in near darkness (98% light exclusion). Etiolated shoots on reaching the desired 6 cm height were banded and then placed under 80% shade. Shoots were then weaned to light over a period of 1 week.

Banding was achieved by firmly pressing a 3 × 3 cm Velcro™ strip around the base of the etiolated or light grown shoots. To further maintain uniformity and regulate growth, shoots in all treatment groups had their apical bud removed once the shoot reached a height of ~12 cm. At the end of the 3-week treatment period cuttings were harvested. Cuttings had two leaves each, larger leaves were trimmed and the freshly cut bases dipped in 6,000 ppm of IBA dissolved in 50% aqueous ethanol for 10 sec. After the IBA solution had dried, cuttings were stuck into a horticultural grade perlite medium and sprayed weekly with Banrot® [15% Etridiozole - (5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole), 25% Thiophanate-methyl (dimethyl [(1,2-phenylene) bis (iminocarbonothioyl)] bis [carbamate])] as a precaution against fungal attack. Cuttings were rooted under intermittent mist (6 sec every 8 min) from 6:00 AM – 9:00 PM and shaded (65% shade). Data was taken after 4 weeks on the number of cuttings that rooted per treatment group and the number of roots per rooted cutting (NRPC).

Experiment 2. The Effect of Light and Stem Banding Pretreatments and Phenological Age (Over a 9-Week Period), on Rooting in *Quercus Macrocarpa* and *Quercus Bicolor* Cuttings.

On 6 May 2005, 8-year-old, field-grown dormant trees of *Q. bicolor* and *Q. macrocarpa* plants were cutback leaving a 4 cm (1.6 in.) stump above the soil. Treatments were randomly assigned to cutback plants before budbreak. Upon evidence of bud swelling on the cutback stem, half the stumps were etiolated (grown in the dark, LI-189 portable light meter reading of 0.1 $\mu mol \cdot m^{-2} \cdot s^{-1}$) and the other half left uncovered (grown in natural light). On reaching a height of 6 cm, which marked the start of shoot treatment, shoots arising from half the stumps in each group (natural light or etiolation) were banded with Velcro™, while shoots from the other half were left unbanded. Shoots arising from a third of the stumps in each treatment group were allowed to grow for 3, 6, or 9 weeks at which time cuttings were taken and rooted under mist. All shoots had their apical buds removed once the shoots reached a height of ~12 cm. Etiolation of cutback stumps was achieved by inverting a #3 pot (covered with aluminum foil to prevent heat buildup), over the stumps and the inverted pot held in place with a concrete block. Weaning of etiolated field plants to full light was as described by Amissah (2007). Stem banding, harvesting and rooting conditions of cuttings were the same as in Experiment 1 except cuttings were not sprayed with Banrot®. Data was taken after 5 weeks on the number of cuttings that rooted per treatment group and the NRPC.

Experiment 3. Effect of Light and Stem Banding Pretreatments and Phenological Age (Over a 4-Week Period), on Rooting in *Quercus macrocarpa* and *Quercus bicolor* Cuttings.

On 13 April 2006, shoots were removed from dormant previously cutback *Q. bicolor* and *Q. macrocarpa* trees (9-year-old field plants). Plants were randomly assigned one of four treatments (light, light+banding, etiolated, etiolated+banding). Twelve to fifteen trees were used per treatment per cutting age per species. Shoot treatments were the same as in Experiment 2. Cuttings were harvested 2, 3, or 4 weeks after light and stem banding pretreatments and rooted under mist.

Experimental Design and Statistical Analysis of Results. Cuttings were stuck using a completely randomized design (CRD). Data was analyzed using the PROC GENMOD procedure in SAS (SAS Computer Software, 2002–2003). Rooting and number of roots per cutting (NRPC) were analyzed using the binomial and negative binomial regression models, respectively. Means separation was done using Least Square Means.

RESULTS

Light and stem banding pretreatments using stock plants grown in the greenhouse, had no significant ($p > 0.05$) influence on rooting ability and NRPC in both *Q. bicolor* and *Q. robur* cuttings (Table 1). Nevertheless, percent rooting of cuttings averaged 89.4% and 63.8% across treatments groups (Table 1). In Experiment 2, cuttings age ($p < 0.0001$) and light treatments ($p < 0.01$) significantly influenced rooting in *Q. bicolor*, while banding did not. Percent rooting in *Q. bicolor* decreased with increasing shoot age over the 9-week treatment period, and was significantly ($p < 0.0001$) higher in cuttings at 3 weeks (62.3%) relative to those at 6 (10.3%) or 9 weeks (4.7%) (Table 2). The highest rooting percent and NRPC (79.4%, 8.4) was found in *Q. bicolor* cuttings etiolated and harvested at 3 weeks (Table 2). However, in *Q. macrocarpa* only one cutting rooted (data not shown).

Rooting ability in *Q. bicolor* cuttings was significantly influenced by light ($p < 0.0001$), stem banding ($p < 0.05$), and cutting age ($p < 0.01$) treatments in the second field experiment. Rooting response was highest in 2-week-old etiolated *Q. bicolor* (80.5%) and *Q. macrocarpa* (41.2%) cuttings compared to the other treatments (Table 3). Rooting response in etiolated *Q. bicolor* cuttings decreased after the third week with significant differences found between etiolated cuttings treated for 2 and 4 weeks ($p < 0.001$) and between those treated for 3 and 4 weeks ($p < 0.05$) (Table 3). Mean NRPC in *Q. bicolor* was significantly ($p < 0.05$) higher in etiolated cuttings at 2 weeks (10.0) compared to light-grown (5.2) cuttings (Table 3). In *Q. macrocarpa*, cutting age ($p < 0.01$) and light treatments ($p < 0.0001$) significantly influenced rooting. Rooting percent over the 4-week treatment duration, decreased with increasing cuttings age and was highest in cuttings treated for 2 weeks (24.2%) declining to 16.1% and 11.0% by the 3rd and 4th weeks, respectively (Table 3).

DISCUSSION

Percent rooting was generally high in cuttings taken from greenhouse-grown *Q. bicolor* (89.4%) and *Q. robur* (63.8%) stock plants. Light and stem-banding pretreatments in the greenhouse did not significantly influence rooting ability, NRPC or length of the longest root (LLRT), unlike in cuttings taken from stock plants in the field, where etiolation significantly enhanced rooting. In a review on light and propagation, Stoutemeyer (1961) also noted that cuttings taken from stock plants grown under glass, usually rooted much more freely, compared to the same species grown outdoors. The difference in rooting, he speculated, could be a result of greater humidity or higher temperatures under glass, or the screening out of ultra-violet rays by the glass. Similar observations have been made by Maynard and Bassuk (Maynard and Bassuk, 1990). Possibly the difference in rooting ability between cuttings from the two environments could be a result of variations in mechanical perturbation (MP) — due to wind movement in the two environments, a phenomenon known as thigmomorphogenesis. Research has shown that responses to MP are species-specific, usually resulting in decreasing stem elongation, increasing stem thickness and lignification (Larson, 1965; Telewiski and Jaffe, 1986).

Generally, percent rooting in *Q. macrocarpa* cuttings were low compared to those of *Q. bicolor* treated in the same way (Table 3). Similar differences in rooting ability have been reported between oak species (Griffin and Bassuk, 1996). The low rooting response in *Q. macrocarpa* could be attributed partly to this species having a "narrower window" of rooting opportunity. Rooting in *C. retusus* has been reported to occur only during a narrow 2-week period in spring (Stoutemeyer, 1942). It is our observation that cuttings taken from field-grown plants mature faster than those from greenhouse-grown plants and may therefore respond better to shorter treatment durations (< 3 weeks). Consequently, shortening the treatment duration from 3 to 2 weeks in Experiment 3, improved the rooting response in *Q. macrocarpa* cuttings. Rooting ability in *Q. bicolor* decreased as cuttings aged from 62.3% at 3 weeks to 10.3% at 6 weeks and finally 4.7% by the 9th week. Morgan et al. (1980) observed a similar pattern in *Q. virginiana* cuttings.

Overall, etiolated cuttings rooted significantly ($p < 0.05$) better and had more NRPC than those grown in natural light. Etiolation was shown to be an important pretreatment for rooting field-cuttings (Tables 2 and 3). Other researchers have reported increased rooting ability in etiolated cuttings (Maynard and Bassuk, 1992; McGuighan et al., 1996).

CONCLUSION

Our research findings show that cutting propagation of oaks is possible. Differences in rooting ability were observed between cuttings (of the same species), taken from plants grown under glass and those grown outdoors. Etiolation notably enhanced rooting in field-grown cuttings, although not in greenhouse-grown shoots. Percent rooting decreased as cutting age increased in *Q. macrocarpa* and *Q. bicolor*.

Table 1: Experiment 1. The effect of light and stem banding treatments on rooting, number of roots, and length of the longest root in *Quercus bicolor* and *Quercus robur* cuttings.

	Rooting (%)	Total no. of cuttings	Ave. no. roots/cutting	Length of longest root (cm)
Quercus bicolor				
Etiolated+banding	91.2a	11.6a	4.8a	68
Light+banding	87.8a	12.1a	3.9a	74
Light only	89.2a	14.5a	4.0a	37
Quercus robur				
Etiolated+banding	64.9a	9.6a	3.7a	37
Light+banding	62.7a	11.1a	3.0a	51

Treatments means were not significantly ($p > 0.05$) different from each other within species. Etiolated + banding: refers to cuttings that were etiolated and then banded; Light + banding: refers to cuttings that were grown in natural light and then banded and Light only: refers to cuttings grown in natural light without banding.

Table 2: Experiment 2. Effect of treatments on rooting and number of roots per cutting over a 9-week period in *Quercus bicolor*.

	Cutting age/treatment duration (weeks)					
	3		6		9	
	Treatments					
	Light	Etiolated	Light	Etiolated	Light	Etiolated
Rooting (%)	48.3b	79.4a	7.7e	13.2c	0	8.0e
Rooting/cutting age[y] (%)	62.3a	10.3b	4.7c			
Ave. no. roots/cutting[z]	4.7b	8.4a	3.5b	3.9b	0	6.6ab
Total number of cuttings	56 (29)[x]	46 (32)	50 (4)	46 (8)	45 (0)	64 (5)

[x] Number of cuttings that rooted. The different letters represent treatment means significantly different ($p \leq 0.05$) from each other between treatments; means were separated using least square means.

[y] The value represents the weighted average of percentage rooting.

[z] Average number of roots per cutting.

Table 3: Experiment 3. Effect of treatments on rooting and number of roots per cutting as cuttings aged over 4-week period.

	Cutting age/treatment duration (weeks)					
	3		6		9	
	Treatments					
	Light	Etiolated	Light	Etiolated	Light	Etiolated
Quercus bicolor						
Rooting (%)	37.8bc	80.5a	32.8bc	74.3a	22.8c	45.8b
Rooting/cutting age[x] (%)	58.4a	55.4a	31.8b			
Ave. no. roots[z]	5.2b	10.0a	5.3b	7.4a	5.8b	6.4ab
Cuttings no./ treatment	84 (31)[y]	79 (62)	60 (20)	72 (57)	86 (21)	55 (24)
Quercus macrocarpa						
Rooting (%)	13.3b	41.2a	5.73c	26.1a	6.8bc	14.1b
Rooting/cutting age[x] (%)	24.2a		16.1b		11.0b	
Ave. no. roots[z] *	3.1	7.5	2.7	7.0	3.4	4.1
Cuttings no./ treatment	136 (16)	87 (40)	83 (3)	87 (28)	73 (9)	99 (9)

[x] The value represents the weighted average of percentage rooting.

[y] Number of cuttings that rooted.

[z] Average number of roots per cutting;

*Treatments had no significant ($p > 0.05$) effect on the number of roots per cutting. The different letters represent treatment means significantly different ($p \leq 0.05$) from each other between treatments; means were separated using least square means.

PROTOCOL FOR ROOTING *QUERCUS* CUTTINGS

After plants receive their required chilling

Duration Sequence

Day 0: Cutback of dormant plant
Day 7: Bud break /etiolation
Day 14: Banding of shoots at 6 cm for 2 weeks
  Pinch shoots at 12 cm
Day 28: Treat cuttings (IBA) and mist
Day 58: Harvest/pot-up (shade)

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