

STRANGULATION PRE-TREATMENT EFFECT ON THE DEVELOPMENT AND ROOTING OF FASCICLE CUTTINGS OF *PINUS RADIATA*

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ABSTRACT

The effect of strangulation of the stems of 2- and 7-year-old *Pinus radiata* D. Don trees on the development and rootability of fascicle shoots was examined. Strangulation of the stems of juvenile *P. radiata* may be beneficial, in terms of subsequent development of shoots from needle fascicles, and speed and percentage of rooting. Both the development and rootability of needle fascicle shoots declined with increased spacing of strangulation along the stem.

Strangulation of stems of 7-year-old trees was done at different crown positions, using clones that were easy and difficult to root as stem cuttings. Fascicle bud development was better in the upper part of the crown but strangulation had no significant effect. Needle fascicle development varied with clone, and there was no significant difference between easy- and difficult-to-root clones. None of the fascicle buds from 7-year-old trees had rooted 90 days after setting.

Keywords: vegetative propagation; pre-treatment; fascicle cuttings; rooting; *Pinus radiata*

INTRODUCTION

Pines are generally regarded as being amongst the more difficult plants to root from stem cuttings, except for a few species including *Pinus radiata*. Experiments with cuttings of *P. radiata* have shown that the rooting ability of cuttings decreases with increasing age of the tree from which they were obtained (Allsop 1950; Cameron 1968; Field 1934; Fielding 1954, 1964; Jacobs 1939; Libby & Conkle 1966; Pawsey 1950; Sherry 1942; Thulin & Faulds 1968; Menzies *et al.* 1988). Often only a small number of stem cuttings can be obtained from a clone, especially if there are only a few ramets available.

The problem of obtaining large numbers of propagules can be overcome in *P. radiata* by using needle fascicles, which can be collected in large numbers over several years. Needle fascicles from juvenile trees can be rooted reliably under suitable environmental conditions,

require less space than stem cuttings, but may be more expensive than larger cuttings grown outdoors because of the cost of rooting facilities (Menzies *et al.* 1988).

There can be problems in obtaining even-sized fascicle cutting material in a given time period. Fascicle shoots are stimulated to develop by removal of the terminal bud (Isikawa & Kusaka 1959). However, the apical fascicle shoots develop more quickly than those near the base of the branch or stem, and soon suppress further development of the basal fascicle shoots because of apical dominance (Cooperrider 1938). There can also be problems in obtaining suitable bud development and subsequent rooting of fascicle shoots from older trees, as occurs with stem cuttings from older trees (Isikawa & Kusaka 1959; Toda 1948a, b).

Rooting of pine needle fascicles has been carried out in numerous experiments (Andrews 1980; David 1964; David & Launay 1966; Hare 1965; Kummerow 1966; Kummerow & Schmidt 1967; Libby & Conkle 1966; McDonald & Hoff 1969; Rudolph & Nienstaedt 1964; Sargento & Barker 1978; Struve & Blazich 1984; Wells & Reines 1965; Yli-Vakkuri & Pelkonen 1976), although few have examined physical pre-treatments to induce a large number of even-sized needle fascicles from the stem and branch in a given period.

This paper reports the results of three experiments designed to study:

- (1) The effect of stem strangulation on stimulation and development of needle fascicles on 2-year-old seedlings;
- (2) Clonal variation in and effect of crown position on development of needle fascicles after strangulation on 7-year-old hedged trees;
- (3) Effect of strangulation on rooting of needle fascicle cuttings from 2-year-old seedlings and 7-year-old hedged trees.

METHODS

The ortets used in this study were 2- and 7-year-old *P. radiata* seedlings and hedged trees growing in the nursery at the Forest Research Institute, Rotorua, New Zealand. The 2-year-old seedlings were used to test the effect of strangulation spacing along the ortet stem on fascicle shoot development and subsequent rooting, compared to a non-strangulated control, and to test the effect on developing needle fascicles from the strangulated ortets which were pinned down to the ground and covered with pumice soil, 1 cm thick.

The 7-year-old hedged trees were divided into two groups of clones that were easy- and difficult-to-root by conventional stem-cutting propagation methods. Four ramets of each of eight clones planted in rows were selected from the Long Mile Clonal Archive. Clones 2, 14, 16, and 228 were classified as easy-to-root and Clones 5, 7, 93, and 107 as difficult-to-root. Strangulation treatments were carried out on the first-order branches at four different crown positions (upper, upper middle, lower middle, and lower) and development of the fascicle buds was studied.

To stimulate development of fascicular shoots, terminal buds of the ortets used were removed, and stems were strangulated with 0.5-mm copper wire at intervals of 3, 6, 9, and 12 cm, respectively. The control treatment had the terminal bud removed, but there was no strangulation. The cuttings were collected after 40 days by removing the required length of treated stem, with the fascicle shoots attached. They were sealed in polythene bags and placed in a refrigerator for up to 12 hours until the fascicle buds were excised for setting. Fascicles with pre-developed shoots were excised from the plant stems of the 2-year-old

seedlings and from the branches of the hedged trees with a sharp scalpel blade and placed in a shallow bowl of 0.5% Benlate fungicide solution for 10 minutes. They were then set into seed trays (rooting medium 50:25:25 by volume peat, pumice, and perlite) in a randomised complete block design using four replications, each with 20 fascicle cuttings. The trays were placed in a sealed polythene tent in a temperature-controlled glasshouse (min 7°C, max 25°C) until they rooted. The cuttings were set on 27 December 1988, and rooting success was assessed 90 days later.

RESULTS

Elongation of Needle Fascicle Shoots in Juvenile *P. radiata*

Number of elongated fascicle shoots

Analysis of variance (ANOVA) was done on the effect of soil cover and the effect of strangulation on the number of elongated fascicle shoots. There was no significant difference in the number of elongated fascicle shoots on pinned-down seedlings between the uncovered and soil-covered treatments, but the effects of strangulation treatment, strangulation spacing, and the interaction between the two treatments were significant at the 1% level. With the non-strangulated control, there was no elongation of shoots below 3 cm from the top (Table 1). Stem strangulation increased the number of elongated shoots, with the 3-cm spacing producing a better response than the 6-cm and 12-cm spacings. Only needle fascicles from the strangulation treatment at 3-cm spacing showed bud development beyond 9 cm from the top. The strangulation treatment at 12-cm spacing was similar to the other two treatments in the number of developed shoots at 3 cm from the top, but with no fascicle shoots below 6 cm from the top.

Length of induced fascicle shoots

The effect of strangulation on the length of fascicle shoots was similar to that on the numbers of fascicle shoots (Table 2). With the non-strangulated control, fascicle shoots grew for an average length of 5.7 mm on the 0–3 cm length of stem from the decapitated top but no fascicle shoots were observed below that zone. With the strangulation treatment at 3-cm intervals, fascicle shoots developed uniformly with an average length of 3.7–4.9 mm from the bottom to the top in each of four strangulated zones. With strangulation treatments at 6 cm and 12 cm spacing, the needle fascicles did not develop at 9–12 cm and 6–12 cm, respectively.

Rooting of Fascicle Cuttings from Juvenile *P. radiata*

Fascicle cuttings were taken from stems strangulated with 0.5-mm copper wire at spacings of 3, 6, and 12 cm respectively, and a non-treated control. An analysis of variance for the percentage of rooted cuttings 90 days after setting showed that the effect of strangulation on rooting was highly significant ($p < 0.001$).

There was no significant difference in rooting percentages between the 3-cm and the 6-cm strangulation treatments (Table 3). However, cuttings from these treatments showed higher rooting than those from the 12-cm treatment or the control. Even the 12-cm spaced treatment gave a higher rooting percentage than the control.

TABLE 1—Average number of elongated fascicle buds per tree

Strangulation spacing (cm)	Distance from the top (cm)			
	0–3	3–6	6–9	9–12
3	11 a	9 c	6 e	5 f
6	11 a	8 d	5 f	0 h
12	10 bc	4 g	0 h	0 h
Control	10 ab	0 h	0 h	0 h

Means followed by the same letter are not significantly different (LSD test $p=0.05$)

TABLE 2—Average length of elongated fascicle buds (mm)

Strangulation spacing (cm)	Distance from the top (cm)			
	0–3	3–6	6–9	9–12
3	4.9 bc	4.5 c	3.9 d	3.7 d
6	4.9 bc	4.7 c	3.8 d	0 f
12	5.4 ab	2.7 e	0 f	0 f
Control	5.7 a	0 f	0 f	0 f

Means followed by the same letter are not significantly different (LSD test $p=0.05$)

TABLE 3—Effect of strangulation on rooting of fascicle cuttings from juvenile *Pinus radiata*

Strangulation spacing (cm)	Mean rooting (%)	5% LSD-test
3	91.4	a
6	86.2	a
12	63.8	b
Control	51.3	c

Means followed by the same letter are not significantly different.

Strangulation Treatments on 7-year-old Hedged Trees

The results of an ANOVA on the number of developed fascicle shoots for four different strangulation treatments at four crown positions using four easy-to-root (No. 2, 14, 61, and 228) and four difficult-to-root clones (No. 5, 7, 93, and 107) were as follows.

Effect of strangulation

On average only four fascicle buds were stimulated per branch, but there was no significant difference between the four strangulation treatments (Table 4). None of the

TABLE 4—Average number of fascicle shoots developed on branches of 7-year-old hedged trees after strangulation at four heights in the crown

Strangulation spacing (cm)	Crown position			
	Upper	Upper middle	Lower middle	Lower
3	8.0	6.9	0.9	0.4
6	7.9	6.8	3.6	0.6
12	8.0	4.9	2.4	1.1
Control	10.1	3.7	1.2	0.0

strangulation treatments was effective in stimulating fascicle bud development and there were no significant interactions between strangulation treatments and crown position. It was apparent that strangulation was not effective regardless of position in the crown.

Clonal variation

The effect of clonal variation was highly significant at the 1% level. Clone 14 developed most fascicle shoots, while Clones 5 and 61 developed fewest shoots (Table 5). However, there was no significant difference between the easy and difficult rooting groups of clones as shown by the result of an orthogonal contrast analysis.

Crown position

The effect of crown position on development of fascicle shoots was significant ($p=0.01$). More fascicle shoots developed higher up the crown (Table 6).

Effect of strangulation on rooting of fascicle cuttings

None of the fascicle cuttings from 7-year-old hedged trees formed roots within 90 days of setting.

TABLE 5—Average number of fascicle shoots developed

Easy-to-root clones		Difficult-to-root clones	
Clone No.	Mean	Clone No.	Mean
14	10.3 a	7	5.6 b
228	4.0 bc	93	4.9 b
2	3.9 bc	107	2.0 cd
61	1.3 d	5	1.3 d

Means followed by the same letter are not significantly different (LSD test $p=0.05$)

TABLE 6—Crown positions in the average number of developed fascicle shoots

Crown position	Mean	5% LSD-test
Upper	8.5	a
Upper middle	5.6	b
Lower middle	2.0	c
Lower	0.5	d

Means followed by the same letter are not significantly different.

DISCUSSION

Mass propagation in pine species can be achieved with needle fascicle cuttings. However, the method has been limited partly by difficulty in rooting, especially in obtaining uniform shoot development. Both chemical and physical treatments have been reported to stimulate fascicle shoot production. Chemical treatments with BAP on pine species (Inglis 1984; Mulgrew & Williams 1984; Stiff & Boe 1985), BAP in the presence of either Silwet L-77 or Citowett on *P. radiata* (Aitken-Christie & Coker 1986), cytokinins or gibberellic acid on *P. radiata* (Kummerow & Schmidt 1967; Wells & Reines 1965), and kinetin plus adenine on *P. pinaster* Ait. (David & Launay 1966) have been reported to have stimulating effects on the development of fascicle buds. However, most of the results should be considered tentative as often subsequent rooting was not tested or investigations are continuing.

In stimulating the development of fascicle shoots, physical methods are more practical; these include removal of the apical meristems (Andrews 1980; Kummerow 1966; Libby & Conkle 1966; McDonald & Hoff 1969; Mergen & Simpson 1964; Rudolph & Nienstaedt 1964; Struve & Blazich 1984; Yli-Vakkuri & Pelkonen 1976), pinching the branch tip (Hare 1965), and clipping the terminal (Mergen & Simpson 1964). Elongation of fascicle buds takes place in the form of reconstruction growth after the loss of the terminal bud of a main stem or lateral branch, probably through removal of the apical dominance (Cooperrider 1938; Kummerow 1966). One of the problems with these treatments on pine species is that the fascicles closest to the cut top of stem have more growth activity than the fascicles in a lower position on the same stem (Mergen & Simpson 1964). This study showed that decapitation of the apical meristem and strangulation by copper wire can greatly stimulate development of fascicle shoots down to a lower position on the treated stem. Strangulation at 3-cm intervals was the best treatment to increase the number of uniformly developed fascicle buds 4–5 mm in length over the full stem length of 12 cm.

These results are similar to those achieved by girdling. Girdling involves the removal of a band of phloem tissue, causing an accumulation of photosynthates and growth substances in the stem (Miller & Berryman 1986). Strangulation treatment of the stem would also interrupt basipetal transport through phloem tissue.

It is not known why strangulation pre-treatment of needle fascicles improves rootability. Removal of the terminal buds from cuttings at the time of pre-treatment has an important effect on root initiation (Kozlowski 1962), as has clipping of the terminal buds. Possibly it enhances a growth hormone (Hong 1969) which stimulates growth in the bud before collection and is also beneficial to rooting (Mergen & Simpson 1964). This rooting stimulation could be due to changes in both nutrients and auxins in the treated stem. There have been conflicting reports about these factors. Struve & Blazich (1984) reported a positive relationship between carbohydrate levels in fascicle cuttings and subsequent rooting, but other researchers have been unable to predict a rooting response based on foliar carbohydrate and mineral nutrient analyses (Childers & Snyder 1957; Reins & Bamping 1962, 1964). Rudolph & Nienstaedt (1964) found that application of rooting hormone did not increase rooting of fascicle buds when tried without bottom heat. Roots were not formed when IAA (indole acetic acid) alone was used (Sargento & Barker 1978) and they decreased with NAA (alpha-naphthalene acetic acid) on 7-year-old *P. densiflora* Sieb. & Zucc. (Toda 1948a), but beneficial effects of auxins on the rooting of fascicle shoots have been reported with IBA (indole butyric acid) (Andrews 1980; Kummerow 1966; Mergen & Simpson 1964;

Rudolph & Nienstaedt 1964; Sargento & Barker 1978), IAA (Mergen & Simpson 1964; Yli-Vakkuri & Pelkonen 1976), and thiamine (Struve & Blazich 1984) and with NAA (Thimann & Delisle 1942). The improved rooting stimulated by strangulation may be a synergistic effect of various phytohormones accumulated in the treated stems.

Kummerow (1966) reported that a high rooting capacity seemed to be correlated with the use of large fascicle buds. Buds from the central part of stems at six different positions in the crown of 2.5-year-old trees were significantly larger than those from the upper or lower sections, and they had a higher rooting capacity.

In this study of 7-year-old hedged trees, the development of fascicle buds at four different positions was greater for the upper than the lower position but with no successful rooting, while the rooting percentage of needle fascicles from juvenile stems was greater with the larger needle fascicles from strangulation at 3-cm intervals. These results are similar to those of Kummerow (1966). He found that cuttings of larger needle fascicles had better rooting. Auxin treatment may be beneficial for this older material.

There was no significant difference in the development of fascicle buds between the easy- and difficult-to-root groups of clones, but there were individual clonal differences in fascicle shoot development. This observation of clonal differences for shoot development has also been reported by Hare (1965), who found that fascicle bud development as well as rooting was markedly affected by clone. Strangulation was effective on juvenile ortets but not effective on 7-year-old hedged trees of *P. radiata* regardless of crown position.

A report on similar treatments with IBA plus sucrose on eastern white pine (*Pinus strobus* L.) and other five-needle pines showed that the treatment effects varied with juvenile and adult cuttings (Deuber 1942).

CONCLUSIONS

Strangulation of the stem of juvenile *P. radiata* was effective in increasing the number of developed fascicle buds over a longer length of stem. Strangulation at a spacing of 3 cm along the stem was more effective than wider spacing. Rooting of the fascicle buds was better with the 3- and 6-cm spaced strangulation than with 12-cm spacing or no strangulation.

Strangulation was not effective in stimulating bud development on 7-year-old hedged trees, and there was an average of only four fascicle buds stimulated per length of stem. There was a significant difference between clones in the number of buds stimulated, and more fascicle buds developed on material from higher up in the crown. However, none of the fascicle buds formed roots within 90 days of setting.

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