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VARIATION IN SEED SET ABILITY IN *VICIA FABA* L.

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Thesis submitted for the degree of Doctor of Philosophy

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STATEMENT

This thesis contains no material which has been accepted for the award of any other degree or diploma in any University and, to the best of my knowledge and belief contains no material previously published by another person, except when due reference is made in the text.

SUMMARY

*Vicia faba* is under consideration for inclusion in the farming rotation practised in southern Australia. It would serve as a cropping replacement of the pasture phase. The local potential of the species has not been established and most of the research and agronomic information, has come from Europe. In Europe the species has a reputation for fluctuating yields. This has been partly attributed to its dependence on bees for tripping and pollination. In southern Australia flowering of the crop coincides with the coldest and wettest part of the winter; conditions which are unfavourable for bee activity. It was decided to study variation in *V. faba* for the ability to set seed in the absence of bees and the influence this has on yield. The material consisted of introductions, and inbred and hybrid progenies derived from these introductions.

The term autofertility is used to describe the ability of plants, in the absence of bees, to set seeds when the flowers are not tripped. Self-fertility refers to their ability to set seeds when flowers are tripped or self-pollinated.

The experiments involved (a) an assessment of autofertility among a range of introduced populations and (b) a comparison of auto- and self-fertility among various inbred and hybrid generations. The experiments were conducted from 1974 to 1976 in the field, within a cage that excluded bees.

Among 100 introduced populations, many plants within the populations had a zero autofertility. Low autofertility was found in both small-seeded and large-seeded populations, but in a second study of 20 more uniform populations inbred for two generations there was an indication

that autofertility was negatively correlated with seed weight. The indices used for autofertility were the number of seeds and the number of pods. Among the populations there was no apparent association between yield and seed weight.

Tripping improved pod set and cross-pollination improved it even further. The only exception to this general rule occurred in material that had at the outset a very high autofertility and pod set. No further improvement in pod set was then obtained from tripping and cross-pollination and it is suggested that physiological limits were operating to prevent further increases in yield.

The number of seeds per pod was little affected by the flower treatments and there may be a limited capacity for change in this character.

The beneficial effect of cross-pollination was not considered to be due to overcoming incompatibility but to the process of pollination which ruptured the stigmatic structures and resulted in better pollen growth.

The fertility of inbreds was much lower than that of hybrids. Low autofertility of the inbreds is a consequence of inadequate pollination. This is evident when an increased pod set results from tripping. Inbreeding for one generation was sufficient to result in a response to tripping. Further inbreeding resulted in very low autofertility in some but not all lines. This suggests that inbreds may be selected with a high autofertility. Selection for this character must be carried out on advanced generation inbreds as heterozygosity strongly affects the characters. The material need<sup>d</sup> to be homozygous before selection is attempted.

A higher autofertility for yield, pod and seed set occurred in

hybrids than in related inbreds. In some instances the hybrids also produced more seeds per pod and heavier seeds than the inbreds. Because of their high autofertility the hybrids show a limited response to tripping. Again a physiological limitation to increases is suggested.

The level of self-fertility was a factor limiting the production of seeds after pollination was accomplished. Hybrids show high self-fertility but inbreds varied in the level of self-fertility from very low to as high or even higher than the hybrids.

Variation in the combined levels of auto- and self-fertility was found among the inbreds. Some had both low auto- and self-fertility; others had a low auto- but high self-fertility. Overcoming the tripping requirement in the latter would result in good yields.

With regard to the production of high yielding *V. faba* varieties, the study indicated that breeding might have as its objective high auto- and self-fertility inbred lines which would not be dependent on bees for tripping and pollination. Hybrid varieties have such qualities but are not considered feasible in the current situation. Breeding for both these possible objectives was considered in the discussion and compared with the more normal breeding objectives of producing improved populations which are natural mixture of inbreds and hybrids.

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TABLE OF CONTENTS

	<u>Page</u>
SUMMARY	i
ACKNOWLEDGEMENTS	iv
1.0.0 INTRODUCTION	1
2.0.0 LITERATURE REVIEW	3
2.1.0 Variation within <i>Vicia faba</i> .	3
2.2.0 The yield problem.	4
2.2.1 Cultivation and yields.	4
2.2.2 Cultural and other factors affecting yield.	4
2.2.3 Flowering, pollination and yield.	5
2.3.0 Pollination.	6
2.3.1 Insect pollinators.	6
(a) Type and efficiency of pollinators.	6
(b) Honeybees and yield of <i>V. faba</i> .	7
(c) Weather conditions and bee activity	8
2.3.2 The tripping mechanism.	13
2.4.0 Self- and cross-fertility.	13
2.4.1 Self-fertility.	13
2.4.2 Self- and cross-pollination.	15
2.4.3 Self-incompatibility	15
2.5.0 The breeding system.	17
2.5.1 Natural crossing	17

	<u>Page</u>
2.5.2 Characteristics of inbreds and hybrids in <i>Vicia faba</i> .	18
(a) Autofertility	18
(b) Self- and cross-fertility.	19
2.5.3 Inbreeding depression and heterosis.	19
2.5.4 Consequence of the breeding system.	20
2.6.0 Yield improvement.	20
2.7.0 Statistical methods.	23
2.8.0 Summary of literature review and relevance of overseas findings to the present investigations.	25
2.8.1 Overseas findings and present investigation.	26
 3.0.0 EXPERIMENTAL DETAILS	 28
3.1.0 The Experiments.	28
3.2.0 Site.	29
3.3.0 Climate.	30
3.4.0 The bee-proof cage	30
3.5.0 Material and methods	34
3.5.1 Experiment 1 (1974)	36
3.5.2 Experiment 2 (1976)	41
3.5.3 Experiment 3 (1975)	43
3.5.4 Experiment 4 (1975)	48
3.5.5 Experiment 5 (1976)	50
3.5.6 Experiment 6 (1976)	52



	<u>Page</u>
4.0.0 RESULTS	57
4.1.0 Experiment 1 (1974).	57
4.1.1 Autofertility of the populations.	57
4.1.2 Detailed studies on the restricted group of populations.	61
4.1.3 Conclusions.	67
4.2.0 Experiment 2 (1976).	69
4.2.1 Grouping of lines by seed characters.	69
4.2.2 Autofertility of the inbred lines.	69
4.2.3 The effect of the tripping and cross-pollination treatments.	77
4.2.4 Conclusions.	85
4.3.0 Experiment 3 (1975).	88
4.3.1 Plant growth.	89
4.3.2 Autofertility - Yield and its components.	89
4.3.3 The influence of tripping on yield characters.	94
4.3.4 Abortion of fertilized ovules.	104
4.3.5 Conclusions.	106
4.4.0 Experiment 4 (1975).	107
4.4.1 Autofertility - Yield and its components.	107
4.4.2 The effect of tripping on pod and seed set.	111
4.4.3 Conclusions.	111

	<u>Page</u>
4.5.0 Experiment 5 (1976)	115
4.5.1 Autofertility - Yield and its components.	115
4.5.2 The influence of tripping on seed set.	115
4.5.3 Conclusions.	118
4.6.0 Experiment 6 (1976).	119
4.6.1 Autofertility	119
4.6.2 The effect of tripping	125
4.6.3 Vegetative and floral characters	131
4.6.4 Conclusions	140
4.7.0 Miscellaneous studies	141
4.7.1 Vegetative propagation of <i>V. faba</i>	141
4.7.2 The effect of scarification of the stigma on pod and seed set	145
5.0.0 DISCUSSION	147
5.1.0 Autofertility	147
(a) Autofertility of the material studied.	147
(b) Early and late autofertility.	148
(c) Autofertility and seed size.	148
(d) Autofertility in inbreds and hybrids.	149
5.2.0 Effect of flower treatment.	150
(a) Effect of tripping.	150
(b) Effect of cross-pollination.	151
5.3.0 Characters associated with yield improvement.	152
5.4.0 Plant vigour and yield improvement.	153

	<u>Page</u>
5.5.0 Breeding considerations.	154
(a) The identification of genotypes with high autofertility	154
(b) Self-fertility	156
(c) Relation of progeny to parents.	156
(d) Open-pollinated populations.	157
(e) Inbred lines.	158
(f) Hybrid and production	158
5.6.0 Problems of experimentation with <i>V. faba</i> .	159
REFERENCES	161
APPENDIX	168

LIST OF TABLES

<u>No.</u>		<u>Page</u>
1	Mean monthly minimum and maximum air temperatures for stations in South Australia and U.K.	10
2	Mean number of days in the month in which the maximum daily air temperature exceed 15, 20, 25 and 30°C for the period 1961 to 1973 at Waite Agricultural Research Institute, South Australia.	11
3	Monthly minimum and maximum air temperature (°C) at Waite Agricultural Research Institute averaged for the years 1925 - 1973 and in 1974, 1975 and 1976.	31
4	Monthly rainfall (mm) and pan evaporation (mm, Australian tank) at Waite Agricultural Research Institute as long term averages, and for the years 1974, 1975 and 1976.	32
5	Mean solar radiation ( $\text{MJ.m}^{-2}.\text{day}^{-1}$ ) at the Waite Agricultural Research Institute; averages for the years 1965 to 1973 and in 1974, 1975 and 1976.	33
6	Daily minimum and maximum air temperature (°C) recorded within the experimental cage and at the Waite Agricultural Research Institute meteorological station in August 1975 and 1976.	35
7	Number of autofertile plants in each population	58
8	The autofertility (number of pods) of plants from a sample of population. The plants are ranked for autofertility.	60
9	The number of plants that set pods from early autofertility, late autofertility, from tripped flowers and from cross-pollinated flowers (The maximum number of plants is 18).	63
10	The number of pods and number of seeds per pod on a sample of 25 autofertile plants from flowers that were untreated, tripped and cross-pollinated.	65
11	The effect of flower treatment on pod set and seeds per pod of autofertile plants.	66
12	Seed and plant characters of 20 lines of <i>V. faba</i> .	70
13	Percentage of autofertile plants in small and large seeded lines of <i>V. faba</i> (% values based on 24 plants)	71

<u>No.</u>		<u>Page</u>
14	Correlation between yield and its components in untreated flowers.	76
15	The effect of flower treatment on the mean number of pods set per plot.	78
16	Percentage of flowers forming pods when flowers were untreated, tripped and cross-pollinated.	79
17	The regression equations for pod and seed number when flowers were untreated, tripped or cross-pollinated.	83
18	Mean seed numbers per pod from untreated, tripped and cross-pollinated flowers.	84
19	The regression equations for young and mature pods when untreated, tripped or cross-pollinated.	85
20	Mean number of stems per plant for the open-pollinated, inbred and hybrid progenies.	90
21	Mean yield, pod number <sup>per</sup> plant, seeds per pod and weight per seed for open-pollinated, inbred and hybrid progenies. The values were measured from untreated flowers.	91
22	Mean number of seeds per plant produced by untreated flowers on inbred and hybrid progenies of each seed parent.	92
23	Mean yield (g) per plant produced by untreated flowers of inbred and hybrid progenies of each seed parent.	93
24	Mean values for yield characters from tripped flowers of inbred and hybrid progenies.	98
25	Mean yield (g) per plant produced by tripped flowers of inbred and hybrid progenies.	100
26	Mean number of pods per plant produced by tripped flowers of inbred and hybrid progenies.	101
27	Mean number of seeds per plant produced by tripped flowers of inbred and hybrid progenies.	102
28	Analysis of variance of yield, pod and seed number of population 155 from tripped flowers.	103
29	Mean number of fertilized ovules that aborted per 100 pods.	105

<u>No.</u>		<u>Page</u>
30A	Yield characters of inbred and hybrid progenies - Population 41.	108
30B	Yield characters of inbred and hybrid progenies - Population 95.	109
31	Mean number of flowering stems per plant for inbred and crossbred progenies of determinate populations.	112
32	Mean number of pods and number of seeds per pod produced from untreated and tripped flowers of inbred and hybrid progenies.	113
33	Mean yield, pod number per plant and seeds per pod for I <sub>2</sub> and F <sub>2</sub> progenies (Untreated flowers).	116
34	The mean number of seeds per plant produced from untreated (control) and tripped flowers of I <sub>2</sub> and F <sub>2</sub> progenies.	117
35	Mean yield (g) per plant for progenies of different levels of hybridity.	121
36	Mean number of seeds per plant for progenies of different levels of hybridity.	123
37	Total number of seed produced by parent plants in the production of inbreds and hybrids. (Figures in parenthesis indicate the number of selfed seed).	124
38	Mean yield per plant (g) for parents, inbreds and hybrids -- all untreated flowers -- together with the calculated value for heterosis.	126
39	Mean number of seeds per plant for parents, inbreds, and hybrids.	127
40	The effect of flower treatment on the mean seed set per plant of progenies of different levels of hybridity.	129
41	Mean number of seeds per plant produced by untreated and tripped flowers.	130
42	Percentage heterosis of the crosses based on the seed set of tripped flowers.	132
43	The effect of flower treatment on the mean number of seeds in the pod of progenies of different levels of hybridity.	133

<u>No.</u>		<u>Page</u>
44	Mean number of seeds per pod produced from untreated and tripped flowers.	134
45	Mean plant height, stem number, stem weight and raceme number per plant for progenies with different levels of hybridity.	135
46	Mean plant height (cm) of parents, inbreds and hybrids.	136
47	Mean number of stems per plant for parents, inbreds and hybrids.	137
48	Mean stem weight (g) per plant for parents, inbreds and hybrids.	138
49	Mean number of racemes per plant for parents, inbreds and hybrids.	139
50	The effect of rooting compounds on the rooting of cuttings from different parts of the stem.	142
51	Rooting of cuttings with different number of nodes with and without terminal bud.	143
52	Mean number of roots produced from single node cuttings.	144
53	The number of propagules that develop to pod production.	144

LIST OF FIGURES

<u>No.</u>		<u>Page</u>
1	Daylength and mean daily solar radiation.	12
2	Distribution of plants for the number of days to first flowering.	39
3	Schematic diagram for the production of inbreds and hybrids for Experiment 6.	54
4	Distribution of populations for the percentage of auto-fertile plants in a population.	59
5	Distribution of 32 populations for percentage fertile plants when flowers were imtreated, tripped and cross-pollinated. <sup>ud</sup>	62
6	The relationship between the number of seeds and seed weight.	73
7	The relationship between the number of pods and seed weight.	74
8	Mean yield per plot for lines with different seed weight.	75
9	Some typical responses to flower treatment of lines with low and high autofertility.	80
10	The relation between the number of pods and the number of seeds for each flower treatment. (Each value is the plot mean).	82
11	The relation between the number of young pods and the number of mature pods for each flower treatment. (Each value is the plot mean).	86
12	The relation between inbreds and hybrids for the yield of untreated flowers.	95
13	The relation between inbred and hybrids for the seed set of untreated flowers.	96
14	Distribution of plants for the number of seeds produced from untreated flowers of open-pollinated, inbred and hybrid progenies of population 155 and 680.	97
15	Distribution of plants for the number of seeds produced from inbred and hybrid progenies of population 41 and 95.	110
16	Mean yield, seed number and seed weight of inbred, open-pollinated and various hybrid progenies.	120



1.0.0 INTRODUCTION

This study is concerned with the fertility and potential yield of *Vicia faba* in southern Australia. The species is under consideration for inclusion in the ley farming rotation, with its alternate years of cropping and pasture. The pastures usually consist of annual legumes such as annual medics or subterranean-clover (*Trifolium subterraneum*) (Webber, <sup>et al.</sup> 1976). Grain legumes could possibly replace the pastures in such rotations. They could provide high protein feed grains, improve soil nitrogen and avoid the continuity of disease and pest infestations that occurs if a cereal is grown year after year.

One grain legume being considered is *Vicia faba* known commonly as field beans or faba beans. In the past this crop has been grown in southern Australia only on a very limited scale (not rated a mention in the Statistical Register of South Australia). The local potential of *V. faba* has not been established and most of the research and agronomic information available has been derived from work in Europe. *V. faba* is grown in other regions of the world but few research reports have been published.

The climatic conditions in Europe, where beans are grown, differ greatly from those of southern Australia. The growing season in Australia begins with the autumn rains and the reproductive phase of the crop coincides with the coldest and wettest part of winter, unlike Europe where temperature and radiation increase during the reproductive phase. It is not known whether the fertility of flowers is greatly affected by winter growing conditions.

The European literature refers to the influence of bees on the yield of *V. faba*. Some of the important bee species present in Europe

do not occur in southern Australia. If the bee species existing here have a low activity during the winter, the yields of faba beans may be adversely affected.

There has been no locally adapted cultivar available in Australia. Recently introductions from the Mediterranean region and other areas where *V. faba* is grown have been made by the Waite Agricultural Research Institute. Little is known of the fertility of these introductions and their dependence on bee activity.

Breeding of *V. faba* has been undertaken in Europe, Egypt and North America. In some programmes, attempts have been made to utilise the hybrid vigour that occurs from crossing inbreds. One of the problems appears to be the poor ability of the inbreds to produce seeds and selection for high yield, using this approach, has met with limited success. Further information of the fertility of varieties when inbred or crossbred would be useful in determining yield improvement programmes.

In this study, therefore, the self-fertility of *V. faba* introductions is investigated together with the effect of inbreeding and hybridization on yield.

## 2.0.0 LITERATURE REVIEW

### 2.1.0 Variation within *Vicia faba*.

*Vicia faba* exhibits a diversity of forms that are classified into 2 subspecies, *paucijuga* and *eu-faba*, the latter consisting of 3 botanical varieties; var. *minor* Beck, var. *equina* Pers. and var. *major* Harz (Muratova, 1931).

The var. *minor* is known commonly as the tick bean and is characterized by small spherical seeds with a mean seed weight of 0.3g. At the other extreme of seed size is the var. *major* also called the broad bean with large flat seeds weighing 1.0g or more. Intermediate between these two groups is var. *equina* or the horse bean with oval seeds and an intermediate seed size. Generally the tick and horse bean are grown as field beans for animal feed and the broad bean type is used for human consumption (Smith and Aldrich, 1967).

Other names such as winter beans or spring beans differentiate between autumn and spring sown types. Both of these names are associated with the small-seeded type (Bond and Fyfe, 1962).

The growth habit of *V. faba* is usually indeterminate, but determinate type mutants with a terminal inflorescence are known (Sjodin, 1971; Lawes, 1973). However, some forms that do not have a terminal inflorescence have been observed in the present study to cease growth as a result of the senescence of the terminal bud. These forms are vegetatively determinate as opposed to those that are floral determinates. The term determinate is used to refer to such vegetatively determinate forms in the present study.

## 2.2.0 The yield problem.

### 2.2.1 Cultivation and yields.

*Vicia faba* has been cultivated since the Bronze age (Schultze-Motel, 1972). However, in spite of its early cultivation there has been little improvement and the crop has a reputation for unreliability with wide fluctuations in yield (Rowlands, 1955; Scriven and Allen, 1961). Yields in Britain vary from 1 to 6 t/ha (Lawes, 1974). The unreliable yield has been responsible for the decline in the area of the crop, particularly in Europe (Rowlands, 1955; Smith and Aldrich, 1967). Between 1962 and 1973 the area in Europe fell by 40% from 902,000 to 534,000 hectares (FAO, 1973). In the same period the area in the world devoted to faba beans declined slightly from 4.83 to 4.73m ha.

### 2.2.2 Cultural and other factors affecting yield.

Yields are influenced by different agronomic practices such as different sowing dates, plant densities, soil conditions and fertilizer applications (Soper, 1952a, 1952b, 1953, 1956; Hodgson and Blackman, 1956, 1957). The plant may produce over a hundred flowers but few of these develop into mature pods. Between 40 and 60% of the flowers may initially develop into pods but many of these abort before reaching maturity. Several workers have reported that only about 20% of all flowers produce mature pods (Soper, 1952b; Rowlands, 1955; Riedel and Wort, 1960; Inoue *et al.*, 1963; Akhundova, 1967; Kambal, 1969a; Graman, 1971). These mature pods develop from inflorescences on the lower part of the stem (Graman, 1971; Poulsen, 1972; Ishag, 1973a;

Adcock and Lawes, 1976). At a podding inflorescence one or two pods are found and these usually arise from the lowest flowers on the raceme (Inoue *et al.*, 1963; El-Tobgy and Ibrahim, 1968b).

The high proportion of flowers failing to develop young pods suggests that pollination may be inadequate (Drayner, 1959) while pod abortion may be due to adverse weather conditions such as low temperatures, storms, drought (Soper, 1952a; Ishag, 1973a) or to physiological conditions such as inter- and intra- ovary competition for assimilates and hormones (Kambal, 1969a).

### 2.2.3 Flowering, pollination and yield.

The sequence of events between flowering and the production of the mature seed has 2 phases, the first, from pollen production to fertilization and the second, from fertilization to the fully developed mature seed (Lawes, 1973, 1974). In the first phase various factors can affect fertilization of the ovules. The factors considered by Lawes (1973) included (a) the availability of adequate viable pollen, (b) the pollination process and (c) the fertilization process.

There has been no record of inadequate viable pollen limiting yield except in the case of male sterile plants (Bond and Fyfe, 1962; Bond *et al.*, 1964a, 1966a). Pollen viability of 80 to 90% was reported by Rowlands (1958) and variation in pollen quantity observed by Drayner (1959). The transport of the male gamete from the anthers to the ovum is, therefore, a more critical factor affecting yield and is reviewed in the following sections.

### 2.3.0 Pollination.

#### 2.3.1 Insect pollinators.

##### a) Type and efficiency of pollinators.

Bumblebees (*Bombus* species) and honeybees (*Apis mellifera*) are the main pollinating insects of *V. faba* in Europe (Poulsen, 1973; Kendall and Smith, 1975). Bumblebees do not occur in Australia (Michener, 1970).

Bees vary in the method in which they visit the flowers. The long-tongued species of bumblebees (e.g. *B. agrorum* and *B. hortorum*) as well as honeybees enter the mouth of the flower when collecting nectar or pollen, but the short-tongued bumblebees (e.g. *B. lucorum* and *B. terrestris*) gain access to the flower by biting holes at the base of the corolla. Some honeybees may also use these holes to gain access to the nectaries (Free, 1970).

The mode of entering the flowers affects the efficiency of the bees in causing pollination. In field bean, visits by honeybees and the long-tongued bumblebees cause a 66% to 71% pod set. In contrast unvisited flowers and flowers visited by the short-tongued bumblebees through holes at the base of the corolla produced 37% to 45% pod set. (Smith et al., 1974; Kendall and Smith, 1975).

The rate of visits to the flowers determines the usefulness of the species as pollinators. Entry through the front of the flowers was observed to be 4.34 visits per minute for honeybees and 7.03 and 10.25 visits per minute respectively for *B. agrorum* and *B. hortorum* (Poulsen, 1973). Free (1962) had reported previously that bumblebees worked at 2 to 3 times the rate of honeybees. In the pollination of the

crop the number of bees foraging is also important (Kendall and Smith, 1975).

b) Honeybees and yield of *V. faba*

In many countries honeybees are the main pollinators especially where bumblebees are few, or completely absent, as in South Australia. The contribution of honeybees to the yield of *V. faba* has been investigated in Britain. Studies on the effect of honeybees have been conducted using cages to confine or exclude bees. The cages were generally about 3m x 3m x 2m (e.g. Riedel and Wort, 1960; Scriven and Allen, 1961 and Free, 1966) and in some cases were smaller (Wafa and Ibrahim, 1960). The results of these studies have to be considered critically because bees do not always work satisfactorily when confined. In addition the cage may affect the environment of the plants. Riedel and Wort (1960) found that twice as many beans were produced on plants in open plots than within cages which excluded bees. The authors did not find any significant difference in the yield of plants caged with or without honeybees (17.6 and 13.1 beans per stem respectively). They explained the absence of a significant difference between plants caged with or without bees as being due to a greater number of beans produced by the upper inflorescences of plants caged without bees. When pollination was inadequate an increased yield from the upper inflorescences compensated for the low yield of the lower inflorescences.

Other authors have reported higher yields when bees were present in the cages. The exclusion of bees from the cages resulted in a 2.8% decrease in the number of mature pods when compared to control open-pollinated plots. Plants caged with bees had a 10.4% increase in pod

set (Wafa and Ibrahim, 1960). Similar results were reported by Scriven *et al.*, 1961). They obtained twice the yield when bees were present in cages than when they were excluded, Watts and Marshall (1961) reported a reduction of 30 to 40% in yield when bees were excluded from the cage. Similar experiments by Free (1966) also confirmed high seed sets and more seeds per pod due to the presence of bees. In Free's experiment, the adverse effect of caging plants was also evident. Plants in the open produced twice the yield of plants in cages.

Two characteristics of plants from which bees were excluded were their extended flowering period and late podding. The lack of pods on the lower part of the stem was compensated for by more pods produced higher up (Riedel and Wort, 1960; Wafa and Ibrahim, 1960; Watts and Marshall, 1961; Free, 1966; and Poulsen, 1972). Riedel and Wort (1960) suggested that the adequacy of pollination may be indicated by the distribution of pods on the stem. Pods clustered on the lower part of the stem are a sign of adequate pollination.

c) Weather conditions and bee activity

Temperature greatly influences bee activity. Honeybees do little pollination when the air temperature falls below 15°C (60°F)<sup>1</sup>. At temperatures below 10°C (50°F) and above 37°C (100°F) bee activity stops completely. The optimum temperature is about 33°C (92°F) (Grout, 1949). A maximum number of bees were reported to forage on red clover at a temperature of 33°C (Wratt, 1968). Windy conditions, approaching

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<sup>1</sup>The temperature in degrees centigrade was converted from the author's values in degrees Fahrenheit.



storms and wet conditions following rain are also unfavourable for bee-activity (Eckert and Shaw, 1960).

It is relevant here to draw a comparison between weather conditions in Europe and those that prevail during flowering in South Australia. The comparison is made to evaluate the likelihood that bee activity will present a problem in South Australia.

In Europe, *V. faba* crops are autumn or spring sown. In England they are predominantly autumn sown between August and mid-October. Spring sowing is from mid-February to March (Bond and Fyfe, 1962; M.A.F.F., 1970). The reproductive phase occurs in the summer between June and August (Soper, 1952, 1956). The average maximum temperatures during the reproductive phase are warmer in England than in South Australia (Table 1).

During the reproductive phase in South Australia (July to September, but in this country, the winter) maximum daily temperatures are around  $15^{\circ}\text{C}$  which are well below the optimum for bee-activity ( $33^{\circ}\text{C}$ ). July and August are the coldest months of the year. The few days during these months when bee activity will occur is evident from Table 2. The maximum temperatures occur only briefly in the afternoon since the daylength is short and solar radiation at a minimum (winter months, Fig. 1). In the northern latitudes the opposite is true. In Aberystwyth and Cambridge (Latitude  $52^{\circ}\text{N}$ ) the day length during the same months is about 16 hours. Solar radiation is also twice that received at Adelaide ( $35^{\circ}\text{S}$ ). Therefore, under the conditions of higher maximum temperatures, longer days and greater solar radiation bee-activity can be expected to be higher than in southern Australia.

TABLE 1: Mean monthly minimum and maximum air temperatures for stations in South Australia<sup>#</sup> and U.K.

MONTH	Minimum °C			Maximum °C		
	WARI	WPRS	PBI	WARI	WPBS	PBI
January	16.2	2.7	0.4	27.8	8.1	6.1
February	16.3	-0.7	0.7	27.4	6.2	6.9
March	15.4	2.5	1.3	25.6	9.4	9.4
April	12.9	3.1	3.9	21.5	12.4	12.4
May	10.5	5.1	6.6	17.7	13.4	16.6
June	8.5	9.8	9.2	15.1	18.2	19.9
July	7.7	10.7	10.8	14.1	18.8	21.1
August	7.9	11.3	10.7	15.1	19.8	21.1
September	9.2	10.8	9.1	17.6	17.1	18.8
October	10.8	10.9	6.6	20.3	15.8	14.9
November	12.6	4.6	2.9	23.2	10.4	9.4
December	14.5	0.7	1.1	25.7	7.2	6.9

<sup>#</sup> WARI Waite Agricultural Research Institute in South Australia; mean for 1925 to 1973.

WPBS Welsh Plant Breeding Station, Aberystwyth; mean for 1919 to 1969.

PBI Plant Breeding Institute, Cambridge; mean for 1958 to 1974.

Note: The reproductive phase of *V. faba* grown in S. Australia is from July through September while that in Aberystwyth and Cambridge occurs in June, July and August.

TABLE 2: Mean number of days in the month in which the maximum daily air temperature exceed 15, 20, 25 and 30 °C for the period 1961 to 1973 at Waite Agricultural Research Institute, South Australia.

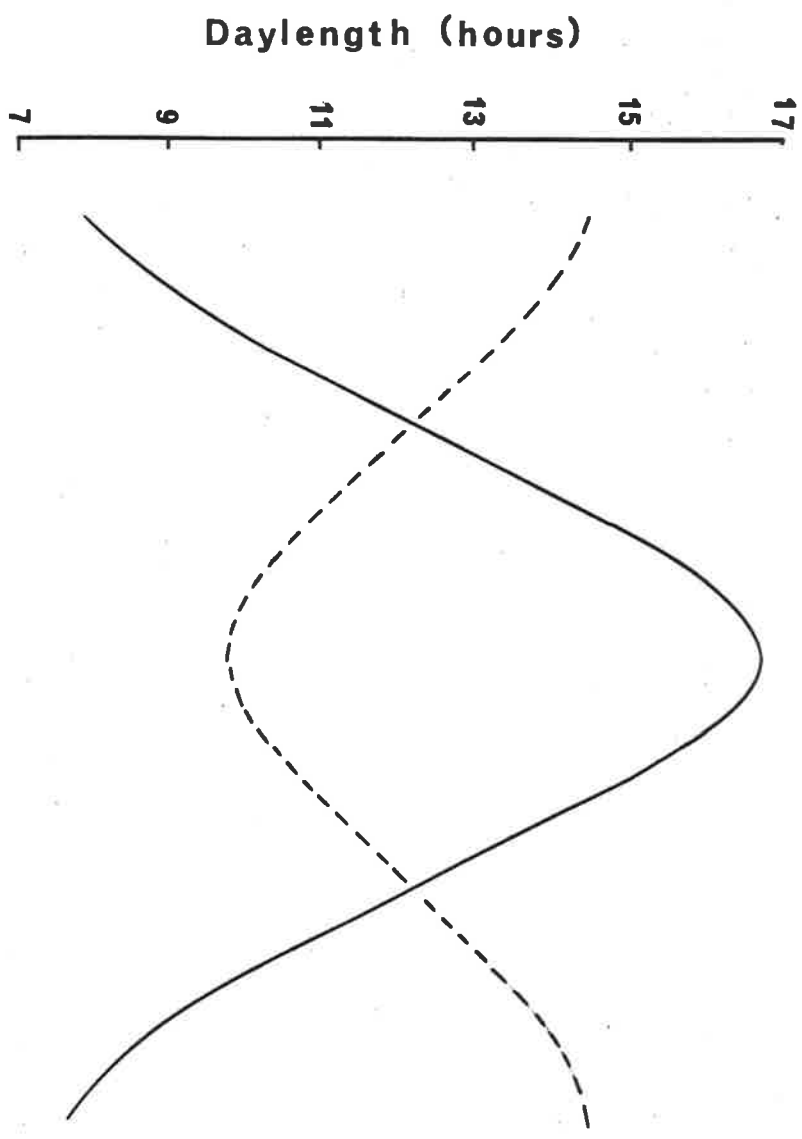
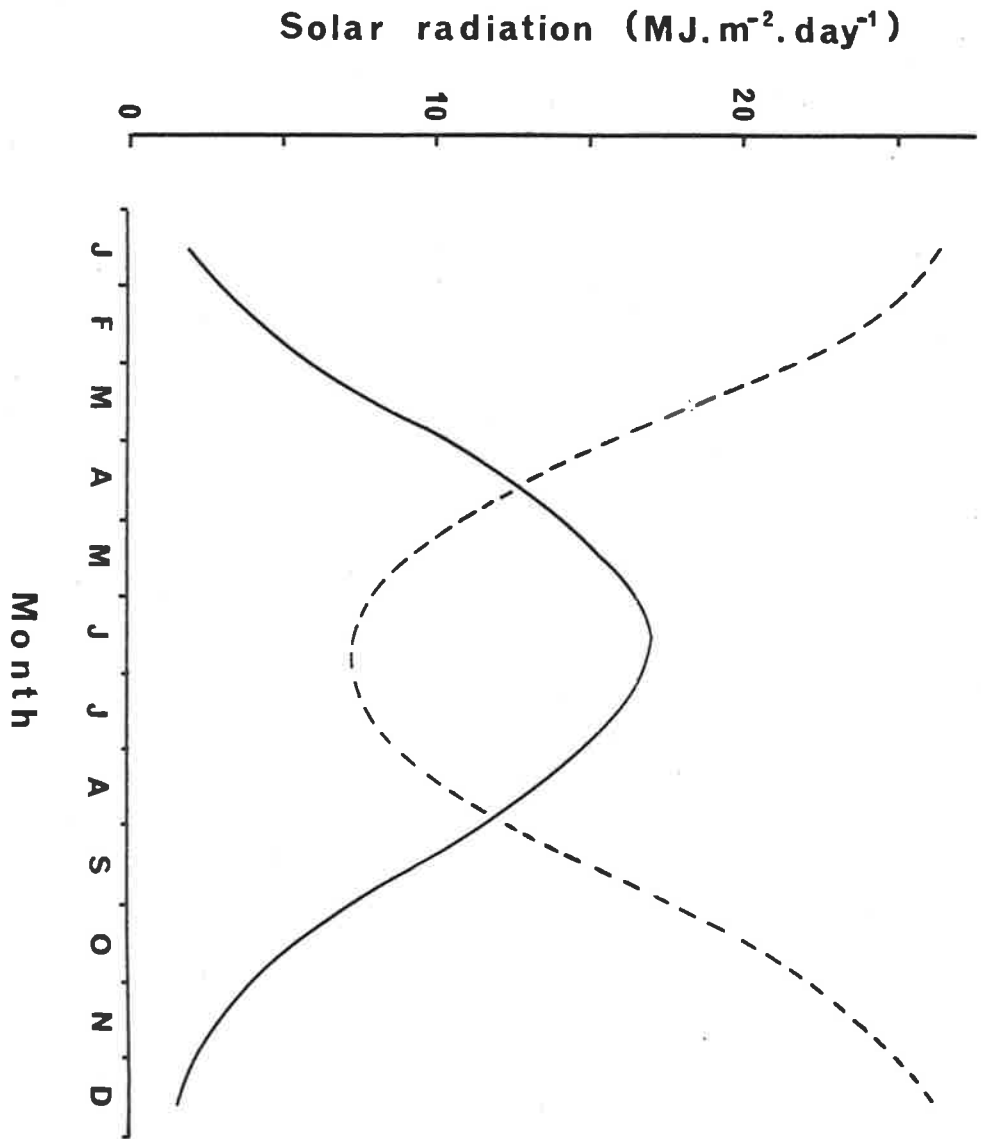
Month	Number of days exceeding			
	15°C	20°C	25°C	30°C
June	17.6	1.5	0	0
July	11.1	0	0	0
August	13.0	1.5	0.1	0
September	20.5	6.7	1.1	0.1
October	28.1	15.5	8.1	1.4

Note: There is little bee-activity below 15°C and optimum temperature of bee-activity is 33°C.

Figure 1

Daylength and mean daily solar radiation.

Solar radiation averaged for the years 1965 to 1973.



### 2.3.2 The tripping mechanism.

The consequence of bee visitation through the front of the flowers is to effect the release of the stigmatic column from the enveloping keel petals. This is referred to as tripping. It may be achieved manually by depressing the keel petals. Pollen is pushed on to the stigma at the same time (Rowlands, 1955; Drayner, 1956). An advantage from tripping has been demonstrated in *Vicia faba* and in general, tripped flowers yield more pods than untripped flowers (Rowlands, 1955, 1958, 1960; Drayner, 1956, 1959; Holden and Bond, 1960; Kambal, 1969a; Lawes, 1973).

In lucerne (*Medicago sativa*) the process of tripping has been reported to rupture the stigma and consequently encourage pollen germination (Armstrong and White, 1935; Brink and Cooper, 1936). A similar effect has been suggested as occurring in field beans. Tripping results in the breakage of tiny papillae on the stigma and their contents affect the germination of pollen (Rowlands, 1958; Holden and Bond, 1960).

Scarification of the stigma may also rupture the stigmatic papillae and influence seed set. Toynbee-Clarke (1974) obtained a better seed set from self-pollination following emasculation than self-pollination by tripping, and attributed the difference to the accidental scarification of the stigma during the emasculation process.

### 2.4.0. Self- and cross- fertility.

#### 2.4.1 Self-fertility.

Self-fertility is a term used to describe the property of some plants able to produce seeds through self-pollination. However, from the

literature relating to *V. faba* it is evident that a distinction needs to be drawn between plants that are capable of setting seeds from untripped flowers and those that required tripping before seeds are produced. Both types produce seeds through self-pollination.

In order to identify plants or seeds set from flowers that were untripped, the terms "spontaneous self-fertility" and "autofertility" were used by Drayner (1956, 1959). Such "autofertile" plants were also known as "self-tripping" by Hanna and Lawes (1967). The term "autofertility" is used in the present study to indicate plants able to set seeds without any tripping of the flowers.

The ability to set seed from self-pollination after tripping was referred to as "self-fertility proper" by Lawes (1973). An absence of pods and seeds from tripped flowers indicated poor "self-fertility proper". The abbreviated term, "self-fertility" is used in this study to indicate the ability of the plants to set seeds after self-pollination.

Various criteria and derived values have been used in studies of *V. faba* to indicate the extent of fertility. The number of pods and seeds produced by the plant, various ratios such as the number of seeds per 100 flowers, seeds per pod, pods per 100 flowers (percentage pod set) have been used (e.g. Drayner, 1959; Holden and Bond, 1960; Rowlands, 1964; Hanna and Lawes, 1967).

Differences in autofertility have been observed among *V. faba* genotypes, some have good seed set without tripping and show little response to tripping, while others with very low seed set in the absence of tripping had improved seed sets when tripped (Drayner, 1959; Holden and Bond, 1960; Rowlands, 1964; Hanna and Lawes, 1967).

#### 2.4.2 Self- and cross- pollination.

The yield improvement resulting from bee-pollination may indicate an advantage of cross-pollination in addition to that arising from tripping. If the amount of self pollen is inadequate or if some form of self-incompatibility mechanism is present, then an outside source of pollen would be beneficial. However, information on the need for cross-pollination has been inconsistent.

An improvement in seed set was found when 4th generation inbreds were cross-pollinated (Drayner, 1959). However, with commercial seeds, hybrids and earlier generation inbreds, Drayner did not find any difference in the seed set from self- or cross-pollination.

Other workers (Holden and Bond, 1960; Toynee-Clarke, 1974) did not find any difference in the number of seeds set from self- and cross-pollination. The latter author used highly inbred material (4th and 5th generation inbreds).

#### 2.4.3 Self-incompatibility.

Self-incompatibility, whether controlled sporophytically or gametophytically, involves a failure of pollen tubes to penetrate the stigmatic surface, a progressive reduction in the growth rate of pollen tubes in the style, or a failure after fertilization has occurred. In many plants the inhibitory action is expressed soon after pollen tubes enter the style (Williams, 1952).

Some evidence of inhibition of the rate of pollen tube growth by the stylar extract of the same flower was presented by Rowlands (1958). However this was not confirmed by other workers. Drayner (1959) did not



find any difference in tube growth of self and foreign pollen grown in pistil extracts on agar media although she noted some inhibition of pollen germination in extracts of the pistil of the same flower. Further studies involving the decapitation of the style at various intervals after pollination also failed to show any difference in the growth rate of self and foreign pollen. She obtained similar seed sets after self- and cross-pollination. Confirmatory results were obtained by Holden and Bond (1960). They felt that the stylar decapitation technique did not discriminate sufficiently between small differences in the growth rate of self and cross pollen and differences in growth rate may occur nearer the ovary. Using black hilum as a genetic marker, they studied the position of seeds resulting from self- and cross-fertilization. A more frequent cross-fertilization of the ovules nearer the stigma would have indicated a faster growth rate of foreign pollen. However, they did not show any difference in the proportion of cross-fertilized seeds at either end of the pod. There was also no evidence to suggest that self pollen was slow in growth since the number of seeds in the pod was the same for self- and cross-pollinations.

Other experiments on the proportion of selfed and crossed progenies following open-pollination did not indicate any tendency for seeds near the stem end of the ovary to be cross-fertilized. This may indicate no difference in the growth rate of self or cross pollen tubes (Hanna and Lawes, 1967). With the exception of Rowlands' report (1958) the studies do not support the presence of a self-incompatibility system.

The possibility of zygotic incompatibility was proposed by Rowlands (1961). He identified 3 factors that influence seed set. They were, (a) the basic fertility of the plant (number of fertile ovules),

(b) the tripping mechanism which favoured cross-pollination and, (c) the restriction of self-fertilization. Tripping did not result in a seed set more than 25% of the basic fertility and he suggested a form of incompatibility affecting the post-fertilization development phase which he called zygotic incompatibility. He proposed a genetic system in which the presence of homozygous recessive alleles resulted in sterility. With this system an increase in the number of genes affecting the character would cause the proportion of fertile ovules resulting from selfing to decrease logarithmically so that for 15 gene pairs, only 2% of self-fertilized ovules would be fertile.

#### 2.5.0 The breeding system.

##### 2.5.1 Natural crossing.

As discussed in the previous sections self- as well as cross-fertilization occurs in *V. faba* and there are no barriers between botanical varieties of *V. faba* from the small seeded var. *minor* to the large seeded var. *major* (Erith, 1930).

Estimates of natural cross-fertilization have been made. They range from 26% to over 65% outcrossing, depending on the cultivar, location and season (Fyfe and Bailey, 1951; Fyfe, 1954; Holden and Bond, 1960; Hanna and Lawes, 1967; Mar'tyanova, 1967; Poulsen, 1974, 1975). Also important are the size and location of the plants in the field (Bond and Pope, 1974). Plant density was shown to affect cross-pollination; plants spaced 6in (15cm) apart had 42%, while those 2ft. (61cm) apart recorded 65% cross-fertilization suggesting that high density reduced the access of bees to the flowers (Holden and Bond, 1960).

More cross-pollination occurred within a row than between rows (Kambal, 1969a). The flowers on lower inflorescence of a plant tended to outcross more (51%) than flowers on the upper inflorescences (33%) possibly due to changes in the external environment and bee-activity as the season progressed (Hanna and Lawes, 1967). Similar tendencies were reported by Poulsen (1975). An extremely low level of crossing (7%) was recorded by Oldershaw (1943). However, it is generally accepted that between 30 to 40% of the crop results from cross-fertilization (Hua, 1943; Fyfe and Bailey, 1951; Fyfe, 1954; Rowlands, 1958; Holden and Bond, 1960; Ivashkina, 1968).

#### 2.5.2 Characteristics of inbreds and hybrids in *V. faba*.

The outbreeding nature of *V. faba* is evident from the entomophilous flower shape and floral mechanisms and the reported levels of outcrossing. Inbreeding of a normally outcrossed species leads to deleterious effects. Most of the evidence on deleterious effects, relates to the fertility of the inbreds and hybrids, there is very little information on their vegetative vigour.

##### a). Autofertility.

Inbreds have a low autofertility and tripping is necessary for seed production (Drayner, 1959). In Drayner's experiments, 4th and 5th generation inbreds were practically sterile in the absence of tripping but their  $F_1$  hybrids had a high seed set and did not respond to tripping. A rapid decline in spontaneous self-fertility (i.e. autofertility in the terminology of this thesis) was also reported by Holden and Bond (1960). They postulated that spontaneous self-fertility was associated with

heterozygosity and was also possibly under the control of specific genes.

b). Self- and cross- fertility,

In her studies of hybrids, Drayner (1959) obtained equal amounts of seeds set from self- and cross-pollination of the hybrids. In the 2nd generation inbreds, there was no difference in the seed set from self- or cross-pollination but in the 4th and 5th generation inbreds, tripping (self-pollination) followed by cross-pollination produced more seeds. She was unable to confirm the advantage of cross-pollination over self-pollination in these inbreds (4th and 5th generation) when emasculated buds were pollinated with self and cross pollen. In other experiments, she found self and cross pollen to be equally effective in producing seeds. Comparing inbred and hybrids she did not find any evidence to show that pollen from inbred plants was less able to secure fertilization than hybrid pollen.

2.5.3 Inbreeding depression and heterosis.

Cytological evidence on inbreeding depression was presented by Rowlands (1958). In studies on inbreds from a large collection of small and large seeded forms of *V. faba* he found abnormalities occurring during meiosis, such as fragments and bridges resulting from stickiness and breakage of chromosomes. There was also a high chiasma frequency per bivalent,  $(3.15 \pm 0.05)$ . He concluded that such abnormalities were consistent with inbreeding a normally outcrossed species as was also found in rye.

Heterosis occurs in *V. faba*. (Bond and Fyfe, 1962; Bond, 1966,

1967, 1968; Abdalla, 1974). Hybrids produce more than their parents and inbreds. In some instances a 20% higher yield than the top parent was obtained.

#### 2.5.4 Consequences of the breeding system.

The breeding system of *V. faba* is intermediate between allogamy and autogamy and this is regulated by the autofertility of the plants (Drayner, 1959; Holden and Bond, 1960). These authors suggested that as outcrossing is about 30 to 40%, one-third of the population can be expected to be hybrids and the remaining two-thirds, inbreds in each generation. The hybrids will tend to produce selfed progenies through spontaneous self-fertility and the inbreds will tend to produce equal proportions of self and crossed progenies. This will result in one-third hybrids and two-thirds inbreds in the following generation. The cross-pollination level of 30 to 40% may thus be an equilibrium point whereby the 2:1 proportion of inbreds to hybrids is maintained.

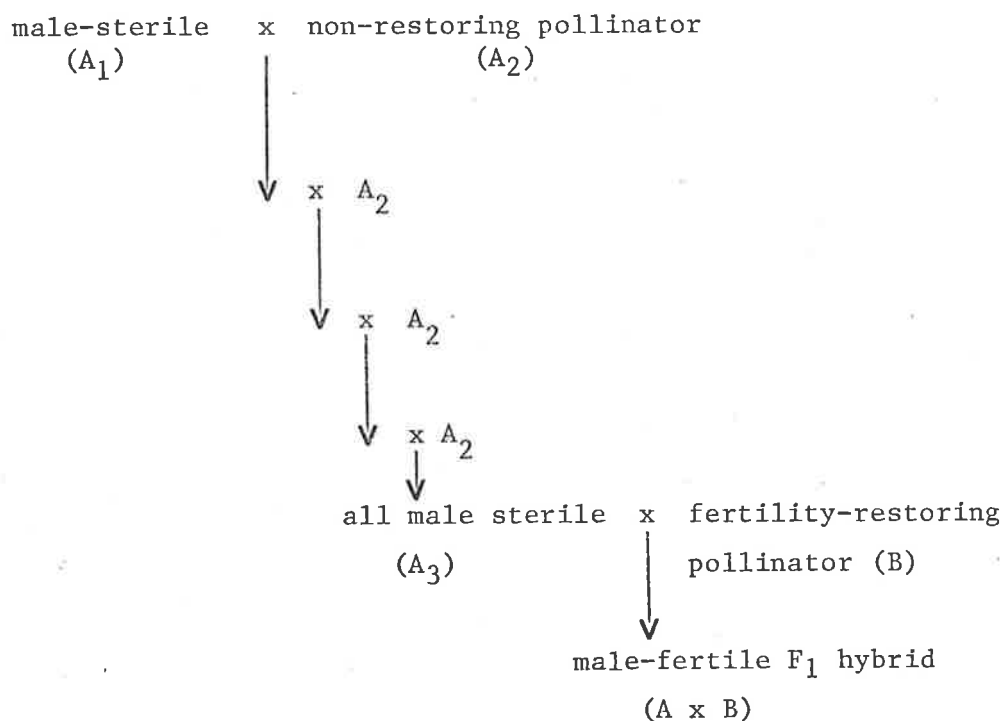
#### 2.6.0 Yield improvement.

A breeding system with an intermediate level of hybridity gives natural flexibility to the population in an evolutionary sense. It also enables the species to survive occasional seasons in which there is a failure of cross-pollination. But such a system is difficult to manipulate in a breeding programme (Drayner, 1959). Drayner felt that the selection of inbred lines from a heterogeneous population and their use as cultivars was unlikely to result in higher yields than the original population in which 30 to 40% of the plants were hybrids. She also suggested that a hybrid cultivar would not be stable but would self-pollinate

readily with subsequent loss of yield in the following generation.

It would be advantageous if high yielding material could be produced that was autofertile and thereby avoid a dependence on bees. As noted above there is considerable variation in the level of autofertility (e.g. Rowlands, 1958; Holden and Bond, 1960; Hanna and Lawes, 1967; Abdalla, 1974, 1975). Hybrids are more autofertile than inbreds (Drayner, 1959) but in some instances inbreds may be more autofertile than hybrids (Hanna and Lawes, 1967).

Selection for autofertility has given inconsistent results. Some workers have successfully increased the autofertility of selected inbreds over unselected material (Holden and Bond, 1960) but others such as Rowlands (1961) have been unable to improve autofertility with 3 generations of intensive selection. He suggested that the selection caused rapid fixation of genes and effective recombination could not occur in the absence of crossing. The exploitation of heterosis through the development of  $F_1$  hybrids was suggested by Bond and Fyfe (1962) and Bond (1967). Studies have indicated that the good yields of hybrids were not dependent on any specific combination of genes which could be fixed in an inbred line (Bond, 1966). The isolation of male-sterile (genetic and cytoplasmic) field bean plants should improve the feasibility of producing hybrids on a commercial scale (Bond *et al.*, 1964a, 1966b). The following scheme is an illustration of Bond's (1968) proposal for the production of hybrids.



The male-sterile is crossed to an inbred ( $A_2$ ) which has a non-fertility-restoring genotype. Their male-sterile progenies ( $A_1$ ) are backcrossed for several generations to the non-restoring inbred ( $A_2$ ) for several generations so that by the time the male-sterile is used in the final cross it is almost identical to Line A in all respects except that it is male-sterile ( $A_3$ ). The male-sterile line is used as the female parent and a fertility-restoring line is used for the pollen parent to produce male-fertile  $F_1$  hybrids.

However not all inbreds can be produced as male-sterile, some are partial restorers of fertility and when used as female parents in hybrid production result in a small degree of contamination of the hybrid seed crop with self seeds. The feasibility of using such mixtures of hybrids and parental inbred seeds have been studied (Bond *et al.*, 1966c). Mixtures of 5:1 of hybrid to inbred yielded as high as pure stands of the hybrid in some crosses but produced less yield in others.

Fertility has to be restored to the  $F_1$  hybrids and suitable fertility-restoring lines are necessary in making the cross. Problems have been encountered in obtaining full-restorers of fertility. Studies have been carried out by Bond *et al.* (1966b, 1966c) on the non-restoring and restoring ability of inbred lines.

In producing the hybrid seeds, the male and female parents are generally grown in alternate blocks and adequate pollinators are necessary to carry out cross-pollination. Small scale production has not experienced any problems with bee-pollination (Bond and Fyfe, 1962; Bond *et al.*, 1966a). However, production on a commercial scale involves large areas. Bond and Hawkins (1967) found a tendency in honeybees to avoid the male-sterile blocks because there was no pollen available to them. Pollination of male-sterile flowers was restricted to chance visitation by bees new to the plots. The bees soon move on to work in the male fertile blocks. Such problems have to be overcome before commercial production of  $F_1$  hybrid seeds is possible.

The development of inbred lines with high self-fertility has been proposed by Lawes (1973). He suggested a procedure of recurrent selection and assortative mating to prevent rapid fixation of genes and to allow for the recombination of desirable genes. This is a simpler approach and seeds could be produced by the grower.

#### 2.7.0 Statistical methods.

In the results to be presented it has been necessary at times to consider derived variables, transformation and ratios. In biological research derived or computed variables are frequently used. Such derived variables are based on two or more independently measured variables



whose relations are generally expressed as ratios, percentages, indices or rates (Sokal and Rohlf, 1969).

One disadvantage of a ratio is its relative inaccuracy when compared with the direct measurement. The measurement of continuous variables are approximations and the ratio of 2 approximations is subject to wide variation. Ratios also have distributions that may be unusual and not normal. Another disadvantage of ratios is that they do not provide information on the relationship between the 2 variables whose ratio is being taken.

Although derived variables present problems they are sometimes necessary to provide an understanding of certain types of biological phenomenon (Sokal and Rohlf, 1969).

The requirement that the error variances be normal is important for many statistical tests (Snedecor and Cochran, 1967). Moderate departures from normality are not serious but large departures can affect the validity of significance tests. Skewness in distributions tends to produce too many significant results (Bartlett, 1947).

In order to conform to the assumptions for statistical tests, various transformation such as angular, square root and logarithmic transformations have been developed. However, it is often difficult to obtain a transformation that can satisfy all the assumptions simultaneously. In some cases the original scale is more relevant and more understandable than the transformed scale. The use of transformation also creates problems in the presentation of standard errors or confidence limits for the estimates of means on the familiar original scale. This is due to the difference in the scale of the transformed and the original data. Where no simple transformation is satisfactory, non-parametric

tests can be substituted for the analysis of variance.

In each instance in the results where ratios or derived variables are used, reasons will be given for their use and alternative presentations considered.

#### 2.8.0 Summary of literature review and relevance of overseas findings to the present investigation.

From the literature reviewed it appears that the major factors in the unpredictable yield of the *V. faba* crop may be (a) the requirement for tripping and (b) the influence of hybridity on fertility.

The first factor is dependent upon foraging bees which to be effective must enter the flowers from the front and depress the keel petals. The amount of bee-activity varies with climatic and ecological conditions thus causing variation in yield.

The second factor is dependent on genetic mechanisms in the species and on its breeding system. These operate to maintain an intermediate level between inbred and hybrid plants in the crop. There has been little evidence to support the presence of an incompatibility reaction between pollen and style but a low autofertility has an effect similar to incompatibility. The inbreds with their low autofertility await bee activity to cause tripping and at the same time receive foreign pollen; the outcome being hybrid progeny. The hybrids are more autofertile than the inbreds and produce mainly selfed progeny. Such differences between the inbreds and hybrids control the proportion of each kind of progeny in each generation to give two-thirds of inbreds and one-third hybrids.

A consequence of such a system when breeding for yield is the difficulty in selecting effectively for high yielding autofertile types. Plant breeders are divided in their views. One school favours maximizing heterozygosity (and autofertility) through the development of hybrids and synthetic varieties. The other school believes that autoferile lines may be isolated through a recurrent selection programme of assortative matings. They believe these lines, although inbred, will be auto- and self-fertile and have a high yield even in the absence of bees. Progress has been achieved in each programme but difficulties are present in the commercial production of hybrids. Development of inbred lines may be a simpler approach.

#### 2.8.1 Overseas findings and present investigation.

It was decided to undertake study of *V. faba* introductions to determine whether their yield was increased by tripping or cross-pollination. Those introductions that have a high tripping requirement may yield poorly in the absence of bees. Others with a high autofertility may be less dependent on bees.

If high autofertility was desirable, one breeding approach would be to produce hybrids as these have been reported to be more autofertile than inbreds and not dependent on tripping of the flowers (Section 2.5.2). However, if the indication is correct that high autofertility may be selected among inbred material, then there may be some success in selection for autofertile inbred lines which are simpler to produce than hybrid varieties.

Hybrid vigour in *V. faba* has been reported (section 2.5.3). Further information is needed on whether the hybrids have a greater

vigour in addition to a higher autofertility than inbreds. If hybrids are basically more vigorous and productive than inbreds they should produce higher yields when both have been tripped. It is of interest to know the extent to which heterosis is expressed when pollination is non-limiting.

One aspect of low yield in *V. faba* is low self-fertility (Section 2.2.0). Self-fertility is affected by (a) the autofertility of the plant and (b) the capacity of the plant to produce seed from self-pollination (self-fertility proper, Section 2.4.1). There may be plants with both a low autofertility and a low self-fertility, plants that have a low autofertility but are highly self-fertile, and plants that are both highly auto- and self-fertile. In the selection for high yields in the absence of bees selection will be against the first two categories even though some plants may have a high self-fertility. This results in an unnecessary limit being placed on the range of material available for further improvement. Recognizing the difference between autofertility and self-fertility could facilitate yield improvement. It is of interest to know whether auto- and self-fertility can be readily identified.

### 3.0.0 EXPERIMENTAL DETAILS

#### 3.1.0 The experiments.

To answer the questions raised in the preceding Section 2.8.1, several experiments were carried out. They may be placed into two groups. One involved an assessment of auto- and self-fertility among a range of introduced populations (Exp. 1 and 2); the second involved comparisons of inbred and hybrid generations and the assessment of auto- and self-fertility in various progenies (Exp. 3 to 6).

The first experiment was set up in 1974 (Exp. 1) to determine the autofertility of a range of introductions and to determine their dependence on bee activity through their responses to tripping and cross-pollination. When the results of this experiment were available it was evident that plants within a population varied widely in autofertility. The difference in hybridity between the plants was considered to be responsible for this range of autofertility. A similar experiment was undertaken in 1976 (Exp. 2) using more uniform inbred populations.

The introduced populations in 1974 had exhibited differences in growth habit; some were indeterminate and the plants produced few stems whereas in others, terminal growth ceased soon after flowering (determinate) and the plants produced many lateral stems. The suitability of each type indeterminate or determinate for South Australian conditions was questioned. In Experiment 3 and 4 the autofertility and the response to tripping of the inbreds and hybrids derived from populations with indeterminate and determinate growth habits were investigated.

In 1976 a follow-up study on the autofertility and tripping response of some inbred progenies derived from Experiment 4 was made (Exp. 5). These inbred progenies had undergone an extra generation of selfing.

Also in 1976 an experiment (Exp. 6) was conducted on the effect of different levels of hybridity on auto- and self-fertility and the expression of these two characters in parental lines and their inbred and hybrid progenies. This experiment provided information on whether the level of autofertility of the inbred parents would deteriorate with further inbreeding and whether the level of autofertility of the hybrids would increase when they originated from crosses made between progressively dissimilar parents i.e. hybrids from plants of the same population as opposed to hybrids from crosses between populations.

### 3.2.0 Site.

The experiments were carried out at the Waite Agricultural Research Institute which is sited on sloping foothills, 122m above sea-level. The Institute is approximately 6.5 Km south-east of Adelaide.

The soil was a red-brown earth (Urrbrae series) which has about 25 cm or more of topsoil of fine sandy loam texture, a prismatic structured clay subsoil and a calcareous deep subsoil with waterworn gravel or stone (Litchfield, 1951).

### 3.3.0 Climate.

South Australia has a Mediterranean type climate with hot, dry summers and cool, wet winters. The cropping season begins in May (Autumn) and ends in November (Spring).

The mean monthly air temperature, rainfall, pan evaporation and solar radiation are given in Tables 3, 4 and 5. These meteorological data were recorded at the Institute's meteorological station located about 200m from the experimental sites.

The experiments were carried out in 1974, 1975 and 1976 in the field. The average temperature during the growing season in these years approximated to the mean for 49 years but rainfall varied considerably from the average. In 1974, 1975 and 1976 the seasonal rainfall (May 1 to November 15) was 523mm, 492mm and 340mm respectively. The long term average for the season was 455mm.

### 3.4.0 The bee-proof cage.

The studies on autofertility and the response to tripping and cross-pollination required plants to be grown in an environment free of bees. It was decided not to undertake experiments on plants in plots in a glasshouse as the environment was too dissimilar from that in the field. Instead a bee-proof cage was constructed in the field to provide an environment similar to that of crop conditions.

The cage measured 15m by 17m by 2m high and was covered with nylon netting (2mm x 2mm mesh). Zippers were fitted at the sides to enable entry and access to the plants. The cage was erected before flowering. It was used for all the experiments on different sites each

TABLE 3: Monthly minimum and maximum air temperature ( $^{\circ}\text{C}$ ) at Waite Agricultural Research Institute averaged for the years 1925 - 1973 and in 1974, 1975 and 1976.

Month	1925-1973		1974		1975		1976	
	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.
January	16.2	27.8	19.9	29.1	14.4	25.3	16.3	26.9
February	16.3	27.4	16.8	26.8	19.2	30.1	18.6	29.2
March	15.4	25.6	18.2	27.9	14.7	24.1	14.6	25.2
April	12.9	21.5	13.7	20.5	13.3	21.5	12.9	21.2
May	10.5	17.7	11.1	17.6	13.0	18.8	10.3	17.3
June	8.5	15.1	9.4	15.5	8.5	15.2	9.0	14.9
July	7.7	14.1	9.2	14.5	10.3	16.6	8.4	15.0
August	7.9	15.1	9.5	15.6	8.8	14.9	9.0	15.8
September	9.2	17.6	9.1	16.2	10.9	18.2	9.8	16.7
October	10.8	20.3	11.9	19.9	11.9	18.4	10.4	17.9
November	12.6	23.2	12.1	22.7	15.1	24.7	12.5	22.9
December	14.5	25.7	14.7	25.0	17.3	27.8	15.7	26.2



TABLE 4: Monthly rainfall (mm) and pan evaporation (mm, Australian tank) at Waite Agricultural Research Institute as long term averages, and for the years 1974, 1975 and 1976.

Month	1925-	1959-	1974		1975		1976	
	1973	1973	Rain	Evap	Rain	Evap	Rain	Evap
January	23.0	243	43.6	231	36.5	194	17.2	214
February	27.5	202	65.2	164	2.2	214	69.6	207
March	20.6	176	1.6	175	100.6	152	1.0	162
April	57.0	112	78.6	76	26.4	105	45.9	110
May	81.5	64	54.4	54	110.0	67	43.6	68
June	74.9	48	48.0	47	27.5	44	62.2	44
July	84.9	47	141.8	47	119.6	63	31.4	59
August	74.1	65	71.0	61	46.6	59	57.6	71
September	60.5	98	78.0	76	69.5	92	56.0	82
October	51.7	148	126.4	111	100.2	106	66.0	105
November	39.5	181	3.8	162	23.2	160	33.6	163
December	31.1	215	18.4	193	6.1	226	31.0	213
Annual total	626.2	1599	730.8	1397	668.4	1482	515.1	1498

TABLE 5: Mean solar radiation ( $\text{MJ.m}^{-2}.\text{day}^{-1}$ ) at the Waite Agricultural Research Institute; averages for the years 1965 to 1973 and in 1974, 1975 and 1976.

Month	1959 to 1973	1974	1975	1976
January	27.0	25.1	27.8	27.5
February	24.0	24.4	23.8	23.6
March	19.4	19.5	18.4	21.3
April	13.0	11.0	13.7	13.0
May	8.8	8.9	7.5	9.2
June	7.4	7.7	8.3	6.6
July	7.2	6.6	7.9	8.7
August	10.4	10.1	10.0	10.5
September	14.7	13.3	14.7	14.4
October	20.2	18.1	13.7	18.7
November	23.7	26.1	22.6	25.0
December	25.9	27.0	27.6	27.6

year.

It is considered that apart from excluding bees the cage did not affect significantly the environment of the plants. Reasons for this belief are:

1. The cage was large in comparison with those used by other workers which have been approximately 3m x 3m x 2m or smaller (e.g. Riedel and Wort, 1960; Wafa and Ibrahim, 1960; Scriven and Allen, 1961; and Free, 1966). The environment within large cages differs little from the outside (Pederson *et al.*, 1950).
2. Light transmission through the cage resulted in radiation levels 80% or more of ambient radiation. There was little restriction of air movement. The minimum and maximum air temperature were similar to those of the nearby meteorological station (Table 6).
3. The plots within the cage were located at least 2m from its edge and were surrounded by non-experimental plants. Border effects were thus minimized.
4. All the results to be presented were obtained from material within the caged environment and environmental effects were not confounded with any treatment effects.

Although the cage was large, the need to use it limited the size of the experiments that could be conducted in any one season and the number of plants in a treatment.

### 3.5.0 Material and method.

General experimental procedures such as land preparation, seeding, transplanting, fertilizer application and cultural operations were similar

TABLE 6: Daily minimum and maximum air temperature ( $^{\circ}\text{C}$ ) recorded within the experimental cage and at the Waite Agricultural Research Institute meteorological station in August 1975 and 1976.

Day	1975				1976			
	Minimum		Maximum		Minimum		Maximum	
	Cage	WARI	Cage	WARI	Cage	WARI	Cage	WARI
1	6.5	7.0	14.5	14.1	9.0	8.5	11.5	12.0
2	9.5	9.5	12.0	12.1	6.5	6.9	12.5	13.2
3	9.0	8.3	14.5	14.0	9.0	8.0	13.5	13.4
4	8.5	8.1	14.0	13.7	7.5	7.7	15.5	15.9
5	9.0	9.4	14.5	14.1	10.5	11.2	14.0	15.3
6	8.5	8.7	12.5	12.8	9.0	8.3	13.0	13.0
7	7.5	7.7	14.0	13.7	8.0	7.9	13.5	13.9
8	7.0	6.4	15.5	16.0	7.5	8.0	14.5	14.9
9	8.0	9.4	17.5	17.7	8.0	8.8	15.5	15.5
10	8.0	10.8	18.0	18.2	9.0	8.8	12.5	13.4
11	7.5	8.4	14.5	14.5	7.5	7.4	12.0	13.2
12	6.5	5.8	14.5	13.9	5.5	6.3	14.0	14.3
13	6.5	6.8	14.0	14.3	9.5	8.6	14.5	13.7
14	8.5	8.2	11.5	11.8	6.0	5.5	16.0	15.8
15	8.5	7.8	11.5	11.7	5.5	9.7	18.5	19.6
16	7.5	7.3	14.0	12.9	11.0	12.8	19.0	19.1
17	6.0	6.8	17.0	16.2	10.5	10.5	13.0	13.8
18	11.0	10.4	17.0	18.2	9.5	9.6	13.5	13.2
19	10.5	12.7	16.0	17.1	5.5	6.9	17.0	16.7
20	9.5	9.8	14.5	14.3	6.0	7.5	19.5	21.0
21	9.5	9.3	12.5	13.3	13.0	14.3	22.0	23.8
22	8.0	8.1	12.5	12.9	8.5	8.4	13.5	14.4
23	7.5	7.1	14.5	14.4	9.0	8.3	11.0	10.7
24	8.0	7.7	17.0	15.9	8.0	7.3	14.5	14.5
25	10.5	9.4	20.0	20.8	6.0	5.8	12.0	11.8
26	12.0	12.3	15.5	15.1	6.5	6.5	14.0	13.8
27	10.0	9.9	15.0	14.6	5.5	5.7	15.0	14.9
28	4.5	5.1	15.0	14.5	6.0	7.8	18.0	18.1
29	9.5	9.4	15.0	15.1	11.5	12.9	20.0	21.2
30	9.5	10.0	16.5	15.5	11.5	13.4	23.5	23.1
31	7.0	8.8	17.0	15.9	3.0	14.2	20.0	20.4

The cage temperature has been rounded off to the nearest  $0.5^{\circ}\text{C}$ .

throughout and will be described only for the first experiment (Exp. 1, 1974).

Details specific to each experiment are given separately. These include the experimental material, design and layout of the experiment, application of flower treatments and the analysis of data.

### 3.5.1 Experiment 1 (1974).

#### a). Material.

The objective of this experiment was to determine the auto-fertility of introduced populations and their dependence on bee-activity for pod set. The populations were open-pollinated introduction obtained by the Waite Agricultural Research Institute as part of a grain legume project. One hundred populations from various countries or regions (e.g. Turkey, Czechoslovakia, India, England, Mediterranean countries) were used. These populations varied in growth habit (indeterminate or determinate) and seed size. The availability of an adequate number of seeds for the experiment influenced the choice of populations.

The size of the bee-proof cage also imposed limitations and a choice had to be made between many plants of a few populations or few plants of many populations. As the purpose was to investigate the productivity of a new crop under local growing conditions, the choice favoured the inclusion of many populations. Thus, 100 populations were used with 18 plants of each.

The accession numbers, origin, seed weight, seed shape and test colour are given in Table 1 of the appendix.

b). Design and layout.

The 18 plants of the 100 populations were grown as single rows of 6 plants in 3 replications. A randomized complete block design was used. The plants were spaced 20cm apart and 50cm between rows. The experiment was surrounded by border plots.

c). Land preparation.

The experimental site was mechanically cultivated with a springed-tined harrow and superphosphate at 27 kg/ha P was applied and mixed into the topsoil. No further fertilizer application was made during the season.

d). Seeding and transplanting.

The experiment was sown on May 2, 1974.<sup>1</sup> A May sowing is customary for crops grown in this region which has a winter rainfall. The seed was inoculated with a commercial preparation of peat culture rhizobia ("Nodulaid", Group E) in a suspension of water immediately prior to sowing. The seeds were sown 3cm deep.

Where necessary replacement seedlings, sown at the same time as the trial, were transplanted to vacant positions taking care not to damage the primary root. No signs of set back and very little wilting occurred after transplanting. Less than 5% of the plants in the experiment were transplanted.

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<sup>1</sup>The sowing dates for all experiments are given in Table 2 of the appendix.

e). Cultural operations.

At intervals during the season the area was hoed to break up the surface crust and to control weeds. No other control was necessary.

f). Flower treatments.

The flowers on 4 stems of each plant were subject to some form of treatment. Inflorescence (racemes) on consecutive nodes were either (a) untreated, (b) tripped or, (c) tripped and cross-pollinated with pollen from another plant of the same population. The latter treatment is referred to as cross-pollination although seeds may have set from self- or cross-fertilization.

The treatment sequence was randomized for each plant, but on a plant the order was repeated up the stems. The 2 lowest flowers on each raceme were treated and the remaining flowers removed. A complete plant treatment involved 12 racemes on each stem (4 sets of 3 treatments).

The various treatments and racemes were identified with coloured rings made from insulated copper wire.

Treatment of the flowers was discontinued after the crop lodged on September 13, 1974 as a consequence of an exceptionally heavy storm with strong winds. Plants were entangled and any attempt at further access to the plots would have caused more damage. The crop was left to mature.

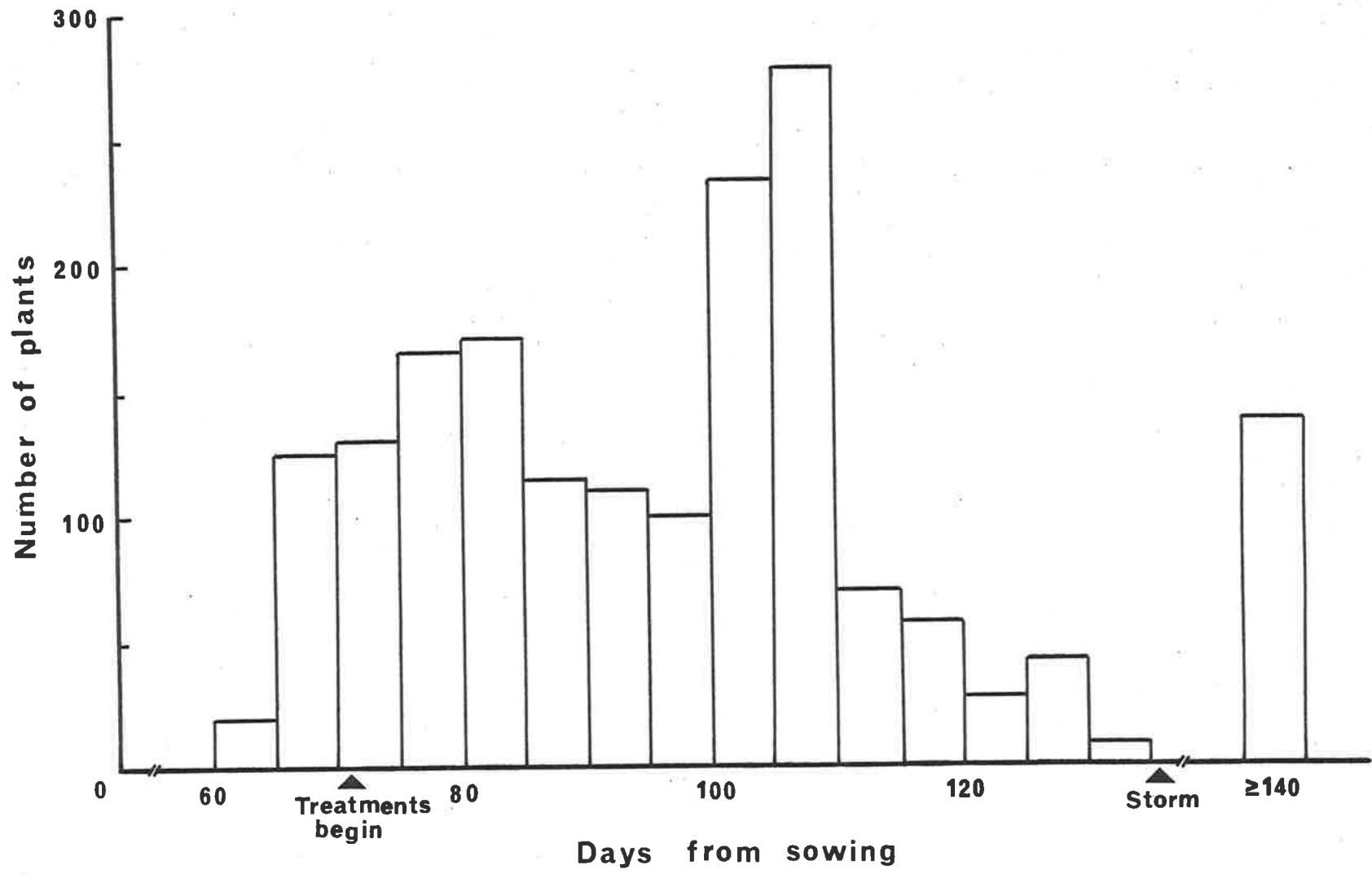
g). Data collection.

The lodging prevented completion of the flower treatments on all but 32 of the populations but it did not affect the assessment of auto-fertility as more than 90% of the plants had commenced flowering before the storm (Fig. 2) and continued to grow and form pods if they were

Figure 2

Distribution of plants for the number of days to first flowering.





autofertile.

The treatments on the 32 populations had been completed before the storm and these constitute the detailed studies on the response to flower treatment.

The experiment was harvested between November 18 and 29 and the pod and seed numbers recorded.<sup>1</sup> The yield was not measured as the seeds at harvest were in various stages of development due to differences in the flowering and maturation period of the population. An assessment of the flower treatment was considered to be more meaningful when based on the number of seeds and pods that developed.

The data on autofertility were determined from the pod set of the plants. A plant was considered autofertile if it carried at least one pod to maturity in the absence of tripping.

The number of seeds per pod was derived from the total number of seeds and the total number of pods produced per plant.

h). Analyses of data.

In all experiments, the original variables are presented together with tests of their significance. Ratios were avoided where possible, however it was meaningful to include seeds per pod as a component of yield.

The analyses were carried out on the University of Adelaide CDC 6400 computer using FORTRAN programmes and the STATSCRIPT package for the analyses of variance (Lamacraft, 1973).

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<sup>1</sup>The harvesting dates for all experiments are given in Table 2 of the appendix.

The comparisons between flower treatments were based on t-tests. A calculated statistic,  $t'$  was used to test the difference between 2 treatment means ( $d$ ) as follows:

$$t' = d/sd$$

where  $sd$  = standard error of the difference between the treatment means.

When the number of observations differed for each treatment, the  $sd$  is calculated from the variance of each. A weighted  $t$  value is used to test  $t'$  (LeClerge, Leonard and Clark, 1962).

The levels of significance used and notations are:

NS = Not significantly different at a probability of  $P = 0.05$ .

\* =  $P < 0.05$

\*\* =  $P < 0.01$

\*\*\* =  $P < 0.001$

Least significant differences (L.S.D.) are given for a probability of  $P = 0.05$ .

Standard errors (S.E.) are the standard errors of the mean unless otherwise stated.

### 3.5.2 Experiment 2 (1976).

#### a). Material.

The results from Experiment 1 showed that there was much variability between plants in a population. It was realised that the populations were recent introductions and may have been heterogeneous. Cross-pollination could have occurred during seed multiplication overseas or during the quarantine phase in Australia contributing to the variability. It was decided to repeat the experiment with inbred material

to obtain greater uniformity within populations. The number of populations studied was reduced to 10 small-seeded (*minor*) and 10 large seeded (*major*) populations to enable detailed flower treatments to be made. The inbred representatives of the populations are referred to as lines. Each line is the bulked inbred progeny of approximately 15 plants. Seed was produced as a result of autofertility.

b). Design and layout.

The 20 lines were grown in separate plots, each plot consisting of a row of 6 plants spaced 17cm apart. The distance between rows was 80cm to enable access and treatment of flowers. There were 4 replications of each line. The experiment was surrounded by border plots.

c). Flower treatments.

The flower treatments were similar to that of Experiment 1 except that only one stem per plant was treated, and untreated flowers on the raceme (i.e. from the third flower upwards) were not removed. Six plants per plot were subject to flower treatment. To ensure good contact between the foreign pollen and the stigma when a cross-pollination was being made, the original plug of pollen was first removed with a toothpick.

d). Discase control.

One line (Line 3) became infected by *Ascochyta fabae* after flowering had commenced but a foliar spray of Benlate (Benomyl 50% W/W) at 130g/100 l applied fortnightly checked the disease.

e). Data collection.

Plants were assessed for the number of young pods, mature pods, yield and seed number. These measurements were recorded for the untreated control, tripped and cross-pollinated flowers and for the remaining flowers on the plant.

f). Analyses of data.

The data were analysed as in Experiment 1 (Section 3.5.1 (h)).

The effect of tripping and cross-pollination on the flowers setting pods (pod set) was analysed separately for each line because the lines differed in the number of flowers treated. The percentage pod set was used only to indicate the relative change in pod set for each treatment (Section 4.2.3).

The relation between the number of flowers treated and the number of pods set, and between the number of seeds and the number of pods for each of the flower treatments was studied using regression analysis (Snedecor and Cochran, 1967).

3.5.3 Experiment 3 (1975).a). Material.

The populations studied in Experiment 1 varied in autofertility and growth habit (indeterminate and determinate). High autofertility was reported to be associated with heterozygosity, and inbreeding results in low autofertility (see Section 2.5.2, Lit. Rev.). The suitability of the indeterminate or determinate growth habit for local cultivation is not known and it was decided to study the fertility of inbreds and hybrids

from both types of populations.

The inbreds and hybrids were produced in the glasshouse in the summer of 1975 (Jan. to Mar.) with 30 plants from each of 10 populations. The inbred and hybrid progenies were produced from common seed parents to allow direct comparison to be made. For each seed parent there was 3 crosses. All the seed parents within a population were crossed to the same 3 pollen parents (as in a top cross) to allow comparison between pollen parents.

An emasculation and cross-pollination procedure was used for the production of hybrids.

The flower buds were emasculated about 5 days prior to flowering (Voluzneva, 1971). Emasculation involved peeling the calyx on one side of the bud to allow the side of the standard to be folded up out of the way. Using a pair of fine curved forceps the 10 anthers were removed through the opening at the top of the keep petals. The forceps were left slightly apart during the process to allow the style and stigma to slide through while the anthers were caught and removed.

Pollination was carried out immediately after emasculation to avoid drying out of the floral parts and the difficulties encountered in reopening the bud at a later date. Pollen was applied to the tip of the stigma with a toothpick or by inverting the pollen plug on the stylar brush of the donor flower directly onto the stigma.

The petals of the cross-pollinated bud were unfolded and set in place to prevent drying of the pistil.

During the crossing programme it became evident that success from cross-pollination was low. There was also inadequate fresh pollen from the male parents to pollinate all seed parents. Many selfed flowers

(assisted by tripping) also failed to produce pods. As a result six populations did not have plants that set an adequate number of inbred and hybrid seeds. The remaining four populations consisted of 2 indeterminate and 2 determinate types.

Each of the two indeterminate populations had at least 12 seed parents each with adequate seeds of the inbred and 3 hybrid progenies. However, the two determinate populations had only 6 seed parents each. These two populations had only one hybrid. Due to the differences in the number of seed parents and the number of hybrids available and in growth habit, it was decided to conduct two separate studies (Exp. 3 and 4).

The first study on the autofertility of inbreds and hybrids (Exp. 3) consisted of 2 small-seeded indeterminate type populations. One population was derived from Maris Bead (coded 155) and the other was an imported commercial tick bean (coded 680). For each population there was one inbred and 3 hybrids derived from each of the 12 seed parents available. The original open-pollinated material was included for comparison.

b). Design and layout.

The seed parent and pollen parents were nested within each population. It was decided to adopt a split-split-plot layout to minimize inter-plot variation. The populations were assigned to main plots and the various progenies (open-pollinated, inbred and 3 groups of hybrids) assigned to 5 sub-plots. The sub-plots were further divided into 12 sub-units for each of the 12 seed parents. The open-pollinated material which could not be related to any seed parent had an equal number of plants

as that of the inbred and hybrid progenies to allow later comparisons. The complete trial contained 1080 plants, i.e. (12 seed parents x 4 progenies x 3 plants) + 36 open-pollinated plants, replicated 3 times for each population.

The plants were spaced 17cm apart in the row and 80cm between rows.

c). Flower treatment.

The treatments consisted of tripping 4 racemes each on the main stem and the first lateral stem. Treatment was applied when at least 5 flowers were opened and could be tripped. All the plants within a replication were treated on the same day. One raceme was treated per plant on each day of treatment.

d). Disease control.

Control of bean rust (*Uromyces fabae*) during the late stage of crop growth consisted of foliar sprays with Plantvas fungicide (75% W.P., 2,3-dihydro-6-methyl-5-phenylcarbanoyl-1, 4-oxathin-4, 4-dioxide) at the rate of 1.3 g/l.

e). Data collection.

The yield, number of pods and seeds were measured separately for each plant and each treatment (untreated and tripped flowers). In addition for the untreated and tripped flowers, the number of fertilized ovules that aborted was determined from pods that reach<sup>ed</sup> maturity, i.e. pods with at least one mature seed.

A visual assessment of the height of inbreds and hybrids was made.



The plants were very tall at harvest and were growing up against the roof of the cage. The number of stems was recorded for each plant.

f). Analyses of data.

As mentioned in Section (b) above, the seed and pollen parents were nested within populations and separate analyses of the populations were necessary.

The first comparison was made between the open-pollinated, inbred and the 3 hybrid progenies for each population.

Source of variation	Degrees of freedom
Replications	2
Progenies	4
Error	8

In comparisons between inbreds and hybrids of each seed parent, the open-pollinated progenies were excluded as it could not be related directly to a seed parent. A split-plot analysis of variance was carried out for this comparison.

Source of variation	Degrees of freedom
Replications (R)	2
Progenies (P)	3
Error a (R x P)	6
Seed parents (S)	11
P x S interaction	33
Error b (R x S = 22 ) (R x P x S = 66 )	88

The Kruskal-Wallis non-parametric one-way analysis of variance based on ranked means (Steel and Torrie, 1960; Colquhoun, 1971) was used to test the difference involving percentage values.

#### 3.5.4 Experiment 4 (1975).

##### a). Material.

For reasons given in Section 3.5.3, this experiment was set up separately from Experiment 3. It differs from Experiment 3 in that the 2 populations from Crete and India (coded 41 and 95 respectively) were determinate in growth habit. In each population the inbred ( $I_1$ ) and hybrid ( $F_1$ ) progenies were produced from 6 seed parents. Within each population, the hybrids share a common pollen parent. The trial consisted of a total of 216 plants (2 populations x 6 seed parents x 2 progenies x 3 plants per replication x 3 replications).

##### b). Design and layout.

The 24 selfed and crossed progenies were grown in the usual 3-plant plots and were placed in a randomized block design with 3 replications.

##### c). Flower treatments.

Racemes were alternately tripped and untripped (control) on the main stem and first lateral stem. Tripping began from the second raceme produced and 2 earliest flowers on the raceme were tripped. The remaining untreated flowers were not removed. There was a maximum of 5 tripped racemes on each stem.

d). Data collection.

This was similar to that of Experiment 3.

Additional data was obtained for the main stem and first lateral on the date of first flowering, nodal position of first flower, number of racemes and flowers per raceme. The number of racemes that failed to develop into flowers (aborted racemes) on each of these stems was also recorded. The number of racemes that produced pods was recorded for all untreated flowers on the plant.

e). Analyses of data.

The statistical analyses of the data involved tests similar to those in Experiment 3. The initial analyses of variance for the entire experiment showed significant differences among progenies within females within populations ( $P < 0.001$ ) in the yield characters. Subsequent analyses was therefore carried out separately for each population to allow further interpretation of the results since the seed parents were nested within populations. The partitioning of the variation was as follows:

Source of variation	Degrees of freedom
Replications (R)	2
Progenies (P)	1
Seed Parents (S)	5
P x S interaction	5
( R x P = 2 )	
Error ( R x S = 10 )	22
( R x P x S = 10 )	

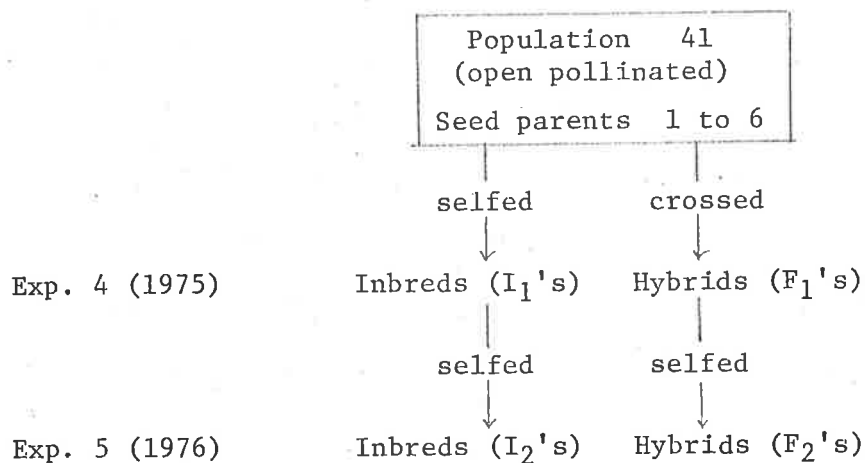
In the comparison between untreated and tripped flowers, only the pod and seed yields of the lateral stem was used. A variable number of flowers were treated on the main stem and on many plants there was not an adequate number of racemes on the main stem for the treatments. Main stem results were, therefore, excluded from the comparison to avoid the effect they may have had on the treatments and the need to use ratios such as the percentage of flowers podded, in order to make the results comparable.

### 3.5.5 Experiment 5 (1976).

#### a). Material.

This experiment was a follow-up study on the autofertility and tripping response of some inbred determinate progenies derived from Experiment 4. In all the other experiments inbreds and hybrids were derived from plants in the open-pollinated populations and it is not known whether these plants in the open-pollinated populations were themselves inbreds. Despite this uncertainty, the progenies have been called  $I_1$ 's as they are from one known generation of selfing. In turn their inbred progenies are called  $I_2$ 's. The crosses between plants of the open-pollinated populations are called Hybrids ( $F_1$ 's) and the inbred progenies of these hybrids are called  $F_2$ 's. The  $F_2$ 's represent one generation of inbreeding, which is less than any of the  $I_2$ 's. In this experiment the  $I_2$ 's are compared with the  $F_2$ 's.

The material used may be schematically represented:



In order to reduce the number of  $I_2$ 's and  $F_2$ 's in the trial so that the experiment could be manageable, only the  $I_2$  and  $F_2$  progenies derived from one  $I_1$  and one  $F_1$  plant of each seed parent of population 41 was used. There were thus six  $I_2$  and six  $F_2$  progenies derived from 6 original seed parents which will be referred to as Parent 1 to Parent 6.

b). Design and layout.

The plot size was doubled that of Experiment 4 and consisted of six plants in a single row. A randomized complete block design with 3 replications was used.

c). Flower treatments.

The flower treatments was applied to 2 stems on each plant. A total of eight racemes were tripped. Each raceme had an untreated control.

d). Data collection.

The usual yield, pod and seed set data was recorded on a single plant basis.

e). Analyses of data.

The analysis of variance was carried out as follows:

Source of variation	Degrees of freedom
Replications (R)	2
Progenies (P)	1
Female (F)	5
P x F interaction	5
(R x P = 2 )	
Error (R x F =10 )	22
(R x P x F =10 )	

3.5.6 Experiment 6 (1976).a). Material.

It became apparent from Experiment 3 and 4 conducted in 1975 that autofertility may be influenced by the parents. The objective of this study (Exp. 6) was to determine if autofertility was increased in crosses that involved progressively dissimilar parents (e.g. inbreds at one extreme to hybrids between different populations at the other) and whether the level of autofertility of the parents was similar to that of their offspring.

In such a study, a completely orthogonal design -- a diallel -- would have been preferable but this would have been extremely difficult to achieve. The parents were single plants; they could not be easily propagated by vegetative means (see Section 4.7.1) and inbred sister lines

had not been inbred for a sufficient number of generations to be considered as being genetically identical. From the nature of the faba bean plant with few pods and few seeds per pod, it is not practical to produce a large number of seeds of 8 crosses on each parent plant to achieve a diallel design.

A design was chosen that would include (a) crosses between plants of the same population ( $H_1$ ), (b) crosses between populations of the same seed size -- a small-seeded with a small-seeded population, a large-seeded with a large-seeded population ( $H_2$ ), and (c) crosses between a small and a large seeded population ( $H_3$ ). These crosses were to have been grown together with the inbred progenies ( $I_2$ ) of each seed parent in a comprehensive study (Fig. 3A). Figure 3A is symmetrical and provides for the range of crosses without being a complete diallel. An attempt was made to produce 8 sets of the design using different parents in the respective populations.

The complete set of crosses represented in Figure 3A was not achieved. Due to the very restricted production of seed from selfing and crossing the large seeded plants and some small-seeded plants, it was decided to use the available material to answer two questions on yield and fertility.

1. Is fertility and yield higher in hybrids produced between dissimilar parents than between similar parents?
2. Are differences in the fertility and yield of inbreds reflected in their hybrids?

To answer the first question inbred and hybrid progenies of crosses with increasingly dissimilar parents ( $I_2$ ,  $H_1$ ,  $H_2$ ,  $H_3$ ) were studied (Fig. 3B). These progenies had a common seed parent. There were

Figure 3

Schematic diagram for the production of inbreds and hybrids for Experiment 6.

$I_1$  = Inbreds with one generation of inbreeding used as parents.

$I_2$  = Selfed progenies of  $I_1$  plants.

$H_1$  = Crosses between parents of the same population.

$H_2$  = Crosses between different populations but of the same  
seed size.

$H_3$  = Crosses between populations with different seed size.

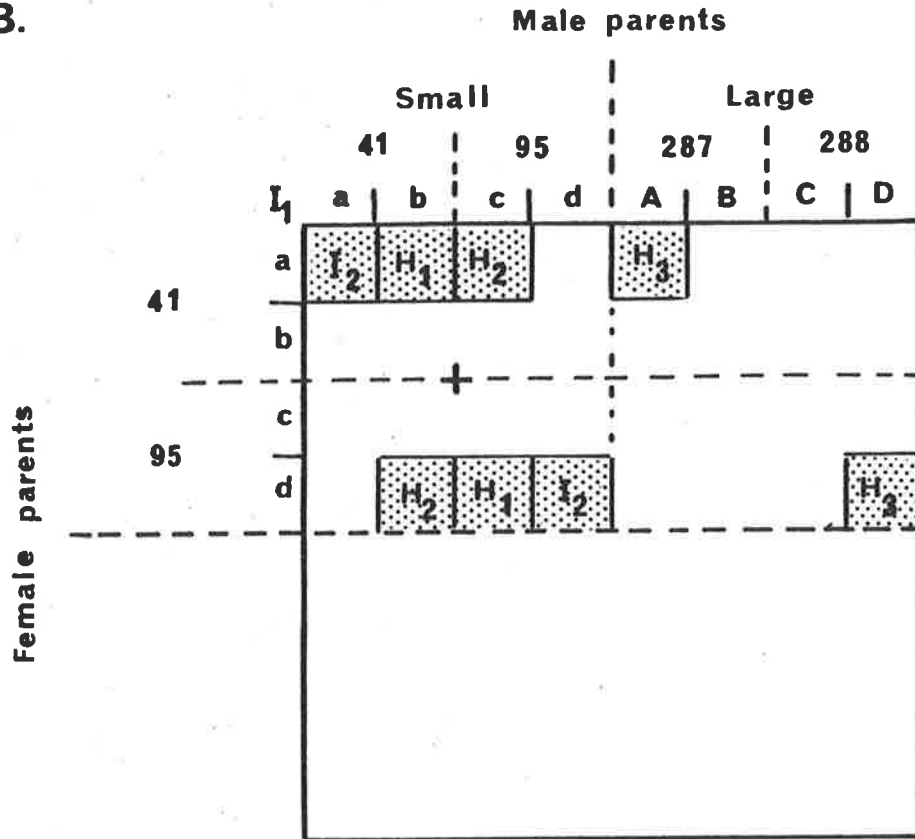


3 A.

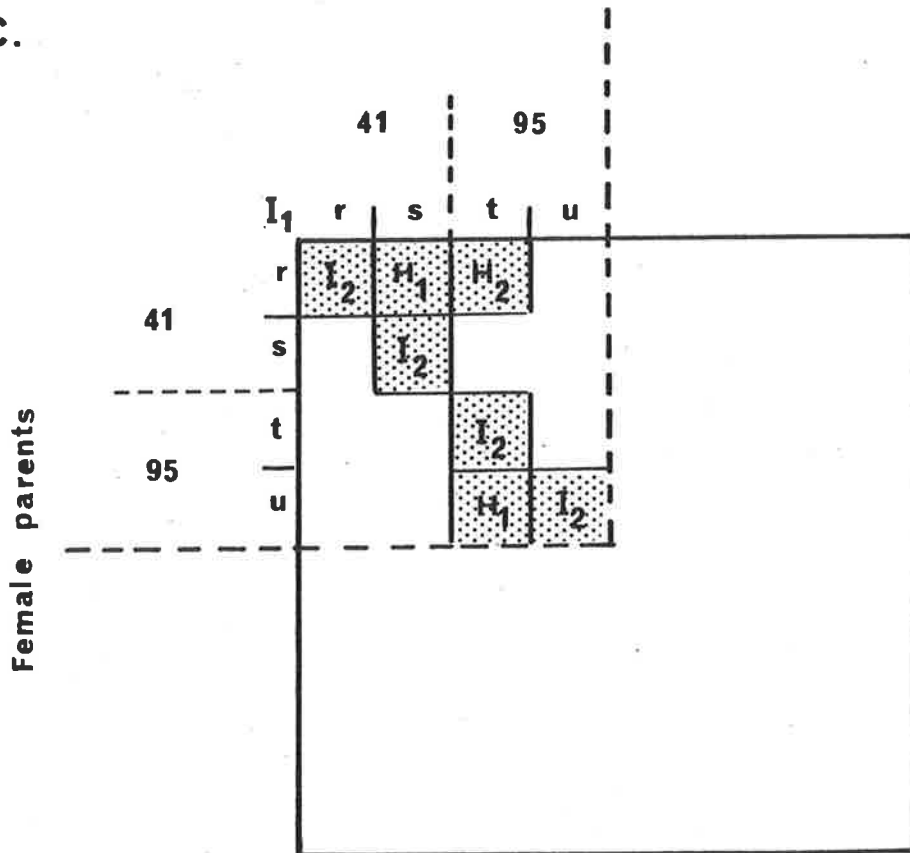
MALE PARENTS

		Small seeded				Large seeded				
		Pop. 41		Pop. 95		Pop. 287		Pop. 288		
		a	b	c	d	A	B	C	D	
FEMALE PARENTS	Small seeded	I <sub>1</sub>								
		a	I <sub>2</sub>	H <sub>1</sub>	H <sub>2</sub>		H <sub>3</sub>			
		b	H <sub>1</sub>	I <sub>2</sub>		H <sub>2</sub>		H <sub>3</sub>		
		c	H <sub>2</sub>		I <sub>2</sub>	H <sub>1</sub>			H <sub>3</sub>	
	Large seeded	d		H <sub>2</sub>	H <sub>1</sub>	I <sub>2</sub>				H <sub>3</sub>
		A	H <sub>3</sub>				I <sub>2</sub>	H <sub>1</sub>	H <sub>2</sub>	
		B		H <sub>3</sub>			H <sub>1</sub>	I <sub>2</sub>		H <sub>2</sub>
		C			H <sub>3</sub>		H <sub>2</sub>		I <sub>2</sub>	H <sub>1</sub>
	D				H <sub>3</sub>		H <sub>2</sub>	H <sub>1</sub>	I <sub>2</sub>	

3 B.



3 C.



progenies from 8 seed parents, 4 from each of populations 41 and 95 in the study. The inbred parental line ( $I_1$ ) from which the seed parents were obtained and the open-pollinated progenies of populations 41 and 95 were included.

To answer the second question the hybrid ( $H_1$ ) was studied with the seed and pollen parental material ( $I_1$ ), and inbred progenies derived from the seed parents ( $I_2$ ) as shown in Figure 3C. The hybrid between 2 small seeded populations ( $H_2$ ) was also included. There were 4 sets of the progenies  $I_2$ ,  $H_1$ ,  $H_2$  derived from 8 parents (4 from each of population 41 and 95).

b). Design and layout.

The regular 3-plant plot was used. The entries were randomly assigned to plots of a randomized block replicated 3 times.

c). Flower treatments.

These were identical to that of Experiment 5.

d). Measurement of vegetative characters.

Plant height was measured 4 weeks prior to harvesting (Nov. 18, 1976). The height above ground of the folded terminal bud on the tallest stem on the plant was measured.

The weight of tops (above ground parts) was measured after drying at 75°C. Most of the leaves had shed by harvest and the weight obtained was mainly that of the stems and petioles.

The number of stems refer to the flowering stems produced early in the season. Small shoots resulting from late rains were not included.

e). Analyses of data.

In the first group compared (Section (a) above) the analysis for the progenies ( $I_2$ ,  $I_1$ , OP,  $H_1$ ,  $H_2$ ,  $H_3$ ) from seed parents of each population is as follows:

Source of variation	Degrees of freedom
Replications (R)	2
Progenies (P)	5
Seed parents (S)	3
P x S interaction	15
(R x P = 10)	
Error (R x S = 6)	46
(R x S x P = 30)	

In the second group all the crosses and their parents ( $I_1$ ) and inbreds were analysed together and a common standard error used in the comparisons made between parents, inbreds and hybrids.

The percentage heterosis for yield and seed set relative to the mid-parent and the high parent was calculated as follows:

$$\% \text{ above mid parent} = \frac{H - MP}{MP} \times 100$$

$$\% \text{ above high parent} = \frac{H - P}{P} \times 100$$

where H = hybrid,

$$MP = \text{mid-parent} = \frac{\text{Parent 1} + \text{Parent 2}}{2}$$

P = higher yielding of the 2 parents

#### 4.0.0 RESULTS

##### 4.1.0 Experiment 1 (1974).

##### 4.1.1 Autofertility of the population.

This experiment was concerned with the ability of the 100 populations to produce pods without tripping, in a bee-proof cage. It was an assessment of autofertility. A plant was considered autofertile if it carried at least one pod to maturity from untreated flowers.

The number of autofertile plants in the populations ranged from 0 to all 18 plants (Table 7). The distribution of populations for autofertility is given in Figure 4. At one extreme, it was found with population 56 that not one of its plants was autofertile whereas at the other extreme in population 84 all of the plants were autofertile. Both populations belonged to the small-seeded type. A majority of the other populations had about 9 out of the 18 plants autofertile. There was no evidence that autofertility occurred more frequently in populations with small than with large seeds and similar distributions were obtained for both types (Fig. 4B and 4C).

Among the plants the level of autofertility varied from 1 to 97 pods per plant. There were very few plants that produced more than 20 pods (only 71 out of the total of 1800 plants). This category of plants was found in 30 populations. The range of autofertility among the 18 plants in a sample of populations is shown in Table 8. Very few populations have plants of high autofertility.

TABLE 7: Number of autofertile plants in each population.

Pop.	No.	Pop.	No.	Pop.	No.	Pop.	No.
1(L)	11	26(L)	11	51(L)	12	76(L)	3
2(S)	8	27(S)	14	52(S)	8	77(L)	16
3(S)	13	28(L)	7	53(S)	7	78(S)	9
4(L)	9	29(S)	9	54(L)	13	79(S)	7
5(S)	11	30(L)	4	55(S)	10	80(L)	2
6(S)	1	31(L)	13	56(S)	0	81(S)	10
7(S)	5	32(L)	7	57(L)	10	82(S)	2
8(S)	10	33(L)	5	58(L)	7	83(L)	9
9(S)	10	34(L)	10	59(L)	9	84(S)	18
10(L)	8	35(S)	8	60(S)	9	85(L)	10
11(L)	7	36(L)	8	61(S)	12	86(S)	10
12(L)	16	37(L)	9	62(S)	12	87(L)	10
13(L)	7	38(L)	9	63(L)	12	88(S)	9
14(S)	10	39(L)	12	64(L)	11	89(S)	13
15(S)	5	40(L)	4	65(L)	13	90(L)	9
16(L)	10	41(L)	5	66(L)	3	91(L)	15
17(L)	11	42(S)	13	67(L)	14	92(S)	12
18(L)	12	43(L)	6	68(L)	10	93(S)	16
19(L)	9	44(L)	2	69(L)	8	94(L)	8
20(L)	7	45(S)	16	70(L)	4	95(S)	10
21(L)	9	46(L)	11	71(S)	7	96(S)	13
22(L)	5	47(L)	1	72(S)	11	97(S)	14
23(L)	10	48(S)	16	73(S)	8	98(S)	9
24(L)	9	49(S)	12	74(L)	4	99(S)	7
25(L)	6	50(L)	6	75(L)	7	100(S)	7

(S) Small seeded type

(L) Large seeded type

Figure 4

Distribution of populations for the percentage of auto-fertile plants in a population.

- A. All populations (100)
- B. Small seeded populations (42)
- C. Large seeded populations (58)

**A. All populations**

**B. Small seeded populations**

**C. Large seeded populations**

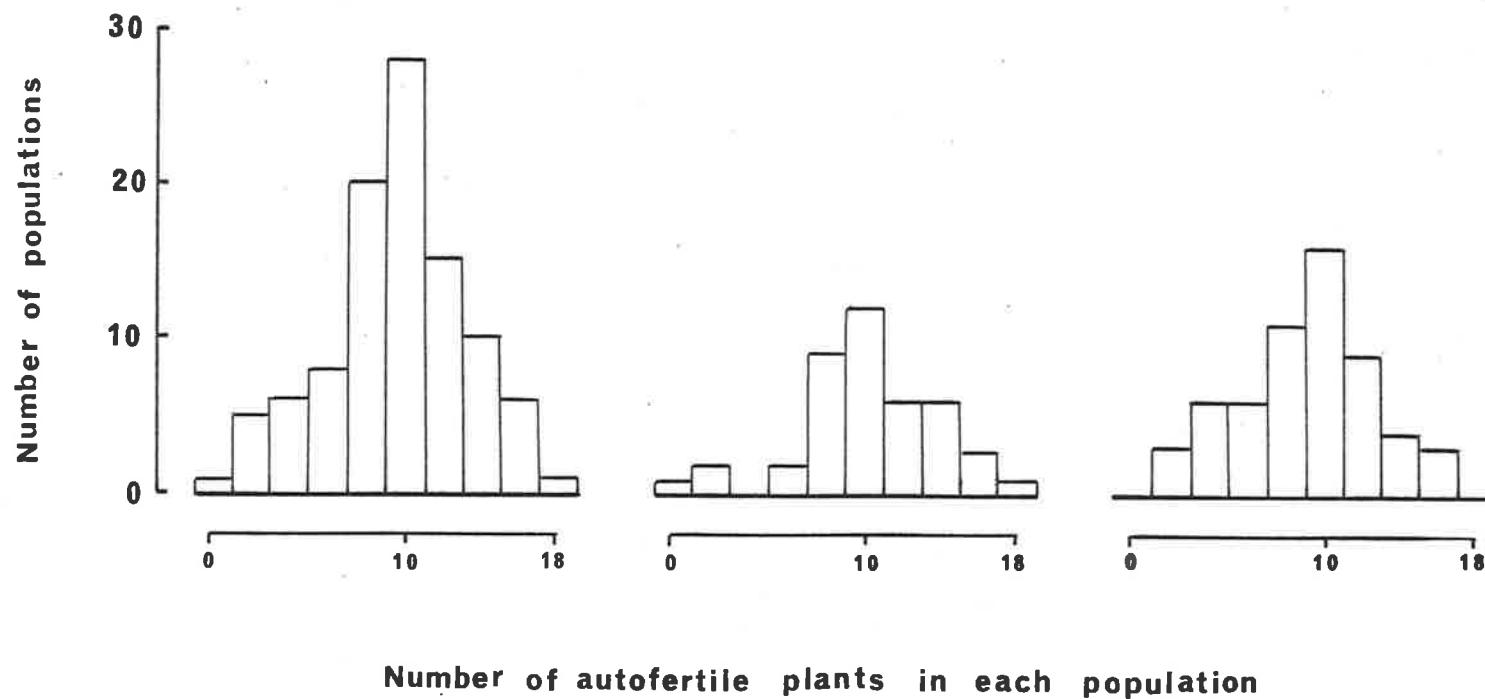




TABLE 8: The autofertility (number of pods) of plants from a sample of population.  
The plants are ranked for autofertility.

Population number	Plants within the population																	
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2
20	0	0	0	0	0	0	0	0	0	0	0	1	2	2	2	3	4	9
36	0	0	0	0	0	0	0	0	0	0	1	2	2	3	3	5	5	7
1	0	0	0	0	0	0	0	1	1	2	2	2	3	3	5	6	17	19
51	0	0	0	0	0	0	0	1	1	1	1	2	2	2	3	4	11	12
62	0	0	0	0	0	0	1	1	1	2	3	3	4	6	7	8	20	58
3	0	0	0	0	0	2	2	2	3	5	10	15	20	37	38	39	48	48
27	0	0	0	0	1	2	4	8	11	13	16	18	24	25	28	34	39	40
45	0	0	1	2	4	9	10	11	13	14	19	20	20	22	30	40	69	71
84	1	2	3	3	4	6	6	7	10	10	10	12	13	16	20	21	25	97

#### 4.1.2 Detailed studies on the restricted group of populations.

##### a). Effect of late flowers and flower treatment on the number of fertile plants.

As mentioned in the Material and Methods only 32 of the 100 populations had been sufficiently treated before the storm of September 13 to provide data on the effect of tripping and cross-pollination.

As the flower treatments were discontinued after the storm, all the control, tripped and cross-pollinated flowers were those produced early in the season. Many plants did not produce pods from the early flowers that were untreated (i.e. the control flowers of the treatments). However at harvest some plants that had not been autofertile during the treatment period produced pods from late flowers high up the stem. There were 174 plants autofertile early in the season (out of 576) but 281 plants were autofertile by the end of the season (Fig. 5, Table 9). Almost all populations improved in autofertility as the season advanced. One population (No. 84) which had 14 autofertile plants early in the season bore pods on all 18 plants at the end of the season. Two populations (65, 73) had the same number of autofertile plants early as well as late in the season.

The improvement in the number of fertile plants early in the season by tripping and cross-pollination treatment of the flowers varied from population to population. In some populations the number of fertile plants increased when flowers were tripped but no further increase was obtained with cross-pollination (e.g. No. 15, 22, 31). In other populations, tripping increased the number of fertile plants and cross-pollination increased the value further (e.g. No. 20, 29, 53). Tripping increased the number of fertile plants from 174 in the control to 383

Figure 5

Distribution of 32 populations for percentage fertile plants when flowers were untreated, tripped and cross-pollinated.

- A. Untreated (early flowers)
- B. Tripped
- C. Cross-pollinated
- D. Untreated (all flowers)

A, B. C are comparable (early flowers)

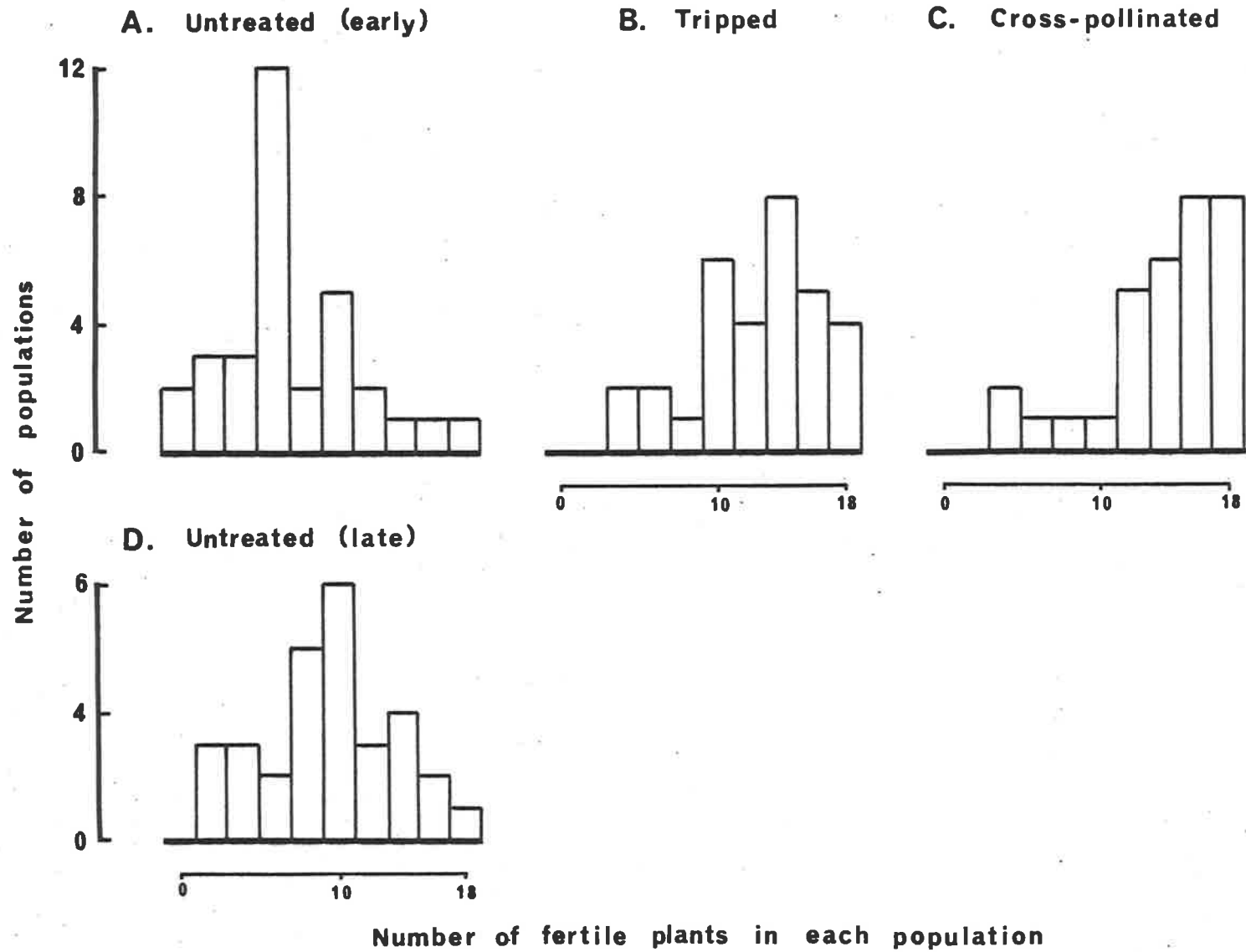


TABLE 9: The number of plants that set pods from early auto-fertility, late autofertility, from tripped flowers and from cross-pollinated flowers (The maximum number of plants is 18).

Population	Untreated early flowers	Untreated late flowers	Tripped flowers	Crossed flowers
2	6	8	15	15
4	4	9	10	13
6	0	1	3	3
15	4	5	9	9
17	8	11	12	12
20	3	7	11	13
22	4	5	7	7
29	8	9	14	17
30	3	4	9	12
31	3	13	14	14
34	1	10	11	12
38	6	9	12	17
39	10	12	15	16
44	0	2	6	12
53	3	7	9	13
58	2	7	10	11
59	5	9	13	15
65	13	13	18	18
67	11	14	15	16
68	8	10	18	18
70	1	4	5	6
73	8	8	13	15
75	5	7	13	15
76	1	3	9	13
77	1	16	17	17
80	0	2	4	4
83	2	9	13	14
84	14	18	18	18
88	5	9	14	16
89	12	13	16	16
91	14	15	16	17
92	9	12	14	17
Total	174	281	383	431

(36% of the total) while cross-pollination gave a further increase in the number of fertile plants by 48 plants (8.3%). There were still 145 plants (25%) that did not bear any pods in spite of the tripping and cross-pollination treatments.

It was not appropriate to relate autofertility to the origin of the populations because of the limited number of populations from each country included in the sample.

b). The effect of tripping and cross-pollination on pod and seed set of autofertile plants.

From Table 9 it is evident that many plants that were not autofertile, bore some pods when their flowers were tripped or cross-pollinated but it is also of interest to know how many more pods and seed were set on autofertile plants as a result of tripping and cross-pollination.

The autofertile plants responded in various degrees to tripping and cross-pollination as may be evidence from the pod number and seeds per pod on a sample of 25 plants varying in autofertility (Table 10). A sample is presented as the complete data, i.e. for 174 autofertile plants is too extensive and can only be presented as means (Table 11). In Table 11 the tripping treatment is compared statistically with untreated flowers and cross-pollinated treatment with tripped flowers because the cross-pollination treatment involves tripping the flowers prior to application of pollen from another plant.

In each group of populations there were significant increases in the number of pods due to tripping or cross-pollination. When all the plants were considered together, both tripping and cross-pollination increased the number of pods set. Autofertile plants therefore, are

TABLE 10: The number of pods and number of seeds per pod on a sample of 25 autofertile plants from flowers that were untreated, tripped and cross-pollinated.

Plant	Popn.	Flowers per treatment	Number of pods			Seeds per pod		
			Untreated	Tripped	Crossed	Untreated	Tripped	Crossed
1	2	18	1	3	6	2.0	3.0	2.5
2	2	28	10	13	11	2.1	2.6	2.4
3	2	24	10	12	17	2.5	2.2	1.8
4	2	24	6	7	9	2.5	2.1	2.1
5	15	12	1	3	5	2.0	1.7	2.0
6	17	26	9	11	11	2.9	2.9	3.6
7	29	16	1	2	6	1.0	2.5	2.3
8	29	22	8	8	8	2.5	2.2	2.4
9	29	20	2	3	4	1.5	3.0	2.5
10	29	24	3	8	14	1.0	1.2	2.2
11	29	22	11	11	13	2.1	2.5	2.4
12	29	14	2	5	5	3.0	2.8	3.0
13	29	20	4	1	3	2.8	4.0	3.3
14	30	18	2	1	7	1.0	4.0	3.4
15	30	16	1	1	1	3.0	2.0	5.0
16	30	22	2	2	3	3.0	3.0	3.0
17	68	18	4	3	2	2.8	2.7	2.0
18	68	24	9	11	6	2.3	2.7	2.3
19	84	22	5	4	5	1.8	3.8	2.6
20	84	10	10	8	9	3.3	3.6	2.9
21	84	18	2	7	9	2.0	1.7	1.9
22	84	18	10	11	16	3.1	3.5	2.9
23	84	12	5	6	10	2.8	3.1	3.4
24	84	18	9	10	13	2.3	2.2	2.4
25	84	10	4	4	7	3.0	3.0	3.3

TABLE 11: The effect of flower treatment on pod set and seeds per pod of autofertile plants.

Flower treatment	Small-seeded	Large-seeded	Both types
<u>Pod Set</u>			
Untreated	4.06	2.57	3.16
	*	NS	*
Tripped	5.58	2.74	3.87
	NS	*	*
Cross-pollinated	6.94	3.42	4.82
<u>Seeds per pod</u>			
Untreated	2.21	2.64	2.47
	NS	NS	NS
Tripped	2.43	2.75	2.62
	NS	NS	NS
Cross-pollinated	2.43	2.87	2.70

Small seeded = 69 plants

Large seeded = 105 plants

NS Not significant at P = 0.05

\* P 0.05

\*\* P 0.01

\*\*\* P 0.001

The above notation is used in the tables throughout this thesis to indicate the level of significance.



capable of higher yields when the flowers are tripped and cross-pollinated.

The number of seeds per pod varied between treatments. As evident from Table 11 the effect of tripping or cross-pollination individually was small but there was significant improvement in the number of seeds per pod when cross-pollinated flowers were compared with the untreated flowers. This could suggest that the number of seeds per pod improved with flower treatment but the difference was insufficient to reach significance at  $P = 0.05$  when the effects were considered separately. The number of ovules in the ovary is limited and consequently increases in the number of seeds per pod is expected to be small.

#### 4.1.3 Conclusion

The main conclusions from this experiment were:

1. Many plants in a population do not produce any pods when the flowers were untreated and not exposed to bee activity. (60% of the plants were not autofertile).
2. Flowers produced late in the season tend to set more pods without tripping than early flowers.
3. There was no evidence in the 100 populations that a higher autofertility occurred in either small or large seeded types.
4. Tripping and cross-pollination of the flowers progressively improved pod set.
5. However even with these treatments 25% of the plants bore no pods. This lack of pods was not a result of the storm, any apparent pathogens or a lack of vegetative agents.

6. Seeds per pod were increased in this experiment by some treatments but there may be a limited capacity for change in this character. Later experiments were only able to establish small differences.

#### 4.2.0 Experiment 2 (1976).

The objectives and nature of this experiment were similar to Experiment 1 (1974) but consisted of inbred lines derived during 1974 and 1975 from 20 introduced populations. Each line was a bulk of inbred seeds produced on approximately 15 plants of each population as a result of autofertility. The experiment was sown on May 16, 1976.

#### 4.2.1 Grouping of lines by seed characters.

On the basis of their seed shape and seed weight, the 20 populations (lines) were separated into 2 groups (Table 12).

- (1) Small spherical to oval shaped seeds with mean seed weight per line ranging from 0.16 to 0.60 g.
- (2) Large flat seeds ranging from 0.77 to 1.58 g (broad bean type).

#### 4.2.2 Autofertility of the inbred lines.

Autofertility of the plants was evident from two assessments. One is based on all the untreated flowers on the plant (Section 4.2.2) whereas, in Section 4.2.3 it is based on the marked untreated flowers that served as controls for the treatments.

##### a). Autofertile plants.

Among the 20 lines, nearly all the plants were autofertile and 97% of the plants set some pods from untreated flowers. Sterile plants occurred in only a few of the lines (7 out of 20 lines) (Table 13). Apparently growing the line over 2 generations of selfing (obtaining seed as a result of autofertility) had eliminated the plants that were

TABLE 12: Seed and plant characters of 20 lines of *V. faba*.

Line No.	Seed Shape	Mean seed weight (g)	Mean No. of stems per plant	Mean No. of Racemes per plant	Days to first flower
1	Sp	0.16	6.3	66.7	96
2	Sp-Ov	0.37	3.4	61.1	92
3	Ov	0.39	1.9	22.8	69
4	Ov	0.41	2.5	65.9	97
5	Ov	0.45	2.7	44.1	70
6	Ov	0.47	3.3	84.1	95
7	Ov	0.50	2.7	60.4	76
8	Ov	0.52	3.4	40.6	73
9	Ov	0.57	3.1	73.9	88
10	Ov	0.60	3.2	86.5	88
11	FB	0.77	3.7	53.3	71
12	FB	0.81	4.2	61.7	79
13	FB	0.89	4.9	72.0	82
14	FB	0.96	3.2	58.2	75
15	FB	1.04	3.5	55.5	75
16	FB	1.06	4.4	68.3	78
17	FB	1.23	4.7	75.1	82
18	FB	1.27	4.6	69.2	80
19	FB	1.36	3.7	81.3	88
20	FB	1.58	4.0	68.6	81

SP<sub>Q</sub> = Spherical

Ov = Oval

FB = Flat and broad

TABLE 13: Percentage of autofertile plants in small and large seeded lines of *V.faba* (% values based on 24 plants)

Small seeded lines		Large seeded lines	
Line No.	% autofertile plants	Line No.	% autofertile plants
1	100	11	100
2	100	12	100
3	100	13	100
4	96	14	96
5	96	15	100
6	100	16	75
7	92	17	96
8	100	18	100
9	100	19	96
10	100	20	100

not autofertile. However, there was some variation in the level of autofertility among the lines.

b). Autofertility and seed size.

In this thesis, autofertility has been defined as the ability of a plant to produce one or more pods in the absence of tripping. A further index of autofertility may be obtained from the number of seeds produced from untreated flowers. When this criterion was used it was found that autofertility was negatively correlated with seed size. This relationship was different from the small and large seeded groups, the negative regression in autofertility being more marked for small seeds than large seeds (Fig. 6). Two lines which were very autofertile produced more than twice the number of seeds of the other lines (lines 1 and 2,  $P < 0.005$ ).

As small-seeded populations may be expected to produce more seeds than large seeded populations it was decided to also assess autofertility using the number of pods. A similar negative relationship between pod number and seed weight was obtained, this was particularly evident for the small-seeded lines (Fig. 7),

c). Yield of the lines from untreated flowers.

The yield per plant is shown in Figure 8. Within each seed size group, lines with high yield were evident. There does not appear to be any relationship between seed size and the yield of the plant. Lines with a low number of seeds set were still able to have a high yield as a result of large seeds e.g. lines 19, 20, the two lines with the highest weight per seed. The highly autofertile line 1 did not have a proportionately higher yield because of its extremely small seed. The

Figure 6

The relationship between the number of seeds and seed weight.

A = Small seeded lines

B = Large seeded lines

The mean number of seeds per plot is the average of 4 replicates each with 6 plants.

The number of seeds is used as an index of autofertility.

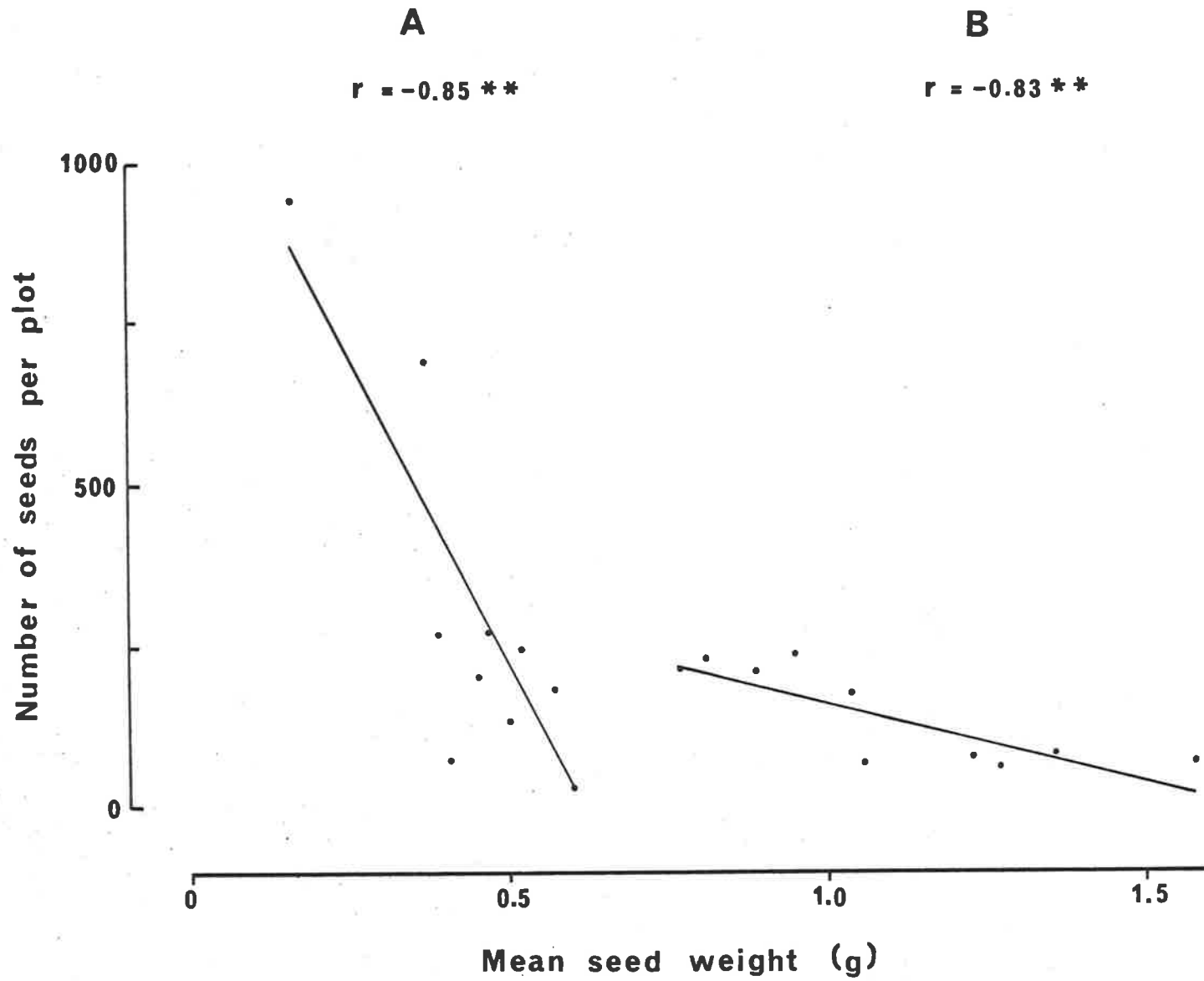




Figure 7

The relationship between the number of pods and seed weight.

A = Small seed types

B = Large seeded lines

The mean number of pods per plot is the average of 4 replicates each with 6 plots.

The number of pods is used as an index of autofertility.

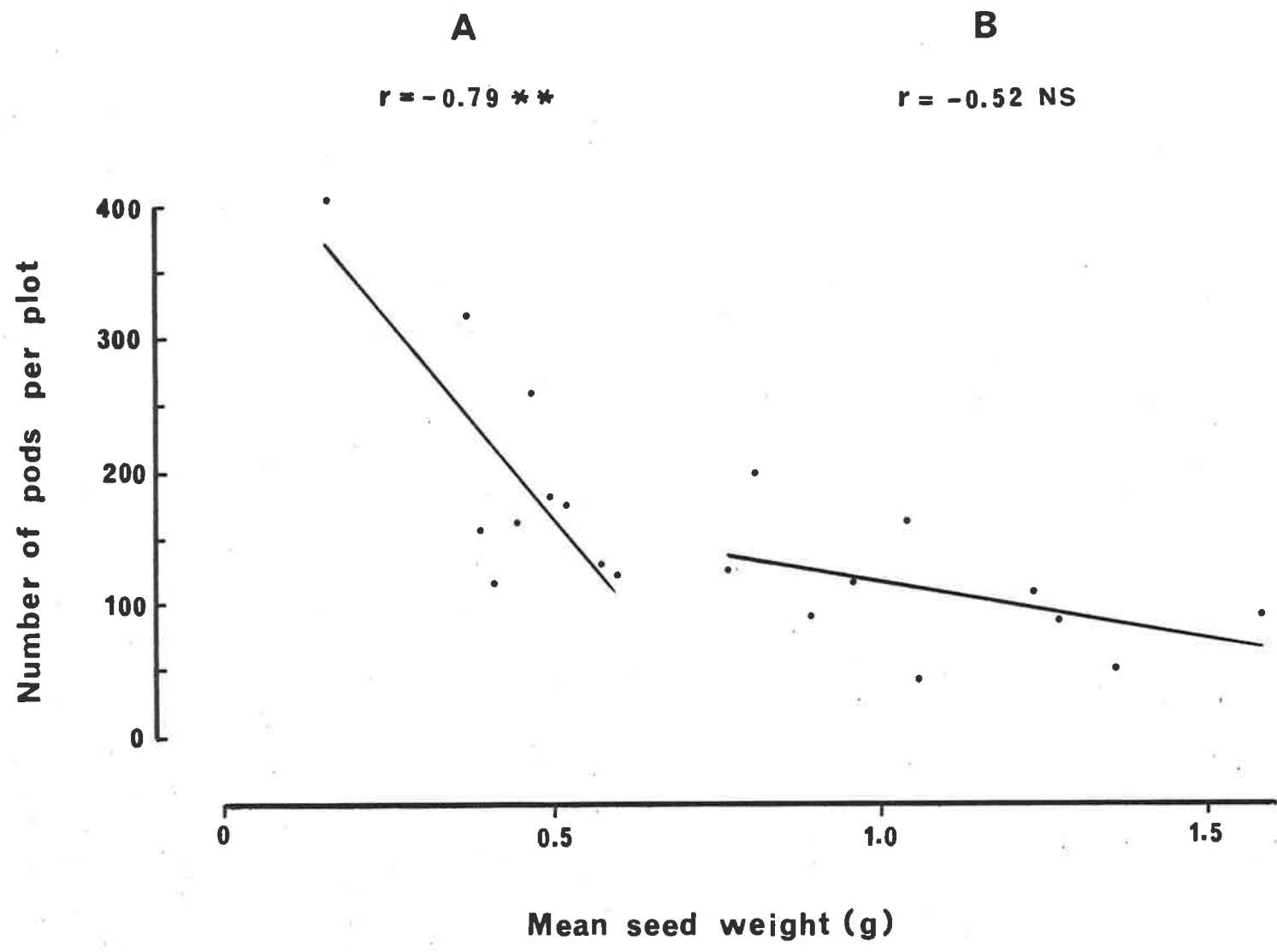
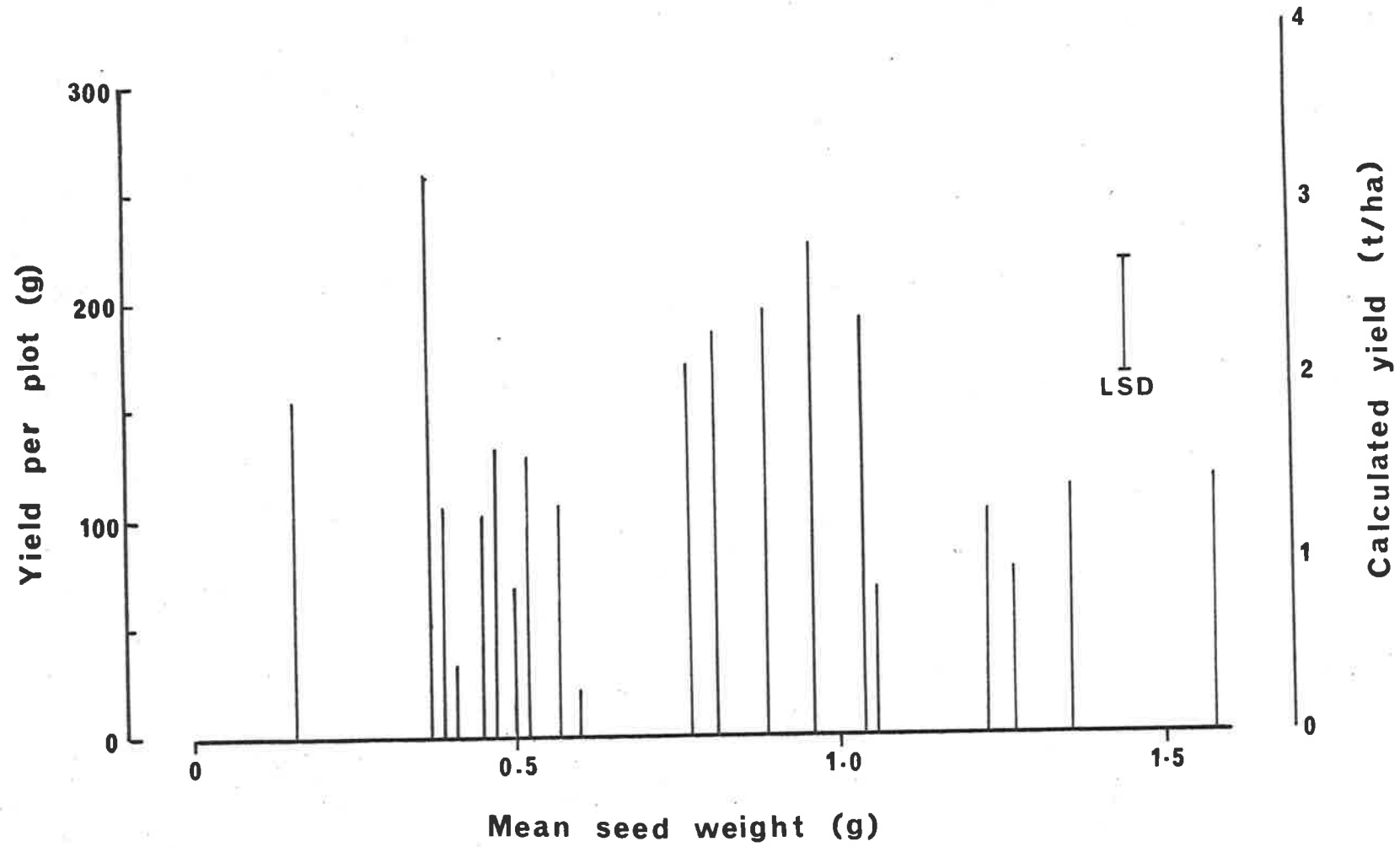


Figure 8

Mean yield per plot for lines with different seed weight.

1 g per plot = yield of 1.225 t/ha (approximately).



yield of the lines ranged from 3.17 to 0.28 t/ha respectively for lines 2 and 10. These yields were based on only 7 plants  $m^{-2}$  compared with the possible optimum of 20 plants  $m^{-2}$ .

d). Components of yield from untreated flowers.

The relation between yield and its various components for all 20 lines is given in Table 14. Yield was not correlated with the weight of seed nor the number of racemes produced by the plant. This indicates that the number of racemes was not a limiting factor to yield. It was common to find that only 1 raceme in 4 bore any pods.

Considering the primary components, the number of mature pods and the number of seeds per pod were closely correlated with yield whereas the weight per seed was not.

TABLE 14: Correlation between yield and its components in untreated flowers.

Character correlated with yield	r
Primary components:	
Number of mature pods	0.59 ***
Number of seeds per pod	0.53 ***
Number of seeds	0.56 **
Weight per seed	-0.01 NS
Secondary components:	
Number of racemes	-0.11 NS
Percent racemes with pods	0.50 ***
Number of pods per podding raceme	0.29 **

There was only a weak relationship between the number of pods per podding raceme and yield. This was because each podding raceme generally produced only 1 or 2 pods and therefore variation in this character was unlikely to have a great effect on yield. Yield was influenced more by the percent racemes with pods.

#### 4.2.3 The effect of the tripping and cross-pollination treatments.

##### a). Pod set.

The results will be presented in 2 forms; as the actual number of flowers that set pods (number of pods set) under the various treatments and as percentages. This second presentation is given to enable comparisons to be made where different numbers of flowers were treated.

In all lines except 1 and 3, there were significant differences across treatments (Table 15). The lines 1 and 3 that did not show an improvement over the untreated flowers by tripping or cross-pollination were highly autofertile with more than 60% of the untreated flowers producing mature pods (Table 16).

This is also seen in Figure 9 where a sample of lines is presented. With the exception of the autofertile lines 1 and 3, in every instance the slope of the line increased between tripped and cross-pollinated flowers.

Although large differences existed between lines in the autofertility of untreated flowers, the good response to the cross-pollination treatment meant that differences between lines were small after cross-pollination. Lines with low autofertility achieved the level of pod set

TABLE 15: The effect of flower treatment on the mean number of pods set per plot.

Line No.	Approx. No. flowers per treatment	Flower treatment			LSD
		Untreated	Tripped	Cross-pollinated	
1	28	20.2	23.0	23.5	NS
2	55	24.2	30.0	47.7	4.2
3	24	15.7	15.7	17.0	NS
4	60	2.7	6.7	34.2	3.1
5	37	7.0	10.7	33.2	3.2
6	60	6.7	13.2	40.0	6.4
7	50	7.0	16.0	39.7	2.7
8	37	9.7	11.5	31.2	15.3
9	60	7.0	13.0	42.0	5.9
10	60	1.0	2.2	27.5	8.4
11	40	7.7	9.0	30.5	5.3
12	48	9.5	14.7	42.5	7.1
13	48	2.2	4.7	23.0	2.3
14	60	6.7	15.7	40.2	6.2
15	50	7.5	15.5	38.7	2.9
16	32	0.5	7.0	24.5	2.9
17	60	2.2	5.7	43.0	7.3
18	48	1.5	3.7	37.7	0.6
19	60	0	5.5	23.2	2.9
20	52	1.5	7.0	38.2	2.8

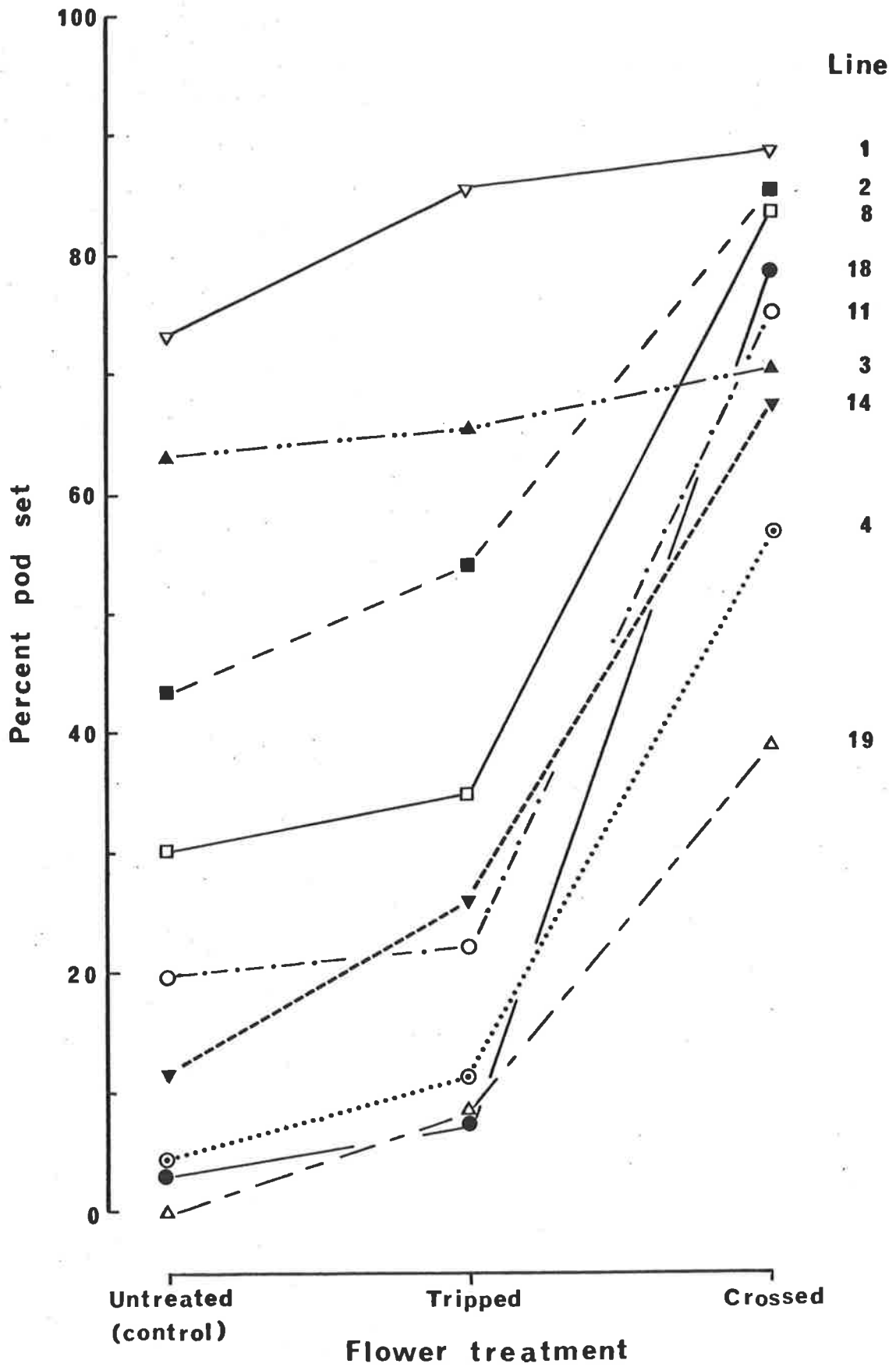
TABLE 16: Percentage of flowers forming pods when flowers were untreated, tripped and cross-pollinated.

Line No.	Flower treatment		
	Untreated	Tripped	Cross-pollinated
1	73.2	85.4	88.7
2	43.4	54.1	86.0
3	63.2	65.6	70.8
4	4.6	11.4	57.1
5	18.6	29.2	89.8
6	11.2	22.1	65.0
7	14.1	31.7	79.5
8	30.3	35.2	83.4
9	11.6	22.1	70.0
10	1.7	3.8	45.2
11	19.8	22.2	75.2
12	19.5	30.7	88.6
13	4.7	9.8	47.9
14	11.2	26.0	67.4
15	14.8	30.7	79.0
16	1.3	21.1	77.3
17	3.8	9.1	72.3
18	3.1	7.8	78.6
19	0	8.4	38.8
20	2.9	13.4	73.1



Figure 9

Some typical responses to flower treatment of  
lines with low and high autofertility.



of lines with high autofertility, after cross-pollination (e.g. lines 16, 17, 18, 20). The lines that had a low autofertility benefited more from cross-pollination than the highly autofertile lines which may have reached their physiological limit for pod set.

In some lines (e.g. lines 10, 13, 19) the percentage of flowers setting pods after cross-pollination was still relatively low in comparison with lines 1, 2, and 3 and some factors other than pollination must be responsible.

It was of interest to determine whether any other component of yield besides pod set was similarly improved by the treatments. Matters investigated were:

(1) Whether tripping or cross-pollination increased the number of seeds in the pods. This was investigated for the material as a whole (Section b) and for individual lines (Section c).

(2) Whether a higher percentage of pods developed to maturity after tripping or cross-pollination (Section d).

b). The relation between pods and the number of seeds.

The effect of the treatments on the relation between pods and seed number was studied using regression. There was very close relation between the number of pods and the number of seeds borne on a plant for each of the treatments (Fig. 10). A comparison of the three regressions showed no significant difference between them for their position (indicated by the y-intercept) and for slope (regression coefficient) as is evident from Table 17.

Figure 10

The relation between the number of pods and the number of seeds for each flower treatment. (Each value is the plot mean).

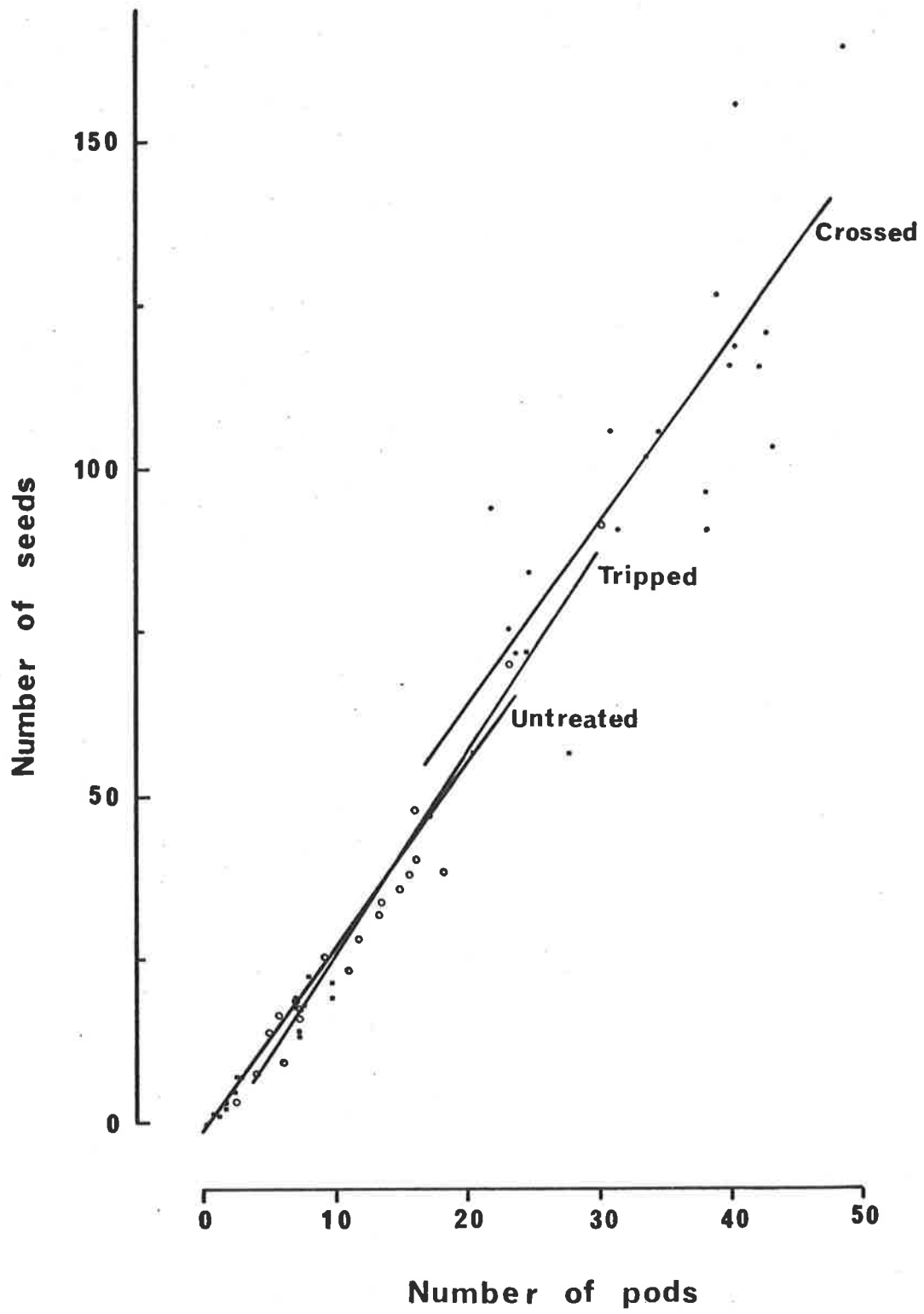


TABLE 17: The regression equations for pod and seed number when flowers were untreated, tripped or cross-pollinated.

Flower treatment	Regression equation ( $y = a + bx$ )	± S.E.	
		a	b
Untreated	$y = -1.78 + 2.81x$	0.99	0.10
Tripped	$y = -4.68 + 3.04x$	1.66	0.12
Cross-pollinated	$y = 6.75 + 2.81x$	15.90	0.46

The results indicate that tripping or cross-pollination of the flowers did not increase the number of seeds in a pod (the regression coefficients are not significantly different) when all the material is considered together.

c). Comparison of individual lines for seeds per pod.

The analysis using the number of seeds per pod showed a significant interaction between the flower treatment and the lines ( $P < 0.001$ ). The tripping treatment did not increase the number of seeds per pod relative to the untreated flowers in most lines but cross-pollination resulted in more seeds per pod than tripping treatment in many lines (Table 18). Some lines had the same number of seeds per pod whether they were untreated, tripped or cross-pollinated. This interaction between treatments and lines could cause the lack of difference between treatments when the lines were considered together in the previous section (b).

d). The relation between young and mature pods.

A comparison between treatments for the number of initiated pods

TABLE 18: Mean seed numbers per pod from untreated, tripped and cross-pollinated flowers.

Line No	Flower treatment		
	Untreated	Tripped	Cross-pollinated
1	2.82	3.05	3.06
2	2.97	3.02	3.44
3	2.42	2.55	2.76
4	2.67	2.86	3.07
5	1.88	2.11	3.06
6	2.62	2.56	2.95
7	2.17	2.39	2.91
8	2.01	2.34	2.93
9	2.66	2.47	2.75
10	1.00 <sup>#</sup>	1.56	2.04
11	2.86	2.77	3.46
12	2.35	2.43	2.83
13	3.12	2.97	3.27
14	2.89	3.01	3.88
15	2.46	2.46	3.26
16	3.50 <sup>#</sup>	2.37	3.40
17	2.25	1.76	2.40
18	2.33	2.01	2.55
19	-	3.12	4.03
20	1.50 <sup>#</sup>	2.36	2.36

LSD between any 2 means = 0.44

<sup>#</sup> calculated from less than 5 pods.

that reach maturity is illustrated in Figure 11. Although a pod, once it had set, normally reached maturity, small but significant differences were obtained between treatments (Table 19).

TABLE 19: The regression equations for young and mature pods when untreated, tripped or cross-pollinated.

Flower treatment	Regression equation ( $y = a + bx$ )	± S.E.	
		a	b
Untreated	$y = -0.170 + 0.846x$	0.296	0.026
Tripped	$y = 0.682 + 0.836x$	0.406	0.027
Cross-pollinated	$y = -0.714 + 0.980x$	1.156	0.032

In the combined analysis, the three regressions were significantly different ( $P < 0.01$ ) for positions and slopes. The survival of young pods developed from cross-pollination was better than from untreated or tripped flowers but as evident from Figure 11 this difference was small.

#### 4.2.4 Conclusion

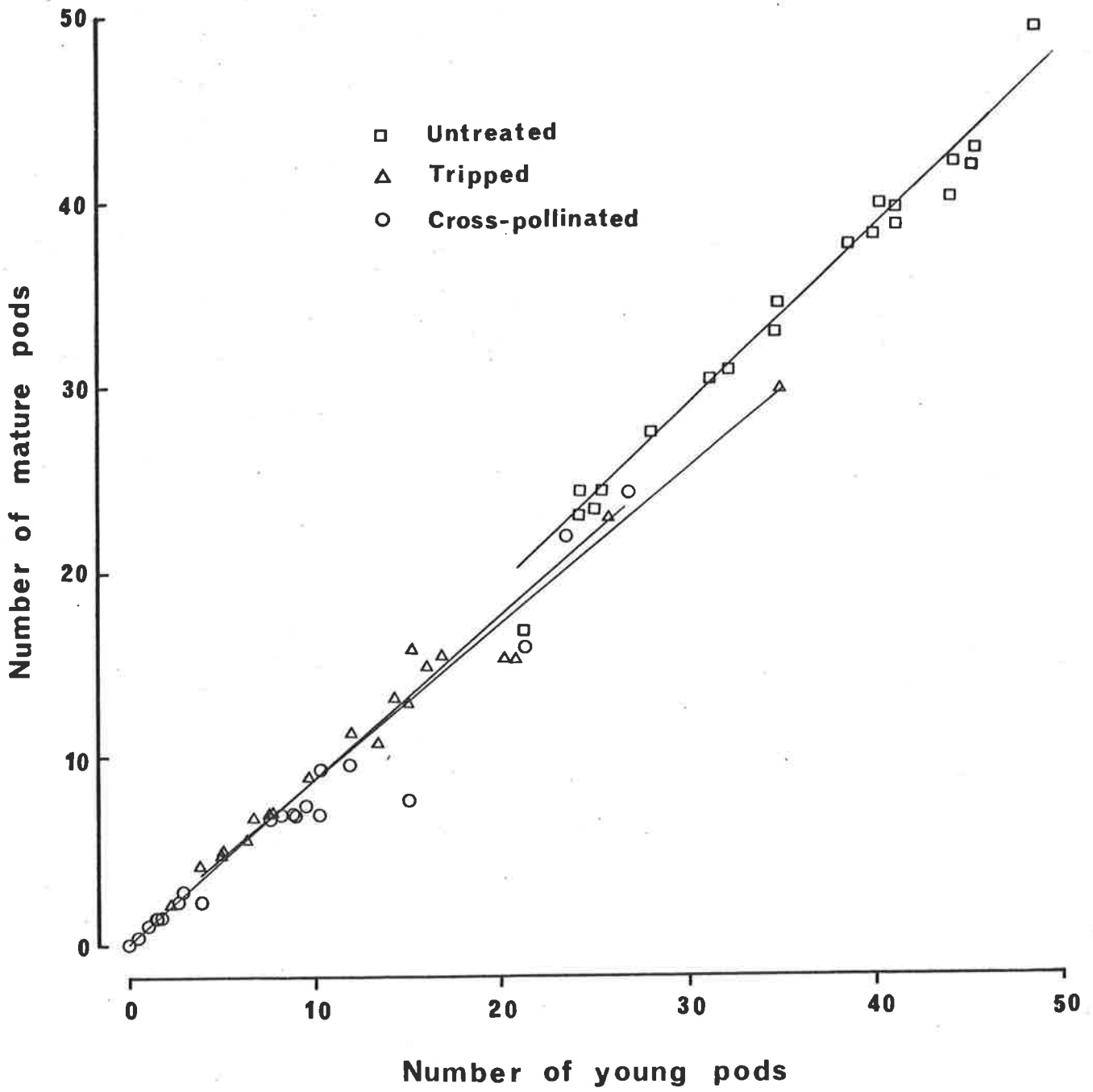
1. For the 20 lines (populations) the level of autofertility as measured by seed number and by pod numbers was negatively correlated with seed weight.
2. There was no apparent relationship between seed size and the yield of the lines.
3. Yield is dependent mainly on the percentage of racemes forming pods.



Figure 11

The relation between the number of young pods and the number of mature pods for each flower treatment.

(Each value is the plot mean).



4. Except in very autofertile lines, tripping and especially cross-pollination increases pod set.
5. The flower treatments did not affect the number of seeds per pod.
6. The proportion of young pods that develop to maturity was slightly higher when flowers were cross-pollinated than when untreated or tripped.

#### 4.3.0 Experiment 3 (1975).

The previous experiments were concerned with treatments conducted on open-pollinated plants or on inbred plants. Beginning with this section the comparison is extended to include treatments on hybrid plants. It has already been mentioned that high autofertility was associated with heterozygosity and inbreeding decreases the level of autofertility (Section 2.5.2 Lit. Rev.). It was of interest to compare the yield, and response to tripping, of inbred and hybrid material. As this was the first time this kind of study was being conducted here, no hybrids were available and it was necessary to carry out a selfing and crossing programme with introduced material in preparation for the experiment. The experiment was conducted in the 1975 season.

Observations on the introduced material in the 1974 experiment revealed differences in growth habit, namely indeterminate and determinate (vegetatively). The growth habit of a variety may influence its suitability for cultivation in South Australia. Indeterminate plants are tall and susceptible to storm damage, and the limited growing season may inhibit any advantage that might be obtained from their extended flowering period. The determinate forms have a low habit, profuse branching and a short flowering period which promotes even maturity of the pods. Due to the difficulties in obtaining sufficient inbred and hybrid progenies (as already explained in Section 3.5.3) a comprehensive comparison of the two types of growth habits and the inbred and hybrid progenies could not be made in a single experiment. Separate experiments were necessary for the indeterminate (Exp. 3) and determinate (Exp. 4) material.

Experiment 3 involved comparisons between inbreds and hybrids of 2 populations (155 and 680). For each population there were 12 inbreds and 36 hybrids produced from 12 seed parents. The hybrids were obtained from crosses of the seed parents with 3 pollen parents. For comparison, the open-pollinated progenies of the original populations were included in the trial (Section 3.5.3(a)).

#### 4.3.1 Plant growth.

The indeterminate habit resulted in plants over 3m tall at harvest. The plants continued to produce flowers to the end of the growing season (November) when moisture stress and high temperature stopped further growth.

There were no visual differences in the heights of the various progenies but significant differences ( $P < 0.05$ ) occurred between the progenies of each population in the number of stems per plant (Table 20). Some hybrids had a higher mean number of stems than the inbreds but this advantage was not consistent.

#### 4.3.2 Autofertility - Yield and its components.

The yield and components of yield for the open-pollinated, inbred and hybrid progenies are given in Table 21.

In each population the hybrids had higher yields than the inbreds largely due to more pods being set. In some crosses there was also a significant improvement in seeds per pod and the mean seed weight, particularly in crosses of population 155.

The open-pollinated progenies, which are anticipated to be a mixture of inbreds and hybrids, showed some indication of having yields

TABLE 20: Mean number of stems per plant for the open-pollinated, inbred and hybrid progenies.<sup>1</sup>

Progeny <sup>2</sup>	Population	
	155	680
Open-pollinated	2.52	2.19
Inbred	2.55	2.21
Hybrid 1	3.04	2.45
Hybrid 2	2.69	2.28
Hybrid 3	2.94	2.71
L.S.D.	0.30	0.28

<sup>1</sup>Average of 108 plants.

<sup>2</sup>The hybrids of population 155 are unrelated to those of 680.

TABLE 21: Mean yield, pod number <sup>per</sup> plant, seeds per pod and weight per seed for open-pollinated, inbred and hybrid progenies. The values were measured from untreated flowers.<sup>1</sup>

Population Progenies <sup>#</sup>	155				680			
	Yield (g) <sup>†</sup>	Pod No.	Seeds/pod	Seed Wt. (g)	Yield (g) <sup>†</sup>	Pod No.	Seeds/pod	Seed Wt. (g)
Open-pollinated	6.96	9.72	2.54	0.253	5.46	8.45	2.54	0.232
Inbred	4.27	7.70	2.16	0.241	2.71	5.04	2.62	0.200
Hybrid 1	7.77	10.97	2.52	0.280	7.08	11.34	2.66	0.229
Hybrid 2	7.83	9.55	2.63	0.300	6.90	9.83	2.99	0.233
Hybrid 3	13.40	15.02	2.87	0.293	4.02	6.17	2.73	0.234
S.E. †	1.06	1.86	0.17	0.008	1.71	1.98	0.05	0.011
L.S.D.	3.44	2.79	0.26	0.027	2.32	3.20	0.16	NS

† The yield values may not exactly equal the product of the 3 components as they were assessed from calculations based on single plants. The values in the table are the overall mean of these calculations.

# The hybrids of population 155 are unrelated to those of 680.

<sup>1</sup> Average of 108 plants.

TABLE 22: Mean number of seeds per plant produced by untreated flowers on inbred and hybrid progenies of each seed parent.<sup>1</sup>

Seed Parent	Progenies				Mean
	Inbred	Hybrid 1	Hybrid 2	Hybrid 3	
<u>Popn. 155</u>					
1	27.2	58.6	46.4	61.8	48.5
2	10.0 (2)	17.0 (2)	26.1	40.4	23.4
3	2.2 (4)	16.3	12.1	18.9	12.4
4	19.3 (1)	23.7	23.1	48.1	28.6
5	29.4	29.7	22.4	48.3	32.5
6	11.2 (2)	29.7	21.6 (1)	41.3	25.9
7	11.9 (1)	15.0 (2)	20.4	28.9	19.1
8	21.1	21.3	30.2 (1)	34.8	26.9
9	15.2	33.9	26.7 (1)	63.3	34.8
10	9.4 (1)	15.2 (2)	15.2	18.1 (1)	14.5
11	23.9 (1)	30.6	30.6	56.3	35.3
12	18.6 (1)	39.9	27.8	65.7	39.0
Mean	16.6	27.6	25.2	43.8	
L.S.D. = 9.3					
<u>Popn. 680</u>					
1	8.6 (2)	20.0	24.4	9.8 (3)	15.7
2	15.4 (3)	28.3	22.0	15.7	20.4
3	5.1 (2)	29.4	28.3 (1)	13.7	19.1
4	10.1 (1)	18.0	37.4 (1)	7.3 (1)	18.2
5	19.4	28.8	32.6	24.7	26.4
6	11.4	40.7	39.2	23.8	28.8
7	22.8	39.7	27.4	16.6 (1)	26.6
8	11.0 (1)	36.0	30.8	21.8 (1)	24.9
9	25.0	34.6 (1)	29.9	26.1	28.9
10	8.3	26.6	27.6 (1)	11.0	18.4
11	11.3 (3)	45.7 (1)	36.4	17.8	27.8
12	10.4 (1)	18.4 (1)	17.2 (2)	12.6 (1)	14.7
Mean	13.2	30.5	29.4	16.7	
L.S.D. = 8.4					

<sup>1</sup>Means were based on 9 plants. Sterile plants were included in calculating the means. Figures within brackets indicate number of sterile plants.

No significant interaction between progenies and seed parents.



TABLE 23: Mean yield (g) per plant produced by untreated flowers of inbred and hybrid progenies of each seed parent.<sup>1</sup>

Seed parent	Progenies				Mean
	Inbred	Hybrid 1	Hybrid 2	Hybrid 3	
<u>Popn. 155</u>					
1	6.12	15.19	15.53	17.50	12.83
2	2.51 (2)	4.96 (2)	7.92	11.25	6.66
3	0.57 (4)	5.08	4.07	6.00	3.93
4	4.50 (1)	5.60	6.88	13.22	7.55
5	7.32	8.22	7.07	15.33	9.48
6	2.91 (2)	8.21	6.33 (1)	13.11	7.64
7	3.79 (1)	3.76 (2)	6.76	9.16	5.87
8	5.94	6.59	9.69 (1)	9.53	7.94
9	3.77	10.29	8.75 (1)	18.85	10.41
10	2.44 (1)	3.63 (2)	4.86	5.26 (1)	4.05
11	6.64 (1)	11.23	10.55	20.42	12.21
12	4.73 (1)	10.48	8.57	21.22	11.25
Mean	4.27	7.77	7.83	13.40	
L.S.D. = 3.93					
<u>Popn. 680</u>					
1	1.77 (2)	4.52	6.19	2.26 (3)	3.69
2	2.45 (3)	5.62	5.03	3.11	4.05
3	0.80 (2)	6.20	6.76 (1)	3.34	4.28
4	2.74 (1)	4.30	8.24 (1)	1.72 (1)	4.25
5	4.10	6.80	7.24	6.16	6.08
6	2.57	10.04	9.02	5.74	6.84
7	5.08	10.32	7.08	4.51 (1)	6.75
8	2.81 (1)	9.19	8.09	5.55 (1)	6.41
9	4.88	7.69 (1)	6.47	6.11	6.29
10	1.41	6.05	6.51 (1)	2.48	4.11
11	2.15 (3)	11.15 (1)	8.17	4.16	6.41
12	1.75 (1)	3.79 (1)	3.95 (2)	3.09 (1)	3.14
Mean	2.71	7.08	6.90	4.02	
L.S.D. = 2.49					

<sup>1</sup>Means were based on 9 plants. Sterile plants were included in calculating the means. Figures within brackets indicate number of sterile plants.

No significant interaction between progenies and seed plants.  
Conversion to t/ha = x 0.0735

intermediate between inbreds and hybrids.

The inbreds and hybrids had a common seed parent. The yields and seed set of each seed parent are given in Tables 22 and 23. In both the populations studied, there were significant differences between seed parents and also between progenies (effect of the pollen parent). There was no significant interaction between the seed parent and the progenies.

The productivity (yield and seed set) of some hybrids showed a positive relationship with the inbred parent (Figs. 12 and 13).

For each group of progeny a range of autofertility as measured by seed set was evident (Fig. 14). The inbreds tended to have a large proportion of plants that had low autofertility whereas the hybrids showed a less skewed distribution with many plants in the high fertility classes. The open-pollinated progenies consisted of a wide range of fertility types.

#### 4.3.3 The influence of tripping on yield characters.

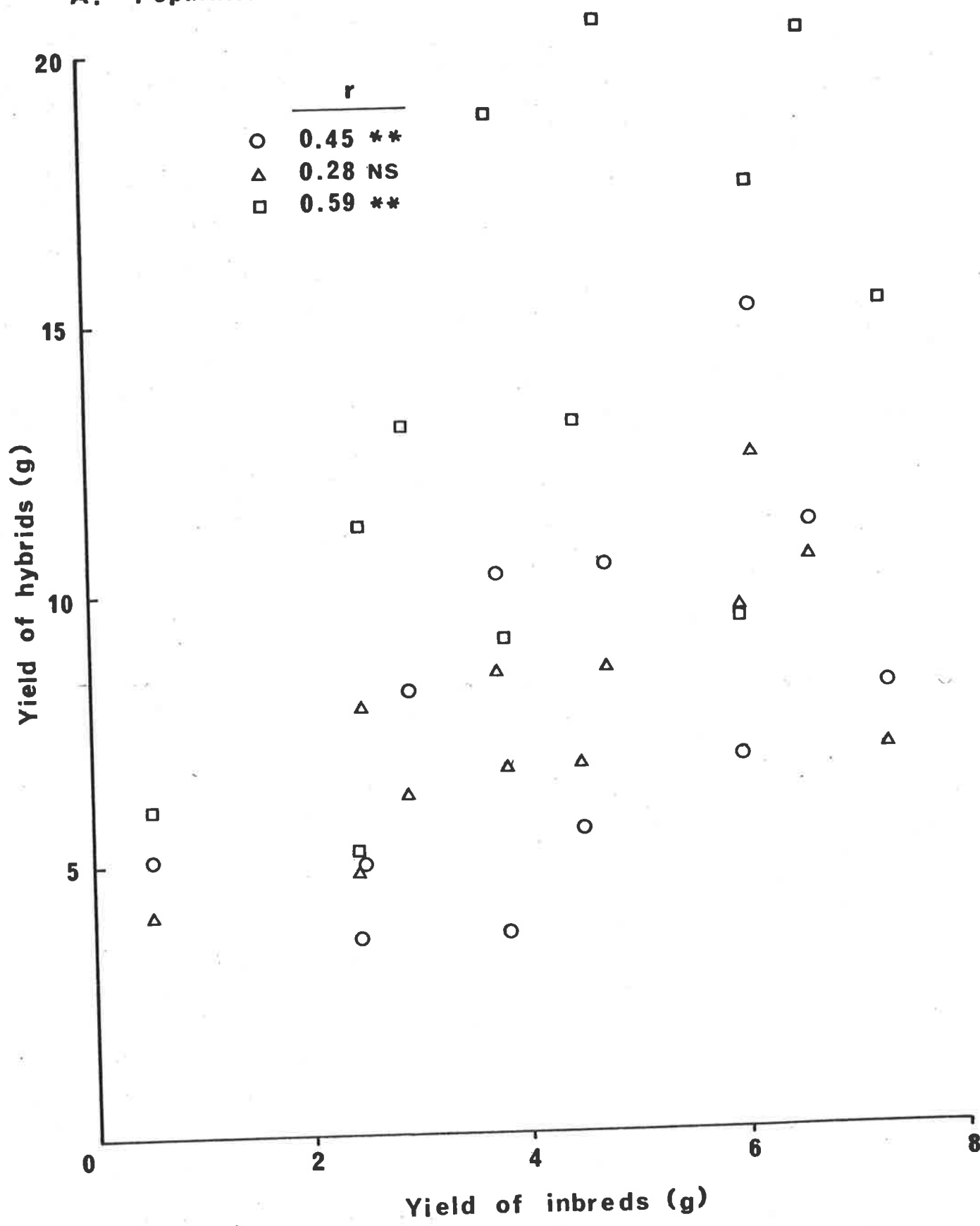
The mean yield, number of pods and the number of seeds set per plant from tripped flowers (40 flowers from 8 racemes per plant) are given in Tables 24 to 27. The overall mean of the 5 groups of progenies is given in Table 24 while in the other tables the means for the inbred and the hybrid progenies of each seed parent are given.

The populations differed in the relative performance of the inbred and hybrid when flowers were tripped. In population 155 the analysis show no significant difference between progenies for yield. However, the variance-ratio for yield had a probability of  $P = 0.055$ , when compared with an error, (replication x progenies) which was very close to the arbitrary level of 0.05 used as the critical level in the test of significance.

Figure 12

The relation between inbreds and hybrids for the yield of untreated flowers.

A. Population 155



**B. Population 680**

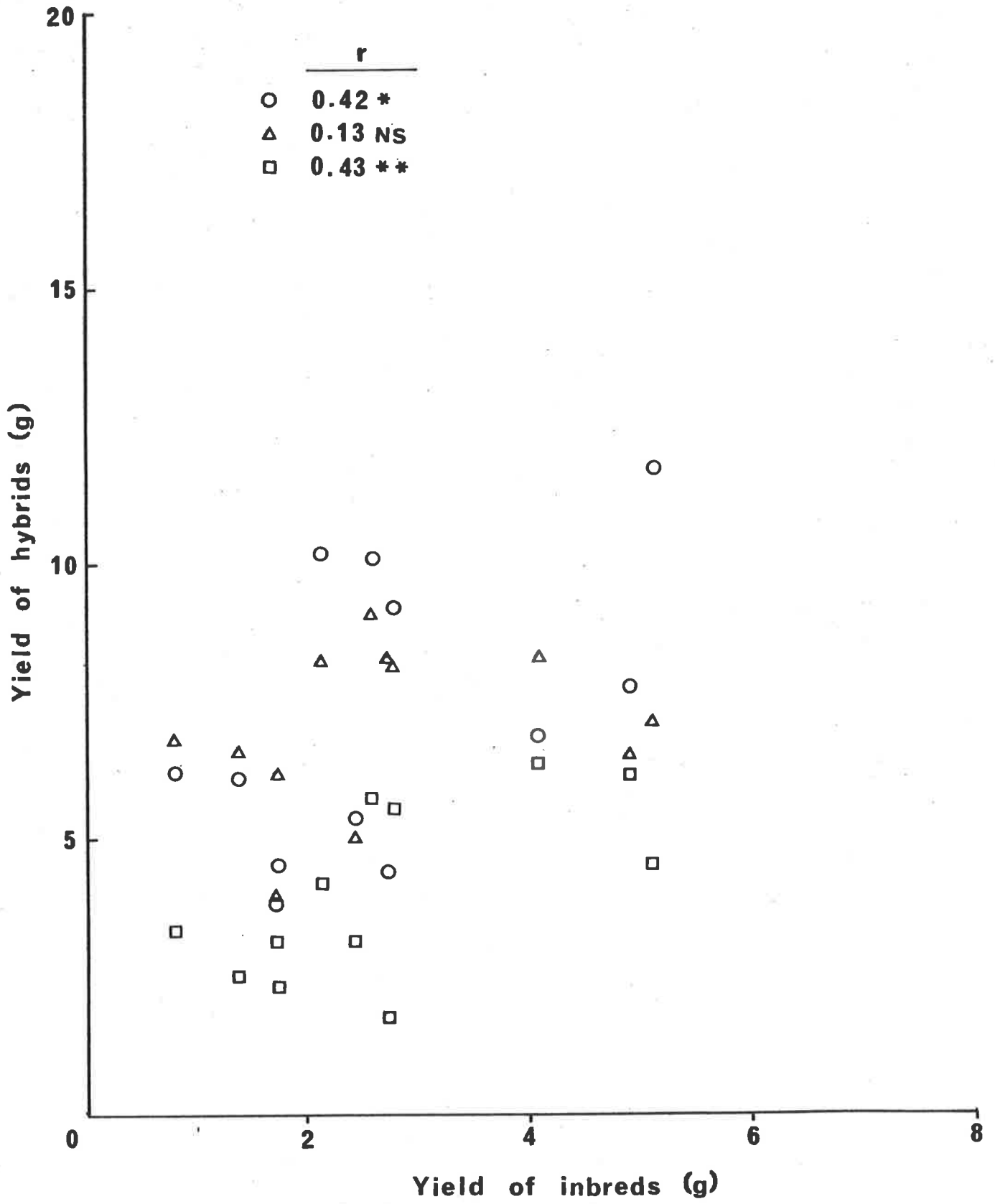
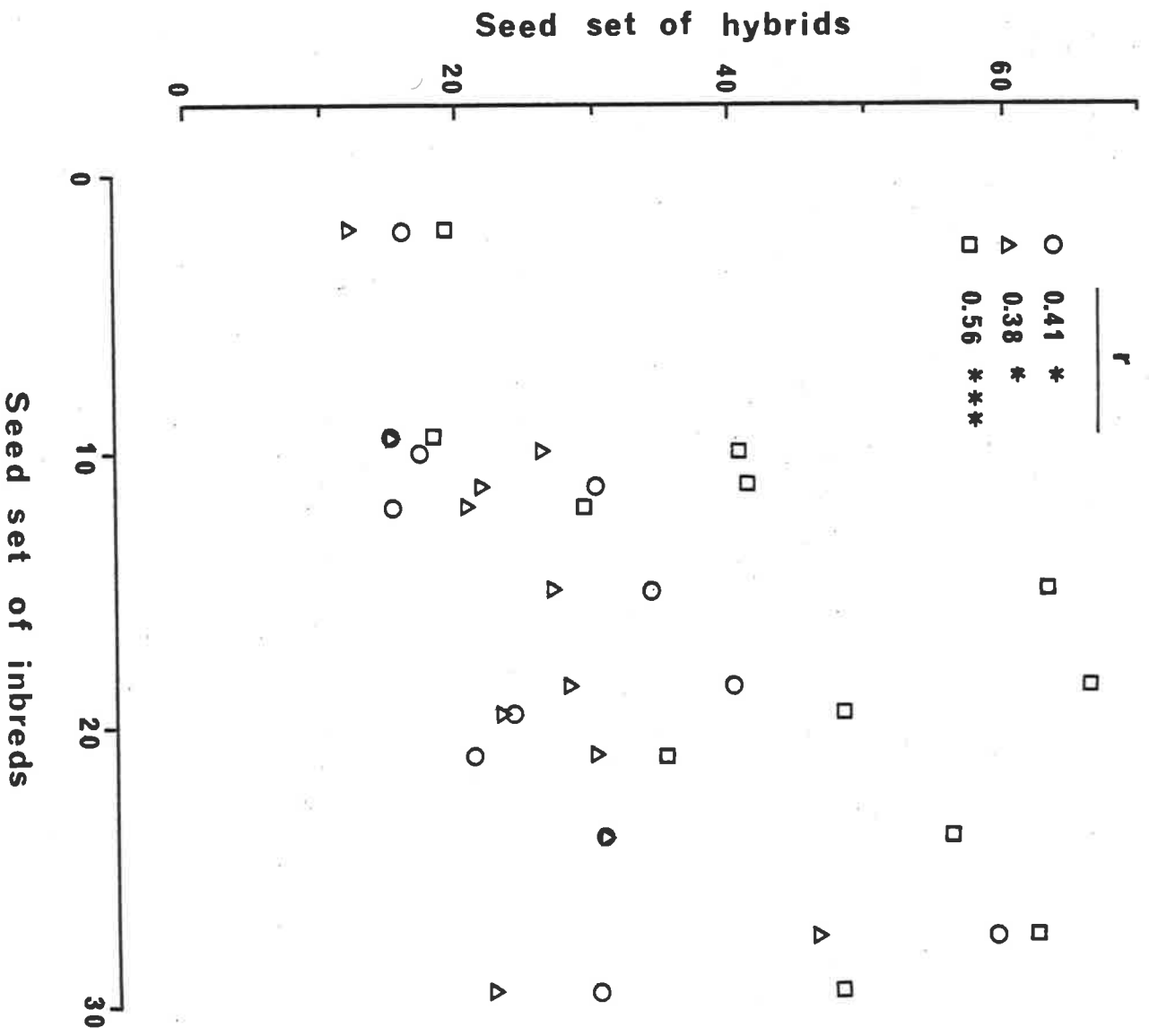


Figure 13

The relation between inbred and hybrids for the seed set  
of untreated flowers,

A. Population 155



### B. Population 680

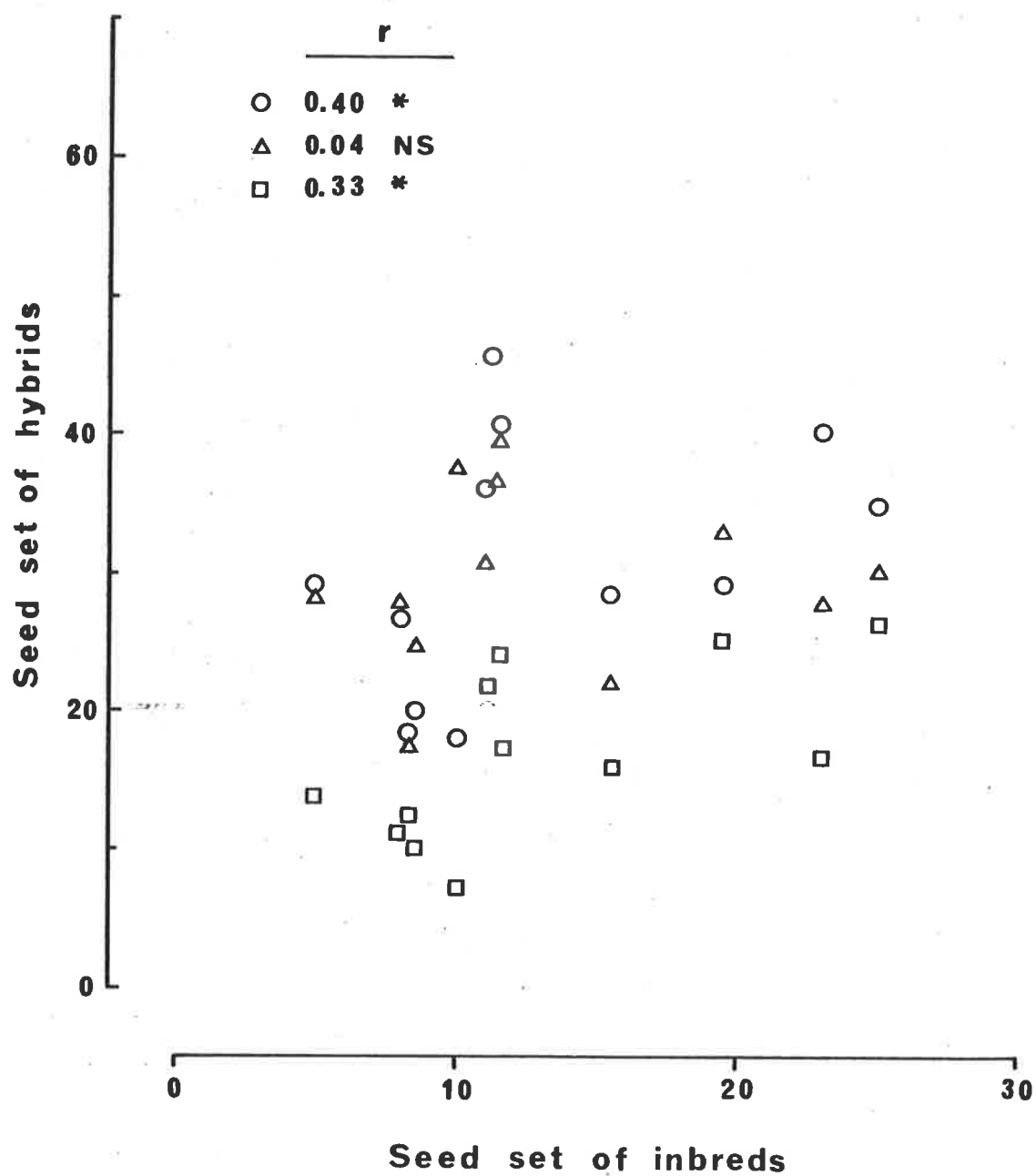




Figure 14

Distribution of plants for the number of seeds produced from untreated flowers of open-pollinated, inbred and hybrid progenies of population 155 and 680.

A = Progenies derived from population 155

B = Progenies derived from population 680

Hybrids of A and B are not related.

Each distribution consists of 108 plants.

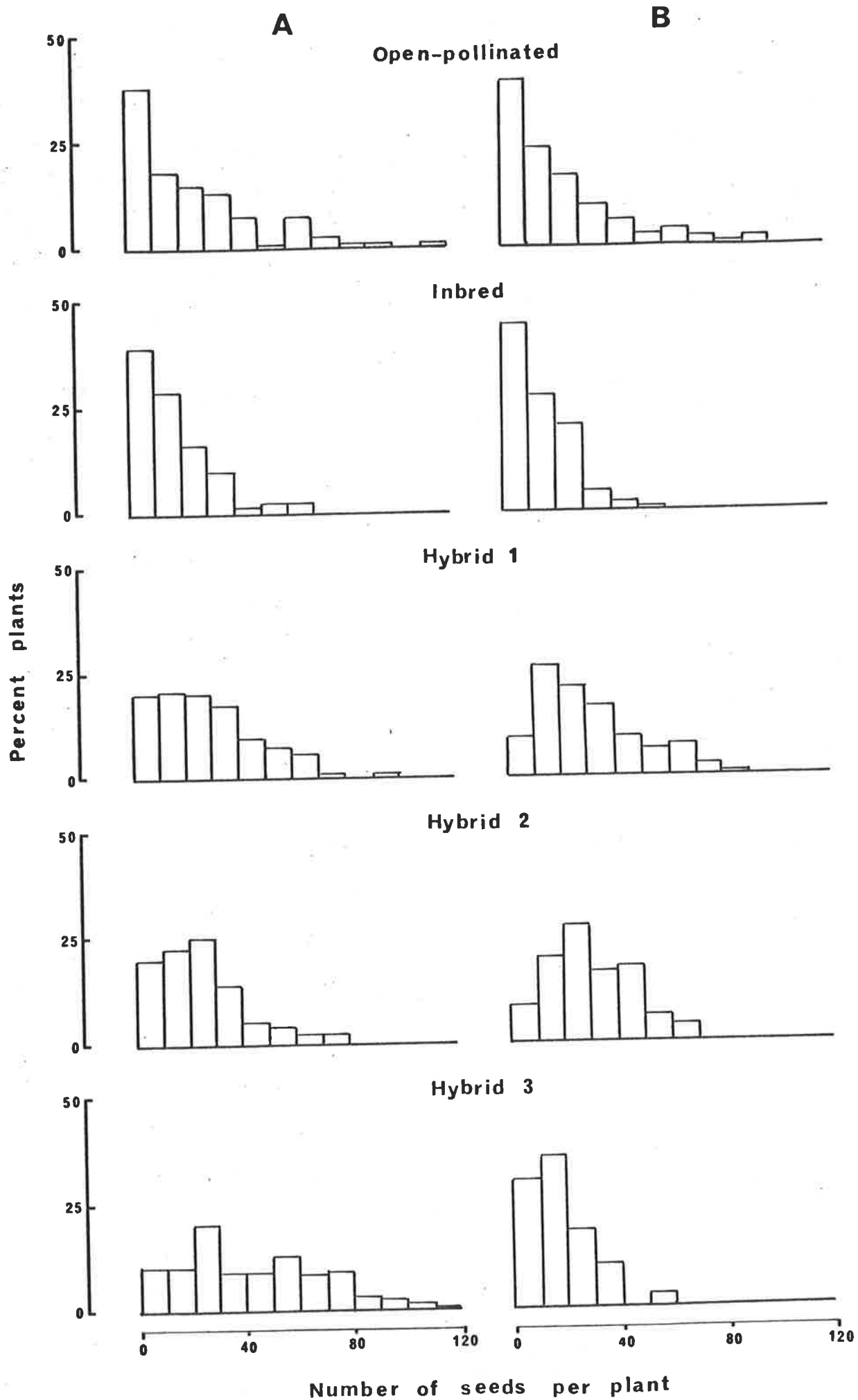


TABLE 24: Mean values for yield characters from tripped flowers of inbred and hybrid progenies.<sup>1</sup>

Progenies	Population	155			680		
		Yield (g)	No. of pods	No. of seeds	Yield (g)	No. of pods	No. of seeds
Open-pollinated		3.97	3.82	11.7	3.16	3.25	9.8
Inbred		4.27	4.77	13.0	1.97	2.14	6.6
Hybrid 1		6.73	6.43	20.4	3.62	3.73	11.7
Hybrid 2		5.18	5.19	15.2	3.69	3.90	12.3
Hybrid 3		5.78	5.00	16.7	2.60	2.45	7.6
L.S.D.		1.58 <sup>#</sup>	1.31	4.8	0.80	0.71	2.1

<sup>1</sup> Average of 108 plants each with 8 racemes tripped

<sup>#</sup> Probability of variance-ratio = 0.055

The hybrids in this population showed a limited advantage over the inbred and open-pollinated progenies. One hybrid (Hybrid 1) was significantly superior to the inbred and open-pollinated progenies for yield, pod and seed number when flowers were tripped.

In the other population (680) the open-pollinated progenies were better than the inbred but were not significantly different from Hybrids 1 and 2. These two hybrids were also significantly higher yielding than the inbreds.

In the detailed analyses of the progenies of each seed parent no results for open-pollinated plants are presented as these plants do not relate directly to any seed parent. The error and L.S.D. used in the comparisons between inbred and hybrid progenies differs slightly from that given in Table 24 due to the exclusion of the open-pollinated group.

In population 155, there were no significant differences between progenies for yield, pod and seed numbers although for many seed parents the hybrids show a higher pod set from tripped flowers than the related inbred (Tables 25-27). The large replicate effect and the loss of precision in testing the main plot factor (progenies) in the split-plot layout used for this experiment was considered responsible (Table 28).

In population 680, differences between the inbred and hybrids were found when flowers were tripped. The inbreds were inferior to the hybrids in yield characters from untreated flowers and this persisted even with tripping of the flowers. It may be noted that the yields of the inbreds in this population were generally lower than those in population 155. Such low vigour may have limited any benefit in yield likely to be derived from tripping of flowers.

TABLE 25: Mean yield (g) per plant produced by tripped flowers of inbred and hybrid progenies.<sup>1</sup>

Seed parent	Progenies				Mean
	Inbred	Hybrid 1	Hybrid 2	Hybrid 3	
<u>Popn. 155</u>					
1	5.07	7.70	5.82	5.74	5.80
2	1.50	4.44	3.17	3.31	3.11
3	2.38	5.20	5.40	3.91	3.88
4	4.66	4.28	5.41	4.54	4.60
5	7.51	8.23	7.78	6.92	6.88
6	2.64	6.96	4.32	5.35	5.02
7	3.40	5.01	5.98	6.49	4.58
8	6.26	10.01	4.60	4.23	5.57
9	5.10	8.56	6.39	9.22	6.81
10	3.22	4.23	4.78	3.28	3.67
11	5.60	9.87	4.43	9.73	7.12
12	3.94	6.31	4.10	6.58	5.19
Mean	4.27	6.73	5.18	5.78	
N.S.					
<u>Popn. 680</u>					
1	3.18	2.64	4.03	2.49	3.24
2	0.53	2.56	4.14	2.00	2.53
3	0.11	1.18	2.06	0.68	1.46
4	3.60	3.69	4.45	2.92	3.54
5	2.95	4.51	2.28	3.10	3.07
6	4.62	4.25	6.84	3.18	4.56
7	2.11	3.22	5.49	1.59	3.22
8	2.17	6.71	3.60	4.63	4.06
9	1.58	3.28	3.06	2.96	2.58
10	0.83	3.92	3.26	2.32	2.30
11	0.34	5.21	3.64	2.86	3.33
12	1.64	2.22	1.44	2.43	2.18
Mean	1.97	3.62	3.69	2.60	
LSD = 0.88					

<sup>1</sup> Average of 9 plants each with 8 racemes tripped.

No significant interaction between progenies and seed parents.

TABLE 26: Mean number of pods per plant produced by tripped flowers of inbred and hybrid progenies.<sup>1</sup>

Seed parent	Progenies				Mean
	Inbred	Hybrid 1	Hybrid 2	Hybrid 3	
<u>Popn. 155</u>					
1	6.56	8.11	6.89	6.00	6.89
2	1.67	4.11	3.33	3.22	3.08
3	2.33	4.67	4.67	3.33	4.64
4	5.00	5.33	4.67	3.56	4.64
5	9.56	8.56	8.33	5.78	8.06
6	3.22	6.78	4.00	4.78	4.69
7	3.78	4.44	5.11	5.44	4.69
8	5.44	8.22	5.44	4.44	5.89
9	6.33	8.33	6.67	7.78	7.28
10	3.22	4.00	4.89	2.89	3.75
11	5.44	8.44	4.22	7.00	6.28
12	4.67	6.11	4.00	5.78	5.14
Mean	4.77	6.43	5.19	5.00	
N.S.					
<u>Popn. 680</u>					
1	4.44	3.22	4.67	2.22	3.64
2	0.56	2.78	4.00	1.78	2.28
3	0.11	1.44	2.11	0.78	1.11
4	3.67	4.00	4.78	2.67	3.78
5	2.89	4.00	2.22	2.89	3.00
6	4.44	4.44	6.33	3.22	4.61
7	2.11	3.22	6.00	1.67	3.25
8	1.78	6.11	3.56	3.56	3.75
9	2.22	3.78	3.67	3.33	3.25
10	1.00	4.11	3.56	2.44	2.78
11	0.56	5.22	3.89	2.67	3.08
12	1.89	2.44	2.00	2.22	2.14
Mean	2.14	3.73	3.90	2.45	
LSD = 0.59					

<sup>1</sup> Average of 9 plants each with 8 racemes tripped

No significant interaction between progenies and seed parents.

TABLE 27: Mean number of seeds per plant produced by tripped flowers of inbred and hybrid progenies.<sup>1</sup>

Seed parent	Progenies				Mean	
	Inbred	Hybrid 1	Hybrid 2	Hybrid 3		
<u>Popn. 155</u>						
1	18.9	26.1	19.4	18.8	19.6	} LSD =5.1
2	4.9	12.9	9.0	10.4	9.4	
3	7.1	14.9	14.6	11.1	10.9	
4	15.2	15.4	14.0	12.2	13.9	
5	22.0	27.8	23.4	19.3	20.7	
6	8.2	21.8	12.7	16.4	15.3	
7	9.8	15.2	16.1	18.6	13.2	
8	16.4	26.9	15.6	12.8	15.9	
9	16.1	26.0	19.7	27.4	20.7	
10	9.7	13.6	14.0	8.9	11.1	
11	14.7	25.8	12.1	24.3	18.6	
12	13.0	18.7	12.1	19.9	15.6	
Mean	13.0	20.4	15.2	16.7		
N.S.						
<u>Popn. 680</u>						
1	11.9	9.0	13.8	7.6	10.5	} LSD =4.1
2	1.8	7.9	13.0	5.4	7.8	
3	0.4	4.3	7.0	2.2	4.9	
4	11.4	12.1	14.2	8.9	11.2	
5	9.2	13.3	7.7	8.9	9.2	
6	15.6	15.6	22.4	10.1	15.2	
7	6.3	9.9	19.2	5.1	10.5	
8	5.8	19.6	10.2	12.4	11.6	
9	5.9	11.9	11.0	10.0	9.0	
10	2.9	12.3	11.6	6.7	7.6	
11	1.2	17.0	11.7	8.3	10.5	
12	6.3	7.8	5.6	6.0	7.2	
Mean	6.6	11.7	12.3	7.6		
LSD = 2.4						

<sup>1</sup>Average of 9 plants each with 8 racemes tripped

No significant interaction between progenies and seed parents.

TABLE 28: Analysis of variance of yield, pod and seed number of population 155 from tripped flowers.

Source of variation	d.f.	Yield		Pods		Seeds	
		M.Sq.	P	M.Sq.	P	M.Sq.	P
Replication (R)	2	229.9	0.041	119.6	0.058	1051.2	0.075
Progeny (P)	3	115.3	0.128	59.2	0.171	1049.1	0.067
Error a	6	40.5		25.2		255.6	
Seed parent (S)	11	77.1	<0.001	85.8	<0.001	700.0	<0.001
P X S	33	15.2	0.641	9.2	0.554	96.3	0.724
Error b	88	17.1		9.7		116.2	

M.Sq. = Mean Square

P = Probability of Variance-ratio



In both populations, significant differences were found between the seed parents. Some of the seed parents show low productivity relative to others in spite of the tripping treatment given to the flowers.

In the top-cross type of test considered in the experiment significant differences were found between tester and between genotypes being tested.

In both populations and for each yield character studied the interaction between the seed parents and various progenies was not significant.

The pod set of tripped flowers cannot be directly compared with those of untreated flowers in this experiment. While tripping was carried out over a period of 2 to 3 weeks, the pod set from untreated flowers occurred from the commencement of flowering till the end of the season. Differences in environmental conditions would contribute to any difference that may be found. In experiments to be described later, there were tripped and untreated control flowers to allow for direct comparisons to be made (Section 4.4.2).

#### 4.3.4 Abortion of fertilized ovules.

The number of fertilized ovules that aborted in the inbred and hybrid progenies from pods set from untreated and tripped flowers are given in Table 29.

The various progenies did not differ in the number of ovules aborted whether flowers were untreated or tripped. The seed set from untreated and tripped flowers results from self-pollination and differences in the proportion of aborted ovules among inbreds and hybrids could be expected if homozygosis of deleterious genes were to result from selfing.

TABLE 29: Mean number of fertilized ovules that aborted per 100 pods.<sup>1</sup>

Progeny	155		680	
	Untreated	Tripped	Untreated	Tripped
Inbred	60.5	55.9	45.5	41.6
Cross 1	55.5	44.6	42.2	46.6
Cross 2	62.4	57.6	41.9	43.5
Cross 3	52.3	38.6	39.1	38.2
K-W statistic	0.95 NS	1.36 NS	0.69 NS	0.44 NS

<sup>1</sup>Derived from mature pods of 108 plants within each type of progeny

The abortion of ovules was not influenced by the hybridity of the progenies.

#### 4.3.5 Conclusion.

1. No obvious differences in height or stem number was found due to hybridity.
2. Hybrids were superior to inbreds for yield, number of pods and seeds per pod as a result of autofertility. The open-pollinated progenies were generally intermediate between the inbreds and the hybrids.
3. When flowers were tripped most hybrids continued to outyield the inbreds.
4. There was no differences in the proportion of fertilized ovules that aborted in inbreds and hybrids.
5. The performance of the hybrids appeared to be related to that of the inbred and varied with the particular pollen parent used.

#### 4.4.0 Experiment 4 (1975).

It will be recalled that due to difficulties in obtaining sufficient inbred and hybrid progenies this experiment on determinate material was separated from Experiment 3 on indeterminate types. It was not statistically valid to combine the two studies. In this experiment two, determinate type populations (41 and 95) were used. Each had 6 inbreds and their related hybrids (common seed parent). No open-pollinated progenies were included.

The comparisons between inbreds and hybrids were similar to those of Experiment 3 except that the effect of tripping could be directly studied by comparison of the tripped flowers with its untreated control.

#### 4.4.1 Autofertility - Yield and its components.

The findings in this experiment were similar to Experiment 3 in that the hybrids were superior to the inbreds for yield, and pod number. Moreover, the hybrids also produced heavier seeds than the inbreds. (Table 30). There was a tendency for the hybrids to set more seeds per pod in population 41 but this was not apparent in population 95.

The distribution of plants for autofertility follows the trend found in Experiment 3. The number of seeds produced in the absence of any flower treatment was again used as an index of autofertility. Among the inbreds there were a large number of plants with very low autofertility whereas the hybrids showed a wide range of autofertility with some plants producing more than 100 seeds (Fig. 15).

Although the comparisons between the inbreds and hybrids showed similar trends as that in Experiment 3, the determinate populations in this experiment appeared to produce much higher yields than the indeter-

TABLE 30A: Yield characters of inbred and hybrid progenies - Population 41

Seed Parent	Yield (g)		Number of pods		Number of seeds per pod		Weight per seed (g)		Number of podding racemes		Pods per podding raceme	
	I <sub>1</sub>	F <sub>1</sub>	I <sub>1</sub>	F <sub>1</sub>	I <sub>1</sub>	F <sub>1</sub>	I <sub>1</sub>	F <sub>1</sub>	I <sub>1</sub>	F <sub>1</sub>	I <sub>1</sub>	F <sub>1</sub>
1	12.2	35.7	19.1	33.7	2.18	2.53	0.29	0.45	13.4	27.4	1.46	1.20
2	16.8	35.8	15.8	35.0	2.26	2.15	0.49	0.47	11.9	27.6	1.29	1.27
3	25.3	44.8	34.1	42.3	1.72	2.47	0.43	0.44	23.8	33.1	1.42	1.26
4	8.3	30.0	10.3	24.9	1.84	2.54	0.53	0.48	9.6	20.2	1.05	1.21
5	8.9	40.2	10.7	27.3	2.15	2.71	0.41	0.55	8.6	24.6	1.22	1.11
6	5.9	49.8	6.9	40.2	1.69	2.37	0.50	0.52	6.3	34.3	1.04	1.17
Mean	12.9	39.4	16.3	33.9	1.98	2.46	0.44	0.48	12.3	27.9	1.25	1.20
L.S.D.	7.0		5.5		0.15		0.04		3.7		NS	

Significant interaction (progenies X seed parent)      L.S.D. between 2 means  
for these characters: Number of seed per pod ..      0.39  
Weight per seed ..      0.08

The above values were based on all untreated flowers averaged for 9 plants.

TABLE 30B: Yield characters of inbred and hybrid progenies. - Population 95

Seed Parent (I <sub>0</sub> )	Yield (g)		Number of pods		Number of seeds per pod		Weight per seed (g)		Number of podding racemes		Pods per podding raceme	
	I <sub>1</sub>	F <sub>1</sub>	I <sub>1</sub>	F <sub>1</sub>	I <sub>1</sub>	F <sub>1</sub>	I <sub>1</sub>	F <sub>1</sub>	I <sub>1</sub>	F <sub>1</sub>	I <sub>1</sub>	F <sub>1</sub>
1	5.1	29.6	6.6	22.3	1.85	2.21	0.38	0.49	7.7	19.9	0.83	1.11
2	9.8	22.4	15.2	21.6	1.70	2.46	0.38	0.43	13.4	19.0	1.11	1.13
3	12.9	38.2	14.8	30.9	2.30	2.41	0.35	0.51	11.8	26.4	1.21	1.16
4	9.3	11.7	7.7	8.7	2.49	2.33	0.49	0.57	7.3	9.4	1.05	1.02
5	8.9	11.2	11.1	8.9	2.45	2.16	0.33	0.50	10.0	11.6	1.54	0.77
6	14.0	33.2	10.9	17.6	2.64	2.87	0.46	0.65	12.0	16.4	0.86	1.05
Mean	10.0	24.4	11.0	18.3	2.23	2.41	0.40	0.52	10.4	17.3	1.09	1.04
L.S.D.	7.9		3.4		NS		0.04		3.1		NS	

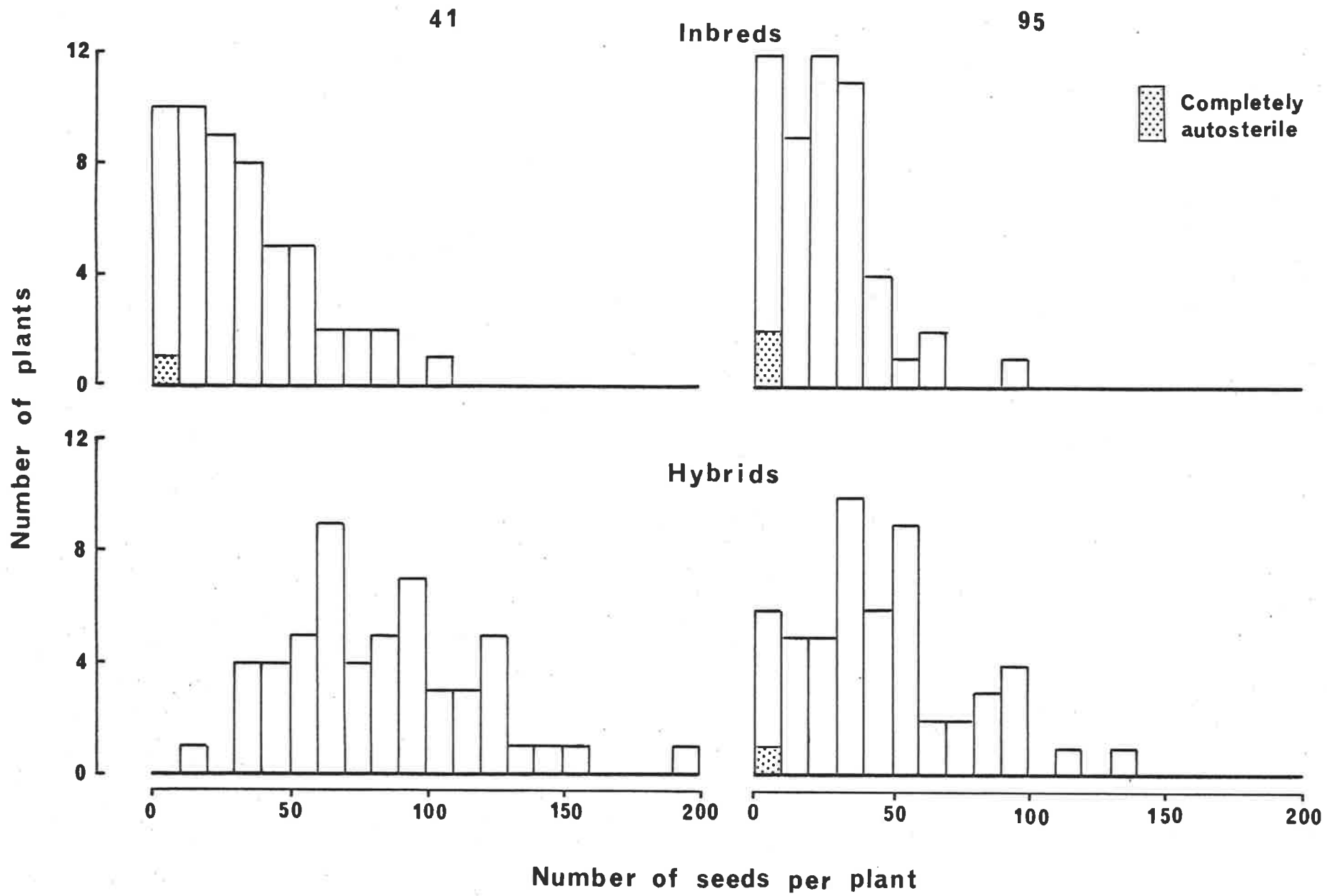
Significant interaction (progenies X seed parent) for these characters:

Number of pods	..	<u>L.S.D. between 2 means</u>	8.4
Pods per podding raceme	..		0.33

The above values were based on all untreated flowers averaged for 9 plants

Figure 15.

Distribution of plants for the number of seeds produced  
from inbred and hybrid progenies of population 41 and 95.





minate populations. Even though the two experiments were not statistically comparable the differences in the means or individual values indicated that the determinate populations were capable of better yields than the indeterminate type. (Compare Tables 21 or 22 with 30). This was in spite of the shorter habit of the determinate types and limited flowering period, which was compensated for by a large number of stems per plant (Table 31). The experiments on the indeterminate and determinate types were managed identically and there is no reason to doubt the advantage of the determinate type.

#### 4.4.2 The effect of tripping on pod and seed set.

In both populations there were more pods produced by tripped than untreated flowers. There was also a significant interaction between the inbred and hybrid progenies with the flower treatment (Table 32). There were large increases in the pod set of inbreds when the flowers were tripped but not significantly different in the hybrids. The inbreds had a strong tripping requirement if they were to achieve the pod set of the hybrids. However in population 41 the inbreds had still significantly less pods than the hybrid even when flowers were tripped which suggested that tripping requirement was not the only factor limiting yield. Other factors such as vigour of the plant or self-fertility could also be involved.

Tripping did not increase the number of seeds per pod in either the inbreds or the hybrids.

#### 4.4.3 Conclusions.

1. The results for comparisons between inbreds and hybrids are

TABLE 31: Mean number of flowering stems per plant for inbred and crossbred progenies of determinate populations.<sup>1</sup>

Seed parent	Population			
	41		95	
	Inbred	Hybrid	Inbred	Hybrid
1	4.00	4.67	5.00	6.33
2	4.44	4.22	5.00	5.78
3	3.89	4.67	3.56	5.56
4	4.22	4.44	4.33	4.62
5	3.22	5.89	5.44	4.56
6	3.89	5.56	5.00	5.33
L.S.D. (any 2 means)	1.33		1.13	

<sup>1</sup>Average of 9 plants. This also applies to other tables of this experiment.

TABLE 32: Mean number of pods and number of seeds per pod produced from untreated and tripped flowers of inbred and hybrid progenies.<sup>1</sup>

Flower treatment	41		95			
	Inbred	Hybrid	Inbred	Hybrid		
<u>Number of pods</u>						
Untreated (control)	0.92	3.57	*	1.41	2.02	*
Tripped	2.59	4.11	***	2.13	2.20	NS
	***	NS		**	NS	
<u>Number of seeds per pod</u>						
Untreated (control)	2.27	2.66	*	2.29	2.41	NS
Tripped	2.48	2.60	NS	2.36	2.67	NS
	NS	NS		NS	NS	

<sup>1</sup>Average of 54 plants with 10 untreated and 10 tripped flowers each

essentially the same as for Experiment 3 ---

- (a) There was no consistent advantage in stem number due to hybridity.
  - (b) Hybrids were superior to inbreds for yield and its components.
2. Inbred<sup>s</sup> have a strong tripping requirement whereas hybrids do not respond to tripping.
  3. The determinate type appeared to be higher yielding than the indeterminate type.

#### 4.5.0 Experiment 5 (1976).

As stated in the Material and Methods, this experiment was set up to compare inbred progenies of known origin. In the experiments so far, although inbreds have been studied, the number of generations of inbreeding in the original parents was unknown. The progenies called  $F_2$  in this experiment would represent plants with only one generation of inbreeding from the hybrid condition. These  $F_2$  are compared with  $I_2$  which would have at least 2 generations of inbreeding. The  $I_2$  and  $F_2$  progenies of population 4 were used.

#### 4.5.1 Autofertility -- Yield and its components.

Most of the  $F_2$  progenies were higher yielding and bore more pods than  $I_2$  progenies (Table 33). All the yield advantage is attributable to pod numbers. There was no advantage in the number of seeds per pod. Differences in the number of seeds per pod were associated with the seed parents.

#### 4.5.2 The influence of tripping on seed set.

The overall effect of tripping on yield could also be studied from the number of seed<sup>N</sup> instead of the components, pod number and seeds per pod. The number of seeds per plant produced from untreated control and tripped flowers is given in Table 34.

Both the  $I_2$  and  $F_2$  progenies responded to tripping. The  $F_1$  material had been previously shown not to respond to tripping as they already produced many pods and seeds in Experiment 4 (Section 4.4.2). One generation of inbreeding from the  $F_1$  to the  $F_2$  had been sufficient to cause a substantial requirement for tripping.

TABLE 33: Mean yield, pod number per plant and seeds per pod for  $I_2$  and  $F_2$  progenies (Untreated flowers).

Seed Parent	Yield (g)		Pod number		Seeds per pod	
	$I_2$	$F_2$	$I_2$	$F_2$	$I_2$	$F_2$
1	25.6	43.0	24.6	28.9	2.22	2.32
2	22.7	17.8	20.1	11.9	1.89	1.79
3	25.2	21.4	23.4	18.1	1.81	1.49
4	21.0	42.6	16.3	29.0	1.98	2.13
5	16.9	26.6	13.1	21.3	2.18	1.84
6	8.0	43.5	7.1	32.2	1.91	1.98
L.S.D.	12.4		8.4		NS	
	(between any 2 means)					

Note: The number of seeds per pod was only significantly different between seed parents.

The above values were based on all untreated flowers.

Each value is the mean of 9 plants.

TABLE 34: The mean number of seeds per plant produced from untreated (control) and tripped flowers of  $I_2$  and  $F_2$  progenies.<sup>1</sup>

Parent	Flower treatment	$I_2$	$F_2$	
1	Untreated	8.8	14.2	NS
	Tripped	22.2	22.1	NS
		**	**	
2	Untreated	1.6	4.6	NS
	Tripped	16.8	9.1	**
		**	**	
3	Untreated	5.1	4.1	NS
	Tripped	11.3	11.8	NS
		**	**	
4	Untreated	5.9	11.9	**
	Tripped	17.0	19.9	NS
		**	**	
5	Untreated	4.9	3.1	NS
	Tripped	9.3	9.2	NS
		**	**	
6	Untreated	1.8	11.9	**
	Tripped	16.7	17.3	NS
		**	**	

<sup>1</sup>Average of 18 plants each with 16 flowers untreated and tripped.

#### 4.5.3 Conclusions.

1. The  $F_2$  progenies had a higher yield and pod set than the  $I_2$  progenies which have at least one extra generation of inbreeding.
2. Seeds per pod was not different for the two progenies.
3. The  $F_2$  as well as the  $I_2$  progenies showed a response to tripping. Evidently a single generation of inbreeding from the hybrid condition results in a response to tripping. Inbreeding depression in this character must occur frequently in normal crops.



#### 4.6.0 Experiment 6 (1976).

The preceding experiments on the effect of hybridity provided a direct comparison of inbreds and their hybrids. In this study, material over a range of hybridity i.e. with different relationships was used. Basically it consisted of second generation inbreds ( $I_2$ ), first generation inbreds ( $I_1$ ), open-pollinated progenies of 2 small seeded populations, together with crosses between plants of the same population ( $H_1$ ), crosses between the 2 small seeded populations ( $H_2$ ) and crosses between the small seeded populations and large seeded populations ( $H_3$ ).  $H_1$ ,  $H_2$ , and  $H_3$  are considered to represent increasing hybridity. The  $I_2$ , and the three hybrids were produced from a common seed parents derived from  $I_1$  material. There were 4 seed parents from each of the small seeded populations which were coded 41 and 95. As the inbred  $I_2$  and the various hybrids were derived from different populations the results are presented separately for each group of seed parents. Further details about the material were given in Section 2.5.6.

#### 4.6.1 Autofertility.

The yield and seed set from untreated flowers for the various progenies are shown in Figure 16. There was very marked increases in yield from  $I_2$  to the hybrids. However, increases between  $H_1$  and  $H_3$  were not consistent.

This was also seen among the individual progenies of each seed parent (Table 35).

Between the hybrids, i.e. between  $H_1$ ,  $H_2$  and  $H_3$  in general the differences were not significant. The  $H_3$  of seed parent No. 8 however,

Figure 16

Mean yield, seed number and seed weight of inbred, open-pollinated and various hybrid progenies.

Progenies are ranked by increasing dissimilarities of the female and male parent.

A = Progenies derived from seed parents from population 41.

B = Progenies derived from seed parents from population 95.

$I_2$  = Second generation inbreds derived from  $I_1$ .

$I_1$  = First generation inbreds.

OP = Open-pollinated progenies of the populations 41 and 95.

$H_1$  = Hybrids within small seeded populations.

$H_2$  = Hybrids between small seeded populations 41 and 95.

$H_3$  = Hybrids between a small and a large seeded population.

$I_2$ ,  $H_1$ ,  $H_2$ ,  $H_3$  are derived from common seed parents ( $I_1$ ) within each population.

□—□ Yield (g) per plant

△—△ Number of seeds per plant

○—○ Weight per seed

There were 36 plants for each of the progenies.

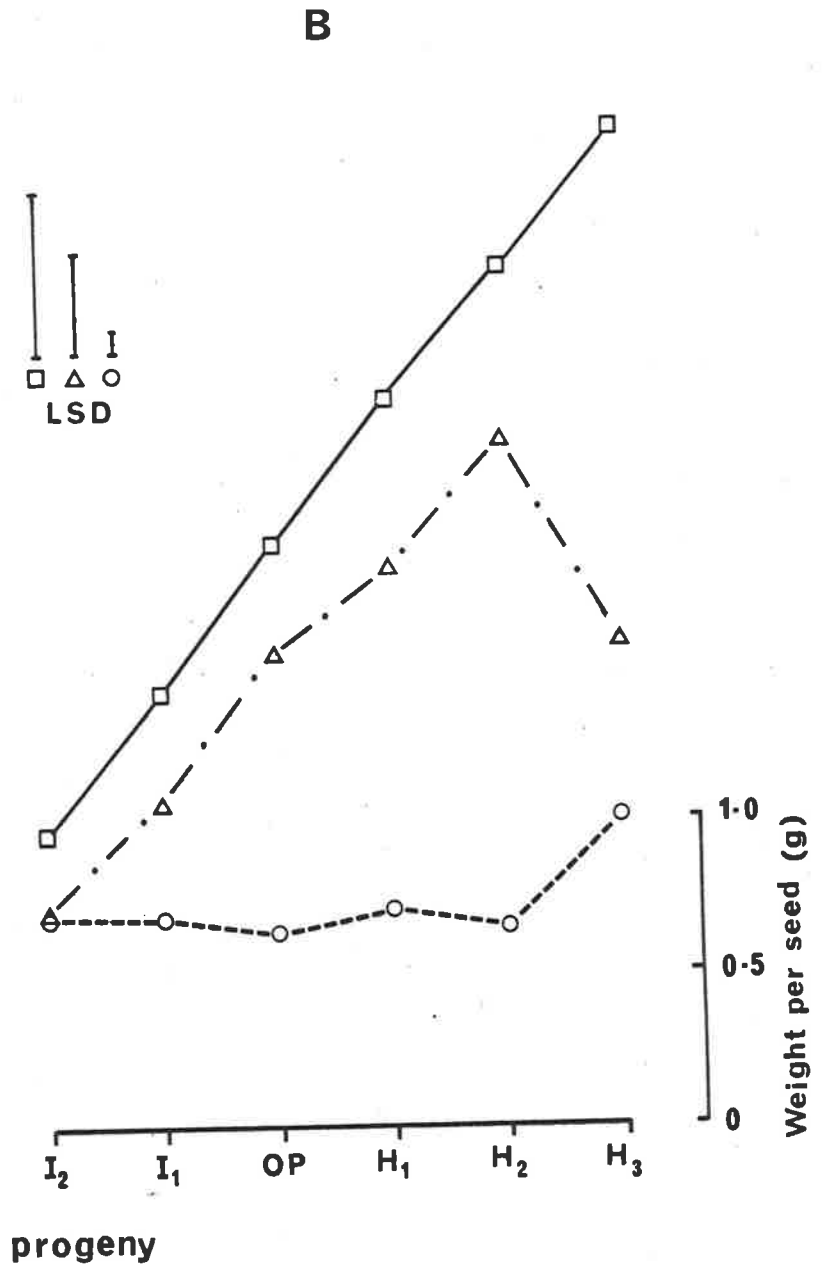
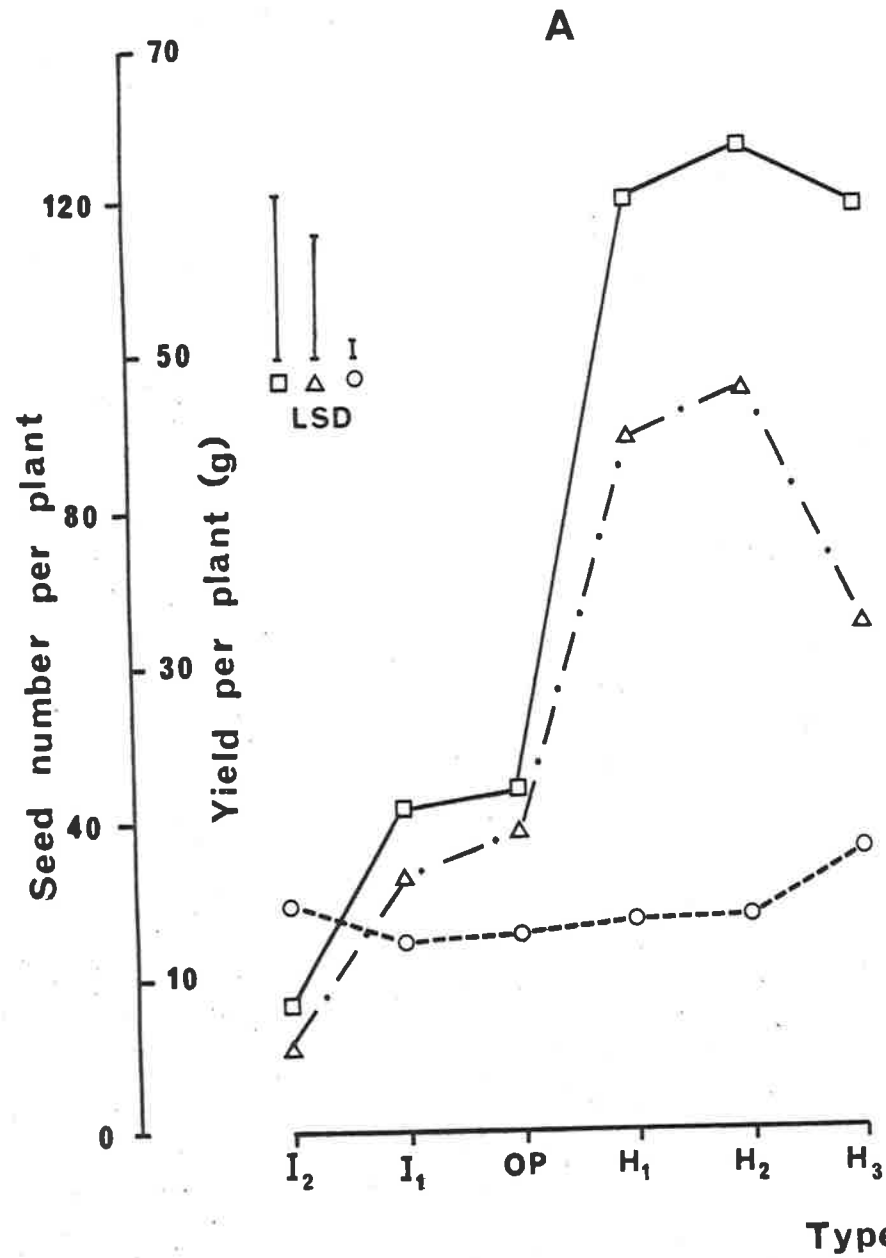


TABLE 35: Mean yield (g) per plant for progenies of different levels of hybridity.<sup>1</sup>

Seed parents	Progeny					
	I <sub>2</sub>	I <sub>1</sub>	OP	H <sub>1</sub>	H <sub>2</sub>	H <sub>3</sub>
<u>Popn. 41</u>						
1	4.8	27.2	22.7	63.6	61.6	43.4
2	4.3	13.6	25.3	54.8	70.0	66.4
3	4.3	3.0	17.3	49.9	59.5	67.7
4	18.4	39.4	22.6	70.3	63.3	60.3
Mean	8.2	20.8	22.0	59.8	63.4	59.5
L.S.D. = 10.1						
<u>Popn. 95</u>						
5	19.8	39.7	39.1	49.3	73.8	75.6
6	20.1	17.5	41.4	48.4	57.4	51.1
7	26.5	34.3	32.1	43.6	49.6	51.2
8	10.0	20.1	37.9	45.8	46.6	83.7
Mean	19.1	27.9	37.6	46.8	55.3	64.6
L.S.D. = 10.1						

<sup>1</sup>Average of 9 plants

I <sub>2</sub>	Second generation inbreds	H <sub>1</sub>	Crosses within populations
I <sub>1</sub>	First generation inbreds	H <sub>2</sub>	Crosses between small seeded populations
OP	Open-pollinated progenies	H <sub>3</sub>	Crosses between small and large seeded populations

The above notation applies throughout this experiment.

No significant interaction between progeny and seed parent.

Conversion to t/ha = x 0.0735

yielded almost twice that of the other hybrids but this was the result of larger seeds rather than higher autofertility in terms of seed set (Table 36).

The yield was closely associated with number of seeds ( $r = 0.90***$ ) but only slightly correlated with seed weight ( $r = 0.33 ***$ ). As shown in Figure 16 the number of seeds follow the yield closely except where seed size was different as in  $H_3$ .

Among the hybrids,  $H_2$  representing a cross between populations of the same seed size tended to have a higher seed set than  $H_1$  whereas with  $H_3$  there was a fall in seed numbers. The drop in autofertility in  $H_3$  may have been due to:

(a) The larger seed size of the  $H_3$  — seed size has been shown to be negatively correlated with seed weight (Exp. 2, Section 4.2.2).

(b) A lower level of fertility in the large-seeded parents. The seed production data of the large-seeded parents used to produce the crosses showed that the pollen parents of  $H_3$  had a very low seed set relative to the other parents (Table 37). This factor of low seed set was the reason for having to modify the experiment from one that would have had the various progenies produced by seed parents of both small and large seeded populations — the present experiment consist of only half of the symmetrical sets of progenies planned (see Section 3.5.6).

When direct comparisons are made between a hybrid and its two parents, it is evident that all the hybrids had higher yields than the top parents even though in several crosses the other parent had very low

TABLE 36: Mean number of seeds per plant for progenies of different levels of hybridity.<sup>1</sup>

Seed parents	Progeny					
	I <sub>2</sub>	I <sub>1</sub>	OP	H <sub>1</sub>	H <sub>2</sub>	H <sub>3</sub>
<u>Popn. 41</u>						
1	7.5	46.8	39.1	97.0	82.6	59.2
2	7.0	28.4	47.1	94.0	123.5	72.2
3	8.5	5.2	32.8	66.6	103.2	77.6
4	18.9	50.2	36.3	98.1	74.8	52.2
Mean	10.6	32.7	38.8	88.8	95.2	65.3
L.S.D. = 15.8						
<u>Popn. 95</u>						
5	28.0	59.2	62.9	65.0	108.0	71.2
6	41.2	31.0	64.4	87.7	98.0	50.8
7	29.4	47.0	60.1	63.6	77.0	54.9
8	12.2	28.2	56.9	72.0	77.9	75.0
Mean	27.7	41.6	61.1	72.1	88.6	62.4
L.S.D. = 13.0						

No significant interaction between progeny and seed parent.

<sup>1</sup>Average of 9 plants

The above values were based on all untreated flowers.

TABLE 37: Total number of seed produced by parent plants in the production of inbreds and hybrids. (Figures in parenthesis indicate the number of selfed seed).

Seed Parent	Pollen parent		
	Cross 1 (H <sub>1</sub> )	Cross 2 (H <sub>2</sub> )	Cross 3 (H <sub>3</sub> )
1 85 (20)	69 (22)	87 (67)	17 (3)
2 136 (53)	81 (47)	93 (24)	0 (0)
3 83 (17)	87 (61)	43 (17)	4 (0)
4 61 (28)	44 (11)	37 (35)	18 (11)
5 57 (37)	87 (67)	69 (22)	13 (4)
6 43 (17)	88 (43)	83 (17)	19 (19)
7 59 (28)	58 (35)	54 (11)	10 (10)
8 80 (51)	56 (37)	10 (2)	19 (15)

The pollen parents for H<sub>3</sub> crosses are large seeded.

All other parent plants are small seeded.

yields (Table 38). The yield advantage of the hybrids when expressed as a percentage of the mid-parent and the better parent showed much variation. Increases up to 2.9 times that of the mid-parent was obtained in one cross (Cross 2) but was small in the other crosses. No specific trends were noted in the performance of hybrids from crosses within a population (Crosses 1 to 8) or crosses between populations (Crosses 9 to 12) and both types of hybrids had various levels of yield increase over the parents.

In a number of inbreds, there was no difference between the yield of the  $I_1$  and  $I_2$ , that is, between the parent and inbred values in Table 38. The additional generation of inbreeding on these inbreds did not result in a depression in yield. Examples include the parents of Cross 12. These and some other inbreds had yields not very inferior to the lowest hybrids.

Again, yield was strongly associated with the number of seeds ( $r = 0.96$  \*\*\*). The data in Table 39 which are the number of seeds and an index of autofertility strongly reflect the yield values given in Table 38.

#### 4.6.2 The effect of tripping.

The data on autofertility, presented in the section above, was based on all the untreated flowers on the plants. In this section, reference is made to untreated and tripped flowers. Here a matching set of flowers was considered so that the untreated flowers are controls for the tripped flowers.

The inbreds with an initial low level of seed set from untreated flowers in general showed a large improvement following tripping. This



TABLE 38: Mean yield per plant (g) for parents, inbreds and hybrids -- all untreated flowers -- together with the calculated value for heterosis.<sup>1</sup>

Cross	Female		Hybrid	Male		% heterosis above	
	Inbred	Parent		Parent	Inbred	Mid-P	High-P
1	5.5	43.5	48.0	3.0	7.7	107	10
2	9.6	6.9	60.1	23.9	18.2	289	151
3	12.0	9.5	73.4	41.7	47.4	187	76
4	12.2	23.9	63.4	36.3	40.1	111	75
5	31.8	45.6	46.5	39.1	21.0	10	2
6	30.5	28.0	47.5	11.9	24.7	130	70
7	23.3	30.6	49.9	42.9	21.8	36	16
8	34.1	41.3	68.4	44.6	45.4	59	53
9	7.7	3.0	56.5	45.6	31.8	133	24
10	18.2	23.9	57.7	28.0	30.5	117	106
11	47.4	41.7	81.0	42.9	21.8	92	89
12	40.1	36.3	56.5	44.6	45.4	40	27

L.S.D. between any two values = 19.2

<sup>1</sup>Average of 9 plants. This applies to all tables in this experiment.

Populations involved in making the crosses.

Crosses 1 to 4, 41 X 41

Crosses 5 to 8, 95 X 95

Crosses 9 to 12, 41 X 95 (has a common parent with each of the Crosses 1 to 8)

This order of presentation is retained throughout this experiment.

TABLE 39: Mean number of seeds per plant for parents, inbreds, and hybrids.

Cross	Female		Hybrid	Male	
	Inbred	Parent		Parent	Inbred
1	8.4	70.3	73.5	4.1	15.3
2	17.6	10.4	78.7	21.2	18.8
3	17.1	14.0	99.1	54.3	59.7
4	19.0	50.1	109.3	47.9	55.8
5	43.1	70.6	59.3	57.8	29.2
6	54.8	55.0	83.1	19.2	41.7
7	27.0	36.9	80.8	59.2	36.4
8	56.6	61.1	111.2	81.8	69.2
9	15.3	4.1	80.7	70.6	43.1
10	18.8	21.2	84.8	55.0	54.8
11	59.7	54.3	111.4	59.2	36.4
12	55.8	47.9	84.9	81.8	69.2

L.S.D. between any two means = 28.7

was most pronounced in the  $I_2$  material (Table 40). However some inbreds did not respond to tripping and had a low seed set. Examples were  $I_2$  of seed parent 7 and  $I_1$  of parents 1 and 3. This probably indicates low self-fertility rather than inadequate pollination. Low self-fertility was also evident in some inbreds that did respond significantly to tripping such as the  $I_2$  of seed parents 1, 3 and 4. Despite their response they continued to have a low seed set.

Some inbreds that benefited from tripping had tripped seed sets as good as or even better than their hybrids (e.g. progenies of seed parents 2, 5 and 8). In these inbreds, a low seed set and autofertility was due to inadequate pollination or a tripping requirement rather than low self-fertility.

Generally for the hybrids,  $H_1$ ,  $H_2$  and  $H_3$  there was little difference between untripped and tripped treatments possibly because the hybrids have a high level of seed set in untripped flowers and a further improvement could not be expected.

Consideration will now be given to the direct comparison of hybrids with their parents and the response to tripping. The results in Table 41 show that the hybrids individually did not respond significantly to tripping although their parents (the  $I_1$ s) and the inbreds of the parents did. The results indicate that the requirement for tripping was influenced by hybridity rather than genotype.

This association of autofertility with hybridity could make selection for autofertile genotypes difficult. The material needs to be inbred sufficiently before the true capability of the plant to set seeds from untreated flowers in the absence of bees is evident. Some of the  $I_2$  with high seed set from untreated flowers may possess characters

TABLE 40: The effect of flower treatment on the mean seed set per plant of progenies of different levels of hybridity.<sup>1</sup>

Seed parent	Flower treatment	Progeny				
		I <sub>2</sub>	I <sub>1</sub>	H <sub>1</sub>	H <sub>2</sub>	H <sub>3</sub>
1	Untreated	0	2.4	17.3	12.1	11.0
	Tripped	4.9	6.4	19.1	12.9	10.1
2	Untreated	2.1	4.4	14.6	26.8	20.8
	Tripped	26.6	32.2	23.6	33.9	20.4
3	Untreated	0.4	0	17.9	21.8	15.6
	Tripped	6.1	2.2	16.7	22.6	19.2
4	Untreated	2.4	8.6	14.9	13.6	9.0
	Tripped	6.0	11.7	16.6	13.8	9.7
5	Untreated	7.1	10.7	12.3	21.2	10.6
	Tripped	16.8	12.4	11.9	24.2	10.0
6	Untreated	9.4	8.4	15.1	20.8	9.7
	Tripped	14.3	14.1	20.8	20.9	12.2
7	Untreated	2.2	3.4	8.7	10.6	6.0
	Tripped	3.2	6.1	13.1	17.2	8.2
8	Untreated	2.8	7.1	15.2	16.4	9.8
	Tripped	33.4	24.1	16.2	21.4	9.2

Untreated refer to the flowers used as control. There were 16 flowers untreated and 16 flowers tripped for each plant.

<sup>1</sup>Average of 9 plants.

TABLE 41: Mean number of seeds per plant produced by untreated and tripped flowers.

Seed	Flower treatment	Female Parent		Hybrid	Male Parent		Inbred	
		Inbred				Inbred		
1	UT	0	5.1	9.1	0.2	0.4		
	T	6.0	10.1	11.7	8.3	8.7	*	**
2	UT	1.0	0.4	20.7	3.1	2.1		*
	T	4.3	2.4	20.1	8.2	10.4	NS	*
3	UT	1.0	0.4	15.0	3.0	6.3		
	T	6.0	8.9	18.9	6.4	14.6	NS	**
4	UT	3.2	9.4	21.7	8.0	3.8		
	T	38.2	31.9	24.9	11.2	13.8	NS	**
5	UT	11.0	13.7	17.3	8.7	3.3		
	T	19.8	18.4	15.1	9.8	14.8	NS	**
6	UT	7.3	11.9	19.8	4.5	10.1		
	T	18.9	14.1	21.8	12.0	16.4	NS	**
7	UT	1.0	1.3	12.0	10.9	5.4		
	T	3.2	5.8	15.1	13.0	16.7	NS	**
8	UT	11.1	10.1	23.3	10.2	11.2		
	T	24.8	29.7	26.6	21.8	13.8	NS	NS
9	UT	0.4	0.2	21.0	13.7	11.0		
	T	8.7	8.3	22.6	18.4	19.8	NS	**
10	UT	2.1	3.1	15.8	11.9	7.3		
	T	10.4	8.2	18.1	14.1	18.9	NS	**
11	UT	6.3	3.0	12.7	10.9	5.4		
	T	14.6	6.4	12.3	13.0	16.7	NS	**
12	UT	3.8	8.0	18.9	10.2	11.2		
	T	13.8	11.2	20.6	21.8	13.8	NS	NS

UT = Untreated control

T = Tripped

conferring high autofertility.

The heterosis of the hybrids in relation to their parents was apparent in the untreated flower treatments, and in the tripped flower treatment. The seed set of tripped flowers indicates the level of self-fertility. Heterosis was strongly expressed when the self-fertility of the parents was low (e.g. Cross 2 and 3), but hybrids from parents showing good self-fertility had little improvement above the mid-parent (Crosses 4, 5 and 8, Table 42). Such a situation might result from a level of seed set that approaches the maximum possible before other major limiting factors come into play.

The number of seeds per pod was not influenced by tripping in the various types of progenies (Tables 43 and 44).

#### 4.6.3 Vegetative and floral characters.

A study was made of the vegetative and floral characters to see if they could account for the high yields of the hybrids.

In general with the exception of plant height, there was no consistent advantage shown by the hybrids over the other progenies. In the comparison involving various inbreds and hybrids ( $I_2$ ,  $I_1$ ,  $H_1$ ,  $H_2$  and  $H_3$ ) the hybrids were taller than the inbreds (Table 45).

For most of the other characters the hybrids  $H_1$ ,  $H_2$  and  $H_3$  were not superior to  $I_2$ ,  $I_1$  and OP and in some were significantly inferior. This is supported by the values in Tables 46 to 49 where details are given of the vegetative character of the parents and their hybrids. The superiority of the inbreds and parents over the hybrids for some of the vegetative characters may be a direct consequence of their lower yields. The stems may have a greater weight in the inbred and parents when less

TABLE 42: Percentage heterosis of the crosses based on the seed set of tripped flowers.

Cross	Hybrid seed set	Mid-parent seed set	Heterosis above M.P. %	High-parent seed set	Heterosis above H.P. %
1	11.7	9.2	27	10.1	16
2	20.1	5.3	279	8.2	145
3	18.9	7.6	149	8.9	112
4	24.9	21.6	15	31.9	-22
5	15.1	14.1	7	18.4	-18
6	21.8	13.0	68	14.1	55
7	15.1	9.4	61	13.0	16
8	26.6	25.8	3	29.7	-10
9	22.6	13.4	69	18.4	23
10	18.1	11.2	62	14.1	28
11	12.3	9.7	27	13.0	- 5
12	20.6	16.5	25	21.8	- 6

TABLE 43: The effect of flower treatment on the mean number of seeds in the pod of progenies of different levels of hybridity.

Seed parent	Flower treatment	Progeny				
		I <sub>2</sub>	I <sub>1</sub>	H <sub>1</sub>	H <sub>2</sub>	H <sub>3</sub>
1	Untreated	No pods	3.12	3.16	2.74	2.83
	Tripped	3.00	3.12	3.14	2.74	2.45
2	Untreated	2.83	2.79	2.81	2.63	2.71
	Tripped	3.26	3.34	2.70	2.77	2.84
3	Untreated	1.50	No pods	2.79	2.64	2.55
	Tripped	1.81	2.10	2.59	2.56	2.79
4	Untreated	2.27	2.43	2.41	2.42	2.35
	Tripped	2.00	2.46	2.51	2.41	2.30
5	Untreated	2.18	2.22	2.49	2.74	2.86
	Tripped	2.38	2.30	2.53	3.09	2.65
6	Untreated	1.98	2.15	1.97	2.56	2.57
	Tripped	2.31	2.25	2.18	2.61	2.40
7	Untreated	2.67	2.09	2.47	2.33	2.46
	Tripped	1.78	2.14	2.68	2.61	2.42
8	Untreated	1.56	2.06	2.28	2.15	2.76
	Tripped	3.00	2.51			

No significant difference between treatments for all progenies except the 2 indicated.



TABLE 44: Mean number of seeds per pod produced from untreated and tripped flowers.

Seed	Flower treatment	Female		Hybrid	Male		
		Inbred	Parent		Parent	Inbred	
1	UT	no pod	2.94	2.95	2.00	2.00	*
	T	3.31	3.12	2.93	2.18	3.28	
2	UT	1.17	1.00	2.83	3.00	1.98	
	T	2.19	2.00	2.77	2.21	2.43	
3	UT	1.50	1.00	2.57	2.22	2.33	
	T	1.78	2.23	2.64	2.06	2.54	
4	UT	2.61	2.51	2.64	2.77	1.65	
	T	3.50	3.35	2.53	3.03	2.24	
5	UT	2.25	2.11	3.01	2.64	2.80	*
	T	2.62	2.46	2.97	2.54	2.57	
6	UT	1.83	2.23	2.43	2.77	2.03	
	T	2.24	1.84	2.26	2.42	2.36	
7	UT	2.17	2.33	2.54	2.30	2.22	
	T	2.27	2.41	2.59	2.57	2.54	
8	UT	2.40	2.34	2.20	2.15	1.71	*
	T	2.66	2.82	2.33	2.66	1.92	
9	UT	2.00	2.00	2.96	2.11	2.25	
	T	3.28	2.18	2.81	2.46	2.62	
10	UT	1.98	3.00	2.56	2.23	1.83	
	T	2.43	2.21	2.52	1.84	2.24	
11	UT	2.33	2.22	2.89	2.30	2.22	
	T	2.54	2.06	2.70	2.57	2.54	
12	UT	1.65	2.77	2.23	2.15	1.71	*
	T	2.24	3.03	2.25	2.66	1.92	

UT = Untreated control

T = Tripped

All UT and T not significant except for the pairs indicated \*

TABLE 45: Mean plant height, stem number, stem weight and raceme number per plant for progenies with different levels of hybridity.<sup>1</sup>

Character	Seed parent group	I <sub>2</sub>	I <sub>1</sub>	OP	H <sub>1</sub>	H <sub>2</sub>	H <sub>3</sub>	LSD
Plant height	A	80.1	79.2	88.6	89.1	93.9	90.1	4.7
	B	76.9	72.1	82.3	84.4	87.6	91.3	6.9
Stem number	A	5.44	4.50	4.06	4.97	4.92	4.81	0.74
	B	4.89	4.86	5.19	5.64	5.21	5.56	NS
Stem Weight (g)	A	61.8	46.5	55.8	49.3	41.3	38.2	8.8
	B	32.9	27.5	28.1	29.0	41.7	36.2	5.1
Raceme number	A	91.4	81.8	76.2	80.8	72.4	66.1	9.8
	B	68.2	66.9	69.0	68.8	79.0	74.3	NS

<sup>1</sup>Average of 9 plants. This applies to other tables in this experiment.

TABLE 46: Mean plant height (cm) of parents, inbreds and hybrids.

Cross	Female		Hybrid	Male	
	Inbred	Parent		Parent	Inbred
1	78	92	96	109	107
2	86	95	93	92	87
3	93	93	100	91	92
4	100	101	102	80	87
5	90	90	94	81	100
6	86	87	95	80	86
7	80	88	82	82	78
8	89	89	97	94	89
9	107	109	105	90	90
10	87	92	96	87	86
11	92	91	99	82	78
12	87	80	93	94	89

L.S.D. between any two means = 10

TABLE 47: Mean number of stems per plant for parents, inbreds and hybrids.

Cross	Female		Hybrid	Male	
	Inbred	Parent		Parent	Inbred
1	4.00	3.89	4.33	2.89	2.33
2	3.22	3.25	4.11	5.44	4.00
3	3.89	4.00	5.44	4.78	5.67
4	4.22	4.44	5.78	5.89	6.56
5	4.37	5.89	4.67	6.89	6.22
6	4.67	4.11	4.78	3.67	3.87
7	5.67	6.44	7.11	5.00	5.22
8	3.56	3.89	4.00	5.56	5.00
9	2.33	2.89	4.14	5.89	4.37
10	4.00	5.44	4.89	4.11	4.67
11	5.67	4.78	6.22	5.00	5.22
12	6.56	5.89	5.00	5.56	5.00

L.S.D. between any two means = 1.27

TABLE 48: Mean stem weight (g) per plant for parents, inbreds and hybrids.

Cross	Female		Hybrid	Male	
	Inbred	Parent		Parent	Inbred
1	64.7	53.6	73.2	89.2	68.4
2	62.7	81.1	44.6	90.0	56.7
3	94.2	95.1	67.4	65.9	69.1
4	87.7	77.4	45.0	31.1	51.8
5	28.1	31.4	29.3	24.2	46.0
6	41.3	40.9	35.0	26.7	29.2
7	55.1	60.1	35.9	32.8	24.6
8	40.6	35.0	36.7	49.0	36.6
9	68.4	89.2	42.0	31.4	28.1
10	56.7	90.0	44.6	40.9	41.3
11	69.1	65.9	49.0	32.8	24.6
12	51.8	31.1	36.1	49.0	36.6

L.S.D. between any two means = 21.0

TABLE 49: Mean number of racemes per plant for parents, inbreds and hybrids.

Cross	Female		Hybrid	Male	
	Inbred	Parent		Parent	Inbred
1	75.1	73.9	87.3	80.2	59.2
2	70.2	86.6	63.9	91.6	61.9
3	90.2	95.6	97.9	96.4	110.7
4	71.6	77.2	74.9	71.3	91.7
5	64.5	75.9	58.3	71.9	71.6
6	72.1	66.8	67.1	45.6	58.0
7	87.7	100.0	82.6	67.1	55.9
8	50.0	54.9	60.1	79.4	69.0
9	59.2	80.2	64.9	75.9	64.5
10	61.9	91.6	77.8	66.8	72.1
11	110.7	96.4	98.4	67.1	55.9
12	91.7	71.3	67.1	79.4	69.0

L.S.D. between any two means = 18.1

metabolic material has gone into the growth of pods and seeds.

It may be concluded that the higher yields, the lower need for tripping and the greater number of seeds per plant exhibited by the hybrids are the heterotic characters and that vegetative characters do not show any evidence of heterosis in this material of *Vicia faba*.

#### 4.6.4 Conclusions.

1. Yield was influenced by hybridity and increased markedly from the I<sub>2</sub>, I<sub>1</sub>, OP to the hybrids. Increasing dissimilarities between parents of some hybrids did not result in those hybrids having increased yields.
2. Heterosis for yield and seed set was expressed strongly over the mid-parent and the high parent in many crosses.
3. There was no relationship between yield of the parents and that of the hybrids.
4. Inbreds but not hybrids generally responded to tripping.
5. A problem of low self-fertility that limited seed set was found in some inbreds.
6. Other inbreds were not affected by further inbreeding and produced yields not much lower than those of hybrids.
7. In all progenies, the number of seeds per pod was not influenced by tripping.
8. In contrast to yield and the number of seeds set, there was no expression of heterosis for vegetative character or in the number of racemes.

#### 4.7.0 Miscellaneous studies.

As with any new crop there are many aspects of its culture or biology that are unknown and needed investigation. These were assessed in supplementary experiments. They included the vegetative propagation of *V. faba* and the effect of scarification of the stigma on pod and seed set.

#### 4.7.1 Vegetative propagation of *V. faba*.

It would have been much simpler to resolve some of the matters considered in this thesis if it had been possible to vegetatively propagate genotypes. For example the low production of seeds per plant would not have been important if the genotypes could have been propagated as clones.

In a study of vegetative propagation, several experiments were conducted on the feasibility of propagating stem cuttings of various length and from different parts of the stem. Mist propagation equipment was used for the experiments. Cuttings were kept in the propagator for 15 days. In one study 24 stems were taken from 12 plants. These were divided into cuttings of the terminal, intermediate and basal portions of the stems. Half of these cuttings served as controls and the other half were treated with Seradix.<sup>1</sup> There was no advantage attributable to Seradix. The cuttings bearing the terminal bud had the highest frequency of rooting and the number of roots (Table 50A). In another study it was found again that the terminal portion of the stem

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<sup>1</sup>Seradix 1, a commercial rooting powder for softwood cuttings containing 4-indol-3-ylbutyric acid (IBA).



TABLE 50: The effect of rooting compounds on the rooting of cuttings from different parts of the stem.

A.

Cutting	Number of cuttings rooted <sup>1</sup>		Roots per cutting		
	Control	Treated	Control	Treated	Mean
Terminal	9	7	12.7	14.2	13.4
Intermediate	6	3	2.8	1.3	2.0
Basal	3	2	1.1	0.9	1.0
5th node Single node	4	2	1.6	0.5	1.0
10th node cuttings	4	4	1.7	4.0	2.8
N.S.					

} L.S.D.  
=2.2

<sup>1</sup>The maximum possible was 12

B.

Cutting	IBA	No. of cuttings rooted <sup>2</sup>			Roots per cutting		
		0	5000	100000	0	5000	10000
Terminal		6	6	6	53.7	58.0	33.5
Intermediate		6	6	6	7.5	62.7	57.7
Basal		3	5	5	3.0	42.8	35.8
LSD between any 2 means = 28.7							

<sup>2</sup>The maximum possible was 6

had the greatest number of roots and did not benefit from the application of a rooting compound, IBA (Table 50B). The lower portions of the stem were found to benefit from treatment with IBA at 5,000 ppm in 50% ethanol but concentrations as high as 10,000 ppm were unnecessary and were no better than 5,000 ppm.

There was no difference in the ability of cuttings with 3 or 5 nodes to produce roots but the presence of the terminal meristem was necessary for good root production (Table 51).

TABLE 51: Rooting of cuttings with different number of nodes with and without terminal bud.

Cutting length	Mean number of roots <sup>1</sup>	
	With terminal bud	Without terminal bud
3 nodes	19.0	4.2
4 "	19.0	13.8
5 "	23.0	8.3
Mean	20.3	8.8
L.S.D. = 6.6		

<sup>1</sup>Maximum of 2

Single-node cuttings were capable of producing roots (Table 52). The cuttings had very little vigour and their success was low when transplanted to the field.

Although these results have shown that cuttings from various portions of the stem can produce roots the only cutting that has a good production of roots and a high rate of success is the terminal portion of vigorously growing shoots. Furthermore the cuttings without a terminal bud were unable to continue growth unless a lateral bud developed.

TABLE 52: Mean number of roots produced from single node cuttings<sup>1</sup>

Character	Position <sup>2</sup> of cutting	Number of roots		
		0	PPM 5000	IBA 10000
Number of cuttings rooted	Node 6	6	4	2
	Node 8	5	3	1
	Node 10	5	1	0
Roots per cutting	Node 6	3.17	3.67	0.33
	Node 8	3.00	2.17	0.67
	Node 10	4.50	0.33	0
	Mean	3.56	2.06	0.33

L.S.D. = 1.31

<sup>1</sup>Mean of 9 cuttings

<sup>2</sup>The node number refers to the position on the stem counting downwards from the first visible node on the shoot apex.

The development of the lateral bud was infrequent and irregular (Table 53).

TABLE 53: The number of propagules that develop to pod production.

Cuttings	Number of propagules			
	Transplanted	Produced lateral shoot	Flowered	Podded
With terminal bud	16	4	16	16
Without terminal bud	11	4	3	2

If propagules can only be successfully obtained from terminal portions of the stem no great advantage in the use of vegetative propagation can be expected with existing techniques. This would represent an insufficient multiplication of material. Further refinements of the technique should be explored as the benefit from vegetative propagation would be considerable. These refinements might involve temperature control of the rooting bed, various concentrations of several growth substances to encourage root and lateral bud development. Improvements in the handling of rooted cuttings and transplanting needs also to be investigated. This may require treatment against root pathogen or environmental "hardening" of the cuttings.

#### 4.7.2 The effect of scarification of the stigma on pod and seed set.

In 1974 the effect of scarification on the pod and seed set of 24 plants was studied. The plants were representatives of 3 populations with 8 plants of each. The plants were grown in 25cm diameter pots in the glasshouse. At the time of treatment none of the plants had set any pods from flowers and the plants appeared autosterile. Treatments were applied to the 2 lowest flowers in each pair of inflorescences. On a plant, one inflorescence was tripped and the other tripped and the stigma scarified by gently stroking with a toothpick. It was then re-pollinated with self pollen from within the keel petals. The results are given below.

Population	Pod number per plant <sup>1</sup>		Seed number per plant	
	Tripped	Scarified	Tripped	Scarified
1	0	1.25	0	3.00
2	0.62	1.12	1.38	2.38
3	0.25	1.88	0.62	6.25
Mean	0.29	1.42	0.67	3.88
± S.E.	0.11	0.32	0.28	0.96

<sup>1</sup> Maximum of 2

The effect of scarification was to cause more rupturing of the stigmatic papillae and membrane than would have resulted from tripping alone. This apparently leads to a better pollen germination and a better pod and seed set.

### 5.0.0 DISCUSSION

The first part of the discussion deals with various aspects of the autofertility found in the introduced populations, inbreds and hybrids. The following section is devoted to the effect of flower treatments on yield. Yield is an important objective in the breeding of *V. faba* and the characters which can possibly affect yield are reviewed. Some considerations are then given to the breeding alternatives that are open to a plant breeder. And finally, it is appropriate to refer to the special problems of experimentation that are encountered with *V. faba*.

#### 5.1.0 Autofertility.

##### a). Autofertility of the material studied.

A wide variation in autofertility was found within and between the 100 introduced populations. This variation was evident in (a) the number of pods produced by a plant (the level of autofertility) and (b) the number of plants in a population that produce pods (number of autofertile plants). Within a population it was possible to find a range of plants from those that did not bear a single mature pod to plants with more than 20 pods. Differences between populations were found in the number of autofertile plants. In some populations all the plants were sterile whereas in others a variable number bore pods.

The variation in autofertility of the introduced populations and the level of autofertility of the plants was possible due to the mixed nature of the populations — inbred and hybrid plants — as a result of open-pollination during the seed multiplication phase.

More uniformity was obtained by using inbred populations (lines) in the second study (Section 4.2.0). In this study nearly all the plants bore some pods but there were variation between the 20 populations in number of pods on a plant that is to say the level of autofertility.

Later studies (Sections 4.3.0 to 4.6.0) using inbred and hybrid plants showed large differences in the level of autofertility. Inbreds were generally of lower autofertility than hybrids.

b). Early and late autofertility.

It is evident that autofertility changed during the season. In the 32 populations, many more plants (an additional 12%) were fertile at the end of the season (27 weeks after sowing) than early in the season (19 weeks). The later autofertility was not due to the late production of flowers as all plants had flowered by the 15th week. It has been reported in England that *V. faba* plants with inadequate bee-pollination of early flowers set more pods on later flowers (Riedel and Wort, 1960) and Poulsen (1975) suggested that late pod set was a survival mechanism should bee-pollination be inadequate. Features of the southern Australian environment are the great variation in the length of the growing season and the frequent sharp curtailment of growing conditions in the spring or early summer. In this environment, therefore, a dependence on late pod formation would be hazardous. Plants with late autofertility in some seasons would be setting pods under very unfavourable hot dry conditions. In contrast, plants with early autofertility are able to set pods over the whole season and should have a greater stability of yield.

c). Autofertility and seed size.

In the 100 populations studied, there was no evidence that small-

seeded populations had a larger number of autofertile plants than large-seeded populations. However, the differences in the level of autofertility among populations was evident in the 20 populations studied in Experiment 2. When both the number of seeds and the number of pods were used as indices of autofertility, a negative correlation was obtained with seed weight. Similar negative relations was obtained by Yassin (1973). It is possible that such negative correlations are fortuitous and a consequence of the small number of populations used in the present and other studies. However, if the association between autofertility and seed size is real, then genes conferring these characters will be linked and selection for high autofertility based upon seed numbers will lead to small seeded varieties. In breeding for high yielding autofertile types it is possible that an optimum seed size in relation to maximum yield could be obtained.

In field beans there is no agronomic disadvantage in small-seeded forms, on the contrary such types are easier to sow and harvest without damage.

Where beans are being used for domestic consumption, small seeds may be a disadvantage.

The population that had the highest autofertility (seed number) was from India but since there were only a small number of populations studied it was not possible to draw any general conclusion on the likely geographic distinction of autofertile material.

d). Autofertility in inbreds and hybrids.

Inbreds were found to have a lower autofertility (pod and seed sets) than hybrids in all experiments. A single generation of inbreeding was adequate to cause a loss of yield and a lower autofertility.



The hybrids were highly autofertile. Hybrid vigour is a phenomenon found in many species when two dissimilar parents are crossed. However, the attempt to achieve increasing dissimilarity in the present study based on crosses between parents of the same population ( $H_1$ ), between populations of the same seed size ( $H_2$ ), and between populations of different seed size ( $H_3$ ) did not result in a consistent effect on autofertility. In one group of crosses a progressive increase in autofertility and yield was achieved but in the other group there were no differences (Section 4.6.0). The larger seed size of the widest cross ( $H_3$ ) may have influenced its yield and seed set causing the lack of improvement. Large seeded populations has been shown to be of low autofertility (Section 4.2.0).

It is also possible that the  $H_3$  hybrids may not have been more heterozygous than crosses between more similar parents ( $H_1$ ,  $H_2$ ) even though the parents differed in seed size. From the results of the present study, the only advantage in using seed parents with different seed size is the possible increase in yield through higher seed weight rather than autofertility.

Thus although it cannot be shown that hybrids from increasingly dissimilar parents have a higher autofertility it can be concluded that an advantage in hybridization of *V. faba* is an immediate improvement in autofertility.

#### 5.2.0 Effect of flower treatment.

##### a). Effect of tripping.

When the introduced populations were tripped there was an increase

in the number of plants that bore pods (Exp. 1). There was also an increase in the pods set per plant when compared to untripped flowers. Similar results were obtained using inbred populations (Exp. 2). Variation in the degree of response was found among the inbred populations (lines). Very autofertile lines showed little improvement in pod set with tripping but lines with low autofertility had a significant increase.

The inbred plants generally show a response to tripping indicating that when they are not tripped their flowers are inadequately pollinated. Hybrids have no requirement for tripping and do not respond to the treatment. The improvement from tripping was due to an increase in the number of pods and seed set but not in the number of seeds per pod.

b). Effect of cross-pollination.

Cross-pollination resulted in large improvements in pod set. There was small improvement in the number of seeds per pod; but this was only significant when compared to the untreated flowers (Exp. 1). These results on seeds per pod together with the ones in the paragraph above would indicate a limited capacity for improvement in the number of seeds per pod.

The high pod set from cross-pollination could indicate that self pollen was less effective than cross pollen in achieving fertilization. It might be suggested that this was evidence of incompatibility, however results from the scarification study (Section 4.7.2) and from other workers (Drayner, 1959; Holden and Bond, 1960; Hanna and Lawes, 1967; Toynebee-Clarke, 1974) suggested that the process of applying pollen rather than self-incompatibility was responsible for the increased

pod and seed set. It is possible that when cross-pollinating, the stigmatic papillae and membrane are more effectively ruptured than when flowers are only tripped. The ruptured stigmatic structures allow closer contact between the pollen and the stigma and provided the moisture necessary for pollen germination. The entrance of a bee into a flower and its foraging may also favour rupturing of the stigmatic surface and a higher pod set.

Rowlands (1958) proposed that post-fertilization abortion of inbred embryos could occur due to homozygosis of deleterious recessive genes. This form of self-incompatibility was not evident in the results. The proportion of fertilized ovules that abort in inbreds was the same as in hybrids.

There was an indication that a larger proportion of young pods resulting from cross-pollination reach maturity. However the effect was small.

In Experiment 1, 25% of the plants did not produce pods even when they were cross-pollinated. Such sterility was not evident in later studies and could possibly have been due to the poor adaptation of some of the introduced material.

#### 5.3.0 Characters associated with yield improvement.

In the detailed experiment (Exp. 6) assessing inbred parents and hybrids, the greater yield of the hybrids was a consequence of a greater number of seeds. Yield and seed number were highly correlated ( $r > 0.9$  \*\*\*) whereas the correlation with seed weight was only slight ( $r = 0.3$  \*\*\*). This emphasises that the greater yield of hybrids was determined largely by a greater number of seeds and only slightly by an increase in weight.

When material being assessed is comparable, and of the same generation, for example the 20 inbred populations in Experiment 2, there was no relationship between yield and seed weight.

When seeking an improvement in yield the number of racemes that bears pods is very important. The total number of racemes on a plant and the number of pods on a podded raceme have small effects on yield. The number of racemes on a plant that bears pods is only 1 in four and the number of pods on a podded raceme is usually one.

The proportion of young pods that reach maturity was very high in these studies in contrast to that reported by Kambal (1969a) in which approximately 50% of young pods aborted. In the present study the problem of pod set was more a matter of inadequate pollination than any physiological limiting factor.

#### 5.4.0 Plant vigour and yield improvement.

No inbreeding depression was evident for the vegetative and floral characters of the plants (e.g. height, stem number, raceme number). The reduction in yield following inbreeding is attributable to low fertility of the plant rather than low vegetative vigour.

It is unusual for a species with a high degree of cross-pollination to show inbreeding depression only in regard to fertility and not vegetative vigour. Most cross-pollinated species such as maize, lucerne, or pasture grasses show a great loss of vegetative vigour from inbreeding. The loss of fertility which is the only obvious effect of inbreeding in *V. faba* favours outcrossing. In this way the species has evolved an alternative breeding system to the incompatibility and loss of vigour that encourages outcrossing in other species.

Vigourous vegetative growth was shown by the indeterminate populations whether inbred or hybrid, and terminal growth continued to the end of the season. Determinate forms however, ceased terminal growth after 15 to 20 inflorescence were produced on a stem. The short growth period of the determinate type is compensated for by the large number of stems. The more restrictive podding period of the determinate type allowed pods to reach maturity by the end of the growing season but with the extended flowering of the indeterminate types, pods formed late in the season did not develop fully. Such differences could account for the higher yields of the determinate populations (Sections 4.3.2 and 4.4.1). Under southern Australian conditions, the indeterminate forms with their tall habit are also susceptible to lodging. It would appear that the determinate habit is preferable in a breeding programme.

#### 5.5.0 Breeding considerations.

##### a). The identification of genotypes with high autofertility.

The association of heterozygosity with autofertility makes it difficult to identify autofertile genotypes. A plant breeder will not know whether an autofertile genotype has an inherent high autofertility or whether it is just more heterozygous. In Experiment 6 some inbreds were found that did not have their autofertility (seed set) lowered by additional inbreeding. These inbreds had undergone two generations of controlled selfing but the original level of inbreeding of their parents was unknown. The matter may be considered theoretically. The approach to homozygosis following inbreeding is indicated by the coefficient  $F$ , devised by Wright (as quoted by Allard, 1964). It is calculated as  $F = \frac{1}{2} (1 + F')$  where  $F'$  is the inbreeding coefficient of the preceding

generation. Homozygosity can be expected to increase by 50% with each generation of inbreeding as given below:

Generations of inbreeding	F
1	0.500
2	0.750
3	0.875
4	0.938
5	0.969
6	0.984
7	0.992
8	0.996
9	0.998
10	0.999

The corresponding decrease in heterozygosity is greatest during the first 3 generations of inbreeding and changes at a very slow rate with further generations. Therefore, if inbreeding depression occurs, it should be greatest in the early generations. The inbreds for which the yields between generations did not differ significantly were considered to be more homozygous -- possibly due to a longer period of inbreeding -- and to have reached their basic level of autofertility.

Since the early generation inbreds retain some heterozygosity, selection for autofertility should be delayed until homozygosity is achieved; only then can a basic level of autofertility be identified. The bulk population method of handling heterozygous and heterogeneous material may be suitable for selecting for autofertility as it allows the material to become homozygous before selection is applied however, the bulk plots would need to be grown in a bee-proof cage to prevent any cross-

pollination.

b). Self-fertility.

The seed set of tripped flowers indicates the level of self-fertility of the plants, as pollen is then not limiting. A range of self-fertility was evident in the inbreds but hybrids generally had a high self-fertility. In Experiment 6, inbreds were found that had seed sets as high as the hybrids after tripping. High self-fertility in plants is a character of value to the plant breeder. When combined with high autofertility, lines would be produced that self-pollinate and produce good seed sets with self-pollination. The identification of self-fertile genotype is simply accomplished in a bee-proof environment. It only requires tripping a uniform sample of flowers on the plants the selection of plants with a high pod set.

c). Relation of progeny to parents.

An inherited influence on autofertility was suggested by the results of Experiment 3. In this experiment the crosses had common pollen parents (top-cross procedure) and common seed parents. Some pollen parents produced hybrids with a higher autofertility than others. The level of autofertility in the inbred progeny of these seed parent was positively correlated with the yield of the hybrids. This suggested that autofertility was genotypically determined.

However, in Experiment 6 where hybrids and their parents and inbreds were studied, there was no correlation between the yields of the parents or inbreds and that of the hybrid. It is not obvious why a correlation was found in Experiment 3 and not in Experiment 6.

Differences in the material could be a factor, Experiment 3 was undertaken on indeterminate types whereas Experiment 6 was on determinate types. Another matter of possible relevance is that the material involved in the correlation in Experiment 3 were not very dissimilar in yield whereas in Experiment 6 the inbreds had yields far below that of the hybrids. The latter situation resembles that frequently experienced in maize where no correlation is obtained between the assessment of inbreds and their hybrids.

d). Open-pollinated populations.

Most land races and cultivars of *V. faba* are open-pollinated populations. Under open-pollination seeds can set from self- or cross-pollination and the composition of a population can be expected to vary with the activity of the pollinators; honeybees in Australia. If bee activity was adequate during flowering, then a balance in the type of progenies from self- and cross-fertilization could be expected. But if bee activity was low in one year, it would result in a large proportion of inbred progenies. When these are grown as the crop the next season they can be expected to be poor in autofertility and very dependent on bee-pollination to cause pod and seed set. Thus the population balance and yield will be determined by bee activity and will vary from year to year. Open-pollinated varieties cannot be expected to be stable unless adequate bee activity is present each year.

Open-pollinated populations could be the objective in population improvement programmes. However, there must be adequate bee activity to ensure a high proportion of hybrid progenies. If bee activity is inadequate in southern Australia during the winter it may be necessary to



have specialised seed production, under irrigation, during the summer or in some other area of the country where winters are less cold.

Alternatives to open-pollinated populations are high-yielding inbred lines or  $F_1$  hybrids. Breeding for these will be discussed in the following sections.

e). Inbred lines.

There has been evidence of inbreds with high autofertility as well as high self-fertility occurring on different plants (Section 4.6.1). Some of these inbreds had high seed sets and yields of 3.5 t/ha from untreated flowers. The fact that these inbreds were not selected, and that they were grown at a density lower than commercial would suggest the further improvements are possible. Density in the experiment was 7 plant  $m^{-2}$  whereas the optimum density appears to be about 20 plants  $m^{-2}$  (Laurence, private communication).

The identification of genotypes for high autofertility and high self-fertility has already been dealt with (see (a) and (b) in this section). In southern Australia the simplest and most practical approach with *V. faba* would be the breeding of inbred lines with a high level of auto- and self-fertility. Inbred lines are easy to maintain, they do not show a depression in vegetative vigour, and the farmer could produce his own seed.

f). Hybrid seed production.

The higher autofertility of hybrids, and the depression in fertility that accompanies inbreeding, suggests that autofertility may be controlled by dominant genes. If this is true it should be possible to breed genotypes in which many of the favourable dominant genes are present

and homozygous. Some inbreds were found with high auto- and self-fertility. However, no inbred was found in the experimental material that out-yielded the hybrids when flowers were untreated. High yield is not dependent on high autofertility alone. Other unknown factors of the plant could also contribute to an improved capacity for seed production. If many genes are involved selection will be effective but slow. Improvement will occur when favourable cross-overs take place in the 6 linkage groups. Therefore, although autofertile inbreds with good yields are possible a more rapid improvement may come from the production of hybrids.

A disadvantage with hybrid varieties is the high cost of seeds and the need for the farmer to obtain  $F_1$  seeds each season. A hybrid seed programme would require specialised seed producers able to maintain the inbreds, an efficient male sterility system with restorer lines and a suitable environment. In South Australia the production of hybrid seeds commercially could encounter problems of inadequate bee-pollination.

#### 5.6.0 Problems of experimentation with *V. faba*.

With any new crop there are many problems of local adaptation that have to be resolved. These problems can be investigated more readily if uniform plant material is available. For instance, when investigating the effects of flower treatments it would have been of great benefit if it had been possible to vegetatively propagate the genotypes. However the number of propagules that could have been successfully propagated from one plant was limited and these were not as vigorous as the original plants. The propagules generally did not produce any lateral stems (Section 4.7.1). Vegetative propagation of excised stem apex in

nutrient media was successfully carried out by Aubry *et al.* (1975) but this method is too slow for current purposes.

In order to maintain some uniformity in the material studied, inbreds and various hybrid progenies were obtained from common seed parents. However, the low seed production of the parents resulted in inadequate seeds of the various progenies from a plant. The large-seeded populations were particularly difficult in this respect. Many plants bore a maximum to 10 to 15 seeds. This represents a very low number when it was desirable to use the seeds in experiments involving several treatments or when making quantitative breeding tests such as topcross or diallel analyses.

Additional features of *V. faba* which complicated experimentation included the great variability present within populations, the intermediate nature of the breeding system with high frequencies of self- and cross-pollination and the uncertainties of year to year bee activity. It is the combination of all these features which made the breeding of *V. faba* a special and challenging task and very different from wheat, maize, sorghum, lucerne or soybeans where well researched breeding programmes have been formulated.

Clearly *V. faba* has a high yield potential in this environment. Its realisation depends on the appropriate breeding programme being undertaken.

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APPENDIX

TABLE 1 *Vicia faba* introductions used in Experiment 1 (1974)

No.	Present Waite code	Previous Waite number	Origin	100 seed weight (g)	Seed Shape	Seed Colour
1	139	CPI 60606	Tur	139.1	Flat	Buff
2	110	CPI 19880	Ind	62.2	Oval	Brown
3	090	CPI 22881	Cze	52.8	Oval	Brown
4	133	CPI 60598	Tur	178.1	Flat	Buff
5	151	CPI 22612	Ita	50.0	Oval	Brown
6	086	CPI 19878	Ind	53.8	Oval	Brown
7	131	WI 5345	Swe	40.8	Oval	Black
8	067	CPI 22613	Ita	55.3	Oval	Brown
9	066	CPI 22894	Cze	58.6	Oval	Brown
10	125	WI 5350	Tur	181.2	Flat	Buff
11	149	CPI 60583	Tur	124.6	Flat	Buff
12	051	WI 5378	Cre	100.0	Flat	Buff
13	147	CPI 60588	Tur	126.4	Flat	Buff
14	138	CPI 22878	Cze	82.3	Oval	Brown
15	107	CPI 19881	Ind	39.8	Oval	Brown
16	082	WI 5342	Cre	121.8	Flat	Buff
17	088	WI 5358	Sic	119.7	Flat	Buff
18	068	WI 5371	Cre	99.7	Flat	Buff
19	056	WI 5365	Cre	152.7	Flat	Buff
20	141	CPI 60599	Tur	172.6	Flat	Buff
21	094	CPI 60572	Tur	138.8	Flat	Buff
22	077	WI 5349	Gre	193.3	Flat	Buff
23	103	CPI 60601	Tur	158.2	Flat	Buff
24	092	CPI 60622	Tur	178.8	Flat	Buff
25	091	CPI 60603	Tur	155.8	Flat	Buff
26	288	JHS 18	Cre	163.8	Flat	Buff
27	031	CPI 22892	Cze	64.6	Oval	Brown

Present Waite code: numbers are preceded by 100 e.g. 139 = 100139.

Origin: The countries are represented by the initial 3 letters, e.g. Tur = Turkey.

No.	Present Waite code	Previous Waite number	Origin	100 seed weight (g)	Seed Shape	Seed Colour
28	089	CPI 60584	Tur	154.6	Flat	Buff
29	035	WI 5340	Cre	65.6	Oval	Brown
30	037	WI 5357	Ita	152.4	Flat	Buff
31	034	WI 5377	Gre	127.6	Flat	Buff
32	065	CPI 60587	Tur	140.3	Flat	Buff
33	070	CPI 60589	Tur	140.9	Flat	Buff
34	039	WI 5375	Cre	134.2	Flat	Buff
35	038	WI 5344	Swe	48.9	Oval	Brown
36	027	CPI 60608	Tur	149.4	Flat	Buff
37	004	WI 5370	Tur	148.5	Flat	Buff
38	286	JHS 16	Cre	197.0	Flat	Buff
39	287	JHS 17	Cre	126.4	Flat	Buff
40	026	CPI 60612	Tur	179.4	Flat	Buff
41	111	WI 5355	Yug	152.1	Flat	Buff
42	100	CPI 22885	Cze	60.5	Spherical	Brown
43	014	CPI 60619	Tur	153.3	Flat	Buff
44	075	WI 5376	Cre	154.9	Flat	Buff
45	120	CPI 22898	Cze	69.5	Oval	Brown
46	015	CPI 60580	Tur	186.4	Flat	Buff
47	061	WI 5374	Swe	97.2	Flat	Brown
48	063	CPI 22882	Cze	78.4	Oval	Brown
49	146	CPI 22890	Cze	47.8	Oval	Brown
50	029	CPI 60607	Tur	150.3	Flat	Buff
51	028	CPI 60585	Tur	177.0	Flat	Buff
52	020	CPI 60609	Tur	79.4	Oval	Brown
53	095	CPI 19879	Ind	75.0	Oval	Brown
54	123	WI 5351	Cre	183.3	Flat	Buff
55	041	WI 5353	Cre	62.4	Oval	Brown
56	043	CPI 22615	Ita	53.2	Oval	Brown
57	080	CPI 60586	Tur	198.8	Flat	Buff
58	087	WI 5362	Ita	181.2	Flat	Buff
59	113	WI 5372	Cre	174.2	Flat	Buff

No.	Present Waite code	Previous Waite number	Origin	100 seed weight (g)	Seed Shape	Seed Colour
60	081	CPI 22876	Cze	61.8	Oval	Buff
61	074	CPI 22893	Cze	60.0	Oval	Buff
62	134	WI 5348	Tur	49.5	Oval	Black
63	136	WI 5364	Cre	167.6	Flat	Buff
64	148	WI 5343	Cre	197.9	Flat	Buff
65	112	CPI 60579	Tur	149.4	Flat	Buff
66	443	NEB 194	Tur	94.2	Flat	Buff
67	305	NEB 17	Syr	117.9	Flat	Buff
68	403	NEB 154	Fra	115.2	Flat	Green
69	441	NEB 192	Tur	154.5	Flat	Green
70	411	NEB 162	Eng	111.3	Flat	Green
71	396	NEB 147	USSR	60.3	Oval	Buff
72	389	NEB 140	Mor	77.5	Oval	Buff
73	565	NEB 338	Egy	60.9	Oval	Buff
74	326	NEB 38	Ira	142.2	Flat	Buff
75	347	NEB 59	Eng	134.6	Flat	Buff
76	405	NEB 156	Fra	139.4	Flat	Violet
77	371	NEB 109	USA	96.6	Flat	Buff
78	464	NEB 218	Tur	82.7	Oval	Buff
79	448	NEB 199	Tur	80.3	Oval	Brown
80	478	NEB 232	Tur	102.1	Flat	Brown
81	601	NEB 374	Eng	49.7	Oval	Buff
82	488	NEB 245	Tur	52.9	Oval	Buff
83	442	NEB 193	Tur	151.6	Flat	Buff
84	383	NEB 133	USA	51.4	Oval	Buff
85	299	NEB 14	Syr	118.2	Flat	Buff
86	345	NEB 57	Eng	38.1	Oval	Buff
87	440	NEB 191	Tur	110.3	Flat	Buff
88	561	NEB 333	Egy	64.4	Oval	Brown
89	521	NEB 287	Leb	56.2	Oval	Buff
90	444	NEB 195	Tur	127.3	Flat	Buff

No.	Present Waite code	Previous Waite number	Origin	100 seed weight (g)	Seed Shape	Seed Colour
91	406	NEB 157	Fra	126.0	Flat	Violet
92	382	NEB 132	USA	66.4	Oval	Buff
93	391	NEB 142	Mor	62.6	Oval	Buff
94	461	NEB 213	Tur	105.8	Flat	Buff
95	373	NEB 113	USA	50.0	Oval	Buff
96	452	NEB 204	Tur	88.2	Flat	Buff
97	524	NEB 290	Egy	87.3	Flat	Buff
98	608	NEB 381	Eng	42.2	Spherical	Brown
99	601	NEB 376	Eng	46.8	Spherical	Brown
100	522	NEB 288	Egy	74.7	Flat	Green

TABLE 2 : Sowing and harvesting dates

Experiment	Year	Sown	Harvested
1	1974	May 2	November 18 to 29
2	1976	May 19	November 26 to December 2
3	1975	May 6	November 21 to 26
4	1975	May 6	November 17 to 19
5	1976	May 19	November 23 to 25
6	1976	May 18	November 15 to 22