

within regions (assuming sites to be representative of localities) the gain equation becomes:

$$\text{gain} = \frac{21 \left(\sigma_f^2 + \sigma_{fr}^2 + \sigma_{fs(r)}^2 \right)}{\left(\sigma_f^2 + \sigma_{fr}^2 + \sigma_{fs(r)}^2 + \sigma_e^2/35 \right)^{1/2}} \quad (\text{Equation 8})$$

This assumes fully random effects. Alternative assumptions regarding fixed effects do not materially alter this expectation.

Appendix Table 1 shows predicted gains after substituting in variance component estimates from Table 2. The gains from regionalising by the two main regions are evident by comparing solutions of Equations 5 and 2, 6 and 3, or 7 and 4 depending on the assumption concerning fixed or random effects. Correspondingly, the gains from regionalising to individual sites are evident from comparing the solution for Equation 8 with those of Equation 5, 6 or 7. The gains foregone by not regionalising can be large and depend strongly on whether fixed or random site effects are involved.

The Progression and Distribution of Graft Incompatibility in *Araucaria cunninghamii* Ait. ex D. Don

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Summary

The progression and distribution of graft incompatibility in *Araucaria cunninghamii* were examined using grafting records and scion assessment data which were available for a total of 199 clones in seed orchards and clone banks up to 15 years after grafting. The species displays a smooth progression in the onset of incompatibility, tailing off to a final overall incidence of 31% by 10 years after grafting. Although no sharp chronological demarcation is apparent, there is some justification for classification into "early" and "delayed" forms on the basis of the extent of scion elongation prior to the manifestation of incompatibility symptoms. Clonal repeatability for scion length at diagnosis of incompatibility was 0.50. The clonal distribution of early incompatibility is bimodal, with approximately 8% of the population highly incompatible, and the remainder highly compatible. Only two clones displaying a high incidence of delayed incompatibility were identified. Other clones clustered around a compatibility level of approximately 75%. For the severely incompatible clones, flowering race and incompatibility type seem to be associated. Clones displaying a high incidence of early and delayed incompatibility were respectively all early and late flowering. The possibility of a relatively simple genetic control mechanism is discussed, and practical implications are considered.

Key words: *Araucaria cunninghamii*, graft, incompatibility, scion, rootstock.

Introduction

Araucaria cunninghamii (hoop pine) is an important plantation conifer in Queensland, where over 44000 ha have been established, and is of potential value as a plantation species in several other countries (NIKLES, 1980). A breeding programme has operated for approximately 30 years (NIKLES and NEWTON, 1983), and all planting stock now used is raised from seed collected in clonal seed orchards.

Some unusual biological features of the species have had a major impact on the development of seed orchard systems (HAINES and NIKLES, 1987b). Like other species of

Araucaria, *A. cunninghamii* has a rigid orthotropic-plagiotropic branching system. As a result of a differential tendency to produce female and male strobili, both orthotropic and plagiotropic ramets have been established in seed orchards, to act respectively as seed producers and pollinators. The wide interclonal variation in flowering season, associated at least partly with provenance, has resulted in the segregation of clones into "early flowering" and "late flowering" "races" for the purposes of allocation to seed orchards.

Graft incompatibility has long been recognised as a problem, but recently has been of particular concern in relation to the use of biclonal orchards for the mass production of superior full-sib families (HAINES and NIKLES, 1987a). Two forms of graft incompatibility in *A. cunninghamii* were described by HIGGINS* (1969, unpublished). The symptoms of "early incompatibility" are profuse budding, very poor elongation and chlorosis of the scion, followed ultimately by scion death. Grafts which undergo greater elongation, but then display reduced growth rate, swelling at the union, chlorosis, necrosis and, finally, death of the scion, have been described as being afflicted by "delayed incompatibility". In terms of the time at which diagnosis can be made, the distinction between these is not sharply defined. Compatible and early incompatible ramets cannot always be distinguished with certainty until three years after grafting, and delayed incompatibility becomes apparent at a range of ramet ages.

Using data now available from a large number of clones grafted in seed orchards and clone banks, the investigation reported here was designed to examine three aspects of the phenomenon:

- the progression of the overall incidence of incompatibility with time;
- the clarity of the separation into early and delayed forms, in terms of the scion lengths at which ramets are diagnosed as incompatible; and

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— the clonal distribution of incompatibility within the plus tree population, in particular in relation to flowering race, scion type, and growth rate.

Materials and Methods

The data examined originate from two clonal seed orchards (Imbil and Taromeo) and six clone banks all located between 26° S and 27° S in south-eastern Queensland. These were grafted with scions of selected phenotypes during the period 1961 to 1983. Although rootstock seedlings were raised from known seed batches, the flowering race of rootstocks used at each clone bank or orchard could not be specified with sufficient confidence to warrant taking this variable into account during interpretation of the data.

The clone banks comprise only orthotropic ramets, while the seed orchards are made up of both orthotropic and plagiotropic grafts. Almost all orthotropic ramets were grafted using a variation of the chip-budding method (NIKLES, 1961) and plagiotropic ramets were predominantly side cleft grafts. An early assessment of the number of grafts that formed a union was made universally. Parameters recorded at the subsequent periodic assessments varied, but included scion elongation, a classification based on a visual appraisal of vigour, health, swelling of the union, and compatibility status. Assessment intervals were generally three to four years.

Accurate measurements of scion elongation, made at intervals varying from two to four years for up to 15 years from grafting, were available for four clone banks. The scion lengths at first diagnosis of incompatibility, as recorded for all incompatible ramets in these clone banks, were used in an attempt to characterise early and delayed incompatibility.

Data from all orchards and clone banks, involving a total of 6777 ramets representing 199 clones, were the basis for the definition of the distribution of compatibility within the plus tree population. For this purpose, the "early compatibility" of each clone — the percentage of ramets not affected by early incompatibility — was calculated as the number of early compatible ramets expressed as a percentage of the number of "takes". "Delayed compatibility" — the percentage of ramets not affected by delayed incompatibility — was calculated as the final number of compatible ramets expressed as a percentage of the number of early compatible ramets. "Overall compatibility" was calculated as the final number of compatible ramets expressed as a percentage of the number

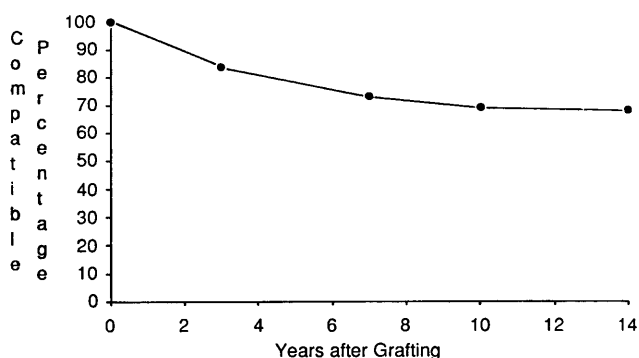


Figure 1. — Decline in the percentage of compatible ramets after grafting of a clone bank (comprising 297 ramets representing 19 clones).

of takes. Estimates based on a denominator of fewer than eight ramets were not used. Estimates of delayed and overall compatibility were calculated from data collected over a period of at least 10 years after grafting. The seed orchards have been thinned periodically. Calculations were made on the assumption that, for each clone, the compatible ramets thinned would, if retained, have become incompatible in the same proportion as did the unthinned compatible ramets of the same health/vigour class.

Data from the Taromeo orchard were used to compare distributions of incompatibility among orthotropic and plagiotropic ramets. In this orchard, the two types of grafts are arranged in alternate rows within which clones are arranged randomly.

The numbers of compatible ramets of the highly incompatible scion clones available in clone banks in which scion elongation was measured were too low to permit the use of scion elongation as a measure of vigour for these clones. As an alternative, the standard normal deviate for volume in open pollinated progeny test (CORTERILL *et al.*, 1983) was calculated as an index of clonal vigour. It is recognised that in the presence of non-additive genetic effects, genotype × maturation state interactions, or genotype × propagule type interactions, these indices of clonal vigour are not completely comparable.

Results

Progression of incompatibility with time

Data from one clone bank, comprising a total of 297 ramets representing 19 clones, have been plotted in Figure 1. This graph, which is typical of the pattern observed in other clone banks, demonstrates that almost all incompatibility has become evident within 10 years of grafting.

Scion elongation at diagnosis of incompatibility

The distribution of scion lengths at the time of diagnosis of incompatibility, for 355 ramets representing 81 clones, is presented in Figure 2. This figure offers some support for the separation of incompatible ramets into at least two groups: those which are diagnosed as incompatible at a scion length of less than approximately 80 cm, and those which undergo greater elongation before incompatibility becomes evident. Separation into two groups was not an artefact of intermittent assessments or variation among sites. The presence of a peak tailing off at 50 cm to 80 cm was a feature of distributions plotted for both individual assessment ages and for separate locations.

Clonal repeatability of the scion length at diagnosis of incompatibility, based on the 355 ramets representing 81 clones, was 0.50 ± 0.06 . The mean scion length at which ramets of a clone were diagnosed as incompatible was not significantly correlated with the final incidence of compatibility within the clone ($r = 0.17$, $p > 0.05$).

Distribution of the incidence of graft incompatibility

On the basis of the above, a scion length of 67 cm (the upper limit of a scion length class used in assessment at several locations) at diagnosis of incompatibility was used to distinguish early and delayed incompatibility. The distributions of early, delayed and overall compatibility within the plus tree population (based respectively on samples of 6463, 4688 and 5146 ramets representing 133, 106, and 119 clones) are presented in Figures 3, 4 and 5. The distribution for early compatibility (Figure 3) is bi-

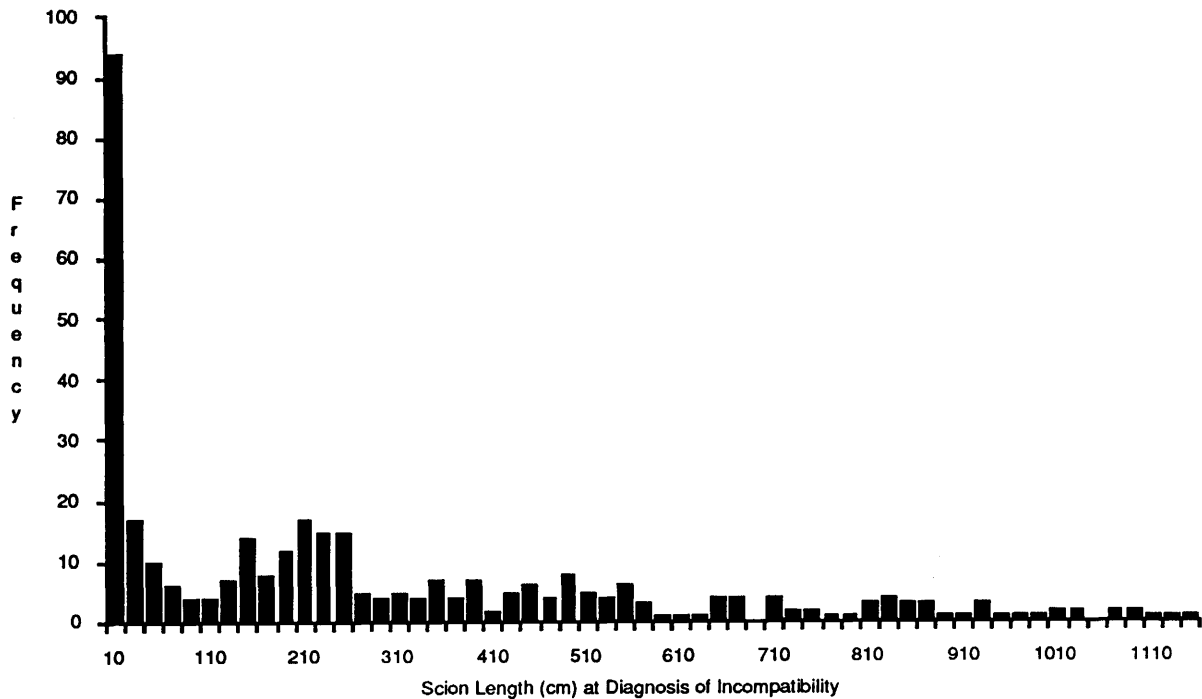


Figure 2. — Distribution of scion lengths at diagnosis of incompatibility for 355 incompatible ramets (representing 81 clones).

modal, with a small group of clones (approximately 8% of the population) highly incompatible, and the rest of the population highly compatible. With respect to delayed compatibility (Figure 4), only two clones are highly incompatible, while the rest of the population clusters around a compatibility level of approximately 75%. The compatibility levels of most clones in each of the highly incompatible groups in Figures 3 and 4 were significantly below the respective mean compatibility level for all clones represented in the distribution. Although the two clones assigned to the highly delayed incompatible group did not differ significantly with respect to compatibility level, highly significant differences among clones within groups were evident for each of the other three groups defined on the basis of Figures 3 and 4.

In terms of the overall incidence of incompatibility (Figure 5), approximately 10% of the population is severely affected, while the remainder of the population is relatively compatible — clustering around 75%. Summed over all ramets of all clones, the final compatibility was 69%.

Sixteen of the clones involved in this study had been grafted at more than one location. In no case was there

an effect of site on the compatibility status group to which the clone would be assigned.

Relationship to vigour

The association between the overall compatibility of a clone and the vigour of its progeny was negligible ($r = 0.07$, 88 d.f.).

Relationship to flowering race

Examination of flowering records for clones contributing to the distributions in Figures 3 and 4 revealed that the 11 clones displaying early compatibility levels of less than 50% were early flowering, while the two severely delayed incompatible clones identified were both late flowering.

Influence of scion type

Distributions of final compatibility for orthotropic and plagiotropic grafts in the Taromeo orchard, for the 49 clones represented as both, are presented in Figures 6 and 7 respectively. The distributions differ principally in that no highly incompatible group is evident in the case of the plagiotropic ramets. The two clones which displayed a high incidence of early incompatibility as orthotropic ramets are relatively compatible as plagiotropic ramets. Of the highly early incompatible clones in the Taromeo orchard, only two orthotropic ramets had been grafted by the side cleft method. One of these was compatible and the other incompatible.

Discussion

At 31%, the overall incidence of incompatibility in *A. cunninghamii* is similar to levels reported for *Pinus eliottii* (28%) and *P. echinata* (33%), but higher than the incidence evident in *P. taeda* (6%) (ALLEN, 1967). The problem is much more severe for *P. radiata* in New Zealand (SWEET and THULIN, 1973; COPES, 1980), and *Pseudotsuga menziesii* (COPES, 1967a, 1967b; KARLSSON, 1970), for which losses can exceed 50%.

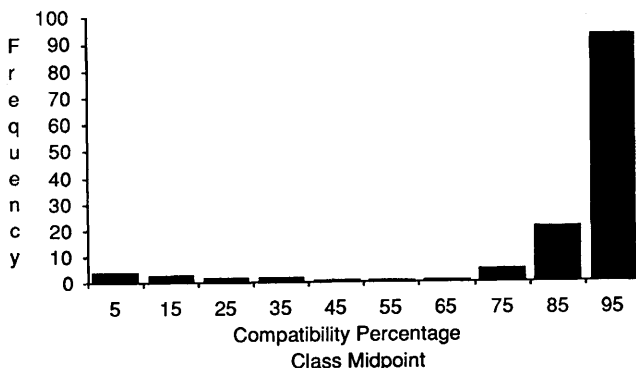


Figure 3. — Distribution of early compatibility percentage among 133 plus tree clones.

The term "delayed incompatibility" frequently has been applied to grafts which suddenly decline after a period of apparently normal growth (Mosse, 1962), a period which may extend to 40 years in some fruit trees (Rogers and Beakbane, 1957). A clear chronological distinction between early and delayed incompatibility has been reported also for some forest trees. In *Pinus monticola* for example, early incompatibility appears soon after the planting out of grafts, while delayed incompatibility becomes apparent after about 11 years (Hoff, 1977). The sudden failure of grafts aged 11 to 17 years is a feature also of *P. merkusii* in Zambia (Mikkola *et al.*, 1979). *P. radiata*, on the other hand, is characterised by a steady progression in the incidence of incompatibility, with losses still occurring at 15 years after grafting (Copes, 1980). Incompatibility in *Pseudotsuga menziesii* also becomes manifest over a long period, although an arbitrary distinction between early and delayed incompatibility has been made, the former being defined as failures occurring between the fourth and 24th months after grafting (Copes, 1967a). Although *A. cunninghamii* resembles the latter two species in the steady progression in losses to incompatibility, there is some justification for a natural separation into different types on the basis of the extent of scion elongation prior to diagnosis.

Consistent with evidence gathered for other woody species (Rogers and Beakbane, 1957), including *Pseudotsuga menziesii* (Copes, 1967b) and *Pinus taeda* (Lantz, 1970), graft incompatibility in *A. cunninghamii* cannot be attributed simply to growth rate differences between stock and scion. Most recent speculation regarding the mechanisms underlying graft incompatibility in plants has centred on the involvement of a recognition phenomenon between

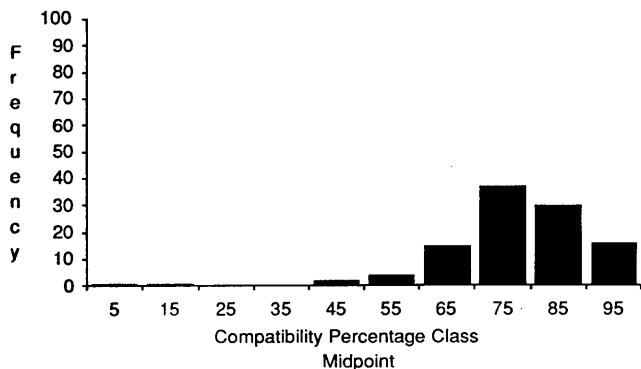


Figure 4. — Distribution of delayed compatibility percentage among 106 plus tree clones.

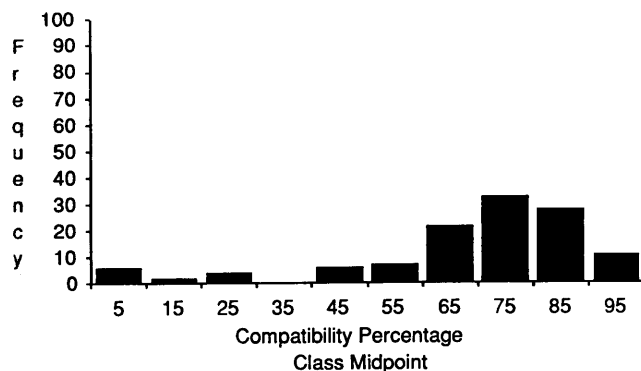


Figure 5. — Distribution of overall compatibility percentage among 119 plus tree clones.

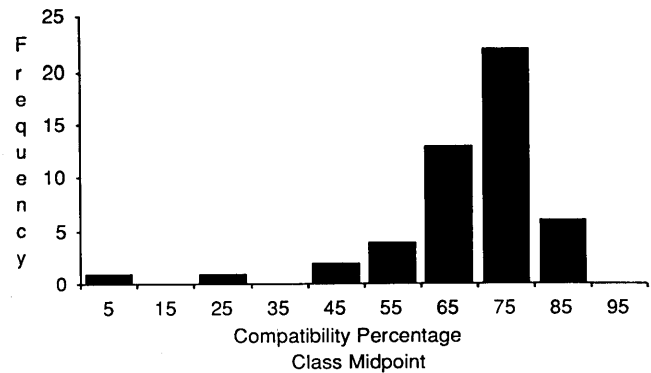


Figure 6. — Distribution of overall compatibility percentage, for orthotropic ramets, among 49 clones in the Taromeo seed orchard.

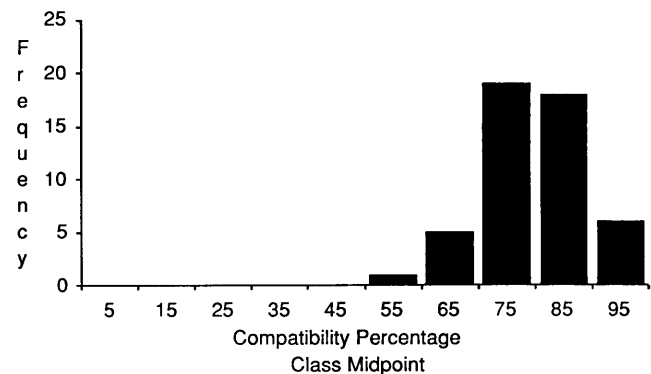


Figure 7. — Distribution of overall compatibility percentage, for plagiotropic ramets, among 49 clones in the Taromeo seed orchard.

sterically complementary molecules, possibly including lectins (Clarke and Knox, 1978; Heslop-Harrison, 1978; Yeoman, 1984). For forest tree species, an underlying genetic basis has been demonstrated by the many reports of higher compatibility between related partners (Slee and Spidy, 1970; Copes, 1973; Van der Siide, 1974; McKinley, 1974; Van Wyk, 1977). *Pseudotsuga menziesii* is the only forest tree for which estimates of genetic parameters relating to graft incompatibility have been made. The distribution of graft compatibility in this species is unimodal, with a peak at a compatibility level of 60% to 70% (Copes, 1974). Control is evidently polygenic, effects are mainly additive, and heritability is very high — 0.81 (Copes, 1973, 1974). This has led to the implementation of a breeding programme for highly compatible rootstocks, based on the crossing of highly compatible parents (Copes, 1981).

Separation into two distinct compatibility level groups for both early and delayed compatibility suggests that the control of incompatibility in *A. cunninghamii* may differ from that in *Pseudotsuga menziesii*. The simplest hypothesis which could be offered regarding a genetic control mechanism is that a single locus has a major influence, with the highly incompatible clones possessing an allele not present in compatible clones. The highly incompatible clones would be compatible only with rootstocks which possess this allele. With this mechanism, and assuming that scions and rootstocks are drawn from the same population, the percentage compatibility of a scion phenotype carrying the incompatibility allele would be equal to the percentage of rootstock phenotypes carrying the allele,

and therefore to the percentage of scion phenotypes with the allele. For early incompatibility, the peaks in the compatibility distribution are approximately in the correct proportions to support such an explanation. The percentage of the population in each group is about the same as the mean percentage compatibility of the group. This evidence is admittedly tenuous, and significant differences among clones within groups indicate that additional factors would be involved.

The relationship to flowering race suggests that severe delayed incompatibility may be a feature of different provenances to those severely affected by early incompatibility, and could involve different genes. For the delayed compatibility distribution, the percentage of the population in each group does not correspond closely to the mean compatibility percentage of the group. The possibility of more than one form of delayed incompatibility, involving different genes which cause failure at different levels of scion elongation, cannot be discounted. Variation in rootstock provenances, together with the probability that the distribution derived for delayed incompatibility has been influenced by the incorrect attribution to incompatibility of losses resulting from other causes, render further speculation unwarranted.

A genetic control mechanism as simple as a single locus system would have important practical implications. Controlled crossing of early-incompatible parents for example, could be used to produce progeny which, when used as rootstocks, would confer a high level of compatibility on early-incompatible scion clones. The probability of incompatibility of an advanced-generation select could be calculated on the basis of parentage, and the appropriate mixture of rootstock types used to optimise compatibility in clone banks.

The existence of a more complex genetic control mechanism, perhaps involving a threshold, would not necessarily prohibit practical solutions, for example rootstock breeding, or the selection and propagation of rootstock clones which are compatible with highly incompatible scion clones.

The production and use of special rootstocks could be warranted at least for the group of clones displaying a high incidence of early incompatibility. Depending on the genetic control mechanism involved, treating this group as a "subline" in the breeding programme (at least for a generation or two, until the relatedness of selections becomes a problem) may offer advantages with respect to seed orchard and clone bank management. Compatible selections produced in this "subline" could be transferred to the main breeding population.

Interacting physiological factors, with respect to which orthotropic and plagiotropic scions differ, presumably are responsible for the absence of severe early incompatibility among the latter, although a grafting technique effect cannot be ruled out. The practical implications of this distinction are that rootstock segregation will not be required for the effective use of plagiotropic grafts in orchards.

Studies currently underway on the interactions between scion clone and rootstock family may provide answers to some of the questions raised here.

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