SEED PRODUCTION IN RADIATA PINE CLONES ON FOUR DIFFERENT SITES

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ABSTRACT

Seed production was studied in cones from 18 clones of radiata pine (**Pinus radiata** D. Don) replicated within and between four contrasting sites. At one site which was phosphate-deficient, number of full seeds per cone, along with cone volume and mean seed weight, was about one-third less than elsewhere. Empty seeds, although never completely absent, were prevalent only in occasional cones. Such cones were common at the phosphate-deficient site, but were not a consistent feature of particular clones. All clones showed inherently high fertility within cones.

There was indirect evidence of a high incidence of ovule abortion: firstly, number of seeds per cone showed much more non-genetic variability than number of scales, and secondly, within-clone correlations between these two characters were not high. Such ovule abortion is believed to resemble conelet drop, which was prevalent within the experiment, in being often not caused by inadequate pollination.

Almost without exception the cones contained totals of 40 or more seeds, suggesting that satisfactory cone development is related to a threshold frequency of either ovule abortion or ovule survival.

INTRODUCTION

It is becoming clear that in radiata pine (*Pinus radiata* D. Don) much wastage of seed production occurs between conelet receptivity and seed filling. The wastage first becomes evident in the abortion of conelets, customarily known as "conelet drop", which mostly occurs soon after pollination, and may involve over half the conelet crop (Sweet and Thulin, 1969). In cones that do mature many of the ovule sites may not contain seeds, and a varying proportion of the seeds that do develop may be empty. However, there is very little concrete information concerning the incidence of missing and empty seeds in radiata pine.

This paper reports a study of the numbers of full and empty seeds together with some other related characters, which was made with a collection of cones from a trial with clones replicated within and between sites. One objective was to obtain circumstantial evidence as to the causes of empty and missing seeds in radiata pine. Another paper (Burdon and Low, 1973) covers cone characters and mean seed weight in the same experiment.

MATERIAL AND METHODS

The Experiment

The experiment is described elsewhere (Burdon, 1971) and a brief account only is given here. Eighteen clones were replicated, as cuttings five yr old from seed, within and between four sites, at Glenbervie, Whakarewarewa (Whaka), Gwavas, and Berwick State Forests respectively. Not all clones were represented on all sites and the number of surviving cuttings within clone-site subclasses ranged from one to six. The cones were collected 12 yr after the planting.

At Glenbervie the trees showed symptoms that are normally associated with acute phosphorus deficiency, and foliar analyses have since revealed phosphorus levels which are typical of this condition (Burdon, in prep.). At Whaka two clones showed signs of an unidentified nutrient deficiency; otherwise growth was very vigorous. At Berwick the latitude was appreciably higher and the rainfall markedly lower than at the other sites.

At three sites a copious supply of pollen was ensured by large blocks of radiata pine growing adjacent to the plots, while the fourth (Whaka), although having less radiata pine growing in the immediate vicinity, is known to receive a significant pollen cloud from plantations in the locality.

On all sites conelet drop was prevalent. Aborted conelets were numerous but many had no doubt vanished, and quantitative study was impossible. However, in addition to outright abortion of conelets, a sprinkling of cones were obviously underdeveloped, being much smaller than the others and very light in relation to their size. Sometimes these obviously abnormal cones intergraded with fully developed ones; this created difficulties in the sampling procedure.

Sampling Procedure

Details of the sampling procedure are given elsewhere (Burdon and Low, 1973). Briefly, we attempted to obtain 10 well-developed cones from each clone on each site, all cones within a clone-site subclass being treated as a homogenous group. However, a highly unbalanced sample was unavoidable, 13 out of the possible 72 subclasses being missing, and all but 17 of the remainder containing less than the intended 10 cones, including two subclasses with single cones each. In all, 405 cones were studied.

Measurements

The measurement of volume, the counting of scales, and the obtaining of mean weight of full seeds, for each cone, are described elsewhere (Burdon and Low, 1973).

For this paper the following additional data were recorded for each cone: weight (g), total number of seeds (full and empty), and number of full seeds.

Calculations were then made of density (g/cc) and of full seeds as percentage of total seeds. In this study the original count of full seeds was rectified by crushing all the putatively empty seeds (i.e., those separated off in studying mean seed weight), and adding the number that proved to be full.

Analysis

Clonal repeatabilities of individual characters were calculated for each site from analyses of variance. Repeatability represents the ratio $V_C/(V_C + V_E)$, where V_C is the between-clones variance component, and V_E is the cones-within-clones (error) component (cf Burdon, 1971). V_C is a parameter of the particular clones at a site. In themselves, the calculated repeatabilities are of limited interest, largely because the sampling of cones was not strictly random. More important are the comparisons of repeatabilities, particularly between characters.

With the highly unbalanced classification, site means were calculated with an adjustment at those sites where clones were missing, and site differences were subjected to approximate tests of significance (Burdon and Low, 1973).

Regression analysis was used to study developmental inter-relationships between seed production characters, namely number of full seeds, percentage of full seeds (untransformed), and total number of seeds, on the one hand; and cone size, cone specific gravity, and scale number on the other. For each site within-clone linear regressions were fitted for one pair of characters at a time. Provided there was no serious between-clone heterogeneity of regression slopes, the pooled within-clone regressions were tested for significance, and the coefficients of determination (R^2) calculated. In addition, the within-clones coefficient of variation (CV) was estimated for each character according to the formula:

$$CV = \sqrt{V_E} \div S$$

where V_E is the pooled within-clones variance, and S is the overall mean for the site in question.

RESULTS

Clonal Behaviour

Calculated clonal repeatabilities for the different characters are listed, by sites, in Table 1. Repeatabilities for these characters are generally much lower than those for mean seed weight, cone volume, and seed number, which ranged from 0.44 to 0.81 (Burdon and Low, 1973).

TABLE 1—Calculated clonal repeatability, by sites, for characters

Character	Site			
	Glenbervie	Whaka	Gwavas	Berwick
Total no. of seeds per cone	0.06 NS	0.53***	0.41***	0.25**
No. of full seeds per cone	0	0.32***	0.33***	0.23**
% full seeds per cone (Arcsin transformation)	0.04 NS	0.12*	0.14*	0.23**

NS = not significant, $P > 0.05$

* = significant, $P < 0.05$

** = highly significant, $P < 0.01$

*** = very highly significant, $P < 0.001$

All characters conformed to roughly normal distributions, except raw percentage of full seeds (Fig. 1). At all sites, in particular Glenbervie, the distributions for this character showed marked tails below the means.

Female fertility was inherently high in all clones, since every clone produced at least one cone containing over 120 full seeds. Moreover, every clone included cones containing over 90% of full seed. At the other extreme, out of the 405 cones only three contained

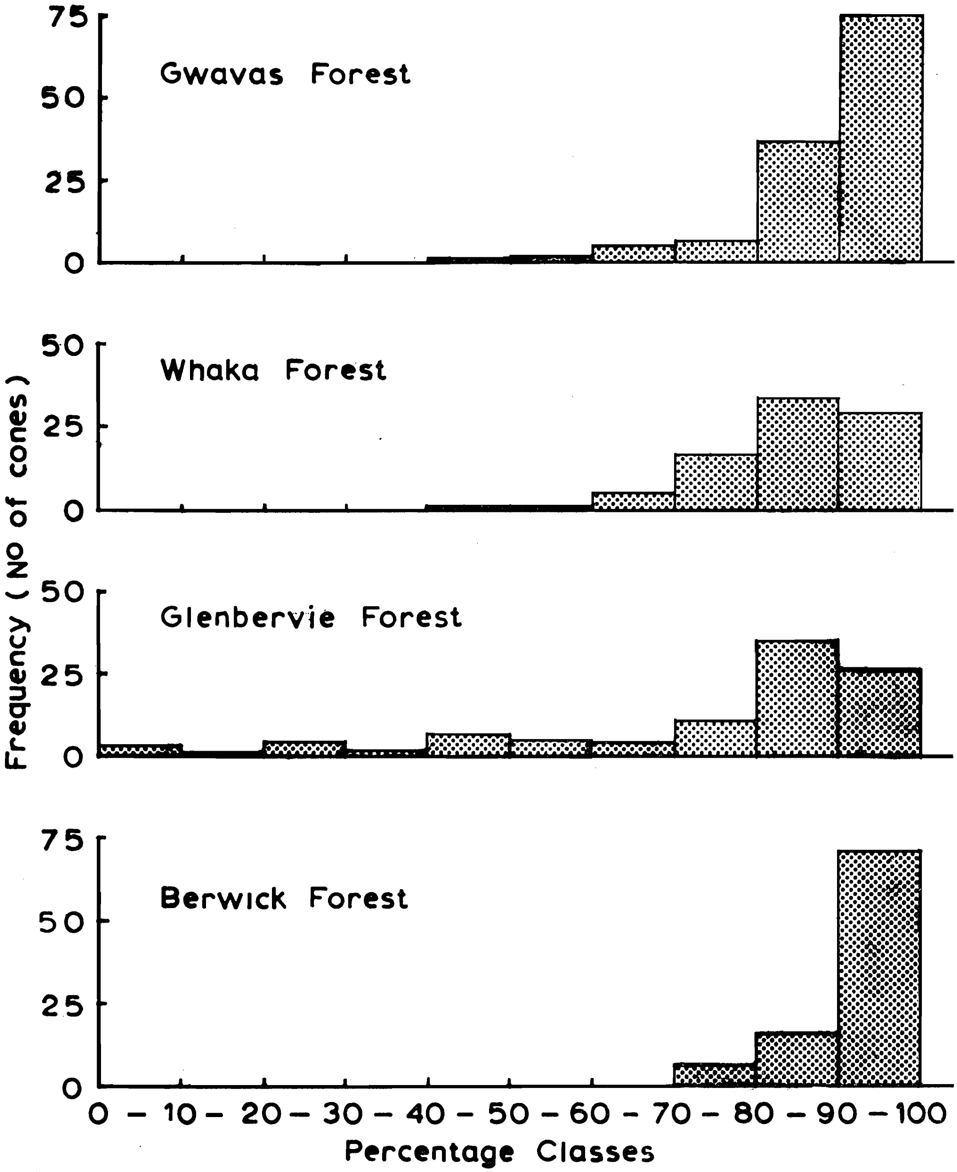

FIG. 1—Frequency distributions of percentage of sound seeds within cones, by sites.

totals of less than 40 seeds, full and empty, the least being 20. Although some cones at Glenbervie contained mainly empty seeds, these were not a consistent feature of particular clones.

Site Differences

Table 2 shows calculated means for the different characters, by sites. Mean seed weight, cone volume, and scale number (Burdon and Low, 1973), are included for comparison.

TABLE 2—Calculated site means for characters

Site	Character					
	Total no. seeds/cone	No. full seeds/cone	% full seeds/cone (untransformed)	Volume (cc)	Mean seed weight (mg)	Scale number
Glenbervie	105 b	81 b	76 b	128 c	23.4 c	196 ab
Whaka	144 a	123 a	85 ab	179 b	33.0 a	209 a
Gwavas	144 a	129 a	89 a	207 a	35.4 a	195 ab
Berwick	129 a	116 a	91 a	208 ab	30.5 b	189 b

Reading down columns, values with two letters in common do not differ significantly at the 1% probability level

Except with scale number, the means at Glenbervie were generally less than elsewhere ($P > 0.01$). Other differences were minor by comparison. Accepting $P > 0.05$ as the criterion of significance (subject to reservations with the tests used), cone volume was less at Whaka than at Berwick, and number of full seeds less at Berwick than at Gwavas.

Within-Clone Variation, and Inter-Relationships with Cone Characters

Results of the regression analyses are summarised in Table 3 which lists, for each site, the coefficients of determination (R^2) for pooled within-clone regressions, the significance of the regressions and the within-clone coefficients of variation (CV) of the characters involved. Pooling of regressions among sites was adopted only in the absence of heterogeneity of regressions and if residual variances differed by a factor of less than two.

Total number of seeds and number of full seeds showed considerable non-genetic variation (CV within clones = 15%-29% and 22%-47% respectively). Volume was similarly variable (CV = 20%-27%), but scale number much less so (CV = 7%-11%). Specific gravity was still less variable (CV = 3%-6%). Among sites Glenbervie showed high coefficients of variation for all three seed production characters.

The R^2 values show that, except at Berwick, where empty seeds were few, cone volume was related more to the total number of seeds than to the number of full seeds.

TABLE 3—Coefficients of determination (R^2) for pooled within-clone regressions and within-clone coefficients of variation (CV) for the characters involved

Regression	Site	R^2	P	CV_x	CV_y
Volume (y) on no. of full seeds (x)	Glenbervie	0.15	* * *	0.47	0.23
	Whaka	0.095	*	0.22	0.20
	Gwavas	0.26	* * *	0.24	0.27
	Berwick	0.21	* * *	0.29	0.21
	Combined	0.179			
Volume (y) on total no. of seeds (x)	Glenbervie	0.27	* * *	0.29	0.23
	Whaka	0.13	* *	0.15	0.20
	Gwavas	0.30	* * *	0.20	0.27
	Berwick	0.21	* * *	0.26	0.21
	Combined	0.222			
Volume (y) on % full seeds (x)	Glenbervie	0.004	NS	0.31	0.23
	Whaka	0.011	NS	0.11	0.20
	Gwavas	0.037	*	0.09	0.27
	Berwick	0.035	NS	0.06	0.21
Volume (y) on scale no. (x)	Glenbervie	0.23	* * *	0.09	0.23
	Whaka	0.094	*	0.07	0.20
	Gwavas	0.27	* * *	0.11	0.27
	Berwick	0.25	* * *	0.09	0.21
	Combined	0.214			
Total no. of seeds (y) on scale no. (x)	Glenbervie	0.20	* * *	0.09	0.29
	Whaka	0.34	* * *	0.07	0.15
	Gwavas	0.30	* * *	0.11	0.20
	Berwick	0.15	* * *	0.09	0.26
	Combined	0.235			
Density (y) on no. of full seeds (x)	Glenbervie	0.16	* * *	0.47	0.064
	Whaka	0.044	NS	0.22	0.033
	Gwavas	0.021	NS	0.24	0.065
	Berwick	0.059	* †	0.29	0.029
Density (y) on % full seeds (x)	Glenbervie	0.38	* * *	0.31	0.064
	Whaka	0.090	*	0.11	0.033
	Gwavas	0.13	* * *	0.09	0.065
	Berwick	0.005	NS	0.06	0.029

† R is negative

Also, cone volume was at least as closely related to total number of seeds ($R^2 = 0.13$-0.30) as to scale number ($R^2 = 0.094$-0.27). The latter two characters, although interrelated, were far from being completely so ($R^2 = 0.15$-0.34); so there is no evidence that the within-clone association between cone volume and total number of seeds is appreciably dependent on the corresponding association between number of seeds and scale number.

Although cone volume was correlated with both total number of seeds and scale number, it clearly showed considerable developmental variation which could not be related to either of these characters.

Within clones density was related more to percentage of full seeds than to actual number of full seeds. The former relationship was particularly marked at Glenbervie ($R^2 = 0.38$) where the percentage of filled seeds varied widely.

DISCUSSION

Clonal Repeatabilities and Gross Site Effects

The lower repeatabilities obtained for number of seeds per cone than for cone size and mean seed weight accord with Fielding's results (1964).

The main site effect was the depression of total number of seeds and percentage of full seeds, along with cone size and seed size, at Glenbervie. This almost certainly resulted directly or indirectly from the phosphate deficiency there, but it was not mediated by any effect on scale number. The indications of smaller cones but greater total numbers of seeds at Whaka than at Berwick are surprising, in view of the positive within-clone association at all sites between these two characters.

Seed Production and Inter-Relationships with Cone Development

Cone development in radiata pine has been described by Sweet and Bollmann (1971). Rapid volume growth occurs during two periods which roughly correspond to spring and early summer in consecutive years. Pollination occurs early in the first period, and rapid ovule growth and fertilisation during the second (B. Henderson, pers. comm. with G. B. Sweet). Final dry weight of the cone is reached about 3 months after final volume, and seed maturation apparently occurs even later. Most conelet drop occurs within 12 weeks of pollination (Sweet and Bollmann, 1970). In *Pinus sylvestris* Sarvas (1962) found most of the ovule abortion and conelet drop to occur during a comparable period.

In this experiment it appears that ovule abortion, as distinct from the occurrence of empty seeds, has been prevalent on all sites. This phenomenon was studied by Sarvas (1962), and generally results in the absence of a seed structure at the base of the wing, save for a small piece of disorganised tissue, within the normally fertile zone of the cone. If the ovule does not thus abort, it appears that a well-developed testa is almost always formed, and this would be counted as a seed, full or empty. It must be stressed, however, that evidence for the widespread occurrence of ovule abortion is entirely indirect. The evidence comprises the much greater non-genetic variability in number of seeds per cone than in scale number and the very incomplete within-clone association between these two characters. Implicit in the argument is the reasonable but unproven assumption that scale number and ovule number are closely correlated within clones. Although ovule abortion was presumably widespread, we have no estimate of its frequency.

The fact that, almost without exception, the cones sampled contained 40 or more seeds, full or empty, suggests that survival of a conelet may be related to a threshold number of surviving ovules. Sarvas (1962) in fact concluded that in *P. sylvestris* the threshold of conelet drop is related more to the number of aborted ovules than to the number of survivors. An effect of this sort could explain why one cone in our sample contained only 20 seeds. However, the sampling procedure was originally directed towards a study of genotypic variation in cone characters, and as such cannot be regarded as a very satisfactory basis for studying possible thresholds of cone development. The occurrence of partly developed cones could be due to injuries, or it could mean that any threshold is not clear-cut.

As stated earlier, conelet drop can be severe, at least in young propagules of radiata pine grown in New Zealand. However, the evidence is very limited as to whether this conelet drop follows from or is closely associated with ovule abortion. Sarvas (1962) attributed ovule abortion and conelet drop primarily to lack of pollination, yet Sweet and Bollmann (1970) found early conelet drop in radiata pine to be influenced very little by pollination, and they postulated that it resulted from nutritional competition within the tree. Moreover, Brown (1970), working with *P. sylvestris* in Britain, found that "cone drop" was partly independent of the degree of pollination.

Ovule abortion may well be an immediate result of nutritional competition with the strobilus, resulting from nutrient competition within the tree, apart from being the consequence of non-pollination. To establish the precise role of ovule abortion in conelet drop could be difficult, because any consequent or associated conelet drop may be determined before degeneration of the ovule becomes readily visible in sectioned material. Within this experiment it is considered unlikely that the apparently high incidence of ovule abortion could be accounted for by unpollinated ovules. Moreover, B. Henderson (pers. comm.) has observed ovule abortion in radiata pine even when the pollen chamber contained pollen grains. The smaller numbers of seeds at the phosphate-deficient site are consistent with nutritional competition being involved in ovule abortion, although other effects could have operated.

If, in fact, conelet drop is governed by a threshold number of surviving or aborted ovules, it would be expected that a low rate of conelet drop would reflect a low rate of ovule abortion in general, in which case the survival of ovules and thence the mean number of seeds per surviving cone would be high. We have already shown a significant positive association between total number of seeds and final cone volume, which is likely to be causal, particularly as cone volume is increasing very rapidly while absolute growth of ovules is greatest. Hence the expectation is that when the rate of conelet drop is low the surviving cones would tend to be large. This pattern, in fact, was observed by Sweet and Bollmann (1971) among the cone crops resulting from three consecutive pollination years. However, we can only say that there is some evidence consistent with an association between ovule abortion and conelet drop.

The within-clone association between cone density and percentage of full seeds accords with the well-known occurrence of occasional cones that are abnormally light and contain no full seed. Since cone dry weight rather than volume is increasing during the period of seed filling this relationship is understandable. By contrast, the weaker relationship between specific gravity and number of full seeds is noteworthy.

The occurrence of empty seeds was attributed by Sarvas (1962) largely to lethal embryo genotypes, which are a typical consequence of inbreeding. In our experiment, however, the incidence of empty seeds among the respective sites suggests that nutrient status can be an important factor. This in turn would suggest that competition with vegetative tissues may also affect the second phase of rapid growth of the cone.

CONCLUSIONS

1. Number of seeds per cone and percentage of full seed per cone have shown lower clonal repeatabilities within sites (0-0.53) than cone volume, mean seed weight, and number of scales (0.44-0.81).
2. Number of seeds and percentage of full seeds, like cone size and mean seed weight, appear to be severely depressed by phosphate deficiency.
3. Number of scales per cone, however, was not correspondingly affected.
4. Abortion of ovules within surviving conelets appears to cause much wastage in seed production; inadequate pollination is considered unlikely to be the sole cause of such abortion.
5. Available evidence suggests that conelet drop occurs if either the number of aborted ovules within the strobilus exceeds, or if the number of surviving ovules falls below, some threshold value.
6. Such a mechanism may account for the high rate of early conelet drop which has been observed in radiata pine in New Zealand.
7. Within clones, the number of developing ovules appears to be an important determinant of ultimate cone volume.

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