von solchen Bäumen wird hier berichtet. Auf 3 Klonen fand man nur sehr kleine Misteln und auf 2 Klonen überhaupt keine. Kontroll-Klone, die man auf Anfälligkeit ausgelesen hatte, waren weniger stark infiziert, als ungepfropftes Baumschulmaterial, das eine hohe Jugendanfälligkeit vermuten ließ. Eine extreme Resistenz zeigten 27 in gleicher Weise geimpfte Sämlingskontrollen; ferner hatte 1 Individuum 6 Infektionen und 3 weitere Individuen mehr als 30 Infektionen. Andere bewegten sich im Mittel um 16. Morphologische und physiologische Faktoren spielen bei diesen Resistenzerscheinungen eine Rolle.

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# Some unusual seedlings of Eucalyptus; their genetic significance and value in breeding

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#### Introduction

Seedling characters have been widely used in applied genetics and plant breeding (HASKELL, 1961) to permit early screening and selection from large populations within a limited space. These are particularly important advantages in forest tree species which take a long time to become adult, and occupy much space in experimental fields. The atypical seedling variants reported here were first noticed when germinating in petri dishes in the laboratory seeds collected from individual trees of Eucalyptus camaldulensis Dehn. and E. tereticornis Sm. selected for use in a breeding Programme. Sirnilar seedlings were also detected later in duplicate sowings in the nursery of the same seed sources in clay pots containing soil. However, their detection in the latter situation was not as easy as against the white background of the moist filter paper in the petri dish. Therefore, had they been raised only in the nursery, such seedlings could readily have escaped notice. The different unusual seedlings recovered from the various seed sources can be broadly classified into the following four kinds -1) twins, 2) abnormal seedlings, 3) pleiocotylous seedlings and 4) albino or chlorophyll-deficient seedlings. These are described and their genetic significance and value in breeding are then discussed.

## Twin Seedlings

The twin seedlings were of two kinds viz. independent twins and conjointed twins. Figure 1 A, B illustrates a pair of independent twins arising from a single seed. One of this

pair (fig. 1, B) is less robust than its mate (fig. 1, A). The two are however free from and unattached to each other. Conjointed twins on the other hand are organically one from the hypocotyl downwards. They have only one primary root but two stems, each with its cotyledons (fig. 1, C,D). Of two twins recovered of this kind, one had a swollen carrotlike hypocotyl tapering down into the primary root below but branching above into two stems, of which one had two cotyledons whereas the other had three (fig. 1,C). When a batch of 25 seeds were softened by soaking in water and then dissected, one seed was seen to consist of two embryos, one smaller than the other (fig. 1, E). It is from such twin embryos that the independent twin seedlings described above must have arisen and so they are the result of polyembryony which has so far not been recorded in the genus Eucalyptus. However, in Eugenia and Syzigium, two other genera of the same family Myrtaceae, nucellar polyembryony is well known (Tiwary, 1926; John-SON, 1936; NARAYANASWAMI & ROY, 1960; and ROY & SAHAI, 1962). The exact origin of the twin embryos in Eucalyptus reported here can be ascertained only from detailed embryological studies. However, the fact that they have so far always been found in pairs and not in larger numbers is significant as will be discussed later.

## **Abnormal seedlings**

These were of different kinds characterized by poor formation, malformation or total atrophy of one or more of the three primary organs of the normal seedlings viz. root, hypocotyl and cotyledons. The rootless type illustrated in fig. 2, A lacked a primary root and had one of its two coty-

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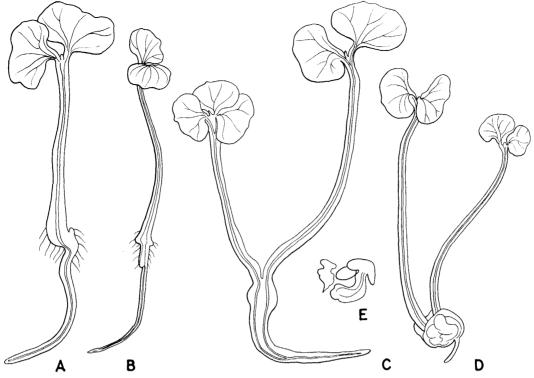


Fig. 1. — A-E. — Twin seedlings and twin embryo.

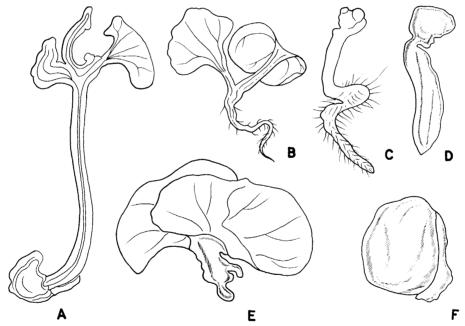


Fig. 2. -A-F. — Abnormal seedlings.

ledons malformed and with a horn-like appendage from its base. There was also a small peg-like apogeotropic structure springing from the base of this seedling. There were others that had no hypocotyl (fig. 2, B) or had no trace of cotyledon (fig. 2, C). A few lacked both cotyledons and roots, but had only a hypocotyl (fig. 2, D). Sometimes only cotyledons were present, and both hypocotyl and root were wanting (fig. 2, E). In one instance the seed coat contained only an undifferentiated mass without a trace of root, hypocotyl or cotyledons (fig. 2, F). None of the abnormal seedlings survived.

## Pleiocotylous seedlings

Seedlings with three instead of the usual two cotyledons (fig. 3, A, B) appeared both in open pollinated and control pollinated progenies of known trees. Rarely tetracotylous (fig. 3, C) and monocotylous (fig. 3, D) seedlings were also picked up. As the tricotylous seedlings grew, their leaves kept on appearing in successive whorls of three each (fig. 4) and not in opposite pairs as is usual for Eucalyptus seedlings. The particular tricotylous seedling illustrated in fig. 4 appeared in progenies resulting from a one-way controll-

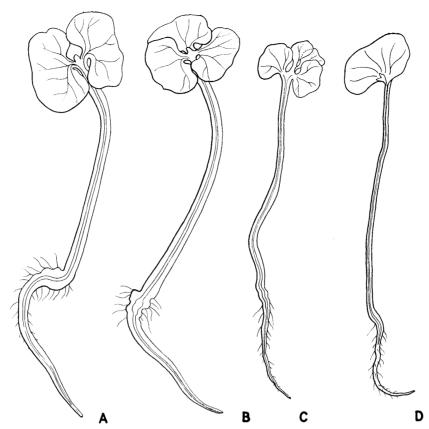


Fig. 3. — A—D. — Typical dicotylous and atypical pleiocotylous seedlings compared.

ed interspecific cross between trees of *E. tereticornis* and *E. camaldulensis*. This tricotylous hybrid seedling was more vigorous than all of its dicotyledonous siblings, two of which appear flanking it in the same picture. This extra growth was either due to inherent vigour or to the initial advantage of extra photosynthetic nourishment made possible by three, instead of the usual two cotyledons, followed by whorls of three leaves at successive nodes in place of the usual pair. In any case, it will be interesting to see if this growth superiority is maintained in later years.

## Albino or Chlorophyll deficient seedlings

Two albino seedlings appeared in a batch of fifty open pollinated progenies of a mother tree of *E. tereticornis*. In another batch of one hundred seeds of the same source, only one albino was picked up. There was a total lack of



Fig. 4. — An older tricotylous seedling flanked by two of its dicotylous siblings.

chlorophyll in all three seedlings which proved lethal, as expected. Their hypocotyls appeared reddish or reddish purple. These albino seedlings died a few days after germination when the cotyledonary reserve foods had been exhausted.

Fifteen successive batches of one hundred seeds each of another open-pollinated mother tree of the same species regularly produced a certain proportion of chlorophyll-deficient seedlings. These, unlike the total albinos described above were pale yellow in colour. They survived longer but gradually turned brown as they approached eventual death after a period of 6 to 10 days.

The different kinds of atypical seedling variants described in the preceding paragraphs and their relative frequency of occurrence are summarized in *Table 1*.

## Discussion

Twin seedlings were found in open-pollinated seed samples of several individual trees of both *E. camaldulensis* and *E. tereticornis*. In a pair of twins, one of the seedlings tended to be poorer in growth than its partner. Whether this was due to its being haploid remains to be ascertained by actual chromosome counts. With this end in view, twin seedlings are reared with great care. The regular appearance of twins but not a larger, variable number of seedlings from some seeds suggests that the two seedlings develop from cells of the embryo sac and are not likely to be of adventive nucellar origin. If some of the twins do turn out to be haploid-diploid (Maheshwari, 1950), then the haploid seedling sporophytes (Stettler, 1966) could be of great value because, by inducing chromosome doubling in them, diploid homozygous plants can be produced at one step and used for

Table 1

Type of atypical seedling	Seeds sown for germination	Seeds germinated	Frequency of atypical seedlings
	(No)	(No)	(%)
Twin seedlings:			
Conjointed twins	1950	912	0.43
Separate twins	4074	3606	0.55
Abnormal seedlings:			
Rootless	1950	1636	0.24
Hypocotyl-less	2074	1714	0.52
Acotylous	300	260	1.15
Acotylous and rootless	1850	1551	0.19
Hypocotyl-less and rootless	200	194	0.51
Undifferentiated into root hypocotyl or cotyledons	650	576	0.52
Seedlings with horn-shaped appendage	1450	500	0.20
Pleiocotylous seedlings:			
Tricotylous	6374	5631	0.71
Tetracotylous	200	196	0.50
Monocotylous	2150	1945	0.25
Albino or Chlorophyll deficient			
Seedlings:			
Albino — total	1800	1588	0.18
Chlorophyll deficient	1550	1302	21.19
Combinations of one, two or more			
unusual features in same seedling or			
seedling pair: Separate twins, one di- the other	1500	1950	0.05
tri-cotylous	1300	1259	0.07
Conjointed twins, one di- the	1450	500	0.20
other tri-cotylous		000	0.20
C. d. *+conjointed twins, one di-	1500	1259	0.07
the other mono-cotylous			0.01
C. d. +separate twins	1500	1259	0.23
C. d. +rootless	1500	1259	0.15
C. d. +hypocotyl-less	1500	1259	0.31
C. d. +acotylous	1500	1259	0.07
C. d. $+$ rootless $+$ hypocotyl-less	1500	1259	0.07
C. d. +tricotylous	1500	1259	0.15
C. d. $+$ tetracotylous	1500	1259	0.15
C. d. + monocotylous	1500	1259	0.39

<sup>\*</sup> C. d. stands for chlorophyll deficient.

heterosis breeding of Eucalypts. In the absence of haploid sporophytes, such homozygous plants can be evolved only gradually by several generations of patient inbreeding, which, particularly in tree species, takes a long time.

The abnormal seedlings recovered recall similar abnormalities induced by irradiations and, like them, are lethal. Presumably, at least some of them represent deleterious and lethal natural mutations of long standing, that have survived in the population because of concealment as recessives in the heterozygous condition.

Three facts observed so far suggest that tricotyly is probably an inherited character, rather than a fortuitous teratological development. Firstly, all the three cotyledons of such seedlings were of similar size and therefore could not have possibly arisen by the splitting of a normal pair of cotyledons. Secondly, such seedlings appear at low frequency in different seed sources. Thirdly, the successive seedling leaves following the cotyledons on tricotylous plants arise in whorls of three and not in pairs as is usual for *Eucalyptus* seedlings. A few tricotylous seedlings of certain seed sources tended to switch from the whorled to

the normal opposite seedling-leaf arrangement but this happened only after the first three sets of seedling leaves had arisen in the unusual whorled fashion. The gene or genes controlling tricotyly therefore seem also to control, at least to some extent, the phyllotaxy of the seedling leaves. It will be some years before the tricotylous plants reported here come to flower-bearing age when the nature of inheritance of this character can be determined. It will be a useful genetic marker valuable in applied breeding work (HASKELL, 1961).

Pleiocotyly, or variation in cotyledon number, has been recorded in the literature, both in Dicots and Coniferae (Haskell, 1954). Under this term are included monocotyly and polycotyly. Among dicotyledonous agricultural crop plants pleiocotyly has been studied in some detail in beet, sugarbeet and cultivated forms of *Brassica*. According to Haskell (1961), the character is polygenically controlled. He reports obtaining one monocot, one tetracot and numerous normal dicot seedlings by sowing seeds harvested from one tree of *Laburnum vulgaris* a dicotyledonous angiosperm. Griffith (1953) found that seeds of the same size

from one tree of the conifer *Cupressus lusitanica* produced seedlings with 2 to 8 cotyledons. Plants with 2, 3 and 4 cotyledons were picked out and planted in an arboretum. Tetracots at first grew slower than di- and tricots, but later tetracots and tricots showed significantly greater heights than the dicots during the year after field planting. This example illustrates the possible use of pleiocotyly for practical ends in forestry.

Among forest trees, albinos and chlorophyll-deficient seedlings have been reported in Conifers and in a few others, but not so far in *Eucalyptus*. The frequency of occurrence of these mutant traits can be used to estimate the degree of natural selfing (see Squillage & Kraus, 1963 on Slash pine) that normally occurs in Eucalypts and of which practically nothing is known at present, even for a single species of this large and important genus (Penfold & Willis, 1961).

#### **Summary**

The incidence of polyembryony, pleiocotyly, abnormal, albino and chlorophyll deficient seedlings is reported in *Eucalyptus*. With the exception of tricotyly this may be the first record of other occurrences for this genus. The possible use of these phenomena in genetical studies and their practical use in *Eucalyptus* breeding are briefly discussed. *Key words: Eucalyptus*, progenies, genetics, aberrant seedlings.

#### Zusammenfassung

Es werden das Vorkommen von Polyembryonie, Pleiocotylie, anomalen weißen und chlorophyll-defekten Sämlingen bei *Eucalyptus* mitgeteilt. Mit Ausnahme der Tricotylie sind diese Erscheinungen erstmals bei dieser Gattung gefunden worden. Über ihre Anwendbarkeit für genetische und praktisch züchterische Arbeiten wird kurz diskutiert.

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# Effect of Partial Measurement on Statistical Precision and Efficiency in a Nursery Study of Pinus ponderosa

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In the assessment of genetic information in the nursery, plot means are frequently used as items in an analysis of variance. Other common practices are measurement of the tallest seedling or the use of mean values based on the two tallest trees per plot in the analysis. Apparently partial measurement is sometimes done for convenience although its statistical soundness remains unverified.

Most nursery experiments consist of multi-tree plots with 10 to 40 seedlings per plot planted in the beds. Time may be saved if not all trees are measured; if done, the amount of genetic information lost should be determined and evaluated. It is also important to analyze what type of sampling (random or deliberate) should be employed to more efficiently derive the necessary genetic information.

This study was undertaken at the Institute of Forest Genetics, Placerville, California, in the summer of 1973 to determine the amount of information lost and time gained by two types of partial measurement.

#### **Material and Methods**

This study consists of measurement of 1-0 ponderosa pine (*Pinus ponderosa*) seedlings in a 28-family half-sib progeny test established March 1972 by Dr. James L. Jenkinson, Forest Genetics staff of the Pacific Southwest Forest Experiment Station, Berkeley, California. The experiment was planted in two adjacent, parallel seedbeds, each 47 feet long and 6 feet wide, which extended east-west and were separated by a path. Spacing between each seedling was  $6 \times 6$  inches. The experiment was a randomized complete block design with 6 replications, each containing one 12- tree north-south plot per family. In addition to the experimental plots, each seedbed contained 10 unmeasured buffer plots between blocks and at the ends of the seedbeds.

The height of each seedling was measured to the nearest 5 mm. and plot means were then computed in 17 different ways, i.e. based on all 12 trees, based on the 1, 2, 3, 4, 5, 6, 8 or 10 tallest seedlings in each plot, or based on 1, 2, 3, 4, 5, 6, 8 or 10 seedlings chosen randomly with replacement in each plot.

Analysis of variance and estimated variance components were computed for each of the 17 sets of height data according to the following tabulation:

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