Propagation of Radiata Pine Plants for Plantation Forestry

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Bareroot seedlings of radiata pine (*Pinus radiata* D. Don) are the main planting stock used in New Zealand at present. These are produced using open-pollinated seed from clonal seed orchards. Control-pollinated seed of the best progeny-tested families is also now being commercially produced. However, since this seed is expensive and still in short supply, vegetative propagation methods have been developed to amplify available planting stock.

Many tree nurseries grow rooted cuttings, either using juvenile stool beds to raise cutting material or making field collections of cuttings from young plantation trees. Stool-bed cuttings are more expensive to produce than seedlings, if the cost of seed is ignored, because of the cost of maintaining the stool beds and of manual collection and setting. Currently, however, the high cost of control-pollinated seed makes stool-bed cuttings cheaper to produce than seedlings. Improvements in stem form have been demonstrated with cuttings compared with similar genetic quality seedlings, particularly with field-collected cuttings planted on fertile farm sites. This has created a demand for field-collected cuttings, even though they cost nearly double the price of stool-bed cuttings.

One New Zealand company is using micropropagation to produce plantlets for establishing clonal plantations, and embryogenesis technology is being evaluated by another company. Currently, these technologies are expensive, but can give high multiplication rates, and also have the advantage that juvenility can be maintained by cold storage or cryopreservation during clonal field testing.

Integration of these tree improvement and propagation technologies is allowing New Zealand to advance towards clonal forestry with radiata pine.

INTRODUCTION

The area of plantation forest in New Zealand now exceeds 1.3 million ha. The planting rate has varied over the years, with planting booms in the late 1920s, early 1930s, 1960s, 1970s, and 1980s (Ministry of Forestry, 1993). The rate of new planting decreased in the late 1980s due to increased land values, the sale of state forest assets, and taxation changes. However, new planting has since increased again, and the area of new forests planted in 1994 probably exceeded 100,000 ha.

Radiata pine has been the main species planted, followed by Douglas-fir, hardwoods, and other softwoods (Table 1). This general trend is expected to continue, although there is increased interest in Douglas-fir, eucalypts, cypresses, and acacias.

Plantations have traditionally been established using seedlings raised in bareroot nurseries. For radiata pine, sufficient seed for New Zealand use is produced in seed orchards, while for other species, most seed is collected from seed stands or imported. More than 7000 kg of open-pollinated radiata pine seed was collected from seed orchards in 1993 (Vincent, 1993). A limited quantity of better genetic quality seed is produced by control-pollination amongst the best progeny-tested families in the breeding programme, but the amount available is less than 5% of the total seed collected (Vincent, 1993). This seed is also very expensive, and so vegetative propagation methods have been developed to amplify plant numbers, so that a larger forest area can be planted with this superior material. There are a number of amplification options available, which differ in multiplication rate, time required to produce planting stock, and cost (Menzies et al., 1985a). Cutting options available include both the use of juvenile stool beds to raise cutting material and field collection of cuttings from young plantation trees. These methods are used by a number of nurseries in New Zealand to produce more than 10 million cuttings annually. Another option is to use micropropagation (Horgan and Aitken, 1981; Aitken-Christie and Thorpe, 1984; Smith, 1986), and one company in New Zealand is using this technology to produce up to 3 million plants annually, as well as using cuttings (Gleed, 1991). Embryogenesis is a more recent technology and methods developed at New Zealand Forest Research Institute (NZ FRI) are now being evaluated in pre-commercial production by a New Zealand forest company. These techniques will be discussed in this paper.

Table 1. Area of stocked exotic forest in New Zealand as at 1 April 1992.

	All estate area (ha)	%	Age class 1-5 yr
Radiata pine	1,176,539	90	93.0
Douglas-fir	66,625	5	3.4
Other softwoods	38,717	3	0.7
Hardwoods	25,760	2	2.9
Total area	1,307,641	100	100.0

Source: Ministry of Forestry (1993)

PRODUCTION OF BARE-ROOT SEEDLINGS

Most seedling crops in New Zealand are raised as 1-year-old crops, with seed being sown in spring (Oct.-early Nov.) and the seedlings being lifted the following winter (June-Aug.).

The two most critical aspects affecting quality of nursery-grown seedlings are spacing in the nursery bed and conditioning by root pruning. Nursery machinery has been developed in New Zealand to mechanise these aspects and ensure that high quality plants can be reliably produced (Menzies et al., 1985b). Spacing is controlled by use of a precision vacuum-drum seed sower, while root conditioning is done with an undercutter/wrencher and a lateral root pruner. The undercutter/wrencher is used to cleanly sever the taproots in summer at a depth of 10 cm. This

encourages lateral root growth while slowing down top growth. A wide tilted blade on the undercutter/wrencher is used subsequently at monthly intervals to break off any sinker roots growing down below the undercutting depth and at the same time aerating the soil. Lateral roots growing across the rows are severed by the rolling coulters of a lateral root pruner once or twice during the wrenching period. This conditioning concentrates new root growth close to the seedling root collar and results in a "balanced" seedling in terms of root to shoot ratio.

Other machinery includes equipment for distributing solid fertiliser accurately between the rows of seedlings and boom sprayers for application of liquid fertilisers, herbicides, fungicides, and insecticides.

This mechanisation means that all nursery operations can be done from the back of a tractor except for the final lifting operation, where hand lifting is preferred.

Lifting and handling practices now include careful hand lifting to minimise root damage, and trimming of lateral roots to 10 cm long. Roots are kept moist by spraying or dipping them in water. Seedlings are packed into rigid containers on the nursery bed, rather than in a packing house, to avoid prolonged exposure. Packing seedlings on their sides prevents tap root damage. The cardboard containers act as storage containers, and also planting boxes out in the field, so the seedlings are not exposed from the time of lifting until they are planted. Seedlings should be stored for no more than 72 h at cool temperatures (2C to 4C).

PRODUCTION OF STOOL-BED CUTTINGS

Production of stool-bed cuttings has been described by Menzies, et al. (1985a) and Dibley and Faulds (1991). Seed is sown as early in the spring as possible. Stock plants are topped in Feb., when they are 4 to 5 months old. The side shoots subsequently develop into stem cuttings, with an average of four cuttings per plant. In subsequent years, the hedged seedlings are topped with a hedge trimmer in late spring, at a height of between 100 mm and 300 mm. Stock plants have been maintained for up to 5 years at the NZ FRI nursery.

Cuttings from stool beds are generally set in raised beds for the production of bareroot plants. They are collected and set in early to mid-winter. The optimum cutting size is 70 to 100 mm in length and >3 mm in diameter. No rooting hormones are required. Smaller cuttings can be set but, because they may be subject to frost lift and soil splash, are best protected with 30% shade cloth.

Cuttings wilt after setting and need irrigation in hot dry conditions. They recover within 4 weeks and begin to gain height and elongate in spring before rooting in early summer. Commencing in mid-summer, the cuttings are conditioned by undercutting, wrenching, and lateral root pruning. The rooted cuttings are ready for planting in winter, 1 year after setting. A yield of at least 80% acceptable plants can be expected.

FIELD-COLLECTED CUTTINGS

Production of field-collected cuttings has been described by Menzies et al. (1985a). Cuttings can be propagated readily from 3- to 4-year-old radiata pine trees in the forest. Plantations suitable for cutting collection have been established with seedlings from special seedlots from the best open-pollinated seed orchard clones (Arnold and Gleed, 1985) or from control-pollinated crosses. Collection from field trees also allows some selection of vigour and form of the parent tree. Tips of lateral

branches with a length between 100 and 150 mm, a minimum diameter of 6 mm, and dense, fully-elongated, healthy needles are collected.

The cuttings are collected and set in early winter during the period of slowest growth, and before the photoperiod starts to lengthen. They are set outside at a depth of 5 cm in raised nursery beds. Overhead irrigation is necessary during warm or dry windy weather, particularly during the first few weeks after setting.

Rooting of field-collected cuttings occurs in spring, a little later than with stool-bed cuttings. Slightly fewer will root successfully, with about 75% forming acceptable root systems.

In trials, field-collected cuttings have performed as well as or better than seedlings of similar genetic quality. On fertile sites, cuttings have shown similar growth rates, but better form than seedlings, providing the physiological age of donor trees has been less than 5 years old. These differences have shown up especially with cuttings taken from donor trees aged 3 years or more, but also with cuttings from stool beds (Fig. 1). The better form of cuttings from field-collected plantations has led to an unsatisfied demand for this type of planting stock.

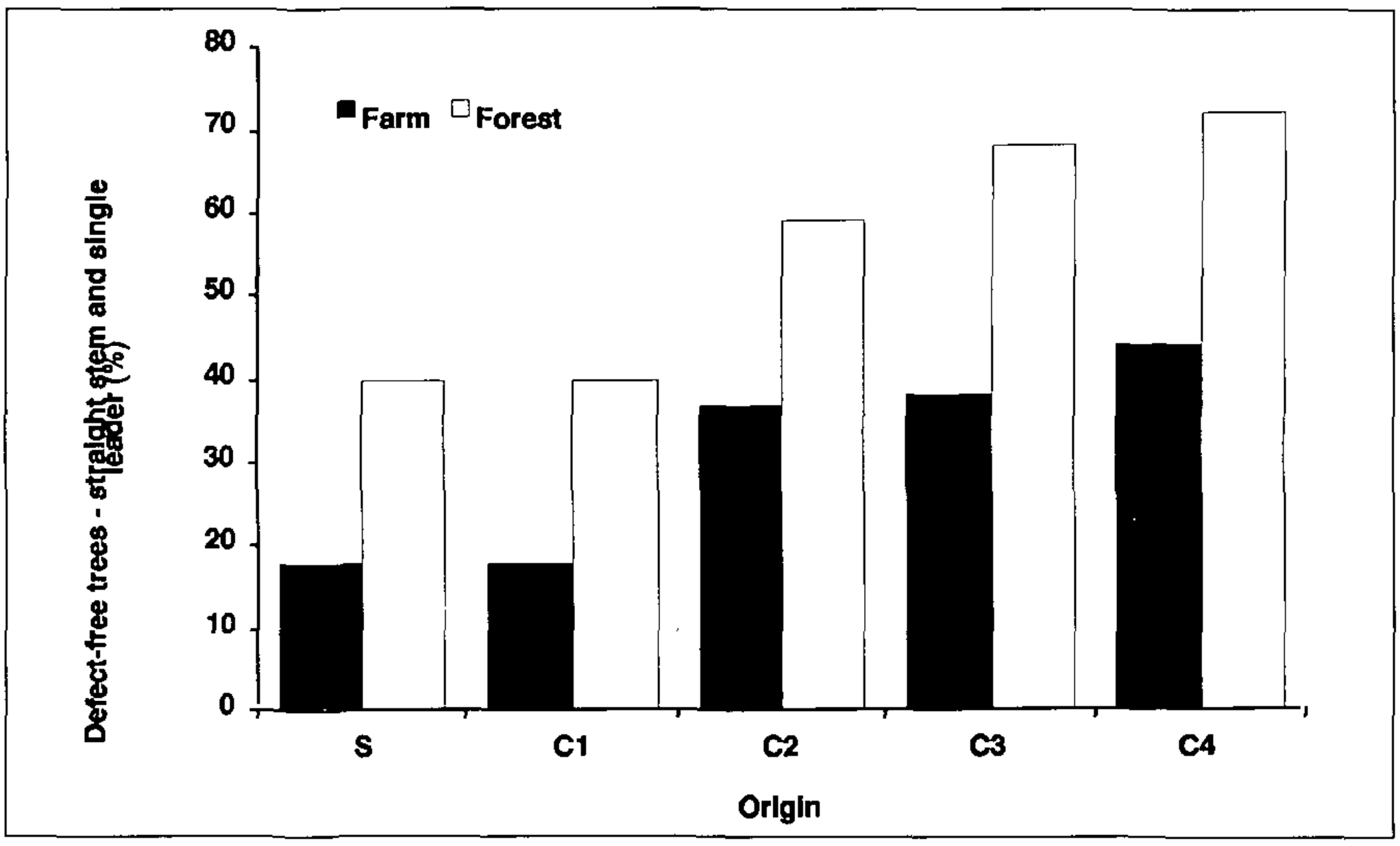

Figure 1. Percentages of defect-free trees at the low pruning stage grown from (a) cuttings from 1- to 4-year-old parent trees and (b) seedlings, on farm and forest sites (Menzies et al., 1991). S = seedling; C1 = cutting from 1-year-old donor tree; C2 = cutting from 2-year-old donor tree; C3 = cutting from 3-year-old donor tree; C4 = cutting from 4-year-old donor tree.

JUVENILE MICROPROPAGATION (ORGANOGENESIS)

While macropropagation methods require the use of large pieces of tissue, micropropagation employs very small plant parts, tissues, or cells. Micropropagation methods include induction of adventitious buds on cultured cotyledons and entire embryos, induction or stimulation of adventitious or axillary buds on cultured shoot tips, regeneration of adventitious shoots and complete plants from unorganised callus and cell cultures, or outgrowth and division of the seedling shoot (epicotyl).

Micropropagation methods have been developed for a variety of radiata pine explants, including embryos, cotyledons, and seedling shoot tips (Horgan and Aitken, 1981; Aitken-Christie and Thorpe, 1984; Smith, 1986). There are four main stages: shoot initiation, shoot elongation, shoot multiplication, and rooting. Tissue is kept sterile through all stages except the last, and is grown in containers on an agar medium containing all the necessary nutrients, hormones, and other substances to support growth. The containers are kept in a controlled environment with artificial lighting. When the shoots are large enough, they are given a hormone treatment to stimulate rooting and set as small cuttings in containers in a greenhouse to form roots. After rooting, they can be lined out in a nursery bed and grown on like seedlings or cuttings (Menzies et al., 1985a).

Tissue-cultured plantlets are currently expensive to produce for two reasons: transfers to fresh media are done manually and the plantlets need to be grown in sterile controlled conditions.

EMBRYOGENESIS

Another propagation technology which is being developed is embryogenesis. Embryogenic cell lines are established from immature seed, and millions of immature embryos of individual genotypes can be multiplied from each seed. These embryos will develop and mature under appropriate conditions, and then can be germinated like natural embryos. The efficiency of this process needs further improvement, but the technology has the potential to produce unlimited quantities of embryos of desirable genotypes, at costs cheaper than current control-pollinated seed prices.

DISCUSSION

There is a wide variation in multiplication rate, time taken to produce planting stock, and cost of the various methods of vegetative propagation. For example, a kilogram of seed will produce enough seedlings to plant approximately 18 ha of

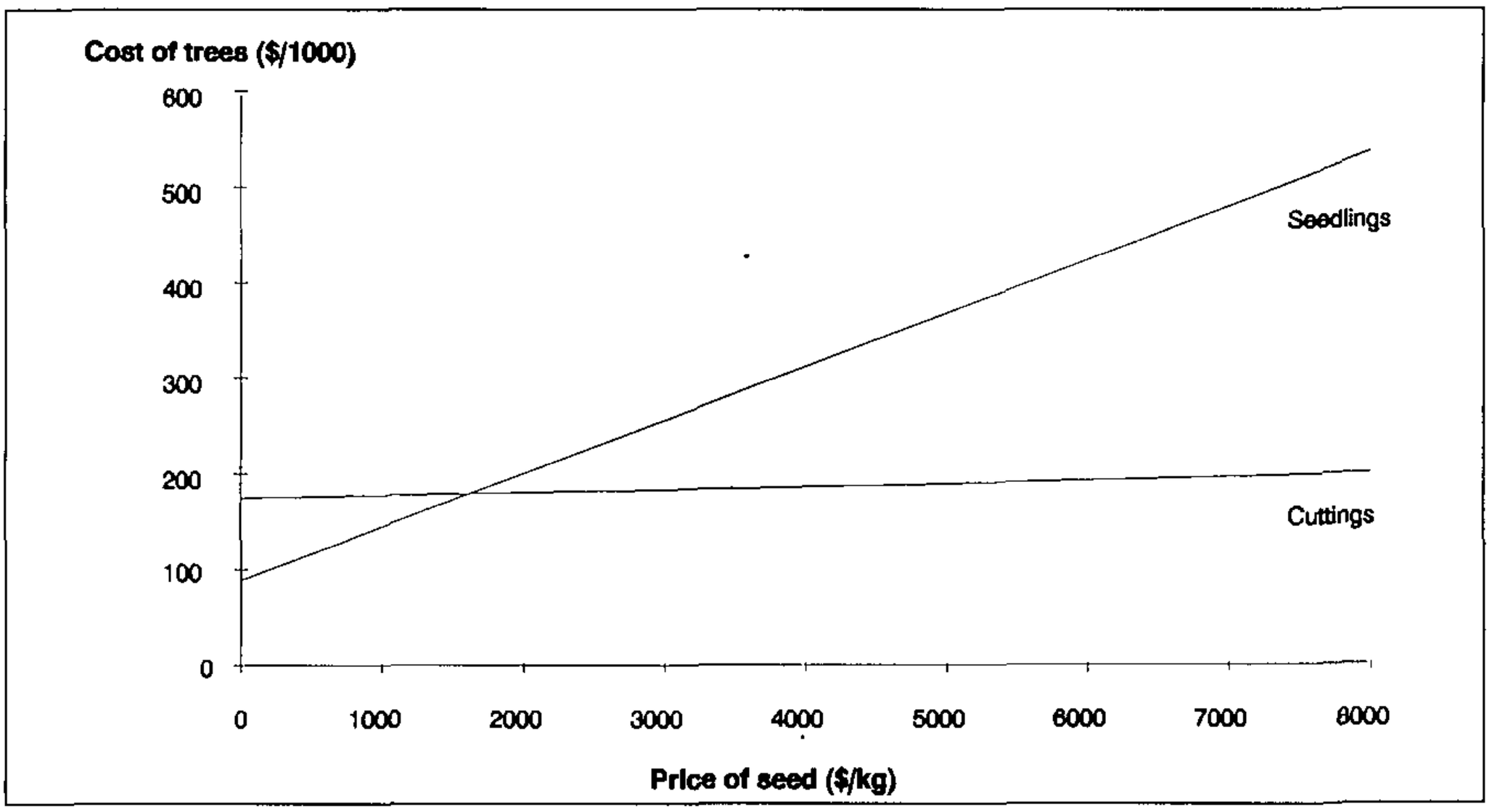

Figure 2. Effect of varying seed prices on the growing cost of seedlings and cuttings (adapted from Dibley and Faulds, 1991).

forest, while the same amount of seed will produce enough cuttings for 360 ha, and enough tissue-cultured plantlets for 3600 ha. However, cutting stool beds will take about 2 years to develop sufficiently to produce this number of cuttings, and a similar laboratory time would be needed for tissue-cultured plantlets.

The approximate growing costs for different options are shown in Table 2. The difference in production cost for open-pollinated and control-pollinated seedlings is the cost of the seed. Disregarding the cost of seed, stool-bed cuttings cost 1.5 to 2 times more than seedlings to produce. However, when the cost of control-pollinated seed is included, stool-bed cuttings of equivalent quality are cheaper to grow. The price of control-pollinated seed has exceeded $6000/kg for the last 2 years and would need to decline to about $1600/kg before seedlings become cheaper to grow than the stool-bed cuttings (Fig. 2).

Table 2. Comparison of growing costs of nursery plants (1994-95).

Stock type	Cost (NZ$)/1000 plants
Seedlings (*open-pollinated seed*)	110
Seedlings (control-pollinated seed)	460
Cuttings from stool beds (control-pollinated seed)	200
Cuttings from field collections	370
Micropropagated plantlets	~700[1]
Cuttings from plantlets	220

[1] Based on Gleed (1993).

Field-collected cuttings are expensive because of the extra costs involved with travel and collection time from plantation trees, and the lower rooting success. However, the form advantages of field-collected cuttings (Fig. 1) have led to an unsatisfied demand for this type of planting stock, despite their higher cost.

Micropropagated plantlets are very expensive because the process is capital and labour intensive. The process does have an advantage of high multiplication rates and this allows the establishment of clonal plantations. Micropropagated clones can be maintained in a juvenile state by cool-storage, while clonal testing is done in field trials (Davies and Aitken-Christie, 1991), and this has the potential to avoid problems of physiological aging. The effective cost of the process could be reduced by using plantlets as stool plants for the production of cuttings (Table 2). This would delay the delivery of planting stock from seed but would increase the multiplication rate with the extra propagation step.

CONCLUSIONS

Reliable methods for vegetative propagation of radiata pine have been developed. These are increasingly being used by forest growers in an effort to establish forests of the best genetic quality at a faster rate than is currently possible with seedlings. At present control-pollinated seed prices, stool-bed cuttings are cheaper to produce. Improvements in stem form have been demonstrated with cuttings compared with similar genetic quality seedlings, particularly with field-collected cuttings planted

on fertile farm sites. This has created a demand for field-collected cuttings, even though they cost nearly double the price of stool-bed cuttings. Micropropagated plantlets are being used by one New Zealand company to establish clonal plantations, despite their high cost.

LITERATURE CITED

Aitken-Christie, J. and **T.A. Thorpe.** 1984. Clonal propagation of gymnosperms, p. 82-95. In: I.K. Vasil (ed.). Cell culture and somatic cell genetics of plants. Vol. 1. Academic Press, Inc., Orlando, Florida.

Arnold, R. and **J.A. Gleed.** 1985. Raising and managing radiata pine vegetative cuttings for production forests. Austr. For. 48(3):199-206.

Davies, H.E. and **J. Aitken-Christie.** 1991. Cold storage technology update, p. 74-75. In: J.T. Miller (ed.). Proc. FRI/NZFP Forests Ltd clonal forestry workshop. NZ Ministry of Forestry, Forest Research Institute Bulletin No. 160.

Dibley, M. and **T. Faulds.** 1991. Production and costs of juvenile radiata pine cuttings, p. 28-34. In: M.I. Menzies, G.E. Parrott, and L.J. Whitehouse (eds.). Proc. IUFRO symposium on efficiency of stand establishment operations. NZ Ministry of Forestry, Forest Research Institute Bulletin No. 156.

Gleed, J.A. 1991. Toward clonal afforestation, p. 61. In: J.T. Miller (ed.). Proc. FRI/NZFP Forests Ltd clonal forestry workshop. NZ Ministry of Forestry, Forest Research Institute Bulletin No. 160.

Gleed, J.A. 1993. Development of plantlings and stecklings of radiata pine, p. 149-157 In: M.R. Ahuja and W.J. Libby (eds.). Clonal forestry II. Conservation and application. Springer-Verlag, Berlin Heidelberg.

Horgan, K. and **J. Aitken.** 1981. Reliable plantlet formation from embryos and seedling shoot tips of radiata pine. Physiol. Plant. 53:170-175.

Menzies, M.I., T. Faulds, M. Dibley, J. Aitken-Christie. 1985a. Vegetative propagation of radiata pine in New Zealand, p. 167-190. In: D.B. South (ed.). Proceedings of the international symposium on nursery management practices for the southern pines. IUFRO/Alabama Agricultural Expt. Stn , Auburn University, Montgomery, Alabama, USA, Aug. 4-9.

Menzies, M. I., J.C. van Dorsser, and J.M. Balneaves. 1985b. Seedling quality—radiata pine as a case study, p. 348-415. In: D.B. South (ed.). Proceedings of the international symposium on nursery management practices for the southern pines. IUFRO/Alabama Agricultural Expt. Stn., Auburn University, Montgomery, Alabama, USA, Aug. 4-9.

Menzies, M.I., B.K. Klomp, D.G. Holden. 1991. Promising future for radiata pine cuttings. Forest Research Institute, What's New in Forest Research No. 212, 4 pp.

Ministry of Forestry. 1993. A national exotic forest description as at 1 April 1992. Ministry of Forestry, Wellington.

Smith, D.R. 1986. Radiata pine (*Pinus radiata* D. Don), p. 274-291. In: Y.P.S. Bajaj (ed.) Chapter X : Forest and nut trees, biotechnology in agriculture and forestry, Vol. 1: Trees. Springer-Verlag, Berlin.

Vincent, T.G. 1993. Supply of improved radiata pine seed. NZ Forestry 38(3):45-48.