The University of Adelaide
Faculty of Agricultural Science

FREQUENCY - DEPENDENT ADVANTAGE IN WHEAT

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Thesis submitted for the degree of Doctor of Philosophy
in
Plant Breeding

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August, 1976
Awarded February 1977

DEDICATION

To my father, Tran Van Lan, for his inspiration, understanding and encouragement.

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

A series of experiments was conducted in 1972, 1973 and 1974 in the field to investigate the effect of frequency-dependent advantage in wheat and its influence on the efficiency of single plant selection.

Frequency-dependent advantage (f.d.a.) is the term used to describe the phenomenon where a genotype growing at a low frequency in a mixture has a higher fitness or yield than when it is grown at a higher frequency in a mixture of the same components.

1. The occurrence and magnitude of frequency-dependent advantage.

F_1 hybrid seeds of the crosses (Warimek * Halberd) and (Wariquam * Halberd) were grown in a stand of Halberd at 5 frequencies: 4%, 6.25%, 11.11%, 25% and 50%. A reduction of 35% to 40% in grain yield of individual plants was observed with both hybrids as their frequencies changed from 4% to 50%. A similar trend with frequency was noted for total grain number and several other plant characteristics, particularly those measured towards the end of the growing season. Halberd plants did not show a corresponding increase in grain yield as their frequency declined from 96% to 50%.

The following season, 76 F_4 lines from the cross (Warimek * Halberd) and 70 F_4 lines from (Wariquam * Gabo) were grown at frequencies of 6.25% and 18.75% in machine sown stands of Wariquam and Halberd, respectively. Again, with almost all these lines, a decrease in grain yield with increased frequency of the genotype was observed. Furthermore, there was a positive correlation between f.d.a. and relative grain yield, suggesting that genotypes with a high mean yield at low frequencies show a greater f.d.a. than lower yielding ones.

It was concluded that f.d.a. was likely to be widespread in wheat.

2. Mechanisms of frequency-dependent advantage.

The relationship between f.d.a. and competitive ability in wheat was investigated by growing F_1 hybrids and their parents in paired combinations at 2 frequencies 6.25% and 25%. The results indicated that a strong competitor had an increased yield at a low frequency when grown in a stand of a weak competitor (thus exhibiting f.d.a.), however, the weak competitor was not significantly reduced when it was grown at low frequency. One hybrid, (Warimek * Timgalen), did not show f.d.a. in either parent, indicating that heterozygosity did not always lead to f.d.a.. The strong competitor, Timgalen, also showed f.d.a. in a mixture of other varieties, suggesting that the mechamism which resulted in f.d.a. in this genotype was specific to this genotype.

The physical basis of f.d.a. was studied in an experiment designed to enable assessment of relative competition for aerial and soil factors. The results indicated that f.d.a. was mainly involved with soil factors. Furthermore, there was a positive correlation between grain yield per plant and distance between the test plants suggesting that f.d.a. was probably due to the interaction between roots of the same genotype in the exploitation of the environment in a slightly different way to that by other genotypes.

3. The effect of frequency-dependent advantage on single plant selection.

(i) in F_2 populations: The distributions of F_2 plants of the crosses (Warimek * Halberd) and (Wariquam * Halberd) were compared with those of the F_1's and the parents, Warimek, Halberd and Wariquam. It was expected that the F_2's should show a larger variance and a more skewed distribution than the F_1's and parental populations as a result of genetic segregation and f.d.a.. Surprisingly, the results indicated that the variances of grain yield per plant were not different between F_1, F_2 and parental populations.

(ii) in mixtures of genoytpes: The relationship between the yield of F_5 single plants from 25 F_2 derived lines of the cross (Warimek * Halberd) grown in replicated mixtures and the yield of the same lines grown in pure stands was compared. In the same experiment, each line was also grown at 2 frequencies, 4% and 16%, in a mixture of the parents, Warimek and Halberd, to compare the frequency responses of these lines with their performance in mixtures and in pure stands.

It was found that the yields of the 25 lines were significantly higher at the 4% than at the 16% frequency, thus they exhibited f.d.a.. The correlation between yield in mixture and yield in pure stand varied from one replicate to another, but when yields of the 25 lines in mixtures were based on the mean yields over all the replicates, the correlation between yields in mixtures and yields in pure stands was positive and highly significant (r = 0.749***). Conversely, the correlation between the f.d.a. of a line and its performance in a pure stand was low (r = 0.001).

In the study reported here, three important features were apparent, concerning single plant selection for yield:

(a) the effect of f.d.a. was small in a heterogeneous population and was unlikely to influence single plant selection.

(b) micro-environmental variation was the main factor influencing the phenotypic expression of single plants,

(c) there was a positive correlation between yield in mixture and yield in pure stand. This correlation was obtained when:

- the mixtures and pure stands were grown at the same density and planting method. Thus, when there were no genotype-density and genotype-environment interactions between the two treatments.
- the yield in mixture of each genotype was based on the mean of a number of plants in replicated plots, and the effect of micro-environment was thereby minimized.

Thus when the effect of micro-environment could be reduced, it should be possible to select for high yielding genotypes, based on the yield of single plants in mixtures.

The effect of f.d.a. in population dynamics was studied by computer simulation, using the model of mixed selfing and outcrossing with selection developed by Hayman (1953). It was shown that a combination of f.d.a. and natural crossing could maintain genotypic frequencies in a stable equilibrium.

This study is applicable to 4 aspects of plant breeding as it;

(i) explains the performance of individual plants which result in the maintenance of heterogeneity in Composite Crosses.

(ii) suggests that multiline varieties will not need frequent re-constitution.

(iii) emphasizes the likely problems with infrequent rogues (off-type genotypes) in a variety during seed multiplication.

(iv) may have a role in reducing the efficiency of single plant selection although the results in section (3) indicated that it was probably the micro-environment which has the largest influence on the phenotypic expression of single plants in a segregating population.

ACKNOWLEDGEMENTS

I wish to express my sincere gratitude to my supervisors, Dr. A.J. Rathjen and Dr. R. Knight, for their advice, guidance and criticism throughout this study.

I would also like to thank Mrs. G. Bishop and Miss S. Chambers for advice in the statistical and computing fields.

Many members of the Agronomy Department assisted with the field work and I appreciate, in particular, the help of Mr. J. Chigwidden and Mr. J. Lewis. Thanks are also due to Mr. B. Palk for the photography and to Mrs. J.B. Schilling for typing the thesis.

Finally, I am grateful to the Commonwealth Government of Australia for financial support through a Colombo Plan Postgraduate Scholarship.

CONTENTS

I. INTRODUCTION

Often, the purpose of a plant breeding program is to select a genotype with a higher yield than that of existing varieties. In the pedigree method of selection, the first step is a careful choice of parental combinations likely to give rise to high-yielding progeny. Following the initial hybridization, superior individuals are selected in successive generations. Finally homozygous genotypes may be released as new varieties.

For self-pollinated crops, it is essential to identify genotypes which are potentially high yielding in pure stands in the early generations, preferably at the F_2, for if they are lost, they cannot be recovered in the subsequent generations. Effective selection in the early generations would also enable the breeder to screen large numbers of genotypes with the minimum of time, cost, labour and space.

Since vegetative propagation is not practicable with self-pollinated cereals such as wheat and barley, selection in an F_2 population is essentially on a single plant basis. Individuals are often selected on their morphological characteristics, however the phenotype of a plant may be strongly influenced by the environment in which it is grown.

This thesis is concerned with one of the factors affecting the phenotypic expression of plants when they are grown as a component in a mixture of a number of dissimilar genotypes, namely frequency-dependent advantage. Frequency-dependent advantage is the term used when a genotype at a low frequency in a population has a higher fitness or yield than it has when present at a high frequency.

Plant breeders recognize other factors that influence the outcome of selection. These arise when a single F_2 plant is used as a predictor for the performance of the same genotype or related genotypes when these are grown over the whole range of conditions in which the crop is cultivated. These include:

1. The relative performance of genotypes in relation to their physical environment:

(i) Interactions between genotypes at different plant densities: Single plants are often grown at low densities to facilitate inspection and to produce more seed for the next generation. However, the appearance and performance of a plant at low density depends on its ability to utilize the environmental resources in the absence of neighbours. Plants may be selected which possess characters with a high expression at a low density (e.g. tall, leafy and high tillering capacity), but these may not be useful when the same genotype is grown as a crop.

(ii) Genotype - environment interactions over sites and seasons: The relative performance of genotypes changes with climate and soil conditions. As these conditions vary with seasons and sites, genotypes selected at a particular site in one season may not perform well at other sites or in other seasons.

(iii) Micro-environmental variation: The term micro-environment used here refers to edaphic variations within a plot. These effects arise as a result of local variation in soil fertility, soil pathogens, topography and water supply. In a population such as an F_2, each genotype is grown in a particular micro-environment. Since the micro-environment differs from plant to plant, this variation may be sufficient to mask the genetic differences between genotypes.

2. Genetic changes:

Heterozygosity may result in hybrid vigour but this higher performance is not an advantage as the cultivars which are eventually produced are homozygous. Heterozygosity may also lower the efficiency of selection since a heterozygous plant will segregate, producing genetically variable progeny.

3. Inter-genotypic competition:

At commercial densities, the environmental resources available to individual plants are limited. In a segregating population, genotypes which can compete successfully for these resources may be high yielding. This advantage may be lost when these genotypes are grown in pure stands.

More specifically, and of particular concern in this study, is the possibility that the performance of a genotype in a mixture is influenced by the frequency of like or similar genotypes in the population. Harding, Allard and Smeltzer (1966) found that, in populations of lima bean (*Phaseolus lunatus*), the frequencies of the heterozygotes at equilibrium in a population were higher than would be expected on the basis of the observed rate of outcrossing, and their fitness increased as they became rare. They ascribed this to frequency-dependent selection.

If rarity leads to enhanced fitness, selection for yield in a segregating population may favour rare genotypes at the expense of common ones. When these genotypes are grown in pure stands - according to this theory - the advantage they enjoyed no longer exists. The selection would then be less effective with many other high yielding genotypes being overlooked. This phenomenon is, in fact, an aspect of inter-plant competition, but one that occurs for genotypes which form only a low proportion of a mixture.

This thesis reports a series of investigations on frequency-dependent advantage in wheat and its influence on the reliability of single plant selection in a segregating population.

II. LITERATURE REVIEW.

1. Introduction:

This literature review consists of two main parts. In the first, the problems associated with single plant selection are discussed, with a particular emphasis on inter-plant competition and frequency-dependent selection. In the second part, experimental studies dealing with frequency-dependent selection as a phenomenon in population genetics are reviewed. These illustrate how frequency-dependent selection acts to maintain polymorphisms in natural populations.

2. Single plant yield as a selection criterion:

A breeder of a self-pollinated crop is constantly confronted with the problem of identifying those individuals which possess the greatest number of the genes contributing to high grain yield. Much attention has been given to deciding whether selection should be initiated in early generations with the retention of only the most promising individuals, or whether a large number of the segregating lines should be kept until testing can be performed on homozygous material. Although it has been stressed that as many as possible of the F_2 should be retained (Allard, 1960; Bell, 1963), most plant breeders are severely constrained by limitations in the availability of land and labour so that the proportion of the F_2 that is retained depends on the number of F_3 families that can be conveniently handled. Often, only about 10% of the F_2 is kept for further testing (Allard, 1960). Harrington (1952), for example, indicated that from 5% to 20% of the F_2 population should be retested. Usually the selection intensity is higher for wide crosses than for crosses between closely related parents.

Effective selection for yield in early generations would be of obvious benefit to the plant breeder since it would enable him to screen large numbers of genotypes with the minimum of expenditure of his resources. Also, Shebeski (1967) indicated the genetic limitations that need to be taken into consideration. He showed that the frequency of plants containing a set of desirable alleles decreased with each generation of selfing. For example, in a cross that differs by 25 genes for yield - for wheat, this is an average of just over 1 gene per chromosome pair - only 0.075% of the F_2, or 1 plant in 1,330 may be expected to contain all 25 of the desired alleles. If selection is delayed until the F_4, only 1 plant in approximately 1.8 million would contain all the more desirable alleles in either the homozygous or heterozygous condition. This frequency is further reduced to 1 in 15.5 million in the F_6. It was suggested by Shebeski that, for reasons of maximum efficiency, selection should commence in the F_2 since the frequency of the desired genotypes was the highest in that generation.

For self-pollinated crops such as wheat and barley, where clonal propagation is not practicable, selection in the F_2 is based on single plants. Many authors, however, consider that selection for high yield in the F_2 is not effective (Allard, 1960; Bell, 1963; Elliot, 1958; Harlan, 1949; Harrington, 1932; Hinson and Hanson, 1962; Johnson and Bernard, 1963). Allard (1960) stated that the environmental effect on the yield of a single plant was so large that effective selection was virtually impossible. This general contention is based both on experimental evidence and theoretical considerations of the problems which confound single plant selection. These will now be discussed in more detail.

3. The interaction between genotypes and plant densities:

In a review of competition among crops and pasture plants Donald (1963) observed that plants show extreme plasticity, responding remarkably in size and form to environmental conditions. The yield of a plant at high density is often reduced to 20%-10% of the yield of like plants growing in isolation. In the early generations, single plants are often grown at low density to facilitate observation and note taking and produce more seed for the next generation. When superior genotypes are selected and released as new varieties - or indeed, during the testing stages in latter generations - they are grown at commercial densities which may be 15 to 20 times higher than the low density. When the ranking of the performance of genotypes is not consistent over densities, then single plant selection at low densities has favoured genotypes which possess characters with a high expression at a low density (e.g. tall, leafy and high tillering capacity) at the expense of genotypes which are high yielding at commercial densities.

Interactions between genotypes and plant densities may occur as a result of differential responses of genotypes to the availability of environmental factors affecting growth at different densities (Knight, 1960). He suggested that if the limiting factor was of equal importance to the plants at all densities, such as temperature, then there would be a good correlation between the performance of a genotype at low and high densities. However, if the limiting factor was not equally available to the plants at different densities, such as light, moisture or nutrients, then interactions might occur and the growth of a plant at a low density might only reflect its ability to exploit environmental resources in the absence of competition from its neighbours.

Experimental evidence indicates that large genotype-density interactions occur: Blum (1970), Gardner (1972) and Sakai (1965) with barley; Genter and Camper (1973) and Giesbrecht (1969) with maize; Marani _et al_.(1974) with cotton; and Hinson and Hanson (1962) with soybean.

McGinnis and Shebeski (1968) selected F_2 single plants grown at a low density (approximately 4.9 plants/m^2) and tested the F_3 selected lines at a commercial density (148 plants/m^2) and found that the selected lines had a mean yield equal to that of a set of random selections. However, this finding may not be conclusive. There could have been an interaction arising from selecting the genotypes in one season and testing them in another which confounded their results.

To avoid the effect of genotype-density interaction, it seems desirable to carry out selection at densities that approximate to those of commercial crops.

4. _The interaction between genotypes and environments_:

The change of climate and soil fertility over seasons and sites can have a large influence on the yield of a particular genotype. A selection experiment carried out in one environment may have a limited use for other environments (Horner and Frey, 1957; McGuire and McNeal, 1974; Miller _et al._, 1958; Rasmusson and Lambert, 1961). Allard and Bradshaw (1964) emphasised the importance of interactions, particularly those between varieties and years in reducing the efficiency of selection.

Any conclusion drawn from a study that involves selection in one environment, whether it be a particular year, plant density or soil type, and testing in another environment must always be suspect. The absence of a correlation across environments may not be a result of inefficient selection in the original environment.

The effect of different environments in reducing the efficiency of single plant selection was present in McGinnis and Shebeski's (1968) and Hamblin and Donald's (1974) experiments. McGinnis and Shebeski not only selected and tested their materials at different densities, but these experiments were carried out in different years (1966 and 1967, respectively). Hamblin and Donald selected F_3 single plants in 1967 and tested the F_5 lines in 1969; two entirely different years, climatically. They concluded that single plant selection, on the basis of yield, was ineffective although it was acknowledged that the genotype-environment interaction could have been very marked between the two years.

5. _The effect of micro-environmental variation_:

Since selection in an F_2 population is based on single plants which cannot be replicated, the validity of such selection may be confounded by micro-environmental variation. Genotypes may respond differently to the various micro-environments within a selection block. Frequently, there are small areas within a selection block that are more favourable for plant growth than others. If this variation is not recognised and taken into account, many plants in a good area will be selected and the plants in a poor area discarded, although some of these latter may be genetically superior to those retained from the good area.

Micro-environmental variation was large in Hamblin's (1971) experiments and Johnston (1972) considered it to be the major factor limiting the efficiency of single plant selection in barley. Both authors used several methods to reduce the effect of this variation. Hamblin (1971), for example, used response surfaces and moving averages. In the former method, a series of polynomial equations was fitted to the yield of individual plants according to their position in the field. This was an attempt to estimate a "smoothed" response surface indicating micro-environmental variation. The yields of individual plants were then measured as the deviations of their actual yields from that predicted by the response surface. However, this method was of limited usefulness as there is no biological principle on which to decide the number of terms to include in the polynomial used in the estimation of the surface. Convolutions in the surface were few or abundant depending on the number of these polynomial terms. In the moving average method, the yield of each plant was expressed as a deviation from the mean yield of a number of neighbouring plants. Hamblin, nevertheless, was unable to show that either of these techniques improved the efficiency of selection in his experiment, probably because of the large environmental difference between his year of selection and the year in which the selections were tested.

Fasoulas (1973) proposed the "honeycomb" method of planting to reduce the effect of micro-environmental variation between single plants. Individual plants in a selection block were grown at the centre and corners of a regular hexagonal arrangement. A given genotype was selected if it had a higher yield than its six immediate neighbours. However, since the distance between plants used in this arrangement was 50cm, any advantage through the reduction in soil heterogeneity is likely to have been small. Also, genotypes selected at a low density may not be high yielding when they are grown at a high density as a crop.

Johnston (1972) undertook a study in which it was possible to account for micro-environmental variation. He studied mixtures of barley varieties and was able to grow replicates of individual genotypes. The same varieties were also grown in pure stands to compare yield in mixture and yield in pure stand. He used a method of adjusting for environmental variation between plots which could be applied to allow for micro-environmental variation between plants. A grid of a check variety, Clipper, was grown in every fourth plot in the experimental area. Expected values for individual plots were calculated from the values measured from the nearby check-plots, using a weighting based on the distances between the check-plots and linear interpolation between the plots. Yields of the test material were then expressed as the deviations from the expected values. Johnston was able to show that this method reduced error mean squares and increased the variance ratio for the treatment effects.

6. The effect of heterozygosity:

In a segregating population following an initial hybridization, many plants are heterozygous. Often, a heterozygous plant is phenotypically indistinguishable from a homozygous plant containing the same number of desirable alleles. A heterozygous plant will segregate making re-selection necessary. Both the difficulty in identifying homozygotes and the re-selection lower the efficiency of selection. Furthermore, if heterozygosity results in hybrid vigour, selection of vigorous plants will then result in selection for heterozygosity. As both the level of heterozygosity and hybrid vigour decrease with each generation of inbreeding, selection in these circumstances will be inefficient for the production of homozygous cultivars.

Hybrid vigour has been reported in many crops: Briggle (1963), Briggle et al.(1967 a & b) with wheat; Clay and Allard (1969), Severson and Rasmusson (1968), Suneson (1962) with barley; Hathcock and McDaniel (1973) with oats; and Murayama (1973) and Saini et al. (1974) with rice. The degree of heterosis varied but, in most cases, the hybrid was higher yielding than the mid-parent and in some cases, it surpassed the best parent by as much as 120% (Saini et al., 1974). Results have indicated that the level of hybrid vigour may change with cultural practices. Severson and Rasmusson (1968) found that barley hybrids were only slightly better (3.2%) than the mid-parent at commercial density, but were much better (22.5%) at very low density.

Briggle et al. (1967 a & b) showed that hybrids were consistently better than the parents at all 5 densities they studied. Furthermore, they showed that the yield of the F_1 was higher than that of the F_2 which, in turn, was higher than the F_3. Thus, it can be seen that yield decreased with decreasing heterozygosity. The average heterozygosity in the F_2 is one-half of that in the F_1, but the F_2 individuals vary.

Some of them are as heterozygous as the F_1 and others as little as the parents. Selection in the F_2 may be ineffective if high yielding plants are heterozygous. Similar results were obtained by Bhatt and Derera (1973a) who found that the yield of F_2 derived lines of four wheat crosses was higher than that of F_3 derived lines; and the yield of F_3 derived lines from three other crosses was higher than that of the corresponding F_4 derived lines.

From the theory of inbreeding, it is expected that the number of heterozygotes will be reduced by a half after each generation of inbreeding. The following diagram depicts the expected genotypic frequencies at the locus containing the alleles A_1A_2 in successive generations:

		Genotype	
Generation	A_1A_1	A_1A_2	A_2A_2
F_2	$\frac{1}{4}$	$\frac{1}{2}$	$\frac{1}{4}$
F_3	$\frac{1}{4} + \frac{1}{8}$	$\frac{1}{4}$	$\frac{1}{8} + \frac{1}{4}$
F_4	$\frac{1}{4} + \frac{1}{8} + \frac{1}{16}$	$\frac{1}{8}$	$\frac{1}{16} + \frac{1}{8} + \frac{1}{4}$
F_t	$\longrightarrow \frac{1}{2}$	$\longrightarrow 0$	$\longrightarrow \frac{1}{2}$

However, Allard and Hansche (1965), Allard and Workman (1963), Harding _et al_. (1966), Imam and Allard (1965), Jain and Allard (1960) used census data of single genes to show that, with populations of lima beans, barleys and oats, the proportion of heterozygotes decreased as rapidly as expected in the early generations, but the rate of decline in later generations was slower than expected, and eventually the proportion of heterozygotes in the population reached an equilibrium. In all cases, the heterozygotes had a higher fitness and therefore a selective advantage over the homozygotes and mechanisms, such as frequency-dependent selection, have been advanced to explain this observation. Thus, even when single plant selection is delayed to the F_5 or F_6, this high yield of the heterozygotes may favour them in selection, but the advantage due to hybrid vigour (or frequency-dependent selection) may be lost when they are grown as homozygous cultivars.

From the literature, it is not possible to conclude what effect hererozygosity *per se* will have on the efficiency of single plant selection. On the one hand, the confusion of heterozygotes and homozygotes makes re-selection necessary, on the other, retention of the heterozygotes means that the re-selection may find genotypes superior to the original selections.

7. The effect of competition:

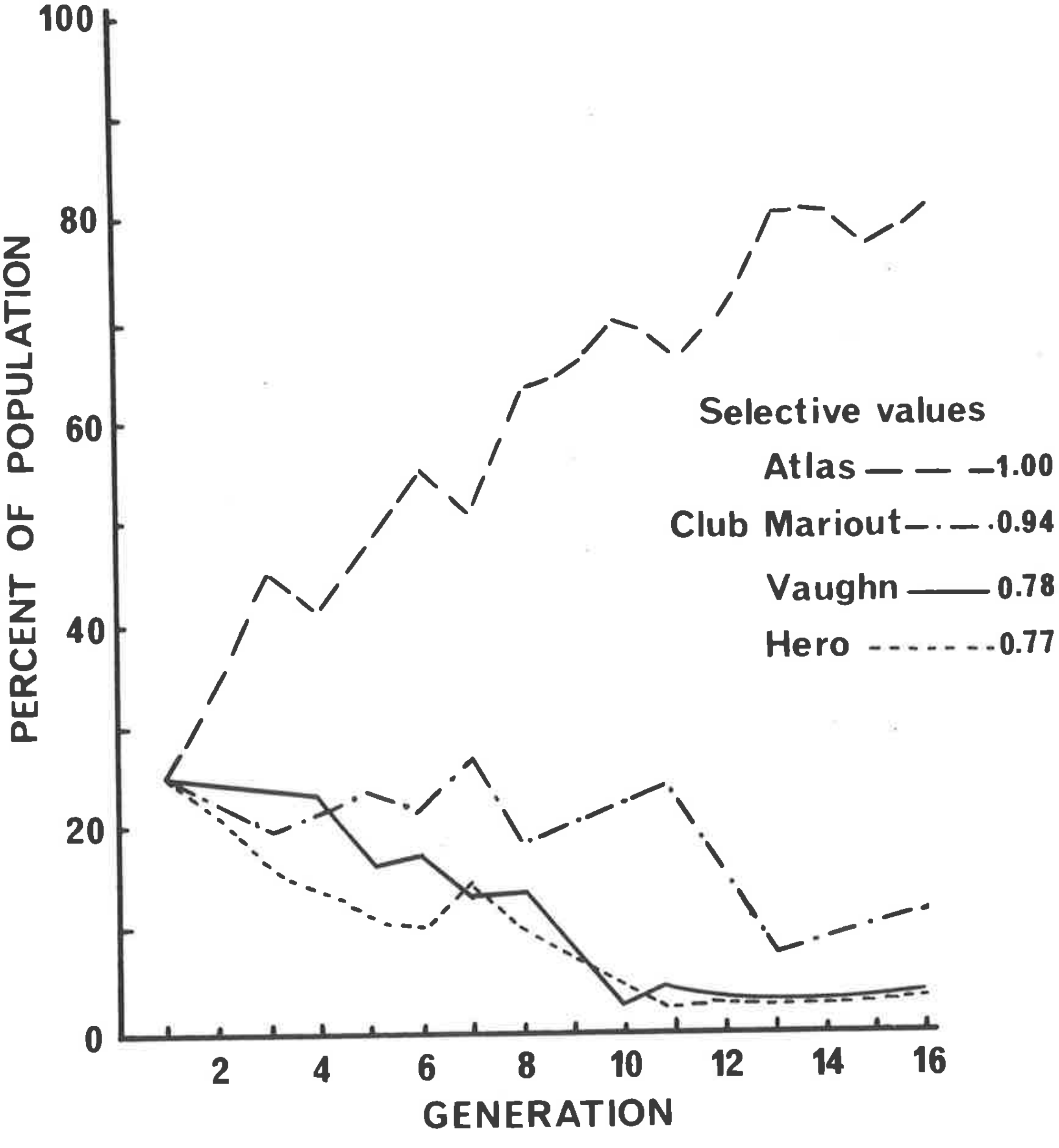
Competition between individuals in a heterogeneous population may be a factor affecting single plant selection. Unless the high yielding individuals in a mixed population are also high yielding in pure stand, single plant selection in the F_2 may favour highly competitive genotypes rather than those with a genetic capability for high yield.

Experiments that demonstrate the effects of competition have been undertaken with mixtures of pure lines (Allard *et al*., 1966; Suneson, 1949). They have shown that one or a few pure lines rapidly became predominant while other pure lines were drastically reduced in proportion.

A typical result is illustrated in Figure 1. In this experiment, all of the four competing genotypes were vigorous and well adapted commercial genotypes (Suneson and Wiebe, 1942). The highest yielding genotype in pure stands over 8 years was Vaughn, then Atlas, Hero and Club Mariout in that order. Nevertheless, after being grown together for 15 years, Atlas made up about 88% of the population whereas Club Mariout, Hero and Vaughn were 10.5%, 0.7% and 0.4%, respectively (Suneson, 1949). Thus, the highest yielding genotype, Vaughn, was the poorest competitor in mixture. Furthermore, Workman and Allard (1964), subjecting Suneson's data to further analyses, showed that the selective values of the poorest competitors, Vaughn and Hero, were only 0.78 and 0.77 respectively that of the best competitor, Atlas (selective value = 1.00). Much greater differences in the fitness values were observed in other experiments. In certain cases, genotypes which were not obviously defective left only about 5% to 10% as many progeny as the best competitor (Allard, Jain and Workman, 1968; Allard, 1965). It is obvious that competition within these self-pollinated species is very great and on the basis of these studies, mixtures involving local varieties might be expected to be reduced, after several years, to one or a few highly competitive genotypes.

There are other results available however, which show that genotypes which survive in a mixed population may also be the highest yielding in pure stand (Allard and Jain, 1962; Harlan and Martini, 1938; Suneson and Stevens, 1953). In one of the most extensive studies of "natural" selection in an agricultural crop, Harlan and Martini (1938) grew a mixture of 11 barley varieties at 10 sites in northern and western United States for between 4 and 12 years without any deliberate selection. After as few as 4 years, one or two varieties became dominant at most

Figure 1: Effect of natural selection on the frequencies of four barley genotypes in a mixture. (After Suneson, 1949).


100
80
60
40
20
0
PERCENT OF POPULATION
2
4
6
8
10
12
14
16
GENERATION
Selective values
Atlas — — —1.00
Club Mariout—·—·0.94
Vaughn ——— 0.78
Hero - - - - - - -0.77

locations. At many sites, the dominant variety was also the local commercial strain. At only 2 locations did the dominant variety in the mixture differ from that commonly grown in that locality by farmers. And in these cases, farmers' preference depended on considerations other than the yielding ability of the varieties. From these results, the following deductions may be made:

(i) Competition among the varieties for the essential factors in the environment must have been intense.

(ii) Competitive ability is related to the environmental conditions under which the population is grown. Different environments at different locations favoured different genotypes.

(iii) The genotype which dominated the mixture was often the local commercial variety.

High yielding lines which became more frequent in the population with advancing generation, can be obtained with composite crosses. Composite crosses refer to hybrid bulk populations formed from many parental varieties grown under "natural" selection - apparently indefinitely. For example, Composite Cross II was a mixture of 378 hybrids from 28 varieties (Suneson and Stevens, 1953). These authors found that the yield of Composite Cross II was inferior to Atlas, the best commercial variety, in the early generations. But after 25 years of selfing without artificial selection, its yield exceeded that of Atlas by 35%. However, it is to be noted that the yield of Atlas decreased over the years and this might have contributed to this apparent improvement. Further evidence in support of the theory that survival in population is related to high yield in pure stands came from the isolation of high yielding genotypes in advanced generations of composite crosses (Suneson, 1956; Lohani, 1976). For example, the top three selections in the F_{24} generation of Composite Cross II exceeded Atlas in yield by an average of 56% in 4 years of replicated trials (Suneson, 1956), whereas none of the isolates made in the F_{12} generation outyielded Atlas.

The differences between competition within mixtures of pure lines and survival in hybrid bulk populations were detailed by Allard (1960). Firstly, segregation will occur in the hybrid bulks, and hence the number and nature of competing genotypes are not expected to be constant from generation to generation. Secondly, only when the majority of individuals is homozygous (about the F_6 to F_8) will the population approximate to a mixture of varieties. Even then, such populations will still contain a much larger number of genotypes competing with each other.

Allard and Jain (1962) have provided evidence on plant height and heading time from Composite Cross V to show that a large number of different genotypes remained in the population after 18 generations of natural selection. There is sufficient evidence to indicate that polymorphisms are common in both natural and agricultural species of inbreeding plants (Allard, 1965; Allard and Hansche, 1964; Allard, Jain and Workman, 1968). These results are in contrast to those obtained from the mixtures of varieties discussed above (e.g. Harlan and Martini, 1938; Suneson, 1949) where it was found that a single genotype often predominated. Allard, Jain and Workman (1968) suggested that the equilibrium observed in populations derived from hybrids might have been a result of a balance between on the one hand, the "loss of variability due to the combination of directional and stabilizing selection" and on the other, a "steady release of new variability resulting from intercrosses between individuals within the population". These authors concluded that "competition among homozygotes rarely if ever occurs in pure form in natural populations because even low levels of outcrossing set the stage for conversion of simple mixtures of limited numbers of homozygotes into a complex and dynamic populational system within a few generations".

In a plant breeding context, competition is an important factor. Whether it will affect the efficiency of single plant selection depends on the relationship between the yield of a particular genotype when grown in mixture as compared to its yield when being grown in a pure stand. The literature considered so far in this section has been mostly concerned with changes in the proportions of the components of the populations with time. Attention will now be turned to considering evidence that the yield of a genotype in a mixture is related to its yield in pure stand.

Jennings and his co-workers (Jennings and de Jesus, 1968; Jennings and Herrera, 1968; Jennings and Aquino, 1968) found with rice, that competitive ability and yield in pure stands were negatively correlated. The high yielding plants in mixtures were good competitors and were low yielding when grown in pure stands. They considered that strong competitive ability was undesirable and was the reason for the limited progress through breeding in some tropical programs. However, the negative relationship between yield in mixture and yield in pure stand in their experiments could be attributed to the gross morphological differences between the components of the mixture. This was recognised when the authors stated that the undesirable effects of competition could be overcome by hand rogueing all tall, leafy and spreading plants from the F_2. This allowed short-statured, small-leafed segregates to achieve their normal seed production. When the effect of competition due to morphological differences was removed, many lines with yields greater than the high yielding parents were present in a random sample from the F_6 generation. In this example, the incident light energy after anthesis was a major environmental factor affecting yield.

Johnston (1972) working with 100 homozygous varieties of barley and F_4 and F_6 generations of 48 F_3 derived lines from the cross, Proctor * C.I. 3576, grown in mixtures and pure stands, respectively, found that there were highly significant correlations between the yield of a genotype in mixtures and its yield in pure stand. Although acknowledging that inter-genotypic competition influenced the yield of some genotypes in the mixture, he concluded that micro-environmental variation was the main factor limiting the efficiency of single plant selection. Furthermore, Johnston elucidated the relationships between the definition and measurement of competitive ability, the yield in mixture and the yield in pure stand. Although competitive ability and yield in pure stand may be negatively correlated (Akihama, 1967; Jennings and Aquino, 1968); progress could still be made through selection if there is a positive correlation between yield in mixture and yield in pure stand. If competitive ability, C_i, of a particular genotype, i, is defined as the difference between its yield in mixture, X_i, and its yield in pure stand, Y_i, (Akihama, 1967), then:

$$C_i = X_i - Y_i$$

Johnston (1972) showed that, from a plant breeding point of view, it is the correlation, r_{XY}, between X_i and Y_i which is a useful value and not that between C_i and Y_i, r_{CY}.

The correlation between competitive ability and yield in pure stand is:

$$r_{CY} = \frac{\text{Cov}_{XY} - \text{Var}_Y}{\sqrt{(\text{Var}_X + \text{Var}_Y - 2\text{Cov}_{XY}) \cdot \text{Var}_Y}}$$

Since $(\text{Var}_X + \text{Var}_Y - 2\text{Cov}_{XY})$ and Var_Y are the variances of C and Y respectively, both must be positive. It can be seen that the correlation, r_{CY}, is negative whenever Var_Y is larger than Cov_{XY}, since the denominator is always positive. In such cases however, the correlation between X and Y is positive if the value of Cov_{XY} alone is positive. This suggests that, despite a negative relationship between competitive ability and yield in pure stand, progress in selection for high yielding genotypes could still be achieved if there was a positive correlation between yield in mixture and yield in pure stand. This result has been ignored too often in discussions of the role of competitive ability in reducing the efficiency of single plant selection in mixtures.

Frequency-dependent advantage is one form of competition, and the above result is of particular interest in this thesis. In a segregating population, many of the individuals are similar, genetically; others may be rare and quite different from the majority. If selection is practised in these circumstances, the efficiency depends on the relationship between the yield of a genotype in a mixed population where it may occur at a low frequency to its eventual yield in pure stand.

8. Frequency-dependent selection:

In the last section, it was demonstrated that there is a seeming contradiction; that on the one hand mixtures of several varieties of an agricultural population of a highly inbreeding species should soon be reduced to one or a few highly competitive genotypes and on the other, that natural populations and bulk hybrids are heterogeneous and in apparent equilibrium. This can be explained by a tendency, noted in several populations, for the selective value of certain genotypes to increase as these genotypes become increasingly rare in the populations (Allard, Jain and Workman, 1968; Harding *et al.*, 1966).

Consider, for example, a population of a self-pollinated crop which differs by only one gene for yield at the A locus. Assuming also that the frequencies of A_1 and A_2 are initially equal to 0.5, then the distribution of genotypes and their frequencies are as depicted in the diagram on page 13.

Theoretically, after "t" generations of selfing, if t is large, the heterozygote will almost be eliminated from the population, and the remaining genotypes are A_1A_1 and A_2A_2, each occurring at a frequency approaching 0.5. However, if frequency-dependent selection does operate, then once the frequency of the heterozygote falls below a certain level, its corresponding increase in fitness is such that it is able to produce twice as many progeny as either of the two homozygotes, and it is maintained in the population. The population therefore remains more heterogeneous than it would have without frequency-dependent selection.

In his early work, Fisher (1930) outlined the general conditions under which a balanced polymorphism will be maintained in a random mating population. Consider again the case of two alleles, A_1 and A_2, at a single locus, which have frequencies "p" and "q", respectively. The distribution of genotypes and their fitnesses may be shown to be:

Genotypes	A_1A_1	A_1A_2	A_2A_2
Initial frequencies	p^2	$2pq$	q^2
Fitnesses	a	b	c
Frequencies after selection	ap^2	$2bpq$	cq^2

If the population is at equilibrium, then the ratios of the gene frequencies are the same before and after selection, such that:

$$\frac{p}{q} = \frac{ap^2 + bpq}{bpq + cq^2}$$

$$\text{or} \quad ap + bq = bp + cq$$

Subtracting each of these from b(p+q), it becomes:

$$p(b-a) = q(b-c)$$

$$\text{or} \quad \frac{p}{q} = \frac{b-c}{b-a}$$

Fisher reasoned that a real equilibrium is obtained if (b-a) and (b-c) are either both positive or both negative; that is, if the heterozygote is either better or worse adapted than both the homozygotes. Although acknowledging that either case could occur in nature, he expressed the opinion that the former is likely to be much more frequent than the latter. If "b" is less than "a" and "c", the equilibrium is unstable and there will be no tendency for such cases to accumulate. However, if "b" exceeds "a" and "c", the equilibrium is stable and such cases will persist. For the population to be in stable equilibrium and heterogeneous, the selective value of the heterozygote should exceed those of the homozygotes.

However, this theory does not explain what happens if the fitness of the heterozygote is related to its frequency in the population.

This model has been generalized and Murray (1972) described the situation as follows:

After selection, the frequency of the allele, A_2, becomes:

$$q' = \frac{bpq + cq^2}{ap^2 + 2bpq + cq^2}$$

and the change of gene frequency is:

$$\Delta q = \frac{bpq + cq^2}{ap^2 + 2bpq + cq^2} - q$$

$$\Delta q = \frac{pq[b-a-q(2b-a-c)]}{ap^2 + 2bpq + cq^2}$$

Since pq and also $(ap^2 + 2bpq + cq^2)$ must be positive, then the sign of Δq is determined by $[b-a-q(2b-a-c)]$. At equilibrium Δq becomes zero and the ratios of the gene frequencies are the same before and after selection, such as was described by Fisher (1930):

$$\frac{p}{q} = \frac{ap^2 + bpq}{bpq + cq^2}$$

or $$ap + bq = bp + cq$$

However, when (1-q) is substituted for p, this becomes:

$$\hat{q} = \frac{b - a}{2b-a-c}$$

Thus, the equilibrium frequency, $\hat{q}$, depends on the fitnesses of the different genotypes and not on the initial frequency, q. It is obvious that for a stable equilibrium to occur ($\hat{q}$ is positive), "b" should be greater than "a" and "c".

However, when "a", "b" and "c" are treated as "weights" which are a function not only of the selection coefficients, mutation rates, migration and inbreeding coefficient of the population, but also of gene frequencies (Lewontin, 1958) it is demonstrated that the stability of an equilibrium depends on the rate of change of Δq with respect to q at or near the equilibrium point, $\hat{q}$. In practice, this is determined by examining the sign of the derivative:

$$\frac{\delta}{\delta q}\ [b-a-q(2b-a-c)]$$

This quantity will be positive if the equilibrium is unstable and negative if it is stable. Thus, through mathematical analysis, it is possible to show that a balanced polymorphism may exist without heterozygote advantage, provided that the selective values of the genotypes change inversely with gene frequencies, hence frequency-dependent selection. This phenomenon may provide stability even though the heterozygote has a lower fitness than the homozygotes, or it may render unstable an equilibrium in which the heterozygote has the highest fitness (Murray, 1972).

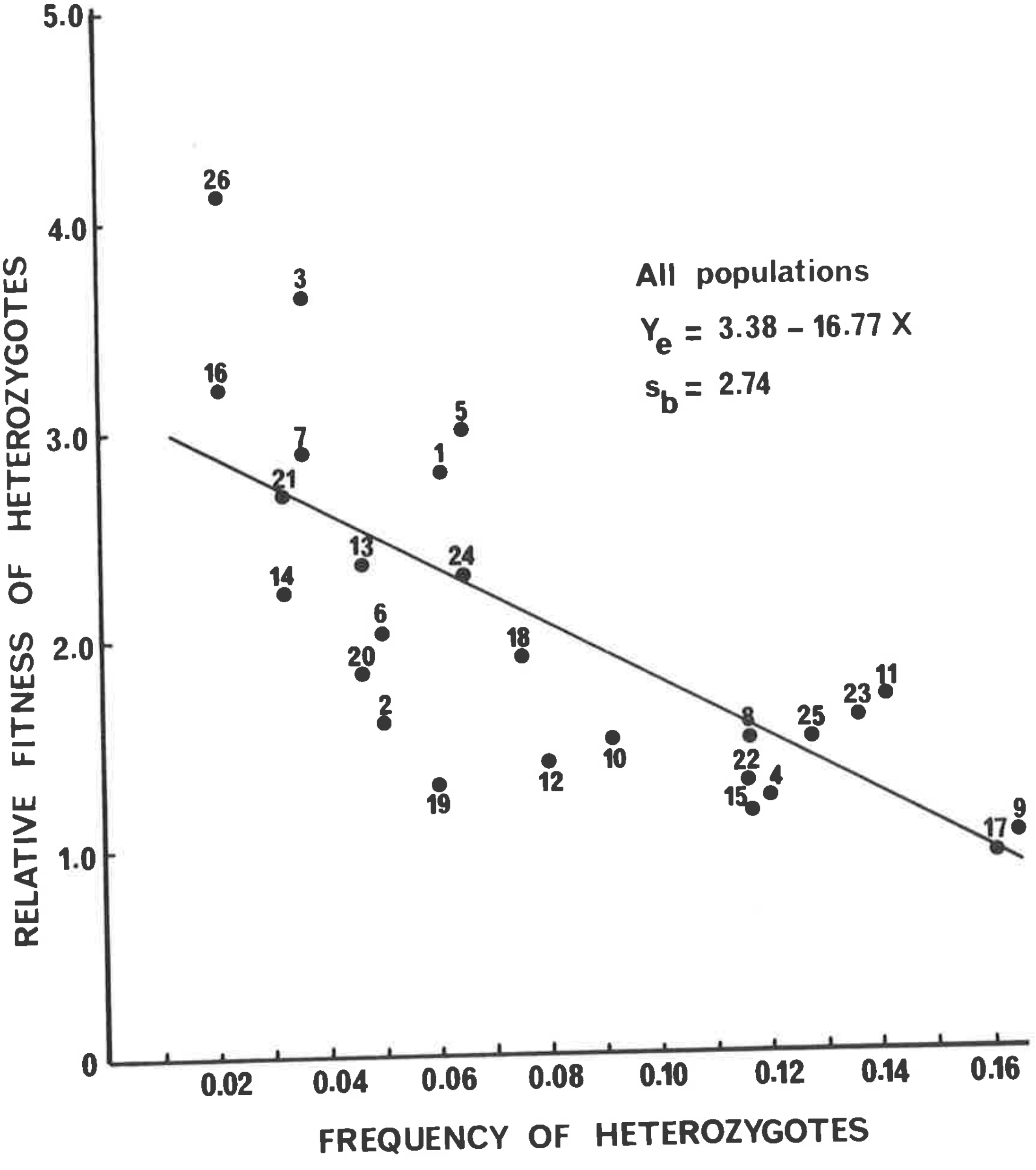
In the example of competition in a mixture of four barley genotypes (Suneson, 1949), Workman and Allard (1964) found evidence to suggest that Club Mariout, Vaughn and Hero had higher selective values when they were in low than when they were in high frequencies. Allard and Workman (1963) examined three populations, 53, 59 and 65, of lima bean segregating at the S/s locus for seed coat pattern from the F_2 to the F_9- F_{11}. They estimated that outcrossing varied from season to season but rarely exceeded 5% with the mean outcrossing percentages for the three populations being 1%, 3% and 4%, respectively. Heterozygotes were found to produce an average of 20% to 30% more progeny than homozygotes. As expected from a population with only about 5% outcrossing the proportions of heterozygotes decreased rapidly in early generations. However, after the F_5, each of the three populations showed an equilibrium in which the proportion of heterozygotes fluctuated narrowly between 10-15% of the population. Allard and Workman (1963) suggested that the results could not be explained on the basis of heterozygous advantage alone. It was estimated that the heterozygotes had greater fitness than either of the homozygotes and that this was more evident in a poor environment. In general, the heterozygotes produced relatively more seeds in a poor season as compared to a favourable season. It was observed that fitnesses fluctuated widely from season to season, including reversals in the selective values of the homozygotes between generations. They suggested that this "cyclic selection" might have been involved in maintaining genetic diversity in the populations they studied.

Harding et al.(1966) used one of Allard and Workman's populations, population 65, to conduct a series of experiments in which the frequencies of the heterozygotes were artificially modified so that the proportions of heterozygotes to homozygotes varied from 2% to 15%. Their results indicated that there was a significant correlation between the frequency of heterozygotes and their selective values. As the frequency of the heterozygotes in the population decreased, their fitness increased until at very low frequencies, they produced about three times more progeny than did the homozygotes (Figure 2). A stable equilibrium seemed to exist about the heterozygote frequency of 0.07. Below this equilibrium, the selective value of the heterozygote increased markedly in relation to those for the homozygotes, thus restoring the equilibrium. Frequency-dependent selection was postulated by Harding et al. (1966) to explain

Figure 2: Relationship between the frequency of heterozygotes at the S/s locus in lima bean populations and their fitness relative to homozygotes. Homozygotes were assigned a relative fitness of 1.0.

Each population was denoted by a number. This represented its composition which was a combination of the year of the experiment, the generations of the bulk from which homozygotes and heterozygotes were isolated and the frequency of heterozygotes in the population.

(After Harding *et al*., 1966)



All populations
Y_e = 3.38 – 16.77 X
s_b = 2.74
RELATIVE FITNESS OF HETEROZYGOTES
FREQUENCY OF HETEROZYGOTES
5.0
4.0
3.0
2.0
1.0
0
0.02
0.04
0.06
0.08
0.10
0.12
0.14
0.16
26
3
16
5
7
1
21
13
24
14
6
18
20
11
23
2
8
25
10
22
12
4
15
19
9
17

the retention of the polymorphism in these populations of lima bean. However, since seed coat pattern is maternally inherited (Allard, 1953) and since the heterozygotes and homozygotes used in the experiment came from the same bulk of an advanced generation (F_4,F_5, F_{13} or F_{14}), any error in identification of the homozygous lines, especially at the 2% frequency, could result in an over-estimation of the fitness of the heterozygotes.

Cockerham and Rawlings (1967), in an investigation using a mating system of mixed self-fertilization and outcrossing, have pointed out the effect on genotypic frequencies at a marker locus when selection involves the rest of the genome. Any force which tends to change the frequencies of the heterozygotes, whether it be selection or outcrossing or otherwise, will tend to do so at all loci. A comparison such as that of Harding et al. (1966), of an observed equilibrium value with that expected from the percentage of outcrossing would lead one to conclude that the heterozygote for the marker locus had a selective advantage over the homozygotes even though the locus itself was selectively neutral. Furthermore, if populations were initiated with different gene frequencies at this locus, they would go to different equilibria. Consequently, selection coefficients for the genotypes would appear to depend on gene frequency at this locus. Such conclusions could be reached when selection at an unmarked locus had a correlated effect, perhaps through linkage, at another neutral marker locus.

The hypothesis of the "neighbourhood effect" (Harding et al., 1966) suggested that frequency dependence of fitness values may be due to different varieties having different micro-environmental requirements (Ayala and Campbell, 1974). Assuming that genotypes differ in their requirement for resources or differ in the efficiency with which they utilize them, the available resources of a certain environment may be more fully exploited when the population consists of a number of different genotypes than when all individuals have the same genotype. The result is a balanced polymorphism, in which genotypic fitnesses are frequency dependent.

Similar results were obtained by Khalifa and Qualset (1974) with wheat. The performance of a tall variety, Ramona 50, and a short variety, D6301, was compared in a study where the genotypes were grown in 9 combinations at 0.125 frequency intervals. D6301 was only 0.75 as tall as Ramona 50, but their relative grain yields in pure stands were 1.00 and 0.80 respectively. At all frequencies, the contribution of D6301 to total yield was significantly below that expected on the basis of the yield in pure stands. This reduction was complimented by a corresponding increase in yield by Ramona 50 over that expected from pure stand performance. The reproductive value of D6301 consistently increased with increasing frequencies, from a relative value of 0.62 at frequency 0.125 to 1.00 in pure stand. On the other hand, Ramona 50 gradually decreased in yield with increasing frequencies, from a relative value of 1.33 at frequency 0.125 to 0.80 in pure stand. The regression coefficients of reproductive values on frequencies were highly significant for both genotypes. It was evident that there was a strong frequency-dependent selection operating in these mixtures. It probably resulted from competition for light between a tall and a short variety as the experiment was grown at a high density (90 kg/ha seeding rate) and at a high altitude (1219m elevation) at one of the locations. Differences in maturity may also have been important (Khalifa and Qualset, 1974) as in this experiment, the better competitor matured a few days earlier than D6301.

The reproductive value of a genotype has also been shown to depend on the genotypes with which it is associated (Allard and Adams, (1969a) with wheat and barley; Sakai, (1961) with rice; and Schutz and Brim, (1967) with soy bean). Furthermore, there is evidence to indicate that the effect on fitness of the genotypic association is often frequency-dependent. Schutz and Brim (1967) found that there was a linear relationship between the yield of a soy bean variety grown in hills or row plots with the number of competitor genotypes in the surrounding hills or rows. Allard (1967) compared the yields of a mass-propagated hybrid bulk population of lima bean, thirty lines derived from the population grown singly, and a mixture of equal proportions of the thirty lines. It was found that the mean yield of the lines grown in pure stands and the yield of the mixtures were lower than the yield of the mass-propagated population by 7.3% and 6.0%, respectively. It was concluded that the advantage of the population was due to favourable interactions which occurred when particular genotypes were present in particular combinations and at proper frequencies. Chapman et al.(1960) found that the total grain yield of mixtures of wheat varieties varied with the relative frequencies of the two varieties in the mixture and this frequency dependency was itself dependent on density. This suggested that the frequency-dependent effect of the mixtures was enhanced at high densities, where competition was most intense.

An explanation of the results on frequency-dependent selection was attempted by Schutz et al. (1968). They considered that "overcompensation" in a feedback mechanism was responsible for the equilibrium observed in Harding et al.'s (1966) experiment. Heterozygotes, in this case, were assumed to be good competitors and the homozygotes, poor competitors. For good competitors to exist with poor competitors in a feedback system at equilibrium, they must have a lower pure stand yield than the poorer competitors in the system. However, the reproductive value of the good competitor is enhanced as it decreases in frequency in a population. A poor competitor, on the other hand, never attains a reproductive advantage as a result of competition, but may exist in a stable feedback system with good competitors, provided it has an adequate pure stand advantage. They claim that an essential feature of the feedback system is the occurrence of over-compensation which results when the reproductive value of the good competitor is enhanced in competition while depressing the reproductive value of the poorer competitor by a lesser amount. In the absence of such an effect, a poor competitor may be eliminated from the population even though it is relatively productive in pure stand.

9. The maintenance of polymorphisms by frequency-dependent selection:

The discussion on frequency-dependent selection thus far has been concerned with its effects on agricultural populations. The literature on this subject also includes experimental studies on the role of frequency-dependent selection in the maintenance of genetic polymorphisms. These will now be discussed.

9.1 : Non-random mating in *Drosophila*;

The mating success of different strains of *Drosophila* flies has been studied by several authors. Petit (1951, 1954, 1968) found that the success of two mutants, "Bar" and "White", of *D. melanogaster*, depended on their frequencies in relation to the wild-type. When "Bar" and wild-type flies were present in equal proportions, "Bar" males were less successful in mating. This disadvantage, however, was reduced as "Bar" males decreased in proportion relative to the wild-type. With "White", the mating success was greater when "White" males were rare or were predominant, and least when they were between 40% and 80% of the population. A stable equilibrium for this system seemed to be at the frequency of 0.4 of the "White" mutant. The other equilibrium point at 0.8 "White" would be unstable because at these high frequencies the mutant allele would become fixed.

Similar results have been found in experiments with other strains of *D. melanogaster* (Rasmuson and Ljung, 1973), and with *D. pseudo-obscura* (Ehrman, 1966, 1967, 1968, 1969; Ehrman *et al.*, 1965; Spiess, 1968) *D. persimilis* (Spiess and Spiess, 1969), *D. tropicalis*, *D. equinoxialis* and *D. willistoni* (Ehrman and Petit, 1968) but not between two sexually isolated strains of *D. paulistorium* (Ehrman, 1966).

Ehrman (1966), found that when two kinds of males and females were present, the mating success depended on their relative frequencies. This frequency-dependent mating advantage was more pronounced for the males than for the females. In *D. pseudoobscura*, this occurred with strains of different geographic origins, mutants and wild-type flies, and flies raised at different temperatures.

In another study, Ehrman (1968) used individuals of three karyotypes: AR/AR, AR/CH and CH/CH (*). In each mating chamber, two karyotypes were represented with frequencies 10:10, 2:18, 18:2. The males of the heterokaryotypes, AR/CH, tended to have an advantage in mating provided that both kinds of males were equally frequent (at ratio 10:10). This advantage was further increased when the heterokaryotype was a minority (at ratio 2:18). When the homokaryotype, AR/AR or CH/CH, was a minority (at ratio 18:2), it was equally or more successful than the heterokaryotype in securing mates. These results suggest that frequency-dependent advantage for mating applies to both homozygotes and heterozygotes. It favours whichever form is the rarer and helps to maintain genetic polymorphisms. This phenomenon was not readily observed among the females. The minority females only sometimes had a slight advantage. This lack of frequency dependency in the females could be due to two reasons:

(i) in Drosophila, it is the female which chooses the partner; hence she could show preference for rare males.

(ii) while a female can only mate once during the period of observation, a male can mate repeatedly.

In the experiments, all the females had mated at the conclusion of the period of observation.

(*) AR = "Arrow head"; CH = "Chiricahua" chromosomal arrangements

Attempts have been made to identify the physical factors involved in the mating advantage of the rare forms. The technique used in most experiments was similar, consisting of two observation chambers, one on top of the other arranged so that the flies could not move from one chamber to the other. In the upper chamber, flies were placed in the ratio of 20 pairs of CH to 5 pairs of AR. In the lower chamber, either there were no flies or 15 pairs of AR. When the double chambers were connected by Saran wrap or by charcoal, materials which were likely to inhibit the passage of odours, the results were inconclusive. With chambers separated by a cheese-cloth, and especially when currents of air flowed between the chambers, there were positive results in favour of olfactory signals being important. When there were no flies in the lower chamber, the rare males had a mating advantage. However, this advantage disappeared when more flies of the rare type were added to the adjacent section. Thus, Ehrman demonstrated that the cue must be air-born and was likely to be chemical (odour) although sound (vibration) was not ruled out. In yet another experiment, Ehrman and Spiess (1969) found that the frequency-dependent mating advantage was nullified when extra males or both sexes of the rare type were added to the adjacent chamber, but not when females alone were added.

Whatever the mechanism may be, frequency-dependent selection - coupled with some degree of heterosis - as displayed by the mating advantage of a minority, could be a powerful force in maintaining genetic variability in populations of *Drosophila*.

9.2 : Competition:

Competition has been postulated by Harding *et al*. (1966) and

Khalifa and Qualset (1974) to explain the frequency-dependent selection observed in populations of lima bean and in mixtures of wheat varieties. Similar effects are evident with chromosomal inversion types in _Drosophila_. The adaptive value of chromosomal variants could be changed by many physical and biological factors: temperature, humidity, food, micro-organisms and population density (Birch, 1955; daCunha, 1951; Levitan, 1951; Spiess, 1950; Lewontin and Matsuo, 1963). The selective value of a particular genotype can also be altered by the presence of other genotypes. Lewontin (1955) used larvae from 22 strains of _D. melanogaster_ and compared their viability both in pure and mixed cultures. Some of the strains were found to have a higher viability in mixed than in pure culture, thus fitness increased with rarity. Lewontin called this relationship "genetic facilitation".

Similar results, in which competition is frequency-dependent, were reported by Ayala (1971), Nasar _et al_. (1973), Petit and Anxolabehere (1968) and especially in a series of experiments by Kojima and his associates. Yarbrough and Kojima (1967) showed that the larval viability of the karyotypes carrying the fast (F) and slow (S) alleles of the esterase-6 locus was frequency-dependent and recorded the change of allele frequencies for a period of 30 generations. Analysis of the selective values associated with this locus showed that there were marked differences between genotypes in the early generations. The rate of change in frequencies of the genotypes was very rapid in the early generations, but during the latter generations, the rate was much lower and analyses showed that approximate selective neutrality existed among the three genotypes. With the use of a computer, it was possible to show that the results could be explained if selective values were allowed to vary with the frequencies. Similar effects were also observed with the differences in viability from egg-to-adult associated with the alcohol dehydrogenase locus in a cage population of *D. melanogaster* (Kojima and Tobari, 1969a) and for several inversion karyotypes in *D. ananassae* (Kojima and Tobari, 1969b; Tobari and Kojima, 1967, 1968).

Attempts have been made to identify competition as the agent leading to frequency-dependent selection (Huang *et al*., 1971). Media were conditioned by using them to rear first-instar larvae of each genotype (FF, FS, SS) until the pupal stage. A second set of first-instar larvae of each genotype was then grown in the conditioned media, until emergence of all adults occurred. There were nine viability coefficients based on the ratios of flies emerging from the media to larvae. The results showed that the relative viability of a genotype was lowest when it was grown in media conditioned by the same genotype. For example, the viability of the SS type was highest in media conditioned by the FF type, intermediate in FS and lowest in SS. It was suggested that individuals of a conditioned genotype either depleted nutrients or left metabolic products harmful to its own genotype. Furthermore, the experiments also proved that fitnesses varied according to allelic frequencies, so rare genotypes had a higher fitness, and the abundant ones had a lower fitness. In a subsequent experiment, Kojima and Huang (1972) found that frequency-dependent selection did not operate when the population density was low. They concluded that frequency-dependent selection could only take place at high population densities, where the level of competition is intense. Results of the contrary have been reported recently by Dolan and Robertson (1975). These authors found that the survival of 6 strains of larvae of *D. melanogaster* was not significantly less when the food had been conditioned by the same strain rather than by another strain, nor did conditioning by one strain have any special effect on the survival of another. It is difficult to point out the difference between the two sets of results since the cornmeal molasses medium used by Dolan and Robertson (1975) was rather similar to that used by Kojima and Huang (1972) and the density for conditioning used by the former corresponded to the highest density used by the latter. The comparison is further complicated since Dolan and Robertson (1975) found that viability in conditioned medium (by the same genotype) was in fact higher than that in fresh medium. These authors cited other examples to suggest that "larval metabolic products do not impede but may even encourage larval development" (Sang, 1949). Until these results are confirmed by further experiments, it seems reasonable to conclude that intergenotypic competition is frequency-dependent.

9.3 : Predator - prey relationships:

Another mechanism which maintains a balanced polymorphism through frequency-dependent selection is the interaction between a predator and its prey. The literature on this subject is large (Ayala and Campbell, 1974; Murray, 1972), however, since the analogy between predator and prey in animals and frequency-dependent selection in plants is remote, the phenomenon will only be mentioned very briefly.

It has been suggested that the common forms of prey are more heavily predated and the rare forms more often over-looked by the predators (Allen and Clarke, 1968; Clarke, 1962; Tinbergen, 1960). The selective value of a phenotype would vary inversely with its frequency.

Clarke (1962) has termed this "apostatic selection". Allen and Clarke (1968) and Tinbergen (1960) used the term "specific searching image" to explain the mechanism by which the predator recognizes prey. The success by a predator in capturing edible prey, results in search for more of the same kind of prey. Thus, the more common form will be hunted more frequently than rare ones.

Frequency-dependent selection can also operate in Batesian mimicry. In this type of mimicry, the models are poisonous, distasteful, or otherwise protected from predators, and are often conspicuously marked. The mimics, however, are innocuous but gain protection from predators by their similarity to the models. Sheppard (1959) pointed out that a mimetic pattern will be at an advantage when the mimic is rare compared with the model but not when it is common, and the results will often be a balanced polymorphism between the mimic and other forms of the innocuous species. He cited data collected by Carpenter on the mimicry of *Pseudacrea eurytus*, a species of butterfly, to show that the frequency of each type of mimic was rigidly controlled by that of the models.

Experiments in which artificial models and mimics were mixed in different proportions (Brower, 1960; O'Donald and Pilecki, 1970) showed that the rare mimics were taken significantly less often than might be expected by their frequency in the total population of models and mimics. A balanced polymorphism in the prey species is maintained, since the rare mimic is protected by its model and hence is avoided by the predator.

9.4 : Evolution of disease resistance:

There is little evidence of the effect of frequency-dependent selection on the evolution of disease resitance. However, in a thought provoking paper, Haldane (1949) wrote: "Probably a very small biochemical change will give a host species a substantial degree of resistance to a highly adapted micro-organism. This has an important evolutionary effect. It means that it is an advantage to the individual to possess a rare biochemical phenotype. For just because of its rarity it will be resistant to diseases which attack the majority of its fellows. And it means that it is an advantage to a species to be biochemically diverse, and even to be mutable as regards genes concerned in disease resistance".

According to the gene-for-gene hypothesis advanced by Flor (1955) to explain the inheritance of resistance in flax and the pathogenicity in flax rust (Melamspora lini), it can be said that for each gene conferring resistance in the host, there is a specific complimentary gene for pathogenicity in the pathogen. Thus, it is possible that for a host to remain resistant, it should possess a rare gene. The probability of the pathogen mutating to a virulent form, becoming common and so able to attack this host is low.

The relationship between host and pathogen is frequency-dependent however, since virulent strains and resistant hosts can multiply freely when other strains and hosts are restricted. Haldane (1949) has cited wheat rust as an example of this. The development of new varieties of wheat resistant to all current strains of Puccinia graminis, whether by mutation or through the effort of the plant breeders, is inevitably followed by the appearance of new strains of rust within a few years.

However, if the gene-for-gene hypothesis is correct, it is likely that host and pathogen have evolved together and all combinations have already been in existence. The spontaneous evolution of host and pathogen could be due to mutation, gene recombination, or migration. Hence finding a virulent strain is probably just a sampling problem, which is made easier when the host is widespread.

The production of multiline varieties is an application of frequency-dependent selection. In a commercial crop, all plants may be of the same genotype and millions of genetically identical plants can be grown over the area of crop. With such a situation, any pathogen which can attack that genotype can attack millions of plants and heavy losses may be incurred. This was particularly the case with wheat and wheat stem rust in North America and more recently, with corn blight in the U.S.A. and stem rust in southern Australia. Multiline cultivars were suggested by Jensen (1952) for oats and Borlaug (1953) as a means of disease resistance. They could be produced by either blending several compatible pure lines of different genotypes (Jensen, 1952) or by backcrossing a large group of donor parents with different genes for resistance to the best available recurrent parent to introduce genetic diversity for disease resistance (Borlaug, 1959). Thus, a multiline variety could be considered as a mixture of many genotypes, each occurring at a low frequency and possessing a different type of resistance to a particular disease. In the presence of a pathogen, the resistant plants in the mixture would be expected to reduce the rate at which infection develops on the susceptible plants by acting as spore traps or as a method of spacing susceptible genotypes and thereby decreasing the probability of a virulent spore landing on a susceptible host-plant. This slowing down of the disease spread is particularly important in grain crops where the grain is produced at the end of the growing season. Browning and Frey (1969) reported that in Iowa, oat grains fill at a rate of 5% per day. If disease spread can be delayed by one rust cycle (about 8 days), a 40% increase in grain yield is possible. An example was cited in which a 1:1 blend of two genotypes, one of which was susceptible, delayed the disease spread in the field by 4 days and increased the yield of the susceptible cultivar by about 20%. In this example, a susceptible variety had a higher yield and fitness at a low frequency in a mixture, than in a pure stand.

10. Conclusions:

If rarity leads to enhanced fitness, then it may influence the outcome of selecting between genotypes in the segregating generations following the intercrossing of two self-pollinating lines. In a segregating population, there is a large number of different genotypes, many of which are similar genetically, while others may be rare and quite different from the majority. If selection is practised in these circumstances, it is possible that the choice of genotypes will be influenced by their apparent frequency, and that their advantage will not persist when those genotypes are multiplied and propagated in pure stands. Selection would then have been less effective and many other high yielding genotypes might have been over-looked.

This thesis reports a series of experiments on frequency-dependent advantage in wheat and its influence on the effectiveness of single plant selection.

Frequently, heterosis has been regarded as the major factor maintaining genetic polymorphisms at a stable equilibrium. Although the effect of heterozygous advantage cannot be disregarded, this thesis will present evidence to indicate that frequency-dependent selection plays a dominant part in the maintenance of such a system in self-pollinated populations.

III OUTLINE OF THE EXPERIMENTS.

A. Introduction

Frequency-dependent advantage in wheat was investigated in a series of experiments. They will be presented in three sections:

1. The occurrence and magnitude of frequency-dependent advantage (f.d.a.) in wheat (Experiments I and II).

2. The nature of f.d.a. (Experiments III, IV, V and VI).

   (i) Is it a result of heterozygosity and hybrid vigour?
   (ii) Does it differ between test genotypes?
   (iii) Does it vary with background genotypes?
   (iv) What is the cause of f.d.a.?

3. The implications of f.d.a. for single plant selection in wheat breeding (Experiments VII and VIII).

   (i) How does f.d.a. influence single plant performance in an F_2 population?
   (ii) What is the relationship between the yield of single plants of several genotypes occurring at low frequencies in two-component and multi-component mixtures and the yield of the same genotypes in pure stands?

The experiments were conducted in 1972, 1973 and 1974 in the field at Roseworthy Agricultural College, South Australia, except for the glasshouse experiment (Experiment IV) and experiment VI, which was undertaken in the field in 1973 at the Waite Agricultural Research Institute.

B. The soil and climate of the experimental sites.

Both the Waite Agricultural Research Institute (W.A.R.I.) and Roseworthy Agricultural College (R.A.C.) are in the cereal belt of South Australia (latitude 34°5 South and longitude 138°3 East). The W.A.R.I. is about 8km South and the R.A.C. is approximately 60km North of Adelaide.

1. The climate:

The climate at both sites is of the Mediterranean type, with cool, wet winters and hot, dry summers. The mean annual rainfall at R.A.C. (1931 to 1971) is 430mm as compared with 626mm at the W.A.R.I. (1925 to 1973).

The mean duration of the growing season is about 6 to 8 months, extending through the winter and spring; from May/June to November/December. Monthly rainfalls for the years of the experiments are presented in Table 1.

It is noted that 1972 was a dry year (90mm below the average at R.A.C.), and 1973 and 1974 were wet years (130 and 210mm above the average, respectively). The W.A.R.I., in general, is wetter than R.A.C. (by approximately 200mm per annum), and 1973 was a very wet year at this location (210mm above the average).

2. The soil:

The soil at R.A.C. is a sandy red brown earth, classified as Dr.2.2.3 in the Australian Factual Key (Northcote, 1965). At the W.A.R.I.

Table 1:

Monthly rainfall (mm) at Roseworthy Agricultural College and Waite Agricultural Research Institute, 1972-1974.

	Roseworthy Agriculture College			Waite Institute
Month	1972	1973	1974	1973
January	39.6	10.4	110.3	34.1
February	44.7	71.9	49.4	53.1
March	0	43.4	45.1	34.5
April	31.0	34.5	73.8	91.1
May	22.6	35.3	72.0	77.8
June	15.7	84.6	24.2	105.6
July	40.6	53.3	72.6	123.3
August	77.2	56.9	35.8	65.0
September	30.7	62.7	47.0	92.5
October	11.2	64.8	87.8	78.8
November	12.7	18.8	8.8	36.3
December	15.7	23.9	14.8	44.6
Total	341.6	560.6	641.6	836.7

Mean annual rainfall at R.A.C. (1931-1971) : 430mm.

Mean annual rainfall at W.A.R.I. (1925-1973) : 626mm.

the soil is a red brown earth of the Urrbrae series (Litchfield, 1951). These types of soil are typical of a large proportion of the soils in the wheat belt of southern and south-eastern Australia.

The heavy soil and high annual rainfall at the W.A.R.I. often lead to poor aeration in the soil following sowing and to poor establishment of wheat (Millington, 1959). This was the reason for conducting most of the experiments at R.A.C.. The land use of each site over the six years prior to experiments is shown in Table 2. The experiments followed either pastures or fallow in the rotation ensuring a high level of soil fertility. Superphosphate was added to the machine sown experiments (experiments IIA and IIB) at the rate of approximately 150kg/ha.

The differences in rainfalls and soil types at the two sites did not affect the interpretation of the results as each experiment was complete in itself. The experiments described in this thesis were designed to compliment each other, with each experiment seeking to answer one or two specific questions.

C. The materials:

This study was conducted with five varieties and the hybrids from the crosses between them. Three of the pure lines, Gabo, Halberd and Timgalen, are commercial wheat varieties in Australia; and the other two, Warimek and Wariquam, are breeders' lines, developed at the W.A.R.I.. Following is a brief description of the five lines.

1. Warimek: This is a selection from the cross (Mexico-120 * Koda), made in 1963 and issued by the University of Adelaide in February,

Table 2:

Details of land use for the 6 seasons preceeding the experiments at each site.

	Roseworthy Agricultural College			Waite Institute
Year	1972	1973	1974	1973
1966	Fallow	--	--	--
1967	Wheat	Barley	--	--
1968	Pasture	Pasture	Barley	Peas(a)
1969	Barley	Wheat	Medic(b)	Barley
1970	Pasture	Barley	Barley	Peas(a)
1971	Fallow	Medic(b)	Pasture	Clovers
1972	Experimental site	Pasture	Wheat	Fallow
1973	--	Experimental site	Fallow	Experimental site
1974	--	--	Experimental site	--

(a) ploughed in September, then fallow.

(b) An annual medic sown and reaped for seed (the common species grown were *Medicago polymorpha*, *Medicago truncatula*).

1971. It is an early maturing variety, with short, erect stature (average height 65cm), and sparsely tillering habit. It has very erect leaves and creamy, awnless, erect spikes. Mexico-120 is a selection from (Yaktana 54 * Norin 10-Brevor) from CIMMYT, Mexico and Koda from (Dundee * Kenya C6042) * (Bobin 2 * Gaza) from the University of Sydney, is related to Gabo.

2. Wariquam: This is from the cross (Mexico-120 * Quadrat), made in 1963 and issued by the University of Adelaide in February, 1971. It is short (average height 65cm), erect, medium to sparsely tillering habit and early maturing. The spikes are creamy, erect, tapering and awned. Quadrat originated from the cross between Ghurka and an unnamed crossbred Currawa * (Major * Gallipoli), made in 1927 at the State Research Farm, Werribee, Victoria and issued in 1941 (Macindoe and Walkden Brown, 1968).

These lines were selected for their high yielding ability and their short stature. Both lines were moderately susceptible to Septoria tritici, but were relatively resistant to leaf and stem rusts, especially in the epidemics of 1973 and 1974, as compared to the local commercial variety, Halberd. Warimek plants were occasionally found to be infected with wheat smut (Tilletia caries).

3. Halberd: This is a commercial variety grown on 60-70% of the acreage devoted to wheat growing in South Australia. It was susceptible to stem rust (Puccinia graminis), during the 1973-1974 epidemics. Halberd is a selection from the cross (Scimitar * Kenya C6042 * Bobin) * Insignia 49, made in 1953 and selected and released

by Roseworthy Agricultural College in February, 1969. It is an early maturing variety, of medium height (average 75cm), with a freely tillering habit. The spikes have brown chaff, and are square, erect and practically awnless.

4. Gabo: This is a result from the cross Bobin * Gaza (a durum wheat) * Bobin, made by University of Sydney and released in 1945. Until 1949, it was resistant to leaf rust and stem rust. It is now susceptible to smut and both leaf and stem rust. Gabo is an early maturing variety, with medium height (average 75cm) and sparsely tillering capacity. The spikes are creamy, compact and almost awnless.

5. Timgalen: Timgalen is a selection from the complex cross (Aguilera * Kenya * Marroqui * Supremo) * Gabo * Winglen, made by University of Sydney in 1958 and issued in August, 1967. Timgalen is an early maturing variety. Its growth is erect, it tillers freely and it is of medium height (average 70cm). The spikes are creamy, tapering, curved and awned. This variety carries 4 different genes for resistance to stem rust and 3 for resistance to leaf rust, including 1 for adult plant resistance (Macindoe and Walkden Brown, 1968). It is highly resistant to these two diseases in the field.

Following a seeding in May or June, all these varieties come into anthesis within a week of each other with Warimek and Gabo being slightly earlier than the others. All varieties mature within a few days of each other.

D. The data:

Plant height and the components of yield as well as yield itself were measured as indicators of frequency-dependent advantage at different stages of growth. Most of the measurements were made on single plants, except where otherwise indicated.

In experiment I, where the most detailed measurements were taken, the characteristics were:

1. Measurements made during the growth of plants including:

   (i) The number of tillers per plant 8 and 10 weeks after sowing.

   (ii) Length of the oldest tiller 8 weeks after sowing (cm).

   (iii) Flag leaf area at anthesis (cm^2), estimated as $A = L \times B \times 0.75$; where A = area of the leaf, L = length and B = width at the broadest section of the leaf. Watson *et al.*(1958) found that the leaf area of barley varieties could be described by the equations $A = L \times B \times 0.76$ in a pot experiment and $A = L \times B \times 0.73$ in a field experiment.

As the position of each plant in the mixture could be accurately identified, it was possible to observe the development of the same plant from emergence to harvest. A pair of trestles and a wooden plank enabled the operator, with some personal discomfort, to make measurements on the growing plants without any disturbance to the crop.

2. Measurements made at maturity: The plants were individually labelled, pulled from the ground and taken to the laboratory. As the

seed was required for further experiments, only air-dry weights were recorded. However, the high daily temperature (25-30°C) and low humidity (50-55%) at the Waite Agricultural Research Institute in the summer were sufficient to eliminate any variability arising from slight differences in moisture content at the time of harvest.

The characteristics recorded in the laboratory were:

(i) Plant height (cm): measured from the base of the main tiller to the tip of its spike, excluding awns.

(ii) Number of fertile tillers; hereinafter referred to as number of spikes per plant.

(iii) Number of spikelets per plant, including infertile ones.

(iv) Number of grains per plant.

(v) Total weight per plant (g): the weight of the whole plant, excluding roots.

(vi) Spike weight per plant (g).

(vii) Grain weight per plant (g).

3. Derived characteristics: In some experiments, the results on 2 derived characteristics were also presented. They included:

(i) Weight per grain (mg): calculated as the ratio of grain weight to number of grains per plant.

(ii) Harvest index (%): calculated as a ratio of grain weight to total weight per plant (Donald, 1962).

E. Statistical methods.

All statistical analysis was performed on a CDC 6400 computer of the University of Adelaide. Programs were written in FORTRAN, STATSCRIPT (Lamacraft, 1969) and using the programs in SPSS (Nie *et al.*, 1975).

The STATSCRIPT program was developed by R. Lamacraft of the Biometry Section at the Waite Agricultural Research Institute. It is written in FORTRAN IV and is designed for analysing data which has been derived from randomized, replicated experiments. Some of the operations that can be conducted on the data set are the analysis of variance, and the calculation of multiple linear regressions, basic statistics such as means and variances, correlations, and derived variates. Multi-way tabulation on any subset or set of the data may also be produced.

Most of the programs for the analysis of the results were written in FORTRAN and STATSCRIPT. SPSS (Statistical Package for the Social Sciences) programs were used for the stepwise regression analysis in experiment VIII.

IV. EXPERIMENTAL PROGRAM AND RESULTS:

A. FREQUENCY-DEPENDENT ADVANTAGE IN WHEAT

The occurrence and magnitude of frequency-dependent advantage (f.d.a,) in wheat were investigated in 2 field experiments. In the first, two F_1 hybrids were grown at five different frequencies in a stand of one of the parents to establish the frequencies at which the effect was obtained and the mean response of a hybrid grown in a parent. The second aimed at measuring the magnitude and variation of f.d.a. in F_2 derived lines, and to compare these to the F_1's and their parents.

1. Materials and methods.

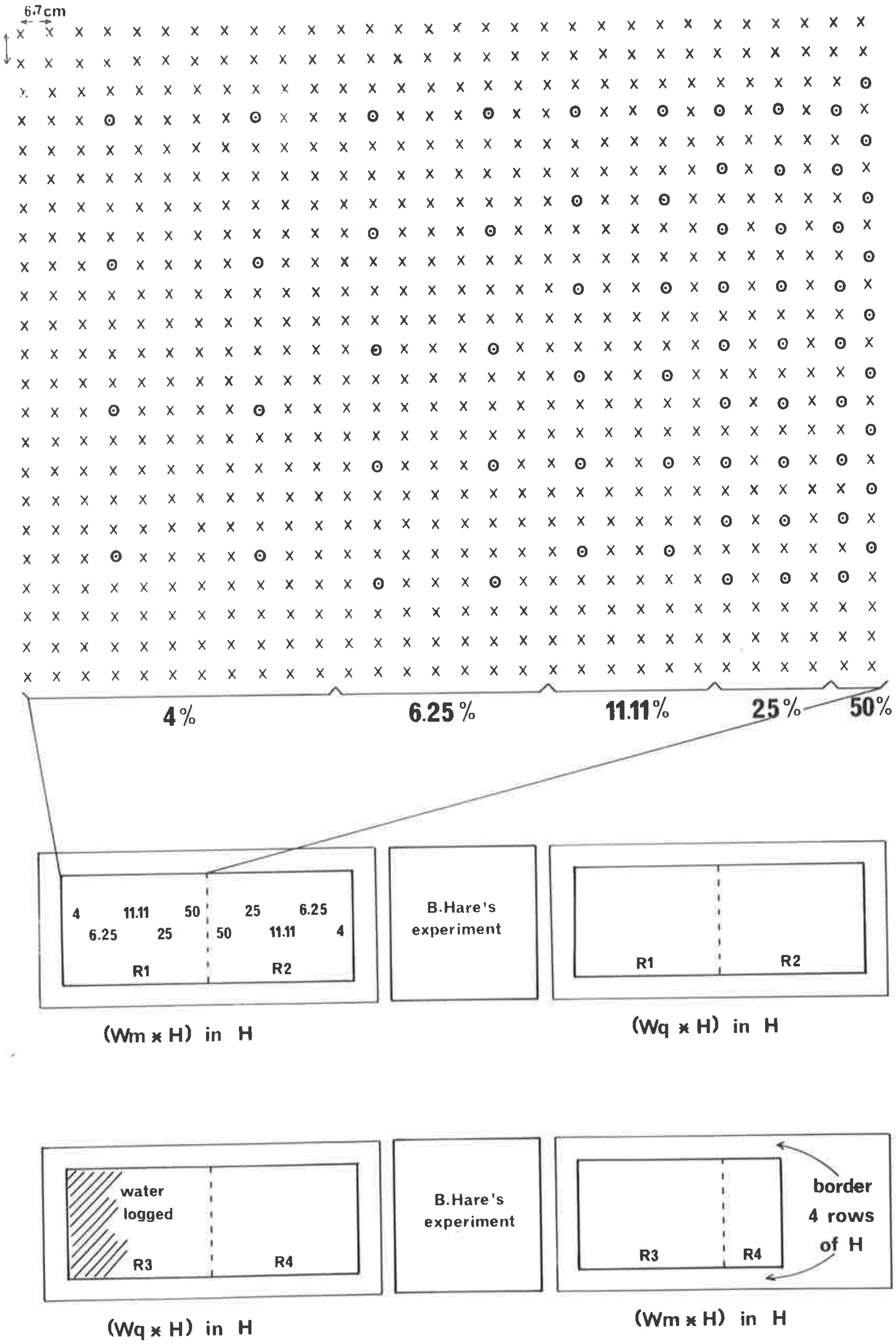
1.1 Experiment I: Experiment I consisted of two sub-experiments of the same design. In experiment IA, F_1 plants of the cross (Warimek * Halberd), subsequently abbreviated as (Wm*H), were grown in a stand of Halberd at five frequencies 4%, 6.25%, 11.11%, 25% and 50%. In experiment IB, (Wariquam * Halberd) F_1's, subsequently abbreviated as (Wq*H), were grown in Halberd at the same 5 frequencies. There were four replicates in each experiment, except the fourth replicate in the (Wm*H) experiment only consisted of the two higher frequencies due to the lack of hybrid seeds. To reduce the number of hybrid seeds required, each replicate was planted with the frequencies in ascending order. Hence, 4% was always on one end of the plot, the intermediate frequencies were bordered by lower and higher frequencies and the 50% was adjoined by the 50% of the adjacent replicate. Each replicate was surrounded on three sides by four rows of Halberd. Figure 3 shows the layout of the experiments and the precise positions of Halberd and the

Figure 3: Field layout of experiment I and a detailed plan of one replicate, showing the positions of hybrid and Halberd at 5 different frequencies of the hybrid.

The position of pure stands of the hybrids and Halberd (B. Hare's experiment) relative to the experiment reported here, is indicated.

x x x Halberd as the background plants.

⊖ ⊖ ⊖ (Wm*H) or (Wq*H) as the test plants.


6.7cm
4%
6.25%
11.11%
25%
50%
4
11.11
50
6.25
25
25
6.25
50
11.11
4
R1
R2
B.Hare's experiment
R1
R2
(Wm × H) in H
(Wq × H) in H
water logged
R3
R4
B.Hare's experiment
R3
R4
border 4 rows of H
(Wq × H) in H
(Wm × H) in H

hybrid at 5 frequencies in one replicate. The five frequencies of the hybrid could be considered as being 1 hybrid in a stand of 5x5, 4x4, 3x3, 2x2 and 1x2 of Halberd, respectively. Due to the lack of hybrid seed, their frequency could not be extended beyond 50%. However, the results may be compared with those of an adjacent experiment (Hare, 1976) in which the same hybrids and Halberd were grown at the same density in pure stands.

The experiments were planted on June 26 and 27, 1972 at Roseworthy Agricultural College. Seeds were sown individually at a distance of 6.7cm in both directions from the adjacent seeds, giving a density of approximately 222 plants per square metre. This density was slightly higher than the average commercial density in southern Australia (approximately 180 plants/m^2). The positions of the hybrids were marked by small wooden sticks to enable observation of their development from emergence to harvest (Plate 1).

At each frequency in each replicate, eight hybrids were chosen at random for observation, except at the lowest frequency where all the hybrids were recorded. Missing plants were replaced soon after emergence by transplanted seedlings to maintain the appropriate frequencies but these were not included in those measured for the results. The same number of Halberd plants immediately adjacent to the selected hybrids were recorded, to determine if these plants might shown an inverse response to that shown by the hybrids. At the two lower frequencies, 4% and 6.25%, the Halberd plants at the second position away from the hybrids were also measured to determine the extent of competition of the hybrids.

Plate 1: Two replicates of (Wm*H) in a stand of Halberd at 5 frequencies, one week after emergence. The positions of the hybrids were marked by small wooden sticks. Low frequencies were in the foreground, high frequencies in the middle and low frequencies again in the background of the photograph (cf. Fig. 3).

The experiment was protected from bird damage by netting.

1.2 Experiment II: During 1972 and 1973, a number of lines from the crosses (Warimek * Halberd) and (Wariquam * Gabo), subsequently abbreviated to (Wm*H) and (Wq*G) respectively, were advanced from the F_1 to the F_4 by the single-seed-descent method. F_1 plants were grown in plastic containers to produce F_2 seeds. For each cross, 156 F_2 seeds were grown in a wooden box (50cm x 30cm and 13cm deep) to hasten growth and development. At maturity, one seed was taken from each of a random subset of 100 of these F_2 plants. The F_3's were grown in plastic containers (25cm diameter and 25cm deep) for seed multiplication. All three generations were grown in a glass house. At the start of the 1973 season, sufficient seeds were obtained from 76 F_4 lines of the cross (Wm*H) and 70 F_4 lines of the cross (Wq*G) for experiments IIA and IIB, respectively.

The F_4 lines, together with the F_1's of both crosses and the four parents were grown at 2 frequencies, 6.25% and 18.75%, at Roseworthy Agricultural College. Experiments IIA and IIB were planted on June 29, 1973. Each experiment was a split-plot design, with the different genotypes as main plots and the two frequencies as sub-plots.

Due to the large area of the experiment, 70 x 30m, it could not be hand sown; instead, the seeds were mixed at the relevant frequencies and sown with a cone seeder. The genotype to provide the background stand of plants, Wariquam or Halberd, was chosen to be sufficiently distinct from the hybrid material for it to be identified at maturity.

The distinguishing features were:

Expt IIA
((Wm*H) hybrid : tall, tip-awned, brown heads.
(Wariquam background: short, strongly-awned, creamy heads.

Expt IIB
((Wq*G) hybrid : medium height, strongly-awned, creamy heads.
(Halberd background : tall, tip-awned, brown heads.

There were 3 replicates in each experiment. Each 2.50m plot consisted of 12 rows 10cm apart. The commercial rate of seedings (70 kg/ha) was used. The layout of the experiments and a detailed plan of one representative plot, showing the possible positions of hybrid plants at 2 frequencies are shown in Figure 4. The cone seeder was constructed to sow two rows from each cell of a seed magazine. In each plot, the four outer rows (rows 1,2, and 11,12) were borders and consisted of only the background genotype. Two of the remaining 8 rows were sown at the high frequency, 18.75%, and consisted of 15 hybrid plants and 65 background plants. The position of the high frequency was randomized between the two pairs of the middle rows (rows 5,6 or 7,8). The remaining 6 rows were sown with the hybrid at the low frequency, 6.25%. Each of these 2 rows consisted of 5 hybrid plants and 75 background plants. Fifteen plants per line (F_4 or F_1) per frequency and per replicate were harvested and bulked.

2. Results.

2.1 Experiment I

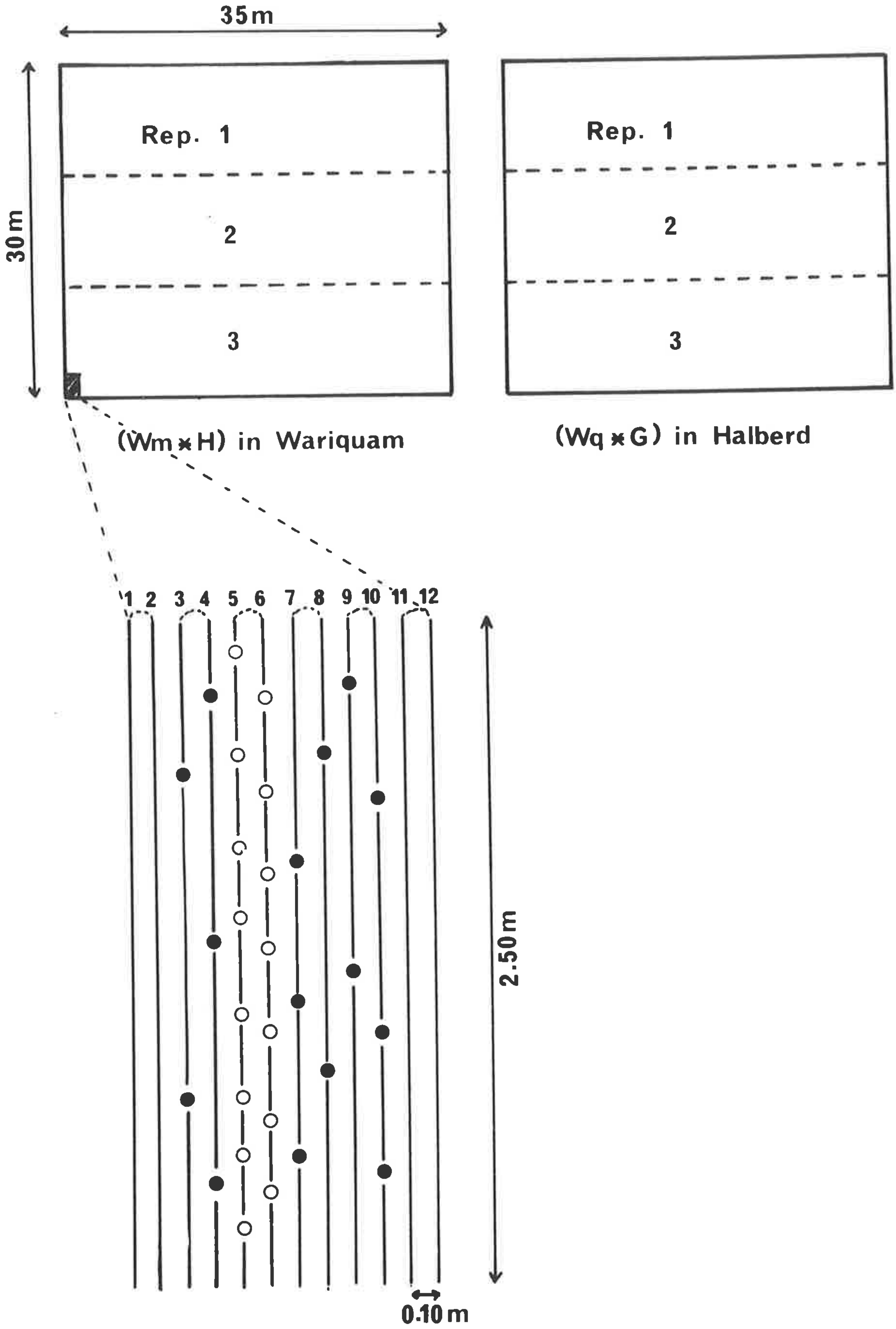
2.1.1 The magnitude of frequency-dependent advantage:

Regression analyses of different characters were performed on the percentage frequencies for the F_1. The mean performance of the

Figure 4: Field layout of Experiment II and a detailed plan of one representative plot, showing the possible positions of the test plants at 2 frequencies.

—————— ——————	Rows 1,2 and 11,12: Border rows of background plants. Each pair consisted of 80 plants.
—○——○— ——○-○——	Rows 5,6 or 7,8: High frequency, 18.75%. Each 2 rows consisted of 15 plants of an F_2 derived line and 65 background plants. The position of the high frequency was randomized between the 2 pairs of the middle rows.
———●——— —●————	Rows 3,4; 5,6 (or 7,8) and 9,10: Low frequency, 6.25%. There were 5 plants of the same F_2 derived line and 75 background plants in each of these two rows.

Open and closed circles have been used for the test plants at high and low frequencies, respectively; but they are of the same F_2 derived line.


35m
30m
Rep. 1
2
3
Rep. 1
2
3
(Wm×H) in Wariquam
(Wq×G) in Halberd
1 2 3 4 5 6 7 8 9 10 11 12
2.50m
0.10m

eight plants per frequency per replicate was used in the regression on frequency.

2.1.1.1 Experiment IA: (Wm*H) in Halberd.

The results for some characters are shown in Figure 5 and a summary of other characters in Table 3. It was found that hybrid plants at low frequencies yielded more grain than those at high frequencies, thus exhibiting frequency-dependent advantage (f.d.a.). The mean yield was 3.55 g/plant at 4% frequency and 2.26 g/plant at 50% frequency, a reduction of about 36.3%. Other characters related to grain yield also showed significant reductions with increasing frequencies; notably, number of grains per plant and spike weight per plant (with reductions of 36.3% and 35.5%, respectively).

The Halberd background plants were unaffected by the enhanced growth of the hybrids when these were at low frequencies. This is evident from Figure 5 and Table 3. It can be seen that none of these regressions was significant and that Halberd plants had similar yields at all frequencies. The mean yield only varied from 1.65 g/plant at the lowest frequency of the hybrid neighbour to 1.45 g/plant at the highest frequency of the neighbour.

2.1.1.2 Experiment IB : (Wq*H) in Halberd.

In this experiment, one of the four replicates was severely damaged by waterlogging and has been excluded. Again, a significant negative linear regression of yields on frequencies was observed (Figure 6a). The regression coefficients for grain yield and other characters for varying frequency are summarized in Table 4.

Figure 5: Variation in the morphological characters, yield components, and grain yield of (Warimek * Halberd) F_1's and adjacent Halberd plants for the five frequencies of the hybrids.

▲—▲—▲ (Wm*H) F_1

△---△---△ Halberd

See table 3 for significant levels of regression coefficients.

(a) Grain yield per plant:

$Y = 3.348 - 0.024\ f$ for F_1, where f is the percentage frequency.

$Y = 1.644 - 0.004\ f$ for Halberd

(b) Total weight per plant:

$Y = 7.857 - 0.053\ f$ for F_1

$Y = 4.190 - 0.009\ f$ for Halberd

(c) Number of grains per plant:

$Y = 99.48 - 0.716\ f$ for F_1

$Y = 56.67 - 0.182\ f$ for Halberd

(d) Number of spikes per plant:

$Y = 3.075 - 0.015\ f$ for F_1

$Y = 1.983 - 0.001\ f$ for Halberd

(e) Plant height:

$Y = 81.58 - 0.103\ f$ for F_1

$Y = 69.93 - 0.005\ f$ for Halberd

(f) Flag leaf area at anthesis:

$Y = 27.72 - 0.138\ f$ for F_1

$Y = 27.82 - 0.091\ f$ for Halberd

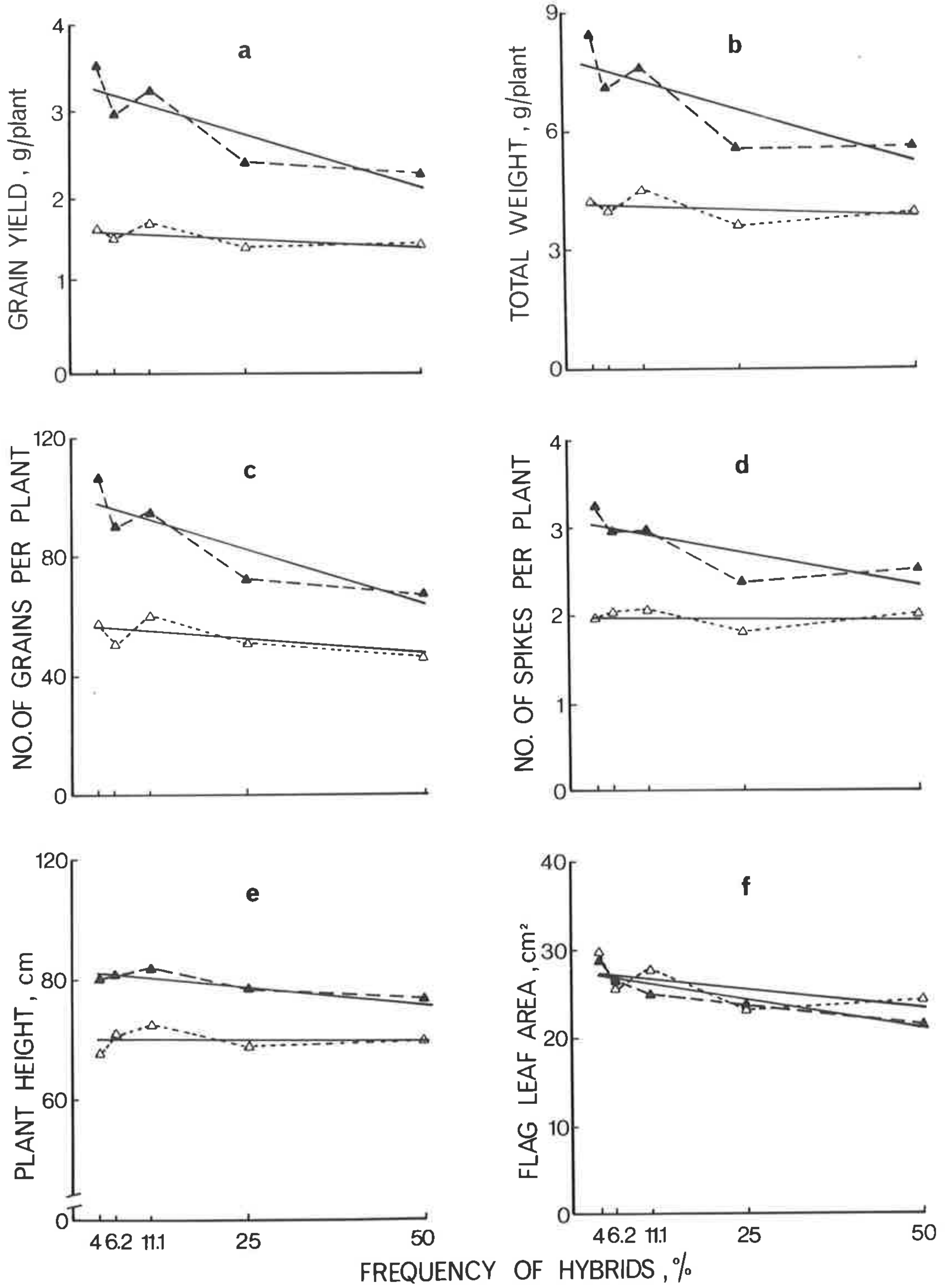

a
b
c
d
e
f
GRAIN YIELD, g/plant
4
3
2
1
0
TOTAL WEIGHT, g/plant
9
6
3
0
NO.OF GRAINS PER PLANT
120
80
40
0
NO. OF SPIKES PER PLANT
4
3
2
1
0
PLANT HEIGHT, cm
120
80
60
0
FLAG LEAF AREA, cm²
40
30
20
10
0
4
6.2
11.1
25
50
FREQUENCY OF HYBRIDS, %

Table 3: Regression coefficients for (Wm*H) in Halberd at 5 frequencies of the hybrid.

Character	F_1 (Wm*H)	Parent (Halberd)
No. of tillers (8 wks)	-0.006	-0.005
No. of tillers (10 wks)	-0.021	-0.001
Tiller length (8 wks)	-0.025	0.005
Flag leaf area (anthesis)	-0.138	-0.091
Total height (maturity)	-0.103	-0.005
No. of spikes	-0.015	-0.001
No. of spikelets	-0.276	-0.032
No. of grains	-0.716 * (a)	-0.182
Total weight	-0.053	-0.009
Spike weight	-0.030 *	-0.004
Grain weight	-0.024 *	-0.004
Weight per grain	-0.004	0.028
Harvest index	-0.030	-0.037

Table 4: Regression coefficients for (Wq*H) in Halberd at 5 frequencies of the hybrid.

Character	F_1 (Wq*H)	Parent (Halberd)
No. of tillers (8 wks)	-0.007	0.001
No. of tillers (10 wks)	-0.019	0.008
Tiller length (8 wks)	-0.020	0.044
Flag leaf area (anthesis)	-0.092 *	-0.041
Total height (maturity)	-0.070	-0.015
No. of spikes	-0.018 **	0.002
No. of spikelets	-0.328 **	0.024
No. of grains	-0.869 **	0.009
Total weight	-0.071 **	0.009
Spike weight	-0.038 **	0.003
Grain weight	-0.031 **	0.002
Weight per grain	0.019	-0.002
Harvest index	-0.009	-0.073

(a) *, **, *** denote significance ($b \neq 0$) at 5%, 1% and 0.1% probability levels, respectively. This notation for statistical significance is used for all tables.

Figure 6 : Variation in the morphological characters, yield components, and grain yield of (Wariquam * Halberd) F_1's and adjacent Halberd plants for the five frequencies of the hybrid.

▲—▲—▲ (Wq*H) F_1

△--△--△ Halberd

See table 4 for significant levels of regression coefficients.

(a) Grain yield per plant:

$Y = 3.853 - 0.031\,f$ for F_1

$Y = 1.591 + 0.002\,f$ for Halberd

(b) Total weight per plant:

$Y = 9.051 - 0.071\,f$ for F_1

$Y = 3.915 + 0.009\,f$ for Halberd

(c) Number of grains per plant:

$Y = 108.05 - 0.869f$ for F_1

$Y = 52.87 + 0.009f$ for Halberd

(d) Number of spikes per plant:

$Y = 3.348 - 0.018\,f$ for F_1

$Y = 1.874 + 0.002\,f$ for Halberd

(e) Plant height

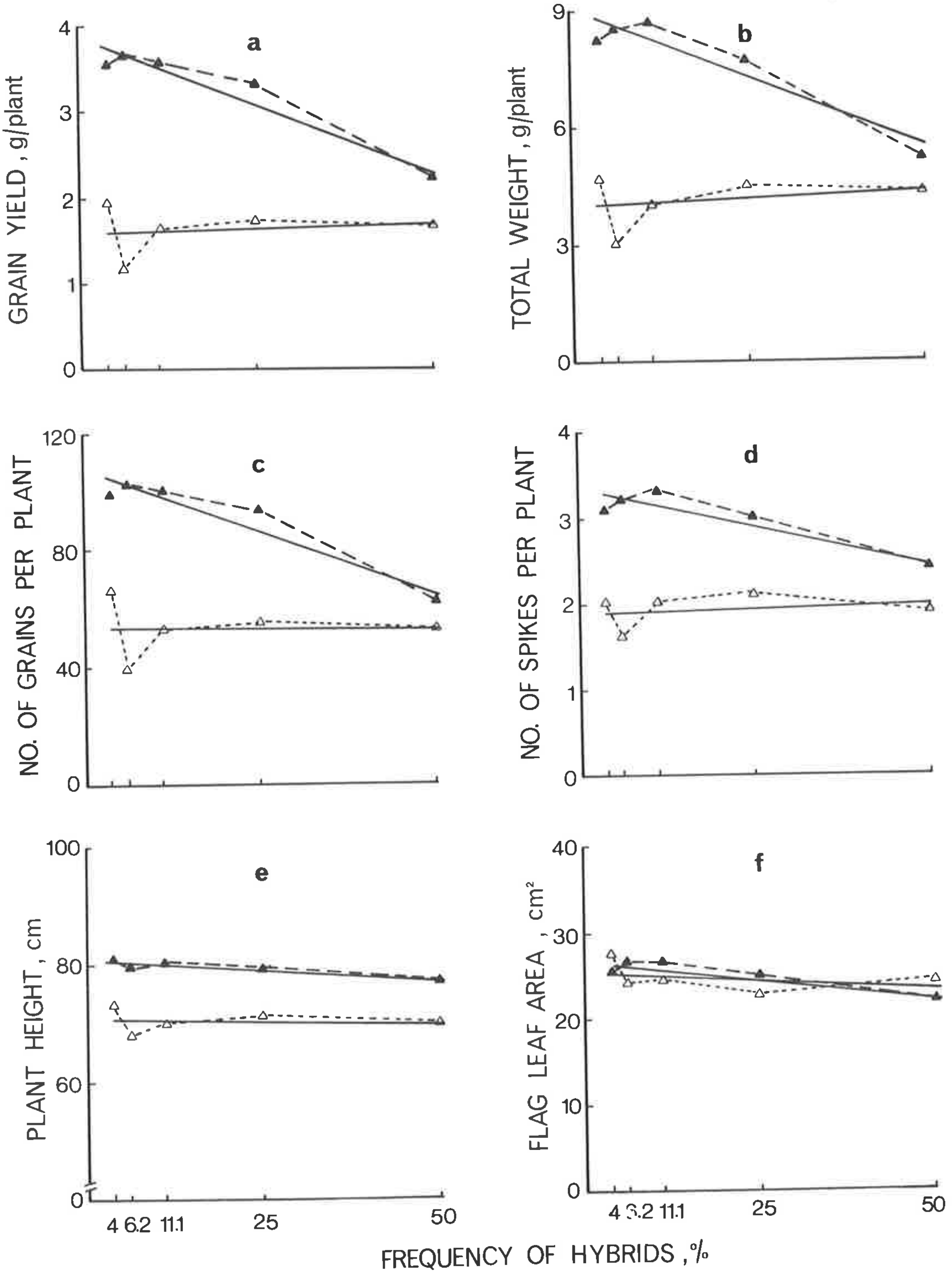
$Y = 80.78 - 0.070\,f$ for F_1

$Y = 70.63 - 0.015\,f$ for Halberd

(f) Flag leaf area at anthesis:

$Y = 26.61 - 0.092\,f$ for F_1

$Y = 25.45 - 0.041\,f$ for Halberd


a
GRAIN YIELD, g/plant
0
1
2
3
4
b
TOTAL WEIGHT, g/plant
0
3
6
9
c
NO. OF GRAINS PER PLANT
0
40
80
120
d
NO. OF SPIKES PER PLANT
0
1
2
3
4
e
PLANT HEIGHT, cm
0
60
80
100
4
6.2
11.1
25
50
f
FLAG LEAF AREA, cm²
0
10
20
30
40
4
6.2
11.1
25
50
FREQUENCY OF HYBRIDS, %

Grain yield was reduced from 3.54 g/plant at 4% frequency to 2.22 g/plant at 50% frequency, a reduction of 37.3%. Most of the other characters also showed a significant regression with frequency; especially those measured late in the growth of the plants, including flag leaf area at anthesis (12.7% reduction), total weight (36.8%), spike weight (35.4%), number of spikes (22.6%), number of spikelets (26.7%) and number of grains per plant (38.2%).

The Halberd plants as neighbours, again, did not differ significantly with different frequencies of the hybrid; varying only between 1.94 g/plant at the lowest frequency to 1.67 g/plant at the highest frequency of the neighbour. Nor did they show any significant change in other characters with changing frequencies.

In both experiments, the characters measured early in the growth of the plants did not differ significantly between frequencies, although they all had regression coefficients with the same sign as that for grain yield.

Since grain size did not differ between frequencies (Tables 3 & 4), the changes in grain yield of the hybrids were mainly due to the changes in numbers of grains per plant. The correlations between these characters were 0.976 for (Wm*H) and 0.977 for (Wq*H), respectively.

Both hybrids showed no change in harvest indices with changing frequencies, indicating that the magnitude of f.d.a. was similar for grain weight and total weight. There was no significant difference in plant height at different frequencies, suggesting that competition for light was unlikely to be an important factor in f.d.a..

2.1.2 Comparison between Halberd and hybrids:

Both hybrids were significantly higher yielding than adjacent Halberd plants, especially at the low frequencies (Figures 5 & 6). Tables 5 and 6 show the mean performance of each genotype at the 50% frequency. At this frequency, the differences between Halberd and hybrids were due to genetic effects alone rather than to any frequency effect, since they occurred in equal proportions. For most of the characteristics, (Wm*H) was significantly different from Halberd at this frequency, but (Wq*H) was only different in height at maturity, weight per grain and harvest index.

2.1.3 Comparison between Halberd plants at positions 1 and 2 at the 4% and 6.25% frequencies of the hybrids.

At the two lower frequencies of the hybrids, there were 4 and 3 Halberd plants between successive hybrids, respectively (Figure 3). Thus, the extent of competition of the hybrids could be estimated by comparing Halberd plants immediately next to the hybrids and those at one position removed. Tables 7 and 8 show the mean performance of Halberd plants at these positions. It can be seen that there was no difference between Halberd plants at these positions, indicating that the competition of the hybrids extended beyond the immediately adjacent Halberd plants and also suggesting that competition for light was not an important factor in f.d.a., since in cereals, it is unlikely that competition for light would extend beyond the immediate neighbours.

Table 5: Comparisons between the means for (Wm*H) and Halberd for various plant characters at the 50% frequency.

Character		Genotype means F_1 (Wm*H)	Halberd	Significant difference
No. of tillers (8 wks)		3.20	3.20	NS
No. of tillers (10 wks)		4.50	5.20	NS
Tiller length (8 wks)	cm	18.90	18.20	NS
Flag leaf area (anthesis)	cm^2	21.23	24.28	NS
Total height (maturity)	cm	76.56	69.92	**
No. of spikes		2.50	2.00	*
No. of spikelets		38.06	27.66	**
No. of grains		67.47	46.03	**
Total weight	g	5.62	3.89	**
Spike weight	g	2.98	1.96	**
Grain weight	g	2.26	1.45	**
Weight per grain	mg	33.25	31.80	NS
Harvest index	%	39.56	35.93	NS

Table 6: Comparisons between the means for (Wq*H) and Halberd for various plant characters at the 50% frequency.

Character		Genotype means F_1 (Wq*H)	Halberd	Significant difference
No. of tillers (8 wks)		3.20	3.10	NS
No. of tillers (10 wks)		4.30	4.90	NS
Tiller length (8 wks)	cm	19.90	19.10	NS
Flag leaf area (anthesis)	cm^2	22.32	24.49	NS
Total height (maturity)	cm	77.16	69.61	**
No. of spikes		2.37	1.91	NS
No. of spikelets		33.21	27.09	NS
No. of grains		61.46	49.04	NS
Total weight	g	5.23	4.32	NS
Spike weight	g	2.94	2.21	NS
Grain weight	g	2.22	1.67	NS
Weight per grain	mg	35.82	31.96	*
Harvest index	%	41.79	37.07	**

Table 7: Comparison between the means for Halberd plants immediately adjacent to and one position removed from the hybrid (Wm*H) at the 4% and 6.25% frequencies.

Characters		Mean performance Adjacent	One position removed	Sig. difference
No. of tillers (8 wks)		3.38	3.27	NS
No. of tillers (10 wks)		5.25	5.25	NS
Tiller length (8 wks)	cm	17.87	17.95	NS
Flag leaf area (anthesis)	cm^2	27.73	25.91	NS
Total height (maturity)	cm	72.06	71.74	NS
No. of spikes		2.08	2.23	NS
No. of spikelets		30.19	32.96	NS
No. of grains		53.83	58.52	NS
Total weight	g	4.44	4.70	NS
Spike weight	g	2.25	2.35	NS
Grain weight	g	1.73	1.80	NS
Weight per grain	mg	32.14	30.76	NS
Harvest index	%	38.96	38.30	NS

Table 8: Comparison between the means for Halberd plants immediately adjacent to and one position removed from the hybrid (Wq*H) at the 4% and 6.25% frequencies.

Characters		Mean performance Adjacent	One position removed	Sig. difference
No. of tillers (8 wks)		3.00	2.92	NS
No. of tillers (10 wks)		4.42	4.40	NS
Tiller length (8 wks)	cm	16.72	17.65	NS
Flag leaf area (anthesis)	cm^2	25.89	25.71	NS
Total height (maturity)	cm	71.81	70.70	NS
No. of spikes		1.90	1.99	NS
No. of spikelets		27.68	27.93	NS
No. of grains		51.93	51.07	NS
Total weight	g	4.20	4.15	NS
Spike weight	g	2.26	2.19	NS
Grain weight	g	1.70	1.66	NS
Weight per grain	mg	32.74	32.50	NS
Harvest index	%	40.48	40.00	NS

2.2 Experiment II.

2.2.1 Experiment IIA : (Wm*H) in Wariquam.

Mean squares from the analyses of variance for several characters are presented in Table 9. Seventy six F_4 lines of (Wm*H) all showed f.d.a. when grown in a stand of Wariquam. They had higher yields at the low than at the high frequency (Figure 7). The mean yield per line was 41.15 g/plot at the low frequency as opposed to 25.57 g/plot at the high frequency. This reduction of 38% was comparable to that found in experiments IA and IB although the range between frequencies was less in this experiment (6.25% to 18.75% as compared with 4% to 50%). The results in Table 9 indicate that the high yield of the hybrids at the low frequency was associated with a greater height, more spikes per plant and a greater total weight.

F.d.a. is also evident in Figure 7 for the parents, Warimek and Halberd, and their F_1. The parents yielded about the same as the mean of the F_4's, whereas the yield of the F_1 was among those for the top 5% of the F_4 lines. The distribution for height of the F_4's, Warimek, Halberd and their F_1 is shown in Figure 8.

The F_4 lines had positive skew distributions for all characters, except height which had a negative skew. Skew distributions of this nature occur commonly in plant competition, especially at high densities (Koyama and Kira, 1956). This phenomenon is discussed more fully in experiment VIII, when the distributions of single plants in F_1, F_2 and parental stands are compared. There was also a highly significant correlation between the f.d.a. and the yielding ability of these lines.

Table 9: Mean squares for the comparisons of 76 F_4 lines of (Wm*H) grown in Wariquam at 2 frequencies.

		Character				
Source of variation	df	Average height	No. of spikes	Total weight	Spike weight	Grain weight
Replicates	2	284.70	2123.60	11229.40	2108.20	321.21
Lines	75	101.89 **	893.64 ***	6871.90 ***	1430.90 ***	487.42 ***
Reps. x Lines (Error a)	150	56.04	265.28	2159.50	385.38	119.77
Frequencies	1	448.03 ***	69190.10 ***	514833.00 ***	101493.00 ***	26683.20 ***
Frequencies x Lines	75	17.25	167.28	1215.40	234.10	68.64
Reps x Freqs x Lines (Error b)	152	17.40	163.94	1284.10	253.97	78.83

Note: The experiment was a split-plot design with the 76 F_4 lines as main plots and the 2 frequencies as sub-plots. In the analysis of variance, the effects for replicates and lines were tested against the interactions between replicates and lines (Error a). The effects for frequencies and the interactions between frequencies and lines were tested against the interactions between replicates, frequencies and lines. (Error b).

Figure 7: Distribution of grain yield per line for the 76 F_4 lines of (Wm*H), the two parents and the F_1 at 2 frequencies in a stand of Wariquam.

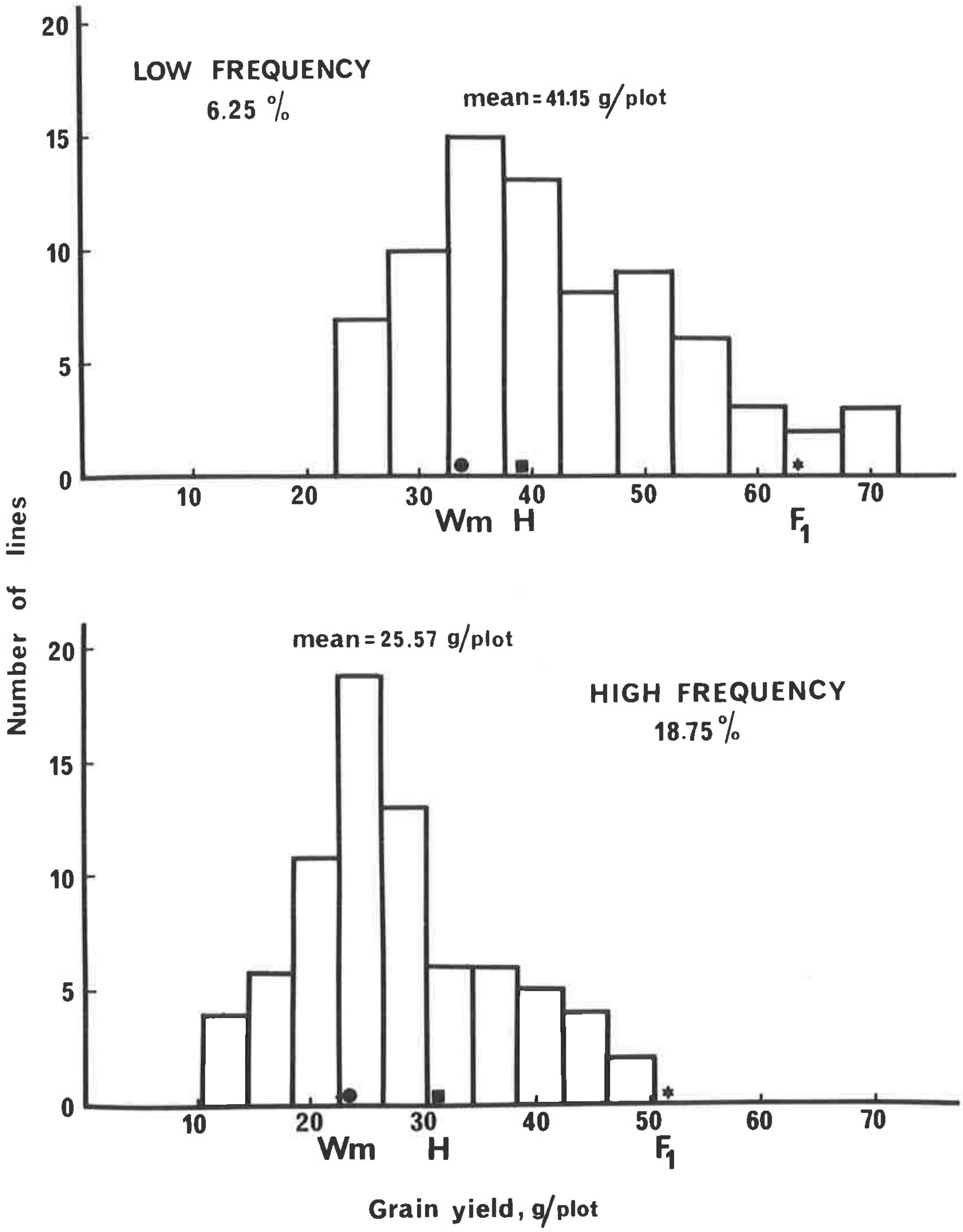
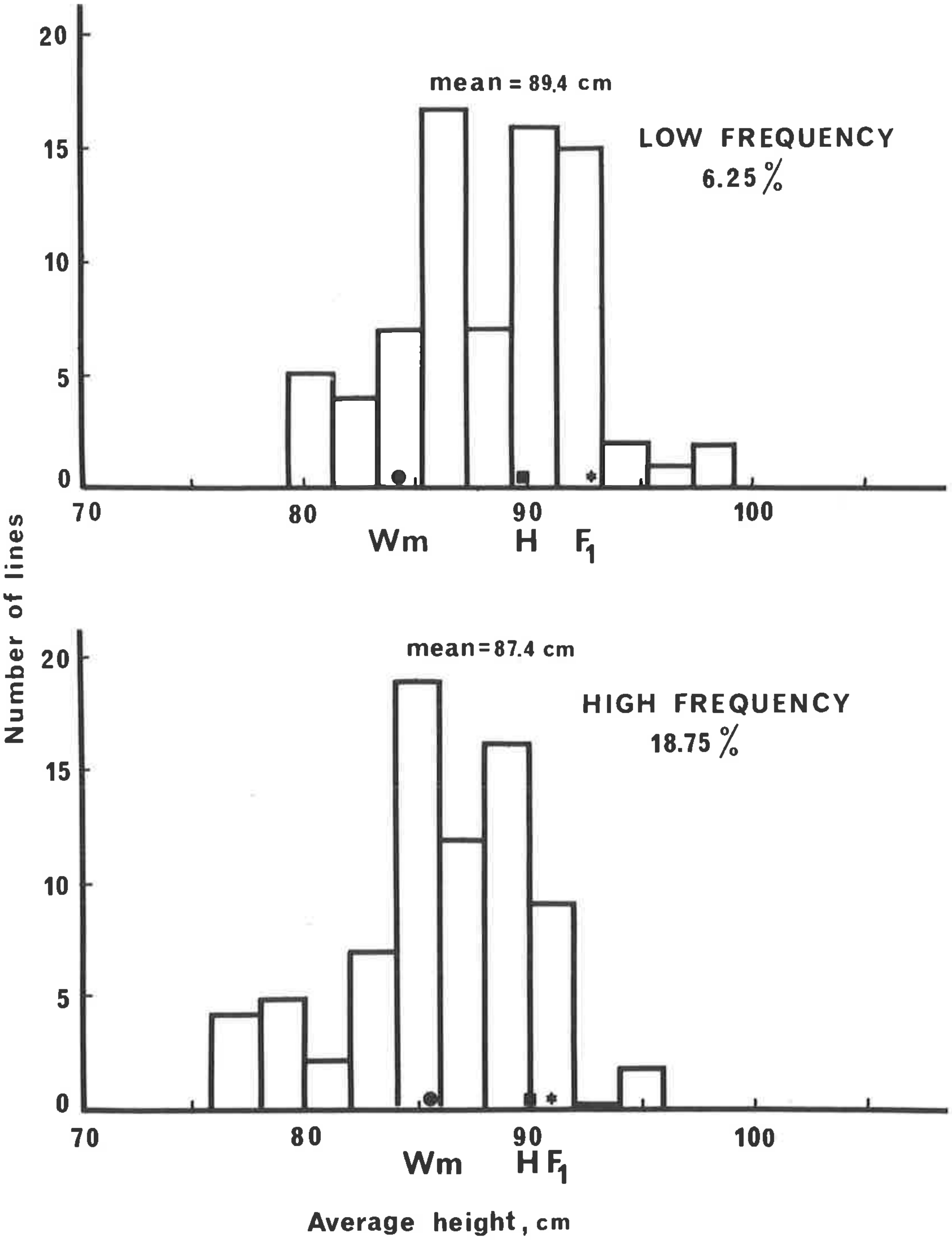

20
15
10
5
0
LOW FREQUENCY
6.25 %
mean = 41.15 g/plot
10
20
30
40
50
60
70
Wm
H
F_1
Number of lines
20
15
10
5
0
mean = 25.57 g/plot
HIGH FREQUENCY
18.75 %
10
20
30
40
50
60
70
Wm
H
F_1
Grain yield, g/plot

Figure 8: Distribution of average height per line for the 76 F_4 lines of (Wm*H), the two parents and the F_1 at 2 frequencies in a stand of Wariquam.



Number of lines
20
15
10
5
0
mean = 89.4 cm
LOW FREQUENCY
6.25 %
70
80
90
100
Wm
H
F_1
20
15
10
5
0
mean = 87.4 cm
HIGH FREQUENCY
18.75 %
70
80
90
100
Wm
H
F_1
Average height, cm

The f.d.a. was estimated by the difference between the yield at low and high frequency, for each line. Yielding ability was assessed as the arithmetic mean of the yields at the 2 frequencies for each line. This estimation was made because all 76 F_4 lines were grown with the same background, Wariquam. The correlation coefficient was 0.47 and was significant at the 0.1% level (Figure 9). The positive correlation suggests that high yielding genotypes tended to be more likely to express f.d.a.

However, the correlation should be regarded with caution since there might be an inherent relationship between the means and the differences of the yields at the two frequencies. This may occur if the yield at one frequency is always substantially larger than that at the other. A numerical example will illustrate this point.

Let the yield at the low frequency be X and the yield at the high frequency be Y and let X and Y have the values:

X_i = 6 7 9 12
Y_i = 2 5 7 8

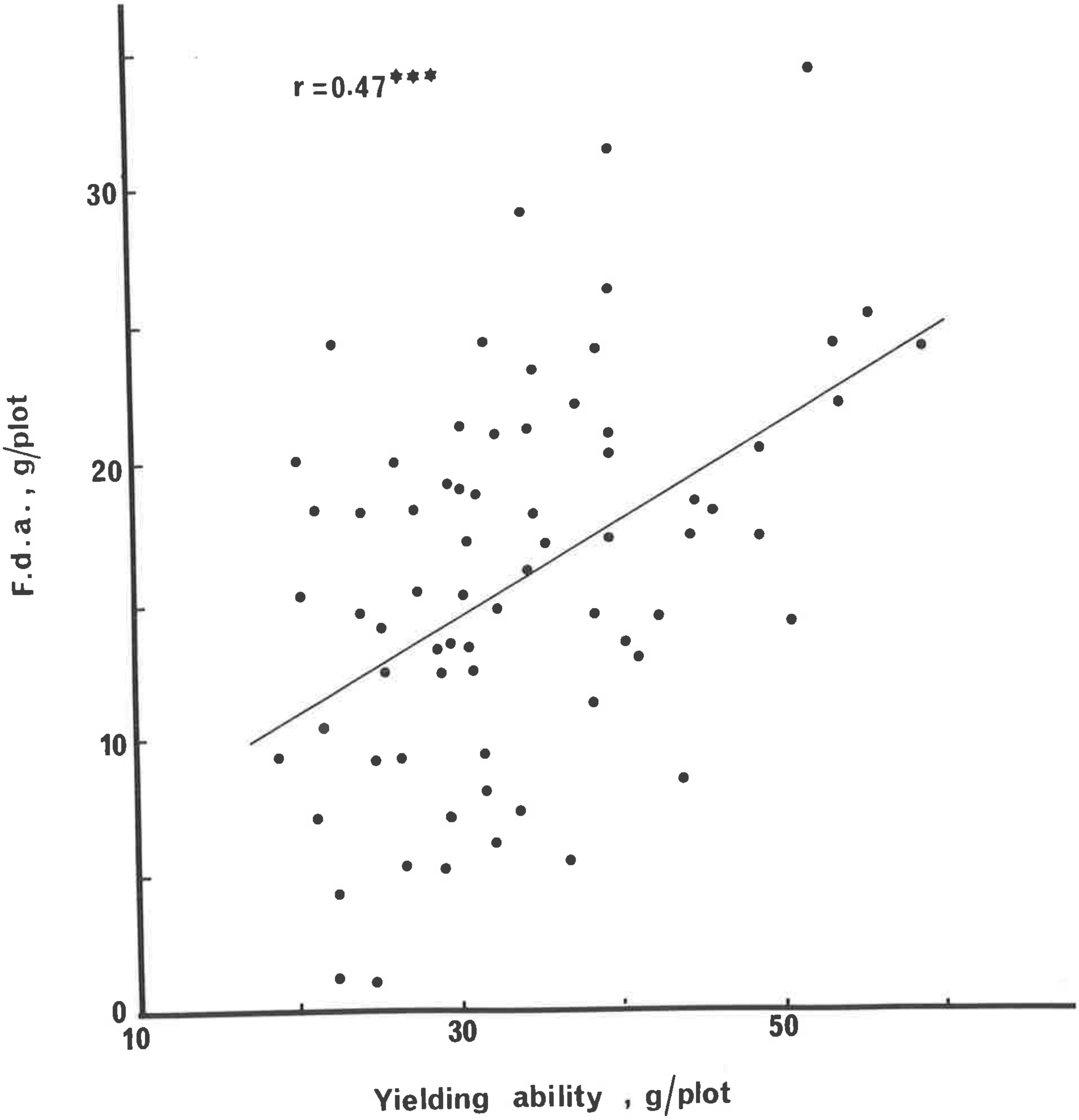
then the differences $(X_i - Y_i)$ are: 4 2 2 4
and the means $\frac{1}{2}(X_i + Y_i)$ are: 4 6 8 10

It can be seen that there is no correlation between the means and the differences in the above example. However, if Y_i's remain the same but X_i's increase by a factor of 10 so that the orders of X and Y are the same as before, we now have:

X_i = 60 70 90 120
Y_i = 2 5 7 8

Figure 9: Relationship between f.d.a. (yield at low freq. - yield at high freq.) and yielding ability (mean of the yield at 2 frequencies) of 76 F_4 lines of (Wm*H) when grown in Wariquam at 2 frequencies.

The fitted line has the equation, $Y = 4.004 + (0.349) X$; where Y is the f.d.a. in gm per plot, and X is the mean yield of the line at the two frequencies. Correlation coefficient $r = 0.47$ ***


r = 0.47***
F.d.a., g/plot
30
20
10
0
10
30
50
Yielding ability, g/plot

then the differences $(X_i - Y_i)$ are: 58 65 83 112
and the means $\frac{1}{2}(X_i + Y_i)$ are: 31 37.5 48.5 64

In this instance, there is a positive correlation between the means and the differences of X and Y. When there is a marked difference between X and Y, subtracting or adding Y_i to X_i had little effect on the ranking of X_i's. Thus, it can be seen that the difference $(X_i - Y_i)$ is nearly equal to X_i and the mean $\frac{1}{2}(X_i + Y_i)$ is approximately one-half of it. Hence, the correlation between $(X_i - Y_i)$ and $\frac{1}{2}(X_i + Y_i)$ would be approximately that between X_i and $\frac{1}{2}X_i$ and would always be positive.

Inspection of Figure 7 shows that the yields at the two frequencies overlap and the correlation, although highly significant, is only 0.47. It seems unlikely that a mathematical relationship is responsible for the positive correlation coefficient, - rather it appears to reflect a biological relationship.

2.2.2 Experiment IIB : (Wq*G) in Halberd.

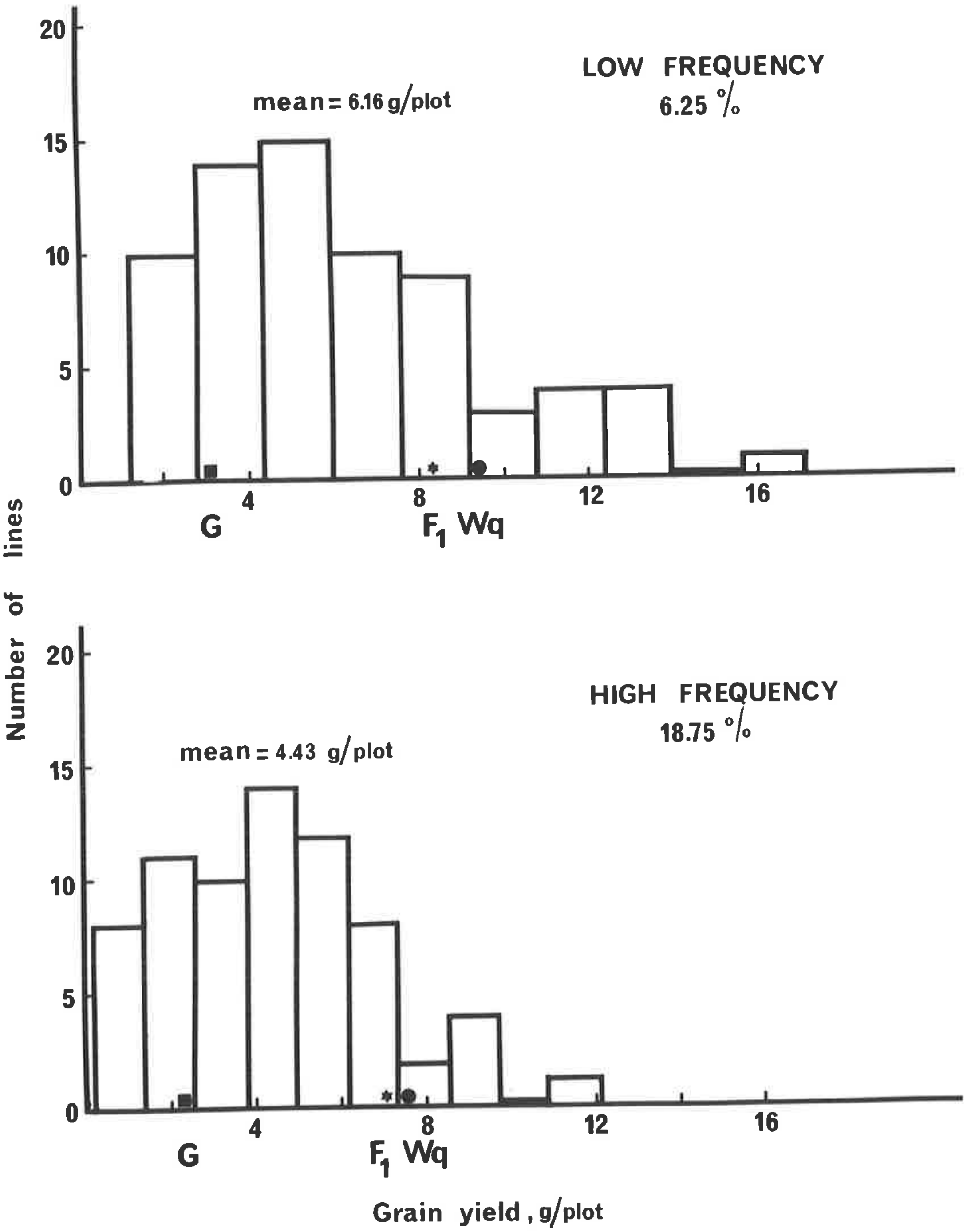
The results in Experiment IIB were similarly analysed (Table 10). Most of the F_4 lines of (Wq*G) had higher yields at the low than at the high frequency (Figure 10). F.d.a. was also expressed by the parents, Wariquam and Gabo, and their F_1. Wariquam was slightly higher yielding than the F_1 and both had yields equivalent to those lines among the 25% highest yielding F_4's. Gabo was similar to the lower 30% of the F_4 lines. The higher yield at the low frequency was associated with a higher expression of other characters at this frequency. The reduction in yield in this experiment from the low to the high frequency was about

Table 10: Mean squares for the comparisons of 70 F_4 lines of (Wq*G) grown in Halberd at 2 frequencies.

		Character				
Source of variation	df	Average height	No. of spikes	Total weight	Spike weight	Grain weight
Replicates	2	190.39	13.95	20.81	25.26	14.75
Lines	69	214.11 ***	180.81 ***	635.58 ***	87.81 **	39.00 **
Reps x Lines (Error a)	138	120.06	91.38	356.56	51.96	22.92
Frequencies	1	1030.40 ***	1113.90 ***	5589.60 ***	679.62 ***	270.21 ***
Frequencies x Lines	69	55.49	41.99	159.91	20.69	8.77
Reps x Freqs x Lines (Error b)	140	63.01	48.00	172.81	24.50	11.38

See footnote at end of Table 9 for an explanation of Errors a and b.

Figure 10: Distribution of grain yield per line for the 70 F_4 lines of (Wq*G), the two parents and the F_1 at 2 frequencies in a stand of Halberd.


LOW FREQUENCY
6.25 %
mean = 6.16 g/plot
20
15
10
5
0
4
8
12
16
G
F1 Wq
Number of lines
HIGH FREQUENCY
18.75 %
mean = 4.43 g/plot
20
15
10
5
0
4
8
12
16
G
F1 Wq
Grain yield, g/plot

28%.

The distribution for height (Figure 11) had a significant negative skew, whereas the distribution for other characters had a strong positive skew.

The F_4 lines had very low yields, averaging 6.16 g/plot and 4.43 g/plot for the low and the high frequencies, respectively, and some plants failed to survive to maturity. One likely explanation is that this was a result of the rust (Puccinia graminis f. sp. tritici) epidemic that occurred in 1973, when the variety, Halberd was heavily infected. Some of the F_4 lines grown in this stand were also severely infected, resulting in a large number of low yielding lines.

The correlation coefficient between f.d.a. and yielding ability was 0.38 and was significant at the 1% level (Figure 12). Although the analysis of variances indicated that the 70 lines had higher yields at the low frequency, the interactions between frequencies and lines were non-significant (Table 10). Figure 12 shows that some of these lines did have non-significantly higher yields at the high frequency.

3. Discussion:

Both of the present experiments were designed to determine whether growing a genotype at different frequencies in a mixture resulted in differences in the yield of single plants of that genotype. In all four cases, frequency-dependent advantage was demonstrated, and increasing the frequency of the genotype from a low (4% or 6.25%) to a moderate frequency (50% or 18.75%) was associated with a reduction in grain yields

Figure 11: Distribution of average height per line for the 70 F_4 lines of (Wq*G), the two parents and the F_1 at 2 frequencies in a stand of Halberd.

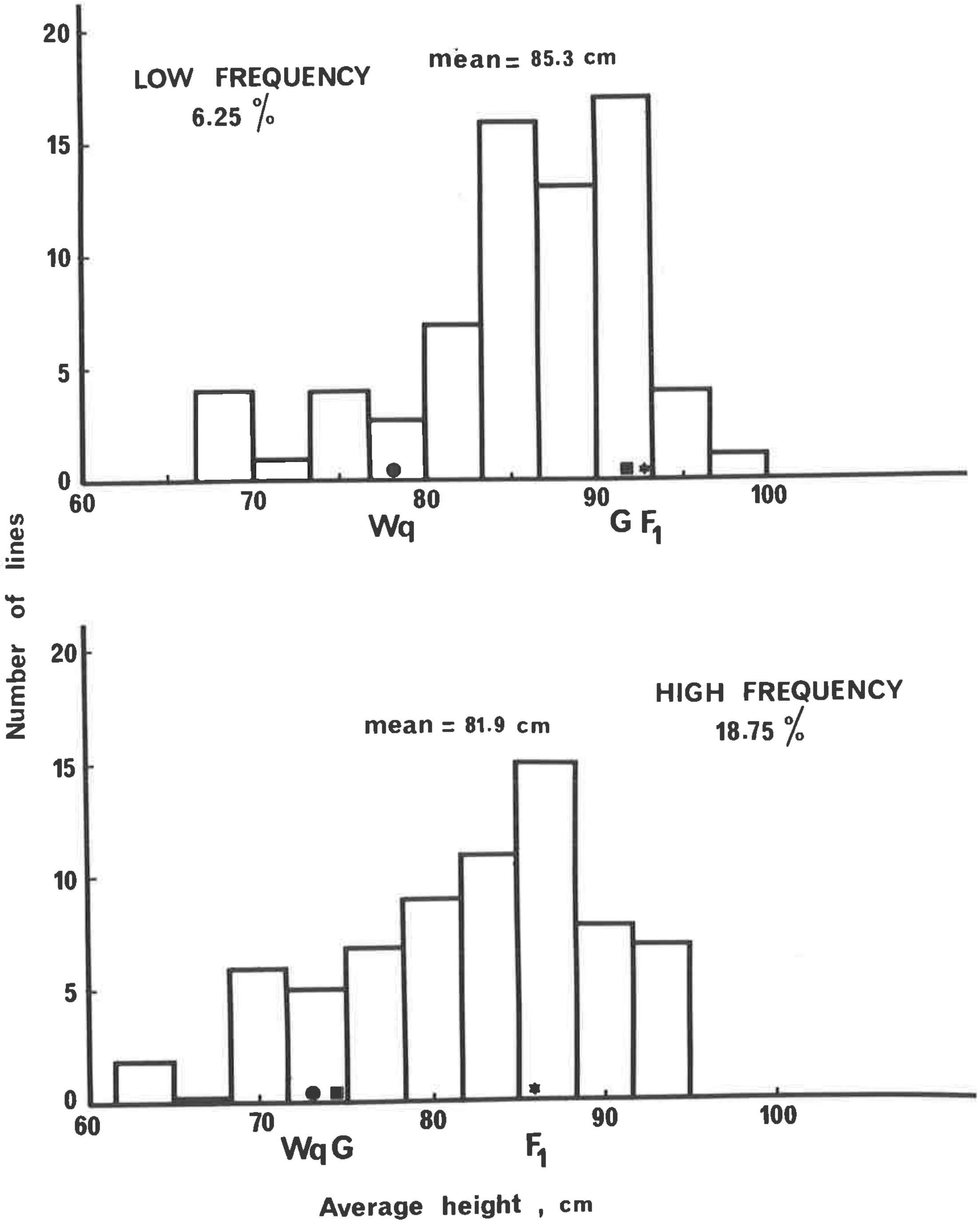
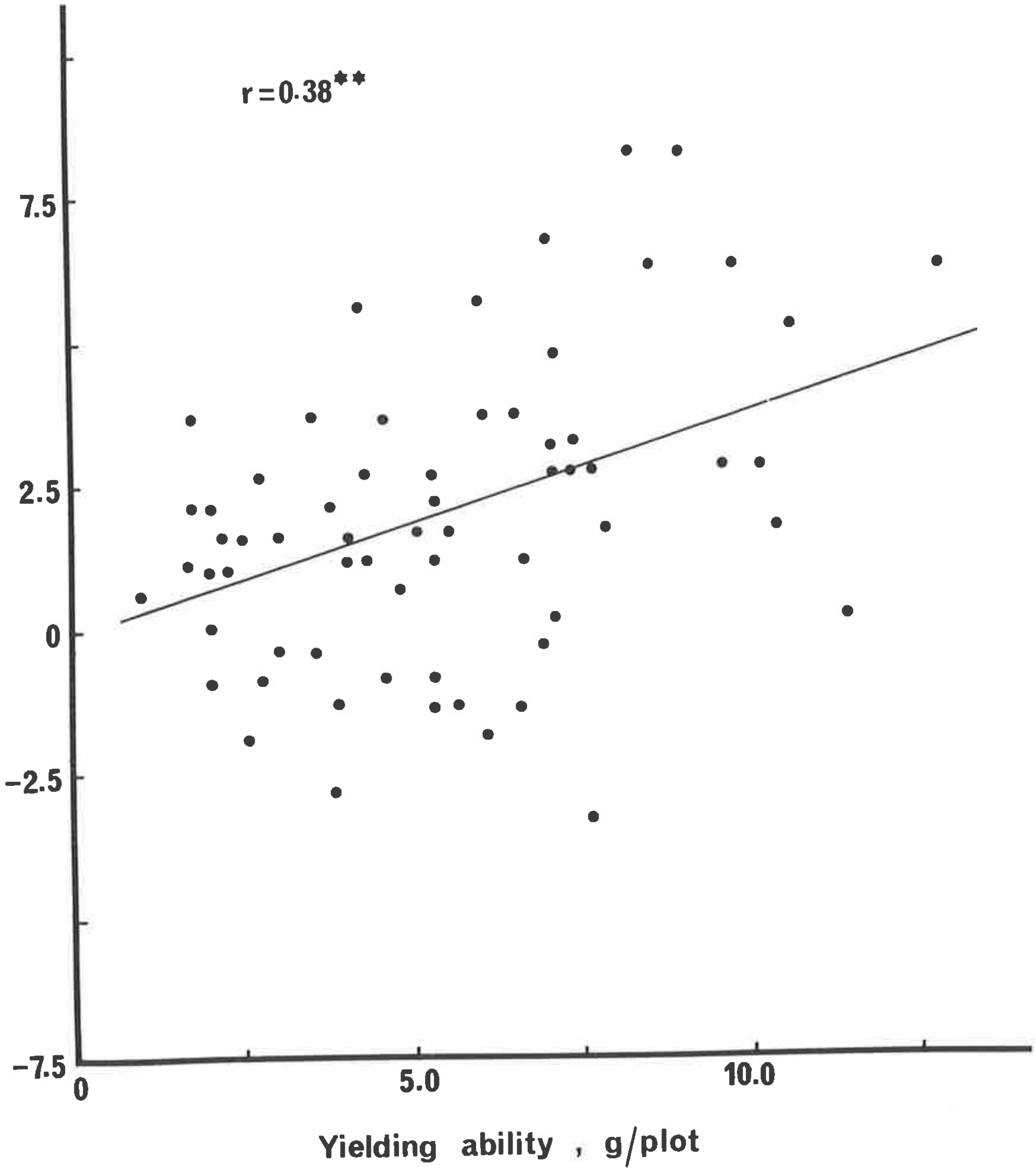

LOW FREQUENCY
6.25 %
mean = 85.3 cm
20
15
10
5
0
60
70
80
90
100
Wq
G F$_1$
Number of lines
HIGH FREQUENCY
18.75 %
mean = 81.9 cm
20
15
10
5
0
60
70
80
90
100
Wq G
F$_1$
Average height , cm

Figure 12: Relationship between f.d.a. (yield at low freq. - yield at high freq.) and yielding ability (mean of the yield at 2 frequencies) of 70 F_4 lines of (Wq*G) when grown in Halberd at 2 frequencies.

The fitted line has the equation $Y = -0.135 + (0.346) X$; where Y is the f.d.a. in gm per plot, and X is the mean yield of the line at the two frequencies. Correlation coefficient $r = 0.38$ **


r = 0·38**
7.5
2.5
0
−2.5
−7.5
F.d.a., g/plot
0
5.0
10.0
Yielding ability, g/plot

ranging from 28% to 38%. As the response from 50% to 4% was linear, it is possible that at even lower frequencies the advantage would be greater. Evidently f.d.a. in wheat is sufficiently common, and of sufficient magnitude, for it to influence the efficiency of single plant selection during wheat breeding.

F.d.a. might be a common phenomenon, as the experiments reported here were conducted under markedly different conditions. The seasons, 1972 and 1973, were quite different. The former had a below average rainfall and a relatively short period of crop growth; during the latter rainfall was appreciably above average. Also, Experiment I was grown as carefully spaced plants in small plots, whereas Experiment II involved a large number of genotypes grown in large contiguous plots resembling a commercial crop. Although each experiment was conducted in only one environment, a similar response to frequency was observed for the F_1 of (Wm*H) in both years, over different ranges of frequencies and in different mixtures.

One major restriction of using hybrid seeds was that they were available in limited numbers. Hence, it was not possible to allocate the different frequencies (Experiment I) in a completely randomized design as this would have required considerably more hybrid seeds for the borders, especially at high frequencies. This was overcome by growing the frequencies in ascending order, although one criticism of such a design was that the differences in yield of the hybrids at different frequencies could have been a systematic effect if there had been a fertility trend in the same direction as frequencies. However, the non-significant regression of the Halberd plants (Tables 3 and 4) with frequency indicated that this was not a factor. Furthermore, the results for each replicate separately showed that the hybrids had higher yields at the low than at the high frequency. It was unlikely that a similar trend in soil fertility occurred in all four replicates, especially as the paired replicates were laid out in opposite directions.

Since grain size did not change with frequency (Tables 3 and 4), the changes in grain yield were associated closely with changes in the number of grains per plant, which, in an agricultural population, is the factor most commonly influencing the fitness of the genotype (Palmer, 1952; Suneson, 1949). Hence, the f.d.a. represents the action of frequency-dependent selection in a single generation.

The results of Harding, Allard and Smeltzer (1966) for lima bean were much more pronounced than for the work reported here. In mixtures of hybrids and parents, the test genotype, a hybrid, had a yield reduction of 66.7% as its frequency increased from 2% to 16%. At the latter frequency, it was found that the fitness of the hybrids was equal to those of the parents. However, as has been pointed out, since seed coat pattern is maternally inherited (Allard, 1953) and since the hybrids and parents in experiments of Harding _et al_. came from the same bulk of an advanced generation, any error in identification of the homozygous lines particularly at the 2% frequency, could result in an over-estimation of the fitness of the heterozygotes. Khalifa and Qualset (1974) found that when two genotypes of wheat were mixed in all combinations at 12.5% frequency increments, grain yield of the stronger competitor was reduced from a relative value of 1.33 at 12.5% frequency to 0.80 in pure stand, a reduction of 39.8% which is comparable to the present findings. However, its yield in pure stand was lower than that of the pure stand of the poorer competitor by 20%.

As a marked f.d.a. effect was evident in experiment I, it was expected that the neighbouring background plants might show an inverse effect; that is a reduced yield when neighbouring a hybrid plant at low frequency. In fact, the Halberd plants showed no significant trend. A partial explanation is that the Halberd plants were not grown at low frequencies, but between 50% and 96%. Hence they were always in competition with a high proportion of like genotypes. This was supported by the non-significant difference in the yield of the two groups of Halberd, one immediately adjacent and the other in one position removed from the hybrid, at the two lower frequencies (Tables 7 and 8). The mean yields were 1.73 and 1.80 g/plant respectively for the (Wm*H) experiment, and 1.70 and 1.66 g/plant for the (Wq*H). Furthermore, there was some evidence that the hybrid did not show an appreciable f.d.a. at high frequencies. Extrapolating from the results depicted in Figures 5a and 6a to pure stand, (Wm*H) would have been expected to yield only 0.94 g/plant and (Wq*H) 0.75 g/plant. In an adjacent, comparable experiment, B. Hare (1976) recorded 1.78 and 1.62 g/plant, respectively, for pure stands of these same hybrids; yields which are nearer to those measured at the 50% frequency in experiment I than those predicted from the regression.

The results of Harding, Allard and Smeltzer (1966) have been re-examined by Allard and Adams (1969b). They showed that the best fit to the data was given by an exponential function which suggests that the linear relationship of fitness on frequency of the heterozygotes was unlikely to be maintained at higher frequencies. Khalifa and Qualset (1974) however reported an effect that was linear between 12.5% and 100%. It was possible that such an effect was due to a strong competition for light between a tall and a dwarf variety grown at a high seeding rate (90 kg/ha), and at a high altitude (1219m elevation) at one of the locations.

It has been suggested (Harding et al., 1966) that frequency-dependent selection was due to competition between plants of the same genotype. This theory could also be advanced to explain the apparently curvilinear relationship between yield and frequency of the hybrids beyond the 50% level and the non-significant regression of the Halberd in this experiment. Referring to the positions of plants in Figure 3, it can be seen that the distances between successive hybrids at the five frequencies were 33.5, 26.8, 20.1, 13.4 and 9.5cm (= $6.7 \times \sqrt{2}$, the diagonal distance), respectively. The regression coefficients of grain yields per plant and distances were positive and significantly different from zero ($p < 0.05$). The correlations between yields and distances were 0.501* and 0.505* for (Wm*H) and (Wq*H), respectively. If the hybrids had been grown at frequencies between 50% and 100% (which would correspond to the frequencies of Halberd), the distances between successive hybrids would have changed only from 9.5 to 6.7 cm. Hence, they were always in close proximity to plants of the same genotype. The physical basis of f.d.a. is further investigated in the next section but it is sufficient to say that the relationship between yield and frequency is inversely related to that between yield and distance between plants of the same genotype and hence depends upon the interaction between these plants in the exploitation of the environment.

During the growth of plants in Experiment I, measurements were made of plant characters which can be used as indicators of vigour. Besides yield, yield components and mature plant height, all the other measurements (Tables 3 and 4) had regession coefficients with the same sign as that for grain yield. In the later stages of growth these measurements were statistically significant. Thus, the f.d.a. would appear to result from a gradual amplification of an early advantage gained by the genotype when grown at low frequency.

It is unlikely that either final plant height or time of maturity is a major factor in f.d.a. Differences in plant height are only expressed after stem elongation, and in a Mediterranean type of climate, where there is ample illumination in the latter part of the season, any advantage to taller plants through additional light interception is likely to be negligible. Also, the f.d.a. was nearly as large in Experiment IIB, where most of the hybrids were shorter than the background Halberd, as it was in Experiment IIA, where the hybrids were taller than Wariquam. Furthermore, in the same experiment, consistent relationships between maturity and f.d.a. were absent.

It is possible that "disease escape or resistance" was an important factor in f.d.a. In experiment IIA, where Halberd was grown as test plants in Wariquam, a rust resistant background, it was relatively rust-free and yielded 2.58 and 2.02 g/plant at the low and high frequencies respectively (Figure 7). In experiment IIB, where Halberd was grown as the background stand, the plants were heavily infected with rust and had very low yield. Since no measurements were made of the background plants in Experiments IIA and IIB, no assessment could be made of the advantage gained through protection from disease of the Halberd plants at low frequencies. In experiment IIA, the background Wariquam could have acted as spore traps and as a barrier to spore movement and hence reduced the rate at which infection developed on the Halberd plants.

The highly significant correlations between the mean yield and the f.d.a. of the lines in experiment II are notable (Figures 9 and 12). F.d.a. is an aspect of relative competitive ability. High yielding genotypes tend to have high f.d.a. and this is likely to be important in the changes which occurred in Composite Crosses. The estimates of f.d.a. for practically all the lines in experiments IIA and IIB were positive, despite the relatively small range in frequencies (effectively 12.5%). The highly consistent increase in yield associated with the lower frequency resulted in the very highly significant variance ratios for frequency in Tables 9 and 10. The absence of any significant interaction in the same tables supports the generality of the conclusion that f.d.a. is widespread in wheat.

B. THE NATURE OF FREQUENCY-DEPENDENT ADVANTAGE

a. MECHANISMS OF FREQUENCY-DEPENDENT ADVANTAGE IN WHEAT

1. Introduction.

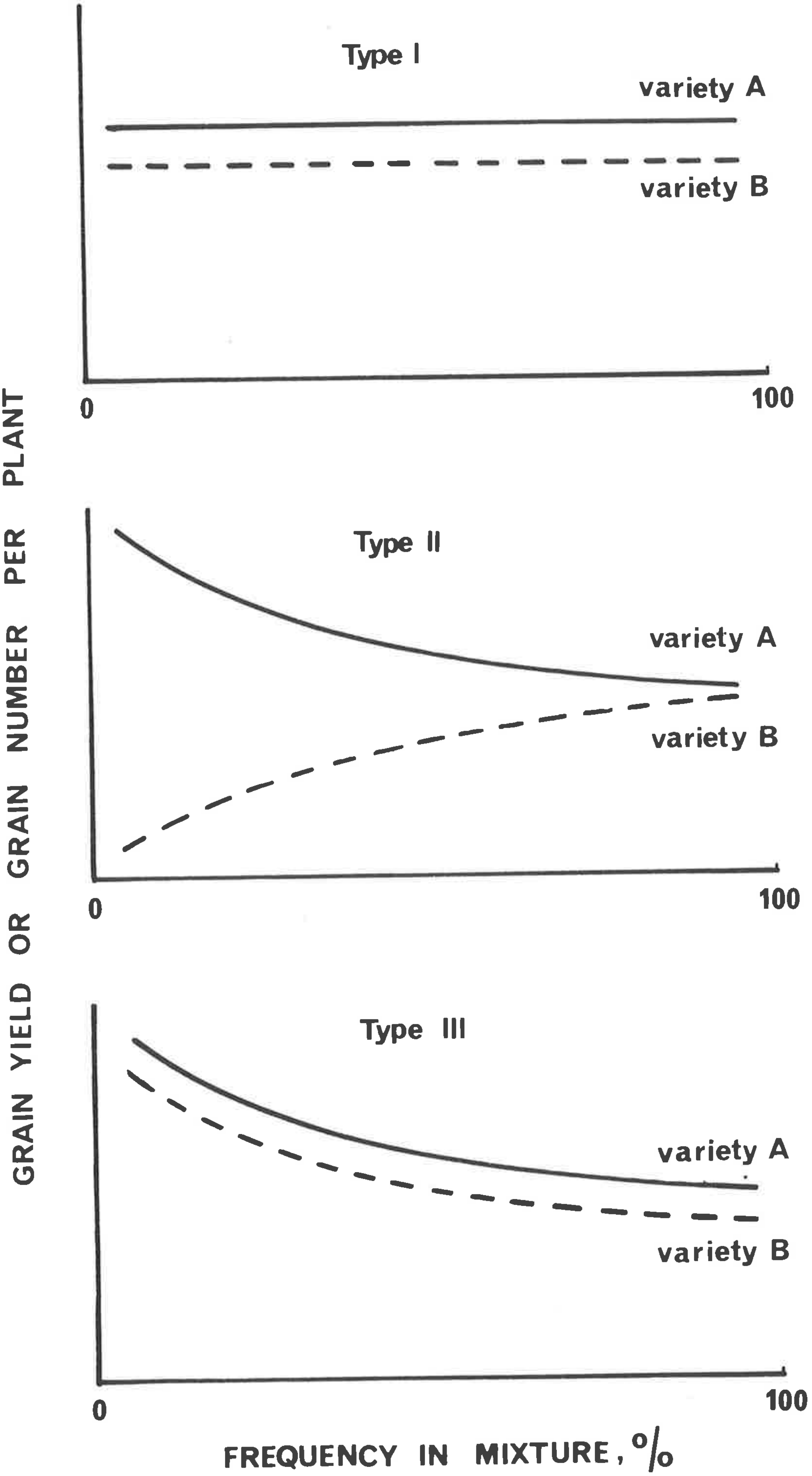
The occurrence of f.d.a. in the previous experiments and the observation that this phenomenon can lead to stable polymorphisms (Harding et al., 1966; Schutz, Brim and Usanis, 1968) encouraged further exploration into the mechanism which confers the advantage.

It has been shown that the performance of an individual in a mixture depends on the genotype with which it is associated (Allard and Adams, 1969a for wheat and barley; Sakai, 1961 for rice; Schutz and Brim, 1967 for soybeans and deWit, 1960 for grass species). All forms of interaction between two or more genotypes, of which f.d.a. is only one, have been called competition, an emotive description which has not helped clarify the subtle relationships which appear to be involved. Obviously, as individual plants become more crowded, the sooner does a plant's exploitation of its surroundings impinge on that of its neighbours'. Following this it is important to distinguish between two aspects of competition. One, where the plants' nutrients or physical requirements are equally useful to both genotypes at the same time. And the second, where the plants are co-exploitative, with one being able to use some resource more effectively at one time than the other. Clements (1904) pointed out that "competition" actually referred to the ability of an individual plant to exploit its own environment insofar as it had been altered by the presence of neighbours.

Both the discovery that natural populations of self-pollinated species are mixtures of many genotypes (Allard, 1965) and the common observation that two component mixtures often slightly outyield the average of the individual pure stands (Trenbath, 1974) suggest that the basic models of the yield of individual plants in various frequencies in a mixed population should be examined. Figure 13 illustrates the three major types of response that can be expected. Simplest of these, Type I, occurs when there is no f.d.a., and one genotype consistently outyields the other. When these yields differ from those measured in pure stands, then the two varieties differ in their "competitive" abilities. If genotypes which expressed a high f.d.a. were also those with a high competitive ability, then the Type II response might be expected. The higher yielding genotype would be expected to be even higher yielding when it occurred at a low frequency when it would have a greater proportion of weakly competitive neighbours. Conversely, the yield at high frequencies of the strong competitor would not be expected to be very different from yield in pure stand, but the weak competitor would be markedly reduced by the presence of a large proportion of such neighbours.

Neither of these models, except by annual fluctuations in selective values, are likely to lead to the stable heterogeneous populations (Allard, 1965; Allard, Jain and Workman, 1968) or mixed agricultural populations (Harlan and Martini, 1938; Suneson, 1956; Lohani, 1970). Type III reactions will often lead to stable heterogeneous populations (Allard and Adams, 1969b), providing that at some (low) frequency the number of seeds produced by the variety B equals that from A at a high frequency. The physical basis of these Type III reactions are likely to be either due to slightly different requirements (particularly at different times) for some resource in restricted supply or differential susceptibility to specific races of plant pathogen. It is also possible that

Figure 13: Hypothetical types of response in grain yield or grain number per plant of two cereal varieties grown together at different frequencies.


Type I
variety A
variety B
0
100
Type II
variety A
variety B
0
100
Type III
variety A
variety B
0
100
GRAIN YIELD OR GRAIN NUMBER PER PLANT
FREQUENCY IN MIXTURE, %

plants produce substances which alter the growth of their neighbours.

The experiments described in this section sought answers to the following questions:

(i) Which of the types of reaction (Figure 13) could be expected in mixtures of strong and weak competitors?

(ii) Did the hybrid of these types show f.d.a. when in mixtures with either parent?

(iii) Was the physical basis of f.d.a. very specific to a particular genotype? Would the advantage be expressed in mixtures of other varieties?

(iv) Was the interaction due to competition below or above ground?

2. Materials and methods.

2.1 Experiment III.

Experiment III consisted of 2 sub-experiments of the same design, each with 3 replicates. In each experiment, genotypes being assessed were grown at various frequencies in a background of other genotypes. They will be referred to as the test and background genotypes, respectively. In experiment IIIA, these were as follows:

Test genotype	Background genotype	Frequency of test genotype
(Warimek * Timgalen) F_1 subsequently abbreviated to (Wm*T)	- Warimek	6.25% and 25%
	- Timgalen	"
	- Mixture *	"
Warimek	- Timgalen	"
	- Mixture *	"
Timgalen	- Warimek	"
	- Mixture *	"

* A mixture of Warimek, Timgalen, Wariquam, Halberd and Gabo.

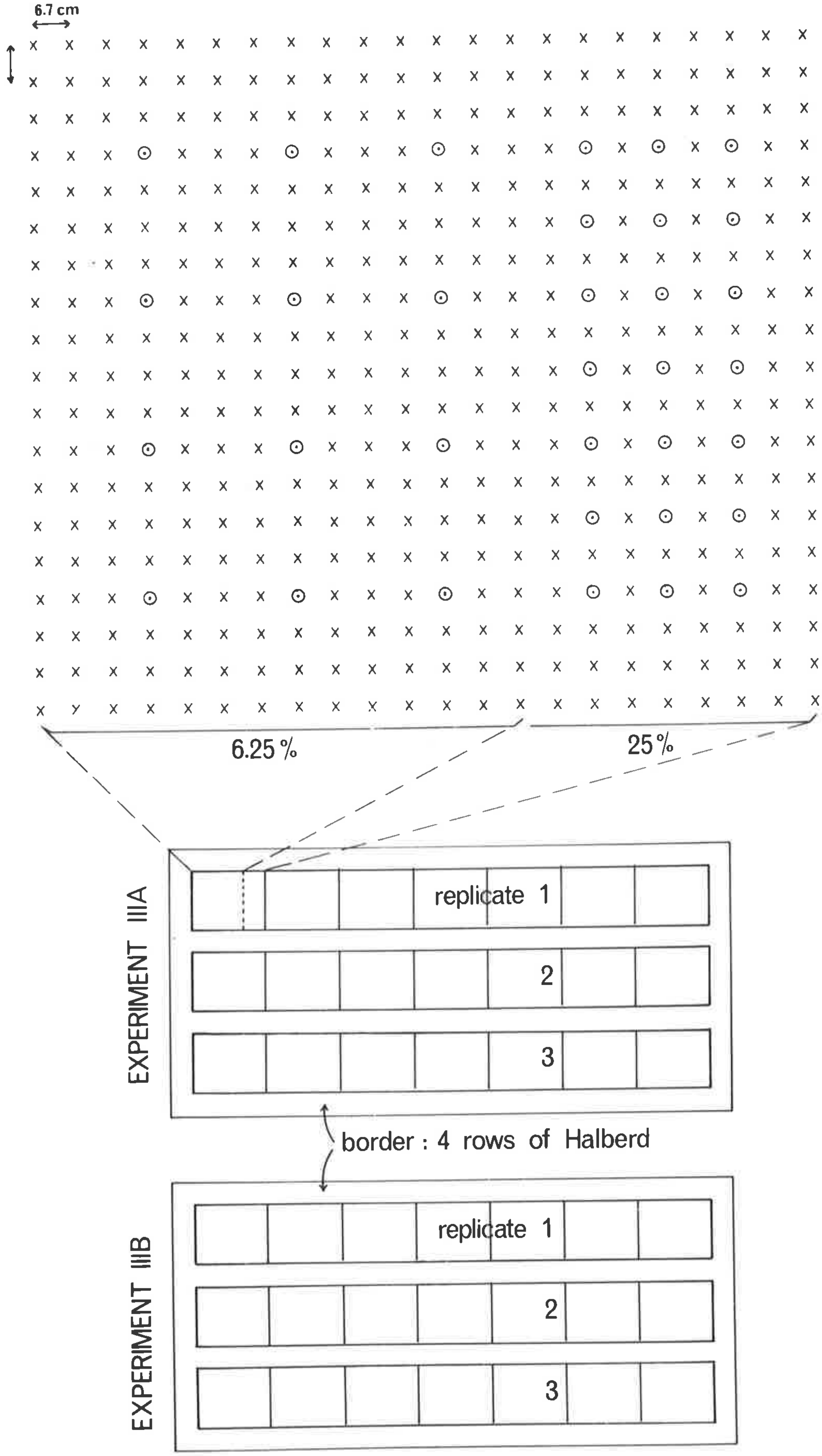
In experiment IIIB, the test genotypes were Wariquam and Gabo, their F_1 hybrid subsequently abbreviated to (Wq*G); and the background genotypes were Wariquam, Gabo and the mixture used in experiment IIIA. The mixture, which consisted of the five pure lines in equal proportions by weight, was sown from a random sub-sample of the bulk.

The experiments were planted on July 12 and 13, 1972 at Roseworthy Agricultural College. Each experiment was a split-plot design, with backgrounds as main plots and frequencies as sub-plots (Figure 14). Seeds were sown individually with a distance of 6.7cm in both directions between plants, giving a density of 222 plants per square metre, which is approximately that used commercially. The positions of the test plants were marked by small wooden sticks to enable their identification through the growing season. Each treatment in each replicate consisted of 12 test plants at the low frequency and 21 test plants at the high frequency, all of which were harvested. The same number of background plants immediately adjacent to the test plants were also harvested. All these were measured for height, number of spikes, number of spikelets, total weight and grain weight.

Figure 14: Field layout of Experiments IIIA and IIIB. At the top is a detailed plan of one treatment.

x x x positions of the test plants (F_1 or parents) occurring at 2 frequencies: 6.25% and 25%.

● ● ● positions of the background plants.


6.7 cm
6.25 %
25 %
EXPERIMENT IIIA
replicate 1
2
3
border : 4 rows of Halberd
EXPERIMENT IIIB
replicate 1
2
3

2.2 Experiment IV.

Experiment IV was conducted in an open sided glass house. Plants of the different genotypes were grown in slot-like containers which represented rows (Plates 2 and 3).. Thus plants occurring in the same row could compete for light, nutrients and water; whereas plants occurring in adjacent rows competed only for light.

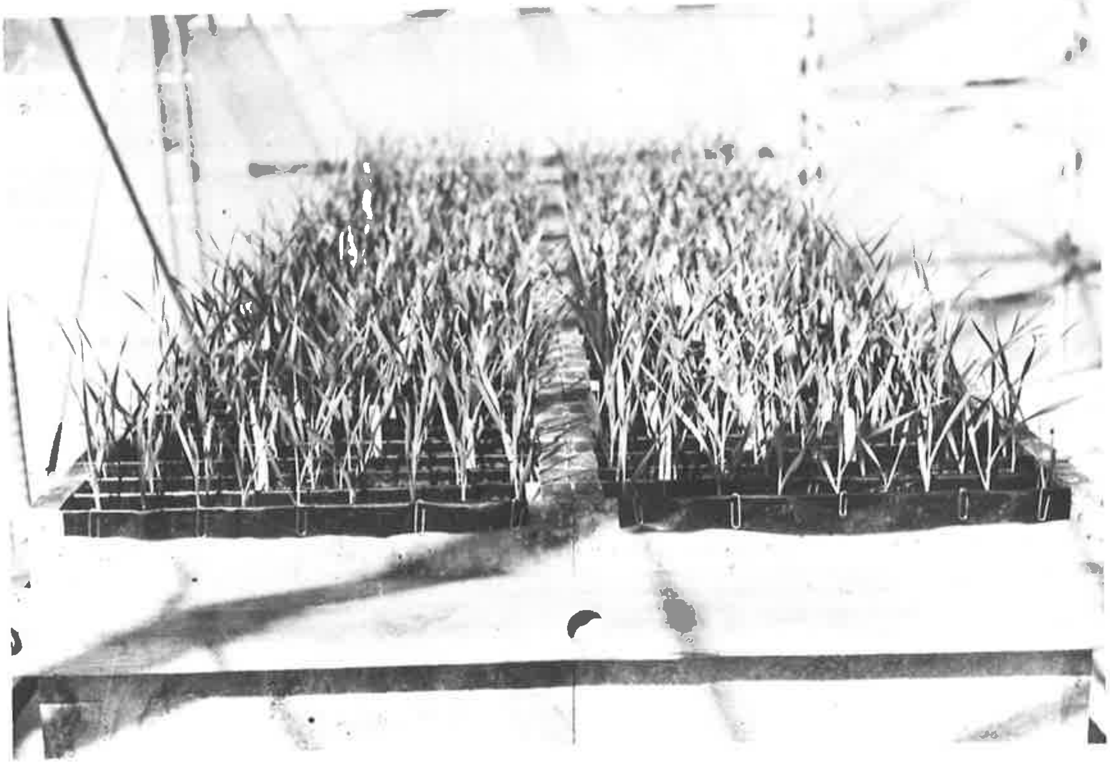
F_1 plants of the cross (Wm*T) were grown in each of the parental stands, Warimek and Timgalen, at 2 frequencies: 11.1% (1 plant in the total of 9 in a container) and 44.4% (4 plants in 9). At the 44.4% frequency of the hybrid, each container with the hybrids was adjoined by a container with only the background genotype.

Seeds were germinated in petri-dishes and planted on July 15, 1973. Nine plants, 5cm apart, were grown in each container in a mixture of 50% sand, 50% peat and all the required nutrients (Baker, 1957). The distance between plants in adjacent containers was also 5cm. Soil moisture was regulated to avoid stress, but no further nutrients were added.

The containers in which the plants were grown have been described by Stern and Donald (1962). The ends and the base of each container were made of wood and the sides of galvanized iron sheeting. Each container had the internal dimensions 5cm x 50cm and 50cm deep and was lined with black polyethylene. A block of 100 containers was held together by clamps.

The experiment, consisting of 4 replicates, was a split-plot design, with the different genotypic backgrounds as main plots and the

Plates 2 and 3: Side views of Experiment IV, 2 weeks after planting. Plants were grown in slot-like containers, dimensions 50cm x 5cm x 50cm, held together by carpenter clamps. The positions of the hybrids were marked by wooden sticks. A dark, fine plastic mesh was used as the border to reduce side lighting. The border has been removed during the photography.



two frequencies as sub-plots. In each replicate and in each background stand, there were 6 hybrid plants at the low frequency and 8 hybrid plants at the high frequency. All hybrid plants were marked by small wooden sticks and all were measured for the characters described in Tables 16, 17 & 18. A parental plant adjacent to each hybrid and in the same container was measured as was a parental plant in the adjoining container.

The whole block of containers was surrounded by a dark, fine nylon mesh which acted as a border by reducing side lighting. It was raised with the growth of the plants.

3. Results

3.1 Experiment IIIA.

3.1.1 The performance of the F_1, Warimek and Timgalen in various backgrounds.

(i) (Wm*T)

The results for (Wm*T) in 3 different backgrounds at 2 frequencies are shown in Table 11. The hybrid had a significantly higher yield in the Warimek than in the Timgalen and the Mixture stands. This higher yield was associated with significant differences in number of spikes and number of spikelets per plant. None of the characters measured showed a significant difference due to frequencies, nor to the interaction between frequencies and stands.

(ii) Warimek.

The results for Warimek are shown in Table 12. Warimek plants did not differ significantly when grown in Timgalen and in Mixture stands,

Table 11: Mean performance of (Wm*T) in 3 backgrounds at 2 frequencies and results of analyses of variance on the same characters.

(a) Height per plant, cm.

	Background			
Frequency	Warimek	Timgalen	Mixture	Mean
6.25%	65.23	65.67	65.27	65.39
25.00%	63.42	64.13	64.56	64.04
Mean	64.32	64.90	64.91	64.71

(b) Number of spikes per plant.

	Background			
Frequency	Warimek	Timgalen	Mixture	Mean
6.25%	3.0	2.5	2.3	2.6
25.00%	2.7	2.4	2.4	2.5
Mean	2.8	2.4	2.4	2.5

LSD (0.05) between backgrounds = 0.4

(c) Number of spikelets per plant.

	Background			
Frequency	Warimek	Timgalen	Mixture	Mean
6.25%	44.5	35.8	34.5	38.3
25.00%	38.4	35.7	33.9	36.0
Mean	41.4	35.7	34.3	37.1

LSD (0.05) between backgrounds = 5.5

(d) Total weight per plant, g.

	Background			
Frequency	Warimek	Timgalen	Mixture	Mean
6.25%	5.99	4.46	4.35	4.93
25.00%	5.08	4.85	3.97	4.63
Mean	5.54	4.65	4.16	4.78

(e) Grain weight per plant, g.

	Background Warimek	 Timgalen	 Mixture	 Mean
Frequency				
6.25%	2.50	1.79	1.74	2.01
25.00%	2.08	1.93	1.61	1.87
Mean	2.29	1.86	1.68	1.94

LSD(0.05) between backgrounds = 0.42

Analyses of variance

Character	Backgrounds	Frequencies	Backgrounds x Frequencies
Height	NS	NS	NS
No. of spikes	*	NS	NS
No. of spikelets	*	NS	NS
Total weight	NS	NS	NS
Grain weight	*	NS	NS

Table 12: Mean performance of Warimek in 2 backgrounds at 2 frequencies and results of analyses of variance on the same characters.

(a) Height per plant, cm.

	Background		
Frequency	Timgalen	Mixture	Mean
6.25%	59.16	62.08	60.62
25.00%	62.64	61.80	62.22
Mean	60.90	61.94	61.42

(b) Number of spikes per plant

	Background		
Frequency	Timgalen	Mixture	Mean
6.25%	2.1	2.2	2.1
25.00%	2.3	2.4	2.4
Mean	2.2	2.3	2.2

(c) Number of spikelets per plant.

	Background		
Frequency	Timgalen	Mixture	Mean
6.25%	30.3	34.1	32.2
25.00%	34.7	37.0	35.8
Mean	32.5	35.6	34.0

(d) Total weight per plant, g.

	Background		
	Timgalen	Mixture	Mean
Frequency			
6.25%	3.41	4.35	3.88
25.00%	4.10	4.50	4.30
Mean	3.75	4.42	4.09

(e) Grain weight per plant, g.

	Background		
	Timgalen	Mixture	Mean
Frequency			
6.25%	1.42	1.76	1.59
25.00%	1.68	1.88	1.78
Mean	1.55	1.82	1.68

Analyses of variance

Character	Backgrounds	Frequencies	Backgrounds x Frequencies
Height	NS	NS	NS
No. of spikes	NS	NS	NS
No. of spikelets	NS	NS	NS
Total weight	*	NS	NS
Grain weight	NS	NS	NS

except for total weight per plant, where it was significantly lower in the Timgalen stand. None of the comparisons for frequencies nor for the interactions between frequencies and stands was significant.

(iii) Timgalen.

The results for Timgalen are shown in Table 13. Timgalen plants had significantly higher yields at the low frequencies than at the high frequencies. It also had a higher yield in the Warimek than in the Mixture stand. The non-significant interaction indicated that the effect of f.d.a. was similar in both stands. The other characters followed the same pattern as grain yield.

It should be noted that the mixture consisted of approximately 20% of each of the five pure lines; hence the actual frequencies of Warimek and Timgalen in this bakcground were approximately 26.5% and 45% for the corresponding frequencies 6.25% and 25% of the test plants, respectively. Table 13e shows that the yield of Timgalen at 25% frequency in the Warimek stand was approximately equal to its yield at the test plant frequency of 6.25% (i.e. actually 26.5%) in the Mixture. However, its yield was significantly increased at a low frequency (6.25% in the Warimek stand) and was significantly reduced at a high frequency (45% in the Mixture).

3.1.2 Frequency-dependent advantage of Warimek and Timgalen.

In this experiment, Warimek and Timgalen were grown both as test plants and as background genotypes. It is possible therefore to consider the yields of Warimek when grown in Timgalen at 6.25% and 25% (test plants)

Table 13: Mean performance of Timgalen in 2 backgrounds at 2 frequencies and results of analyses of variance on the same characters.

(a) Height per plant, cm.

	Background		
Frequency	Warimek	Mixture	Mean
6.25%	72.44	69.44	70.94
25.00%	68.83	68.84	68.83
Mean	70.63	69.14	69.88

(b) Number of spikes per plant.

	Background		
Frequency	Warimek	Mixture	Mean
6.25%	3.5	2.9	3.2
25.00%	2.7	2.6	2.6
Mean	3.1	2.7	2.9

(c) Number of spikelets per plant.

	Background		
Frequency	Warimek	Mixture	Mean
6.25%	53.5	41.9	47.7
25.00%	40.4	37.3	38.8
Mean	46.9	39.6	43.3

(d) Total weight per plant, g.

	Background		
Frequency	Warimek	Mixture	Mean
6.25%	7.86	6.00	6.93
25.00%	5.98	5.20	5.59
Mean	6.92	5.60	6.26

(e) Grain weight per plant, g.

	Background		
Frequency	Warimek	Mixture	Mean
6.25%	3.14	2.20	2.67
25.00%	2,28	1.88	2.08
Mean	2.71	2,04	2.37

Analyses of variance

Character	Backgrounds	Frequencies	Backgrounds x Frequencies
Height	NS	NS	NS
No. of spikes	NS	NS	NS
No. of spikelets	NS	*	NS
Total weight	NS	*	NS
Grain weight	NS	*	NS

and at 93.75% and 75% (background genotypes). Similar considerations may be given to Timgalen grown in Warimek. The graph of the results for grain yield is presented in Figure 15 and a summary of the regression coefficients for other characters in Table 14. The mean per frequency per replicate was used in the regression on frequency.

Table 14: Regression coefficients of Timgalen and Warimek at 4 frequencies.

Character	Timgalen	Warimek
Height	-0.072	0.075 *
No. of spikes	-0.008 *	0.002
No. of spikelets	-0.162 *	0.043
Total weight	-0.031 *	0.011
Grain weight	-0.013 *	0.004

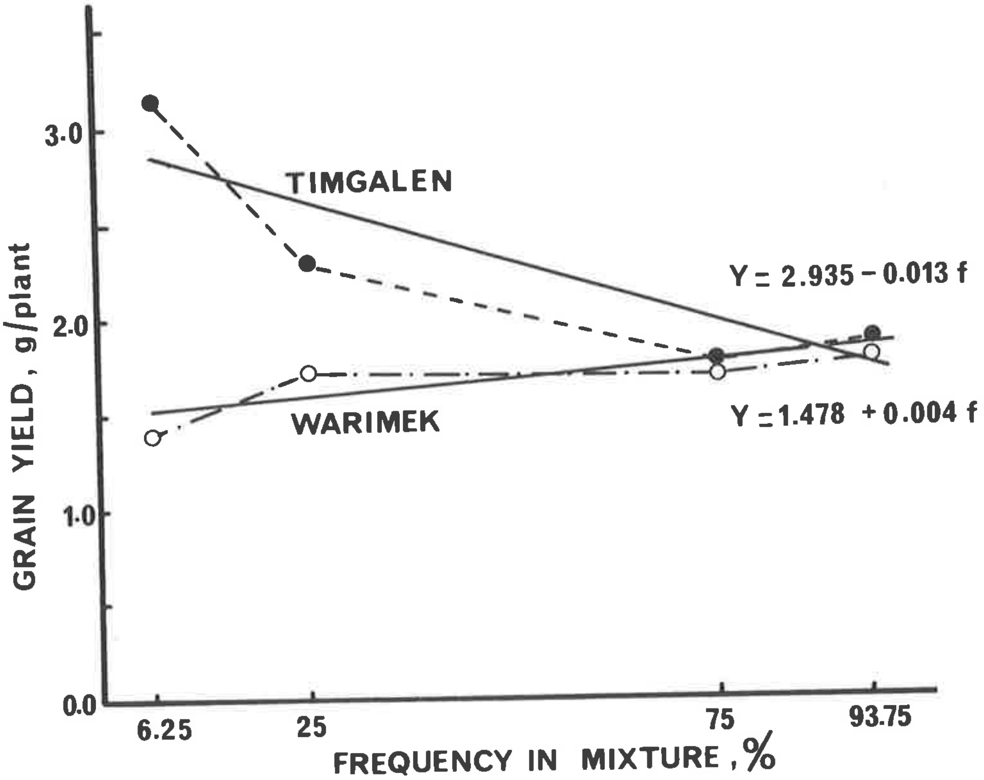
Timgalen plants yielded significantly more grain at low than at high frequencies. The mean yield was 3.14 g/plant at 6.25% frequency as opposed to 1.88 g/plant at 93.75% frequency, a reduction of 40%. Although the linear regression has been calculated, Figure 15 indicates that the reduction in grain yield was largest between 6.25% and 25%. At higher frequencies, the reduction in yield was negligible. Other characters, except plant height, also showed significant reductions with increasing frequencies.

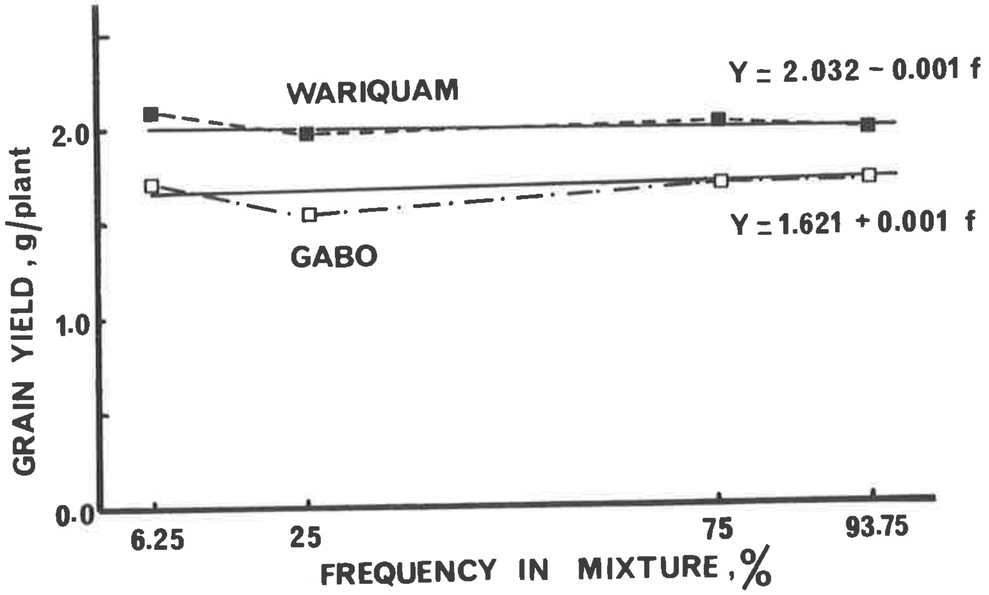
Grain yield per plant and most of the other plant characters of Warimek did not differ significantly with increasing frequencies, although most had small positive regression coefficients.

Figure 15 shows that at a high frequency (93.75%), the yields of

Figure 15: Relation between grain yield per plant of Timgalen and Warimek when each occurred in a stand of the other at 4 frequencies: 6.25%, 25%, 75% and 93.75%.

Figure 16: Relation of grain yield per plant of Wariquam and Gabo when each occurred in a stand of the other at 4 frequencies: 6.25%, 25%, 75% and 93.75%.


GRAIN YIELD , g/plant
3.0
2.0
1.0
0.0
TIMGALEN
WARIMEK
Y = 2.935 − 0.013 f
Y = 1.478 + 0.004 f
6.25
25
75
93.75
FREQUENCY IN MIXTURE , %


GRAIN YIELD , g/plant
2.0
1.0
0.0
WARIQUAM
GABO
Y = 2.032 − 0.001 f
Y = 1.621 + 0.001 f
6.25
25
75
93.75
FREQUENCY IN MIXTURE , %

Timgalen and Warimek were quite similar (1.88 and 1.81 g/plant, respectively) despite the twofold difference at low frequency.

3.2 Experiment IIIB.

3.2.1 The performance of the F_1, Wariquam and Gabo in various backgrounds.

In contrast to the results outlined above, there were negligible differences across frequencies for (Wq*G), Wariquam and Gabo. The differences between background stands were also non-significant.

3.2.2 Frequency-dependent advantage of Wariquam and Gabo.

Wariquam and Gabo, as test plants and background neighbours, could also be considered as being grown at 4 frequencies: 6.25%, 25%, 75% and 93.75% in a stand of one another.

The results for grain yield per plant are shown in Figure 16 and a summary of other characters in Table 15. There was no significant regression on frequency for any character. This is consistent with the results for Wariquam and Gabo in the previous section (3.2.1).

Table 15: Regression coefficients of Wariquam and Gabo at 4 frequencies.

Character	Wariquam	Gabo
Height	0.051	-0.042
No. of spikes	0.001	0.003
No. of spikelets	0.027	0.037
Total weight	0.000	0.002
Grain weight	-0.001	0.001

3.3 Experiment IV.

3.3.1 The performance of (Wm*T) in 2 parental stands at 2 frequencies.

The yield of the hybrid in the Warimek stand was more than double that in the Timgalen stand (Figure 17). The mean yield per plant in the two stands were 2.89g and 1.07g, respectively. The effect due to the background stands was highly significant for all characters, except tiller numbers at 6 weeks (Table 16). The main effects for frequencies in the analysis were non-significant, but the interactions between frequencies and stands were significant for grain weight and number of grains per plant. The hybrid had higher yield at the low than at the high frequency in the Warimek stand, but similar yields at the two frequencies in the Timgalen stand. The results for other characters were similar. Thus, the hybrid grew faster, taller, produced more tillers and more grains in the Warimek than in the Timgalen stand. It showed an f.d.a. in the former stand but none in the latter.

3.3.2 Comparison between F_1 and parents at frequency 44.4%

The performance of the hybrid and each of the parents was compared at frequency 44.4%. This frequency (4 hybrids versus 5 parents) is sufficiently close to 50% to compare the hybrids and parents since the differences were due to genotypic effect rather than to frequency effect. Tables 17 and 18 show the mean performance of each genotype and the results of analyses of variance.

Figure 17: Grain yield per plant of (Wm*T) F_1 in the 2 parental stands, Warimek and Timgalen, and at 2 frequencies, 11.1% and 44.4%.

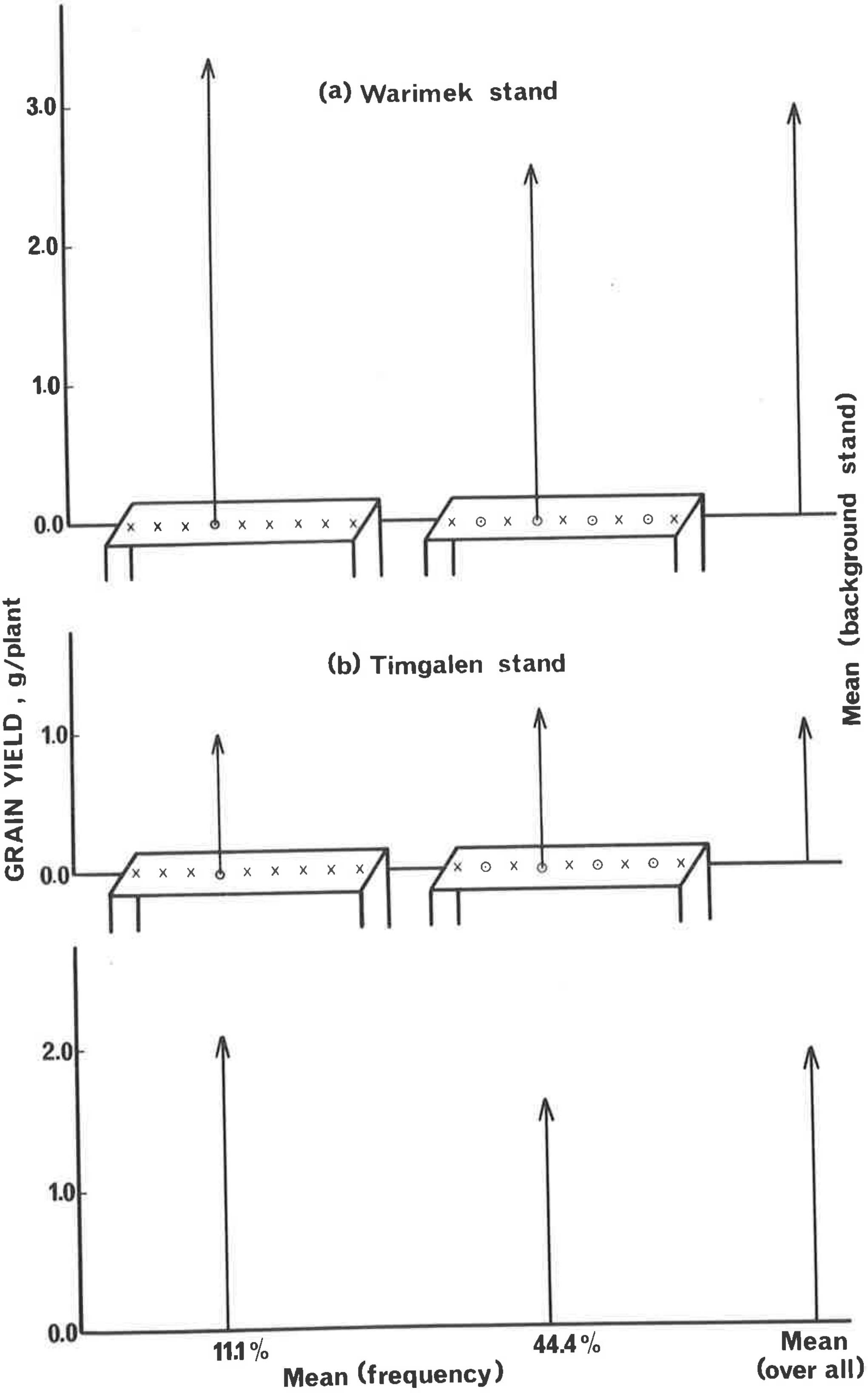

(a) Warimek stand
3.0
2.0
1.0
0.0
(b) Timgalen stand
1.0
0.0
2.0
1.0
0.0
GRAIN YIELD , g/plant
Mean (background stand)
11.1 %
44.4 %
Mean (frequency)
Mean
(over all)

Table 16: Mean performance per plant of the F_1 hybrid (Wm*T) in 2 backgrounds at 2 frequencies, and the results of the analyses of variance on the same characters.

Note: Frequency 1 = 11.1%; Frequency 2 = 44.4%

Stand 1 = Warimek; Stand 2 = Timgalen

(a) Tiller numbers (6wks).

Stand	Frequency		Mean
	1	2	
1	3.2	3.2	3.2
2	2.9	2.9	2.9
Mean	3.1	3.0	3.1

(b) Tiller length (6wks), cm.

Stand	Frequency		Mean
	1	2	
1	43.8	43.1	43.5
2	39.4	40.9	40.2
Mean	41.6	42.0	41.8

(c) Height (maturity), cm.

Stand	Frequency		Mean
	1	2	
1	90.7	88.0	89.4
2	79.5	75.5	77.5
Mean	85.1	81.8	83.4

(d) No. of spikes.

Stand	Frequency		Mean
	1	2	
1	3.2	2.3	2.8
2	1.6	2.0	1.8
Mean	2.4	2.2	2.3

(e) No. of spikelets.

Stand	Frequency		Mean
	1	2	
1	~~54.6~~	40.7	47.6
2	26.3	31.4	28.8
Mean	40.5	36.0	38.2

(f) No. of grains.

Stand	Frequency		Mean
	1	2	
1	74.7	56.7	65.7
2	22.6	24.9	23.8
Mean	48.7	40.8	44.8

(g) Total weight, g.

Stand	Frequency		Mean
	1	2	
1	8.45	6.53	7.49
2	3.11	3.58	3.35
Mean	5.78	5.05	5.42

(h) Grain weight, g.

Stand	Frequency		Mean
	1	2	
1	3.27	2.51	2.89
2	0.98	1.16	1.07
Mean	2.13	1.84	1.98

Table 16 continued: Analyses of variance

Character	Stands	Frequencies	Stands x Frequencies
Tiller no. (after 6 wks)	NS	NS	NS
Tiller length (")	*	NS	NS
Height (at harvest)	*	NS	NS
No. of spikes (")	*	NS	NS
No. of spiklets (")	**	NS	NS
No. of grains (")	**	NS	*
Total weight (")	**	NS	NS
Grain weight (")	**	NS	*

It is evident that the hybrid had significantly higher yield and higher expressions of other characters than Warimek. On the other hand, it was significantly lower yielding than Timgalen plants. The differences in grain yield between genotypes could apparently be attributed to differences in growth rates as shown by the result for tiller length in Table 18.

3.3.3 The effect on Warimek and Timgalen of competition with the hybrid.

For parental plants growing as neighbours to the hybrids, there were two types of competition. When in the same container as the hybrid, they competed for light, nutrients and water; when in adjacent containers, they would have competed only for light. A comparison of parental plants in these two situations gives an indication of the effect of competition with the hybrid.

Table 17: Comparison between (Wm*T) and Warimek at frequency of the hybrid of 44.4%. Means per plant and results of analyses of variance.

Character	Genotype means (Wm*T)	 Warimek	Differences
No. of tillers (6 wks)	3.2	3.0	NS
Tiller length ("), cm	43.2	40.9	NS
Height (maturity), cm	88.8	68.4	***
No. of spikes (")	2.4	1.0	***
No. of spikelets (")	42.1	17.9	***
No. of grains (")	56.8	23.5	***
Total weight ("), g	6.68	2.10	***
Grain weight ("), g	2.52	0.75	***

Table 18: Comparison between (Wm*T) and Timgalen at frequency of the hybrid of 44.4%. Means per plant and results of analyses of variance.

Character	Genotype means (Wm*T)	 Timgalen	Differences
No. of tillers (6 wks)	2.9	3.2	NS
Tiller length ("), cm	40.9	47.9	***
Height (maturity), cm	75.6	76.4	NS
No. of spikes (")	2.0	2.2	NS
No. of spikelets (")	31.4	35.4	NS
No. of grains (")	25.0	36.3	**
Total weight ("), g	3.58	4.90	**
Grain weight ("), g	1.16	1.56	*

(i) Warimek plants adjacent to the hybrids

Warimek plants did not differ significantly when neighbouring the hybrids at two frequencies (Table 19a). However, the plants in full competition with the hybrids had significantly lower yields than those which competed with the hybrids only for light (Figure 18a). The differences were highly significant for all characters measured at maturity (Table 19b).

(ii) Timgalen plants adjacent to the hybrids.

Timgalen plants did not differ significantly when neighbouring the hybrids at two frequencies (Table 20a). In this experiment, both Timgalen and Warimek occurred at high frequencies, 88.9% and 55.6%, respectively, so this lack of significance is not surprising.

Grain yield per plant of Timgalen did not differ significantly in the two types of competition with the hybrids (Figure 18b). Nor was the difference significant for any character measured (Table 20b). However, it is of interest to note that Timgalen showed exactly the opposite effect to that expressed by Warimek. Plants competing with the hybrids both above and below the soil were, in fact, slightly higher yielding than those competing with the hybrids only for light.

In this experiment, it was not possible to show the effect of competition for light alone. Since water was non-limiting, the competition must be due to that for nutrients. Competition with the hybrid for this factor reduced the yield of Warimek by 41.3% whereas the same type of competition increased the yield of Timgalen by 16.5%.

Table 19: Comparison between Warimek plants and results of analyses of variance.

(a) When neighbouring the hybrids at 2 frequencies, 11.1% and 44.4%.

(b) When in 2 types of competition, in the same container or in adjacent containers, with the hybrids.

	(a) Mean per plant at frequency			(b) Mean per plant when competed		
Character	11.1%	44.4%	Difference	Above	Above & Below	Difference
Tiller no. (after 6 wks)	2.8	2.9	NS	2.9	2.9	NS
Tiller length, cm (")	40.1	41.2	NS	41.1	40.2	NS
Height, cm (at harvest)	75.0	74.5	NS	79.4	70.2	**
No. of spikes (")	1.1	1.3	NS	1.4	1.0	**
No. of spikelets (")	20.3	23.3	NS	25.5	18.1	**
No. of grains (")	31.9	31.7	NS	39.1	24.4	***
Total weight, g (")	2.74	2.86	NS	3.38	2.22	**
Grain weight, g (")	1.14	1.06	NS	1.38	0.81	**

Figure 18: Mean yield per plant of Warimek and Timgalem when competing with the hybrid at the 2 frequencies:

- only above ground (plants in adjacent containers).
- above and below ground (plants in the same container).

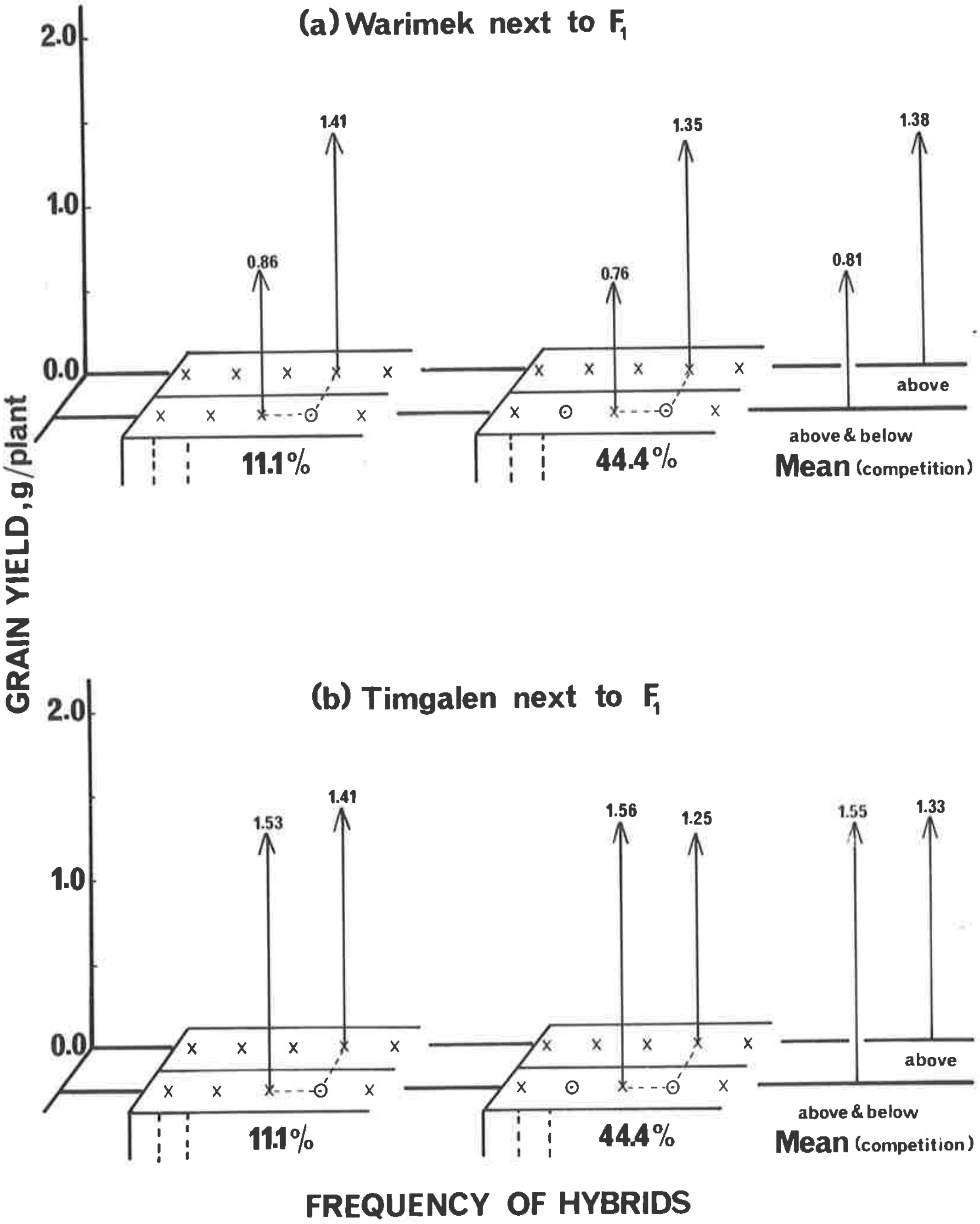

(a) Warimek next to F_1
2.0
1.0
0.0
1.41
0.86
1.35
0.76
1.38
0.81
above
above & below
11.1%
44.4%
Mean (competition)
(b) Timgalen next to F_1
2.0
1.0
0.0
1.53
1.41
1.56
1.25
1.55
1.33
above
above & below
11.1%
44.4%
Mean (competition)
GRAIN YIELD, g/plant
FREQUENCY OF HYBRIDS

Table 20: Comparison between Timgalen plants and results of analyses of variance

(a) When neighbouring the hybrids at 2 frequencies, 11.1% and 44.4%

(b) When in 2 types of competition, in the same container or in adjacent containers with the hybrids.

Character	(a) Mean per plant at frequency			(b) Mean per plant when competed		
	11.1%	44.4%	Difference	Above	Above & Below	Difference
Tiller no. (after 6 wks)	3.1	3.2	NS	3.2	3.2	NS
Tiller length, cm (")	46.2	47.4	NS	46.4	47.2	NS
Height, cm (at harvest)	76.3	74.9	NS	74.6	76.5	NS
No. of spikes (")	2.4	2.4	NS	2.4	2.4	NS
No. of spikelets (")	37.9	37.3	NS	38.1	37.1	NS
No. of grains (")	33.8	32.8	NS	30.5	36.1	NS
Total weight, g (")	4.46	4.63	NS	4.35	4.73	NS
Grain weight, g (")	1.47	1.41	NS	1.33	1.55	NS

4. Discussion.

An important consideration is the relation between f.d.a. and the competitive ability of a genotype. It is necessary to differentiate between two methods of defining competitive ability. One of these is the difference in plant yield between members of the genotype grown at a particular frequency in a mixture (usually 50%) and those grown in pure stand (Akihama, 1967; Trenbath, 1974). When the genotype has higher yield per plant in the mixture, it is regarded as a "good" competitor. The second is measured by growing a genotype in several paired combinations. If this is undertaken for several genotypes, the variety with the highest mean yield is regarded as the "best" competitor. It is theoretically possible to list the genotypes in order of their competitive abilities, with the differences in the genotypes in mean yield per plant being greatest when a "weak" competitor is grown in conjunction with a "strong" competitor.

The latter definition is applicable to Table 11, so that the significant background effect shows that Timgalen is a "strong" competitor and Warimek a "weak" one. This effect was again observed in the glass house (Table 16), which confirmed the choice of these two varieties as being suitable to investigate their frequency responses.

Originally, it had been expected that the f.d.a. would be associated with competitive ability as depicted in the Type II interaction in Figure 13. In fact, none of the present experiments, nor those reported earlier support this expectation. Although Timgalen showed a significant f.d.a. (Tables 13 and 14), Warimek did not show a corresponding decrease, and this occurred despite the considerable difference in competitive ability between the two varieties. In the glasshouse experiment, although the hybrid had significantly higher yield at the low than at the high frequency in the Warimek stand, the yield of Warimek was similar at both frequencies. Thus, it is evident that the genotypes are co-exploitative rather than competitive, with one appearing to be able to use resources more effectively at one time than the other. In experiments IIA and IIB, there were virtually no lines which showed a significant decrease in yield at low frequency. As these were from random selections of F_2's, a large number of them could be expected to be lower yielding in pure stand than the background competitors. Thus, at least in this Mediterranean type of environment, reactions intermediate between the Types I and III seem to be quite common. Similar types of interaction were observed in some genotypic combinations of soybeans tested by Schutz and Brim (1967).

The results are dissimilar to those of Khalifa and Qualset (1974), where, when two genotypes of wheat were mixed in all combinations at 12.5% frequency increments, the stronger competitor consistently increased in yield as it became rarer in the mixture. At the same time, the weaker competitor showed a significant decrease in yield as its frequency decreased. However, it is possible that the competition for light was very intense in their experiments as a result of high seeding rates (90kg/ha) which enhanced the competitive effects between the tall and dwarf plant types. Jennings and Aquino (1968) have shown that competition for light had an important effect in mixtures of tall and dwarf rice varieties.

The F_1 hybrid (Wm*T), was intermediate between the two parents.

It did not show f.d.a. with either parent in the field and in this case heterozygosity (through the interaction of the two genetic systems) does not appear to be an essential factor in f.d.a.. This is consistent with the results reported earlier, which showed that f.d.a. was expressed by F_1's, F_4's and the parents.

The f.d.a. of Timgalen with the mixture (Table 13) indicates that f.d.a. is specific to this genotype, regardless of the background it competed with. Similarly, Allard and Adams (1969a) showed that surrounding a given variety by more than one different genotype had little effect on the test genotype. Since f.d.a. is a common phenomenon, it is likely that the background genotypes would also have f.d.a. of their own. Thus, it is suggested that the mechanism(s) for the f.d.a. of Timgalen was likely to be different from those for other genotypes and that there is a number of alternative mechanisms which will lead to f.d.a. (e.g. differences between genotypes in their requirements for environmental resources or differential responses to plant pathogens). This, in turn, provides an explanation for the persistence of heterogeneity in bulk-hybrid populations.

Frequency-dependent advantage is similar to the over-compensatory interaction in a feed back system, proposed by Schutz, Brim and Usanis (1968), as the mechanism in the evolution of fitness in the bulk populations of barley reported by Allard and Jain (1962). This mechanism also adequately explains the increase in reproductive values of heterozygotes at low frequency in populations of lima bean (Harding *et al.*, 1966). Furthermore, it may ensure that a genotype will be maintained in the population at a low frequency despite a yield disadvantage when compared as a pure stand with other genotypes. In the mixtures of barley genotypes grown by Suneson (1949) for 16 generations, Workman and Allard (1964) found evidence to show that the poorer competitors, Club Mariout, Hero and Vaughn had higher selective values when they were rare than when they were common.

Competition appears to have occurred mainly below ground between the root systems. It has been pointed out earlier that, under a Mediterranean type of climate where there is ample illumination in the latter part of the growing season, final plant height was unlikely to be an important factor in f.d.a.. The results in experiment IV indicated that competition for soil factors was intense in the containers used. This could have accounted for the f.d.a. of the hybrids grown in the Warimek stand in this experiment (Table 16). While direct extrapolation to field conditions may not be fully justified, it does indicate the need to investigate soil factors as well as the aerial ones which have received much more attention. Donald (1958) showed that when both shoots and roots were competing the effect of competition was greater than when only one factor was involved.

Competition from either plants of the same or different species has been found to reduce the size and number of both the nodal and seminal roots as well as their weight, degree of branching and extent of spread in the soil (Troughton, 1962; a review). Lee (1960) showed that, in mixtures of barley, the competitive success of Atlas over Vaughn was due to the former having a better developed root system than the latter. In a subsequent (unreported) test carried out here in the laboratory, it was indeed found that Timgalen had, initially, faster shoot and root growths than Warimek.

Just as competition between roots of different genotypes may lead to competitive success or failure (Lee, 1960), so may competition between roots of the same genotype result in the f.d.a. observed. Weaver (1926) stated that wheat roots had a lateral spread of 15-23cm on all sides. Where the f.d.a. was observed, its magnitude was directly related to the distances between the test plants. In the case of Timgalen grown in Warimek, the distances between successive Timgalen plants at frequencies 6.25%, 25%, 75% and 93.75% were 26.8cm, 13.4cm, 8.5cm and 8.3cm; and the corresponding yields per plant were 3.14g, 2.28g, 1.79g and 1.88g respectively. The regression coefficient of grain yield per plant on distances between plants of the same genotype is positive and significantly different from zero ($p < 0.05$). This result agrees with those found for the hybrids, (Warimek * Halberd) and (Wariquam * Halberd), in experiments IA and IB. Furthermore, the linear regression of yield per plant accounts for more of the variation when the distance between plants of the same genotype is used as the independent variable rather than the frequency. The correlation between grain yield and frequency was -0.57* as compared with 0.66* between grain yield and distance. Inspection of Figure 15 and the relevant figures (Figs. 5a and 6a) in Experiment I suggests that the individual plant yield shows a curvilinear increase to decreased frequency. This, together with the positive correlation between yield per plant and the square of the distance ($r = 0.65$*) between plants of the same genotype suggest that f.d.a. depends upon the distance between these plants and therefore on the exploitation of the environment in a slightly different way from that by other genotypes.

Thus, the available resources of a certain environment seem to be exploited more efficiently by a mixture of several genotypes, than when there is only one genotype. As the frequency of a genotype decreases, there will be fewer competitors for the same resources and that genotype will be at a relative advantage. Trenbath (1974) reviewed published data on biomass productivity of mixtures and concluded that most binary mixtures (1:1 proportions) had higher yields than the mean yields of the monocultures. However, the advantage of the former over the latter was generally small and this can be attributed to the non-significant f.d.a. of the genotypes at the 50% frequency (Figure 15 and discussion of section A) and that the yield of a genotype at this frequency is only slightly higher than its yield in pure stand.

As an alternative to competition for resources, allelopathy can also be involved in the interaction between genotypes. This occurs when a plant produces biologically active substances which enter neighbouring plants and affect their metabolism (Winter, 1961). These chemicals could either inhibit or reduce the efficiency of uptake of factors required for the growth of the neighbours. This allelopathic effect would probably result in the Type II interaction depicted in Figure 13 and would eventually lead to the dominance of a single species (Chou and Muller, 1972). Type III interaction may occur, however, where allelopathy results in autoinhibition. Webb _et al_. (1967) have reported that _Grevillea robusta_, a sub-tropical rain-forest tree in Queensland which does not form natural pure stands, showed very poor growth in commercial plantations. Detailed experimentation indicated that the growth of young seedlings was inhibited by a water-transferable substance apparently produced by the roots of adjacent _G. robusta_ plants. The seeds of this species are numerous, winged and wind-distributed. It is expected that the seedlings would be able to survive in isolation whereas this was prohibited in pure stand. Since _G. robusta_ does not prevent the regeneration of other rain forest species, the autoinhibition may explain the maintenance of floristic diversity in complex tropical forests (Webb _et al._, 1967).

In conclusion, the neighbourhood effect, allelopathy causing autoinhibition and differential susceptibility to specific races of plant pathogens (see discussion of Section A), are the likely mechanisms of f.d.a.. The operation of any of these could lead to the Type III interaction depicted in Figure 13; the first of these, involving the uptake of nutrients, appearing to be the most likely mechanism in wheat in South Australia.

b. VARIABILITY OF THE PARENTAL VARIETIES USED IN THE STUDY.

Experiment V was designed to determine whether genetic differences between the hybrids between the five parents were more apparent with some backgrounds than others and, if this occurred, whether the background could be considered to be a "good" (or "strong") competitor. Competitive abilities of the parents were estimated by comparing the performance of hybrids grown in different background parents. A background genotype was considered a "good" competitor when the hybrids were consistently depressed in that stand. When the hybrids were consistently enhanced, the background was regarded as a "poor" competitor.

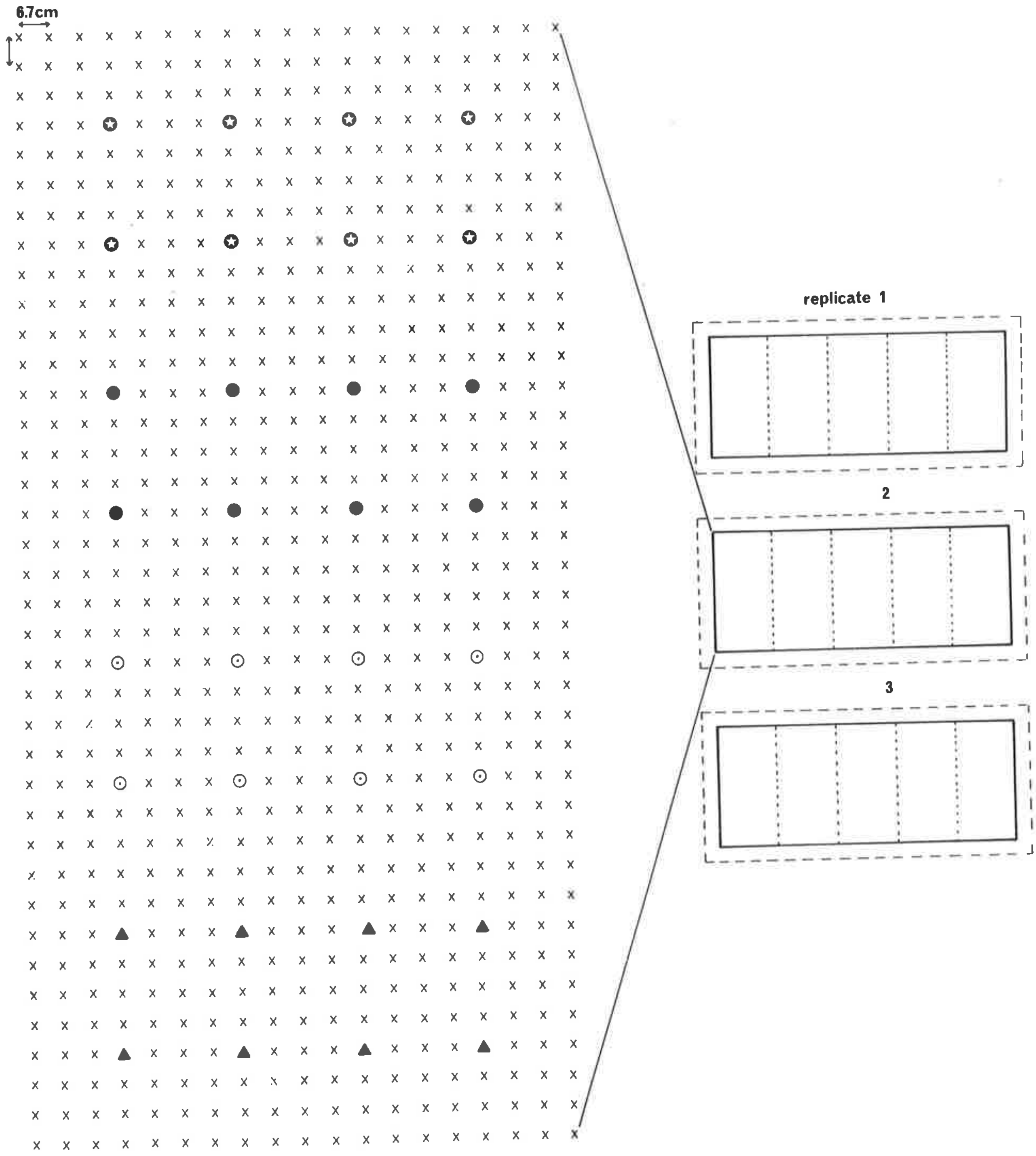
1. Materials and methods:

Experiment V was planted on June 28-29, 1972 at Roseworthy Agricultural College. There were 3 replicates in the experiment. Each replicate consisted of 5 parental stands, Warimek, Wariquam, Halberd, Timgalen and Gabo, and 10 hybrids from the diallel of cross between these five varieties. The 4 hybrids (half sibs) from each parent were grown at a frequency of 6.25% in the parental stand (Figure 19). For example, (Wm*Wq), (Wm*H), (Wm*T) and (Wm*G) were grown in Warimek at frequency 6.25%. Reciprocal crosses were not produced, so the order of the parents in the hybrid's name is used to indicate the background parent. Thus (Wm*Wq) and (Wq*Wm) refer to the same hybrid when grown in Warimek and Wariquam, respectively.

The positions of the parental stands within replicates, and hybrids within stands were allocated at random. Thus in each replicate, every hybrid occurred twice, once in each of its parental stands. There were

Figure 19: Field layout of experiment V showing the positions of 5 parental stands in each replicate. There were 4 crosses in each stand. On the left is a detailed plan of one stand showing the exact positions of the parent and hybrids.

Positions of stands within replicates, and crosses within stands were allocated at random. Each replicate was surrounded by 4 border rows of Halberd.


6.7cm
replicate 1
2
3

8 plants of each hybrid in a stand. The seeding method and plant density were the same as in experiments I and III. The positions of the hybrids were marked by small wooden sticks, painted with a different colour for each cross. At maturity, the 8 hybrids in a treatment were harvested and measured as a bulk. The same number of parental plants adjacent to the hybrids was also measured.

2. Results:

The results were analysed separately for hybrids within each parental stand and for each hybrid in 2 different background parents.

2.1 Comparison between hybrids grown in the same parental stand.

Grain yields per plant of 4 half sib hybrids in each of the 5 parental stands are presented in Figure 20. The results for other characters are summarized in Table 21. The results enable a comparison of the performance of different hybrids when these are grown in a common background at a low frequency.

There were significant differences in yield and in the yield components between hybrids when Warimek, Timgalen and Gabo were used as backgrounds. These did not occur with Wariquam or Halberd. The main character contributing to differences in grain yield was number of grains, and to a lesser extent, number of spikes and number of spikelets per plant.

Figure 20: Grain yield per plant of 10 hybrids in 5 parental stands.

Across each row: Yields of 4 half sib hybrids in the same parental stand.

Down each column: Yields of the same hybrid in each of its parental stands.

On the right are the LSD's at 5% level of significance for the comparisons of hybrids in the same stand.

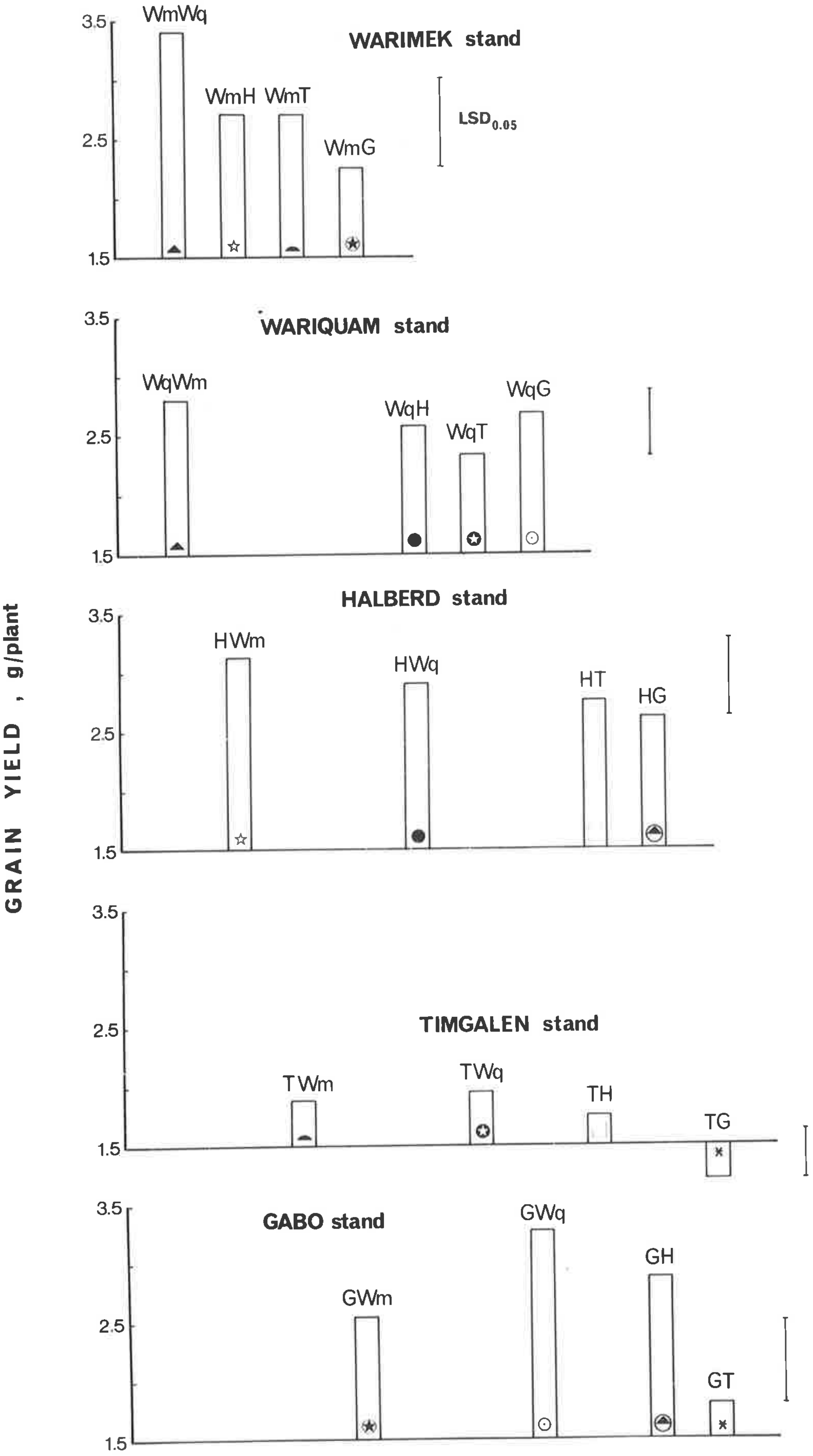

GRAIN YIELD , g/plant
WARIMEK stand
3.5
2.5
1.5
WmWq
WmH
WmT
WmG
LSD$_{0.05}$
WARIQUAM stand
3.5
2.5
1.5
WqWm
WqH
WqT
WqG
HALBERD stand
3.5
2.5
1.5
HWm
HWq
HT
HG
TIMGALEN stand
3.5
2.5
1.5
TWm
TWq
TH
TG
GABO stand
3.5
2.5
1.5
GWm
GWq
GH
GT

Table 21: Means per plant and results of analyses of variance for hybrids grown in the 5 different background stands.

(a) Comparison between hybrids grown in the Warimek stand.

Character	Genotype mean				Diff	LSD(0.05)
	(Wm*Wq)	(Wm*H)	(Wm*T)	(Wm*G)		
Height, cm	65.6	69.5	71.1	71.9	*	4.6
No. of spikes	3.0	2.4	3.1	2.4	*	0.6
No. of spikelets	50.8	40.8	50.2	39.3	*	9.8
No. of grains	85.4	72.9	67.8	62.0	NS	
Total weight, g	7.50	6.14	7.20	5.79	NS	
Grain weight, g	3.37	2.66	2.66	2.22	*	0.75
Weight per grain, mg	39.5	36.5	39.2	35.8	NS	

(b) Comparison between hybrids grown in the Wariquam stand.

Character	Genotype mean				Diff.	LSD(0.05)
	(Wq*Wm)	(Wq*H)	(Wq*T)	(Wq*G)		
Height, cm	66.5	71.8	68.5	73.3	**	3.1
No. of spikes	2.8	2.3	2.9	2.8	NS	
No. of spikelets	46.3	36.0	43.5	44.7	NS	
No. of grains	76.9	67.4	60.7	72.4	NS	
Total weight, g	6.67	5.81	6.44	6.72	NS	
Grain weight, g	2.88	2.56	2.33	2.67	NS	
Weight per grain, mg	37.4	38.0	38.4	36.7	NS	

(c) Comparison between hybrids in the Halberd stand.

Character	Genotype mean				Diff.	LSD(0.05)
	(H*Wm)	(H*Wq)	(H*T)	(H*G)		
Height, cm	71.3	76.9	75.3	72.8	NS	
No. of spikes	2.9	2.9	3.2	2.7	NS	
No. of spikelets	49.0	45.0	49.3	44.8	NS	
No. of grains	84.5	82.0	78.2	71.4	NS	
Total weight, g	7.17	7.00	7.18	6.73	NS	
Grain weight, g	3.10	2.90	2.74	2.63	NS	
Weight per grain, mg	36.7	35.4	35.0	36.8	NS	

(d) Comparison between hybrids in the Timgalen stand.

Character	:	Genotype mean (T*Wm)	(T*Wq)	(T*H)	(T*G)	:Diff.:	LSD(0.05)
Height, cm	:	73.8	66.7	69.5	72.5	: NS :	
No. of spikes	:	2.7	2.7	2.5	2.4	: NS :	
No. of spikelets	:	40.9	38.6	37.3	34.4	: NS :	
No. of grains	:	54.1	52.4	51.4	35.3	: ** :	9.1
Total weight, g	:	5.39	5.31	4.76	4.21	: * :	0.78
Grain weight, g	:	1.87	1.93	1.74	1.19	: ** :	0.33
Weight per grain, mg	:	34.6	36.8	33.8	33.7	: NS :	

(e) Comparison between hybrids in the Gabo stand.

Character	:	Genotype mean (G*Wm)	(G*Wq)	(G*H)	(G*T)	:Diff.:	LSD(0.05)
Height, cm	:	75.9	80.6	76.3	76.6	: NS :	
No. of spikes	:	2.4	2.9	2.7	2.7	: NS :	
No. of spikelets	:	40.5	48.4	46.5	42.3	: NS :	
No. of grains	:	70.0	85.6	78.7	52.6	: ** :	18.2
Total weight, g	:	6.32	8.02	7.17	6.11	: NS :	
Grain weight, g	:	2.55	3.28	2.86	1.80	: ** :	0.70
Weight per grain, mg	:	36.4	38.3	36.3	34.2	: NS :	

2.2 The effect of the background parents on the performance of the hybrid.

The competitive effect of each parental genotype was estimated by a comparison of the performance of each of the 10 hybrids in different background stands. Each column in Figure 20 represents the yields of the same hybrid in its 2 background parents, and the other characters are listed in Table 21.

Cross 1: (Wm*Wq); Stands: Warimek and Wariquam.

There was no significant difference in the performance of (Wm*Wq) in the two stands.

Cross 2: (Wm*H); Stands: Warimek and Halberd.

(Wm*H) did not perform differently in the two stands.

Cross 3: (Wm*T); Stands: Warimek and Timgalen

Except for height, where the hybrid was slightly shorter in the Warimek stand, all other characters had a higher expression in the Warimek than in the Timgalen stand. All of these, except for the number of spikes and the number of grains per plant, were significantly higher (Table 22).

Cross 4: (Wm*G); Stands: Warimek and Gabo.

There was no significant difference between stands for any character.

Table 22: Comparison of (Wm*T) in Warimek and Timgalen stands.

Character	Mean performance in Warimek stand	 Timgalen stand	Difference
Height, cm	71.1	73.8	NS
No. of spikes	3.1	2.7	NS
No. of spikelets	50.2	40.9	*
No. of grains	67.8	54.1	NS
Total weight, g	7.20	5.39	**
Grain weight, g	2.66	1.87	*
Weight per grain, mg	39.2	34.6	*

Cross 5: (Wq*H); Stands: Wariquam and Halberd.

Except for grain size, the hybrid had a higher expression in all characters in the Halberd than in the Wariquam stand. However, the differences were only significant for height, number of spikes and number of spikelets per plant (Table 23). The non-significant difference in grain yield was due to the greater number of grains in the Halberd stand being smaller than in the Wariquam stand.

Table 23: Comparison of (Wq*H) in Wariquam and Halberd stands.

Character	Mean performance in Wariquam stand	 Halberd stand	Difference
Height, cm	71.8	76.9	**
No. of spikes	2.3	2.9	*
No. of spiklets	36.0	45.0	*
No. of grains	67.4	82.0	NS
Total weight, g	5.81	7.00	NS
Grain weight, g	2.56	2.90	NS
Weight per grain, mg	38.0	35.4	*

Cross 6: (Wq*T); Stands: Wariquam and Timgalen.

Again the differences were not statistically significant, except for grain size, where there were larger grains in the Wariquam stand ($p < 0.05$).

Cross 7: (Wq*G); Stands: Wariquam and Gabo.

There was no significant difference in the performance of the hybrid between the two stands.

Cross 8: (H*T); Stands: Halberd and Timgalen.

The hybrid had significantly higher expressions for all characters in the Halberd than in the Timgalen stand (Table 24).

Table 24: Comparison of (H*T) in Halberd and Timgalen stands.

Character	Mean performance in Halberd stand	Mean performance in Timgalen stand	Difference
Height, cm	75.3	69.5	***
No. of spikes	3.2	2.5	**
No. of spikelets	49.3	37.3	**
No. of grains	78.2	51.4	***
Total weight, g	7.18	4.16	***
Grain weight, g	2.74	1.74	***
Weight per grain, mg	35.0	33.8	*

Cross 9: (H*G); Stands: Halberd and Gabo.

No significant difference was observed for any character of the hybrid between the Halberd and Gabo stands.

Cross 10: (T*G); Stands: Timgalen and Gabo.

The hybrid had a higher expression for all characters in the Gabo than in the Timgalen stand. The difference was significant for all characters, except number of spikes and number of spikelets per plant (Table 25).

Table 25: Comparison of (T*G) in Timgalen and Gabo stands.

Character	Mean performance in Timgalen stand	 Gabo stand	Difference
Height, cm	72.5	76.6	*
No. of spikes	2.4	2.7	NS
No. of spikelets	34.4	42.3	NS
No. of grains	35.3	52.6	**
Total weight, g	4.21	6.11	**
Grain weight, g	1.19	1.80	**
Weight per grain, mg	33.7	34.2	*

2.3 The competitive ability of the parents.

The competitive ability of the five parents can be estimated by the difference between the mean yield of the four hybrids in the common background and the mean yield of the same four hybrids in the non-common backgrounds. For example, the competitive ability of Warimek was estimated by comparing the mean yield of (Wm*Wq), (Wm*H), (Wm*T) and (Wm*G) when grown in Warimek with the mean yield of the same four hybrids when grown in Wariquam, Halberd, Timgalen and Gabo, respectively.

If the difference is positive, the variety is a "poor" competitor since the hybrids had a higher mean yield in that background than their mean yield in the remaining backgrounds. If the difference is negative, the variety is a "good" competitor. The order of competitive ability was:

Genotypes :	Timgalen,	Wariquam,	Warimek,	Halberd,	Gabo.
Estimates :	-0.70	-0.26	0.13	0.39	0.44

The background parents were grown at frequency 93.75% so it could be expected that their yields would be similar to those of pure stands (Table 26).

Table 26: Mean performance (per plant) of background genotypes at frequency 93.75% and results of analyses of variance.

	Genotype mean						
Character	Wm	Wq	H	T	G	Diff.	LSD(0.05)
Height, cm	63.5	60.8	69.3	69.4	75.1	***	2.4
No. of spikes	2.1	2.3	2.0	2.2	2.0	NS	
No. of spikelets	34.9	32.6	30.8	32.9	31.6	NS	
Total weight, g	4.64	4.57	4.33	4.13	4.39	NS	
Spike weight, g	2.64	2.56	2.18	2.02	2.10	**	0.34
Grain weight, g	1.98	1.90	1.64	1.39	1.55	***	0.26

Warimek, Wariquam were higher yielding than Halberd, Gabo and Timgalen in that order. The results also confirm the short stature of Warimek and Wariquam relative to the other 3 varieties.

3. Discussion:

Warimek and Wariquam have higher yields than Halberd, Gabo and Timgalen. This is not surprising since Warimek and Wariquam were released recently after being selected for high yields of grain under

South Australian conditions. Halberd, the most widely grown of the local commercial varieties, was higher yielding than Timgalen and Gabo. The two latter varieties were selected by the University of Sydney primarily for resistance to stem rust (Puccinia graminis fsp. tritici) and are not as well adapted to South Australian conditions as the local varieties.

In this experiment, Timgalen was the strongest competitor but it was also the lowest yielder. This effect, which was clearly demonstrated here in section 2.2, was not exceptional since Timgalen was also highly competitive in experiment IIIA in the field and in experiment IV in the glass house. This variety, based on direct measurement (Table 26), is taller than Warimek and Wariquam, about the same height as Halberd and shorter than Gabo. Hence it could not have derived its competitive advantage through the shading of its neighbours. In experiment IV, it was shown that competition for soil factors was intense in the containers used. In another (unreported) test carried out after germination of Warimek and Timgalen, it was observed that Timgalen had initially faster shoot and root growths than Warimek. The faster growth rate of Timgalen possibly enabled it to interfere with the growth of its neighbours, particularly below the soil surface and accounted for the significant reduction in yield of all the hybrids in this stand (Figure 20). The same property enabled this variety to exhibit f.d.a. in experiment III, whereas Warimek, Wariquam and Gabo did not show any effect.

The results in this experiment indicated that the rankings of competitive ability and yield were different between the five parents. The orders, from high to low, would appear to be:

Competitive ability: Timgalen, Wariquam, Warimek, Halberd, Gabo.

Yield: : Warimek, Wariquam, Halberd, Gabo, Timgalen.

Although Timgalen, Warimek and Gabo could be considered "good", "average" and "poor" competitors, respectively, the significant differences between the hybrids in these stands (Table 21) suggested that the expression of genetic differences was only loosely related to the competitive ability of the background. There is some indication that deliberately choosing either a strong (e.g. Timgalen) or weak competitor (e.g. Gabo) as a background will emphasize genetic differences between the test genotypes grown at low frequency in the mixture.

Another aspect which originally led to the design of the experiment was the possibility of reducing the micro-environmental variance in the test genotypes by growing them in homogeneous backgrounds. This was based on the assumption that genetic differences in a segregating population may be confounded by micro-environmental variation induced by differential competitive abilities of a heterogeneous population. This method would have an application if the genetic expression of the test genotypes was consistent in all backgrounds. However, the results indicated that this did not occur, and that the performance of a genotype depended on its competitive ability in relation to that of the background in which it was grown. It is suggested that the yield of a genotype at a low frequency in a homogeneous background cannot be used to predict its yield in pure stand. This theory was tested further in experiment VIII when the yields of F_2 derived lines grown at low frequencies in the parents were compared with their yields in pure stands.

c. FURTHER STUDY ON THE NATURE OF FREQUENCY-DEPENDENT ADVANTAGE.

1. Introduction

In experiments I and III, the test plants at each frequency were grown at regular distances from each other. In experiment II, however, the positions of the test plants within each frequency were random since the seeds had been premixed and planted by machine. In natural populations, as well as in segregating populations where selection is to be practised, it is likely that individuals of the different genotypes are distributed randomly within the mixture. Thus an experiment was conducted to determine whether the response to alterations in frequency was confounded by the arrangement. Two arrangements were compared; "fixed", where the genotype at low frequency was grown at a particular distance from its neighbours, and "random", where the seeds of the two genotypes were premixed and then sown in the order they were taken from the mixture.

2. Materials and methods

Experiment VI was conducted at the Waite Agricultural Research Institute in 1973. It consisted of 2 sub-experiments of the same design. In experiment VIA, Wariquam was grown as the test genotype in a background stand of Halberd at 3 frequencies, 4%, 11.1% and 50%. In experiment VIB, Halberd was grown in Wariquam at the same 3 frequencies. There were 3 replicates in each experiment, and each replicate consisted of 2 treatments. In one, the positions of the test plants were fixed and determined prior to sowing. In the other, the same frequencies were maintained, except that the seeds were mixed and then planted out at random.

The experiment was sown on June 27 and 28. Seeds were sown by hand, using a set of boards similar to those described by Hamblin (1971) and Johnston (1972). These consisted of a plywood board with holes drilled at 6.7cm x 6.7cm spacings and a dibber board with pegs at the same spacing (Plate 4). At sowing, the soil was smoothed and the board with holes was placed in position. The dibber board was placed on top of this, the dibbers were forced into the ground and then removed, leaving holes in the soil of a constant depth of 2.5cm. In the fixed arrangement, seeds of the 2 genotypes were placed in predetermined positions (Figure 21). In the random arrangement, the seeds, which had previously been labelled with non-toxic dyes in the laboratory, were mixed. At sowing the seeds were drawn at random and planted in successive holes. As seeds of the test genotype were sown, their positions were marked by small wooden sticks for identification. Figure 21 shows the arrangements in one such replicate.

3. Results

It was unfortunate that both experiments were damaged by birds; mainly sparrows and parrots, just before harvest so that grain yields were unreliable. However, the results for straw weight and number of spikes per plant are presented here.

3.1 Experiment VIA: Wariquam in Halberd.

Plate 4: A set of boards used to sow experiments VI, VII and VIII.

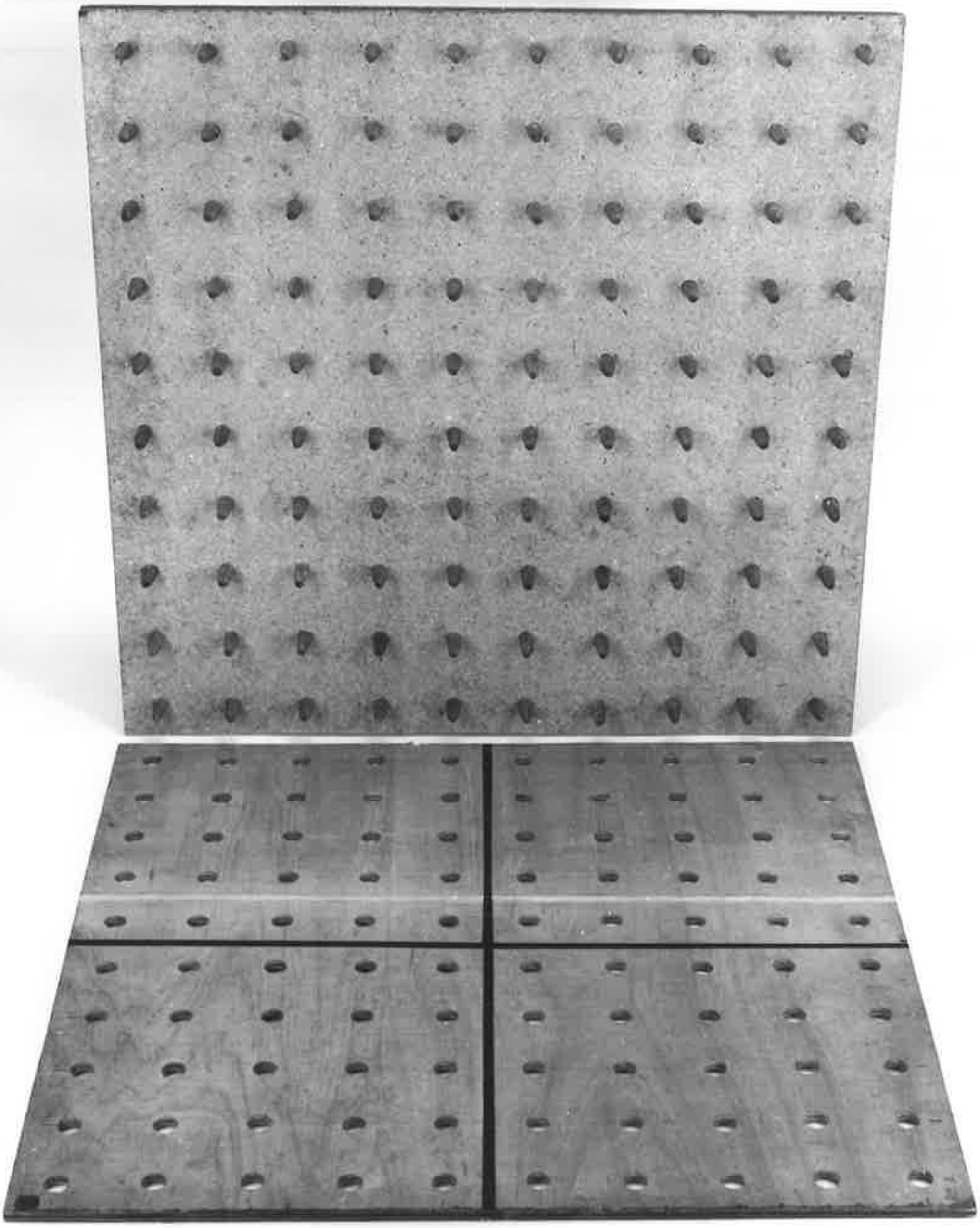


Figure 21: Field layout of experiment VI and detailed plan of one replicate.

x x x positions of the background plants.

● ● ● positions of the test plants in the "fixed" arrangement.

○ ○ ○ positions of the test plants in the "random" arrangement.

Open and closed circles have been used for the test plants in the two arrangements, but they are the same genotype.

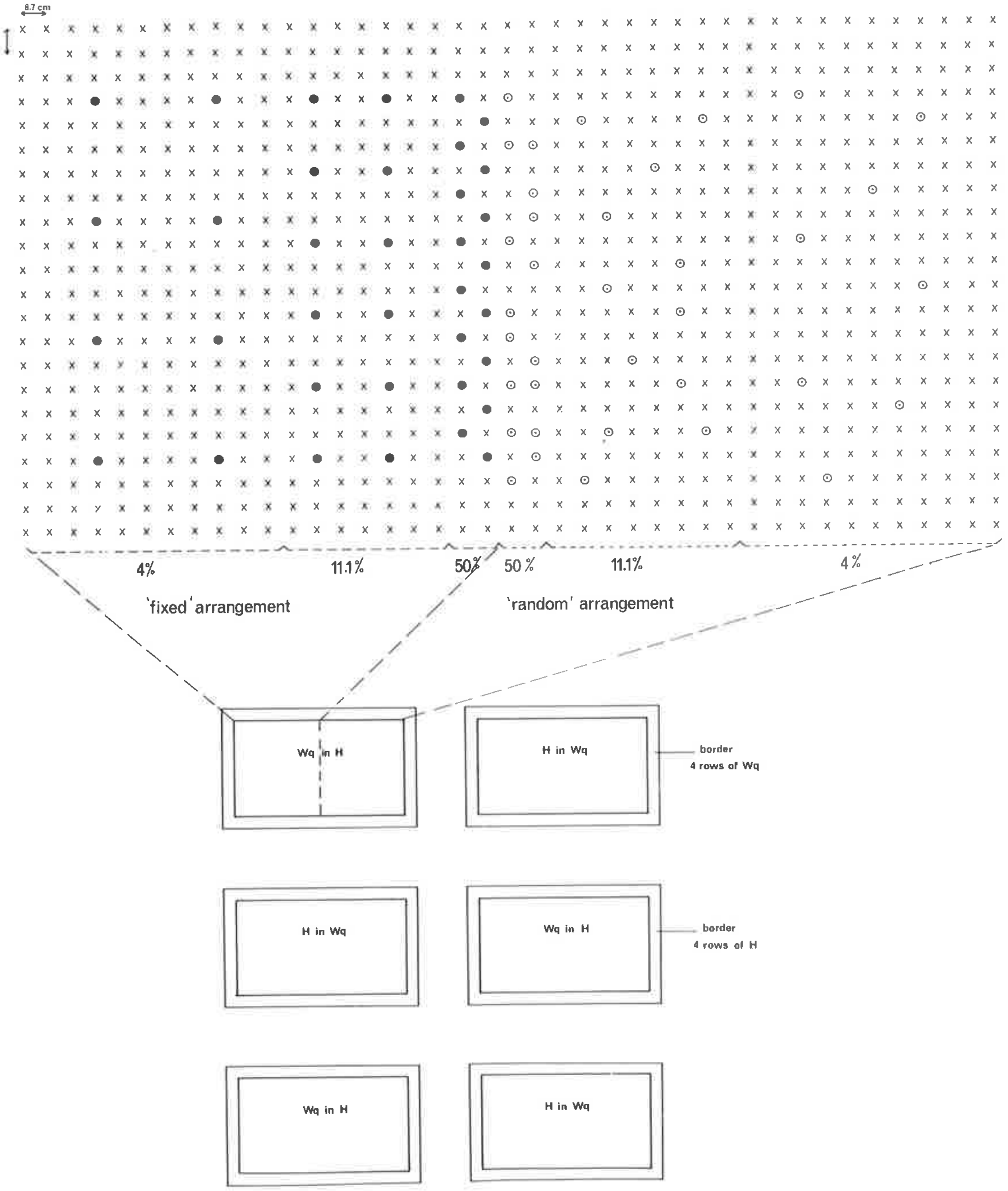

6.7 cm
4%
11.1%
50%
50 %
11.1%
4 %
'fixed' arrangement
'random' arrangement
Wq in H
H in Wq
border
4 rows of Wq
H in Wq
Wq in H
border
4 rows of H
Wq in H
H in Wq

Table 27: Number of spikes per plant, with Wariquam the test genotype.

Frequency	Pattern Fixed	 Random	Mean
4.0%	1.9	2.3	2.1
11.1%	1.8	1.8	1.8
50.0%	1.8	1.6	1.7
Mean	1.9	1.9	1.9

Table 28: Straw weight per plant, g.

Frequency	Pattern Fixed	 Random	Mean
4.0%	2.22	2.28	2.25
11.1%	1.82	1.76	1.79
50.0%	2.00	1.80	1.90
Mean	2.01	1.95	1.98

An analysis of variance with patterns as main plots and frequencies as sub-plots indicated that there was no significant difference between patterns or frequencies, and no interaction. However, in both arrangements, it is noted that Wariquam plants had slightly more spikes and higher straw weight per plant at the lowest frequency, 4.0%, and that the difference between frequencies was slightly greater in the random than in the fixed pattern.

3.2 Experiment VIB: Halberd in Wariquam

It was found that Halberd plants did not differ significantly between frequencies nor between patterns of arrangement. The interactions between frequencies and patterns were also non-significant.

Again there is no evidence that the frequency effect is less with a random pattern than it is with the fixed pattern.

Table 29: Number of spikes per plant, with Halberd the test genotype.

	Pattern		
Frequency	Fixed	Random	Mean
4.0%	1.9	2.2	2.0
11.1%	2.3	2.0	2.2
50.0%	2.1	1.7	1.9
Mean	2.1	2.0	2.1

Table 30: Straw weight per plant, g.

	Pattern		
Frequency	Fixed	Random	Mean
4.0%	2.95	3.45	3.20
11.1%	3.54	3.14	3.34
50.0%	3.13	2.75	2.94
Mean	3.21	3.12	3.16

In 1974, the experiments were repeated at Roseworthy Agricultural College. It was even more unfortunate this time because the experiments were damaged by birds (mainly black crows, Corvus spp.) soon after sowing and had to be abandoned.

No definite conclusions could be drawn from the results. From the limited amount of information obtained, it was possible that the overall response to frequency was independent of the arrangement. If the response to frequency was confounded by the arrangement, it would be expected that the random pattern would show more variability between plants than the fixed arrangement. A comparison of the variances of the fixed and random patterns within each frequency showed no significant differences and no consistent trends.

Neither the results obtained here, nor those derived from a comparison of the fixed pattern of Experiment I with those from the random pattern of Experiment II, suggest that the f.d.a. effects are less in a random than they are with a fixed placement of the genotype at low frequency.

C. THE EFFECT OF FREQUENCY-DEPENDENT ADVANTAGE ON SINGLE PLANT SELECTION

1. Introduction.

An enhanced yield as a consequence of a low frequency could have implications in plant breeding, particularly during the selection of individuals from segregating populations. Simple genetic theory for quantitative characters (amplified below) suggests that when a character such as grain yield is determined by many genes, a segregating population will form a normal distribution with the extreme genotypes occurring at low frequency. The question is whether these genotypes will have an enhanced yield as a consequence of their low frequency when grown in a segregating population. If frequency-dependent advantage does operate in a segregating population, it may reduce the efficiency of single plant selection. An individual selected for high yield will have this attribute as a result of its low frequency as well as its genetic potential. This yield advantage will be lost when the individual is grown finally as a pure stand variety.

Two experiments were designed to evaluate the effect of f.d.a. on single plant selection. In the first, the effect of f.d.a. on the distribution of an F_2 population was investigated. In the second, the relationship between F_2 derived lines as single plants in mixtures was compared with the same lines in pure stands.

2. Experimental procedures.

2.1 Theoretical considerations: In self-pollinated crops, where varieties are usually derived from a single plant in the F_4 or F_5, the plants within a parental or an F_1 generation may be considered to be genetically uniform. Any differences between individuals in such populations may be attributed primarily to micro-environmental variation. In an F_2 population, however, variation is due to a combination of the effects of the micro-environment, genetic factors and the interaction between these, including frequency-dependent advantage. It is therefore expected that the F_2 would have a higher total variance than the F_1 or either of the parents. A comparison of the variances of these populations would give an indication of the variability in the F_2 as a result of genetic segregation and f.d.a..

Secondly, if the micro-environmental variation is random, the performance of individual plants within the F_1 and parental stands are expected to be distributed normally. The genetic variation of the F_2 will be additional to the micro-environmental variation observed for the parents and F_1. If the number of genes involved is large and their individual effects small, this genetic variation should approximate to a normal distribution as it can be described by the binomial expansion of $(p + q)^{2n}$, where n is the number of genes controlling yield and p and q are the frequencies of alleles (here 0.5). Table 31 shows the proportions of individuals in an F_2 population containing 1, 2, 3, ..., 10 desirable alleles when yield is controled by 1, 2, .., 5 genes, respectively. The theoretical distributions of single plants in parental and F_2 stands are presented graphically in Figures 22a and 22b. However, as shown in Figure 22b, plants in regions A and C are at low frequencies and genetically different from those in region B. If f.d.a.

Figure 22: Theoretical distributions of parents (a), F_2(b) and F_2 with the effect of f.d.a. (c).

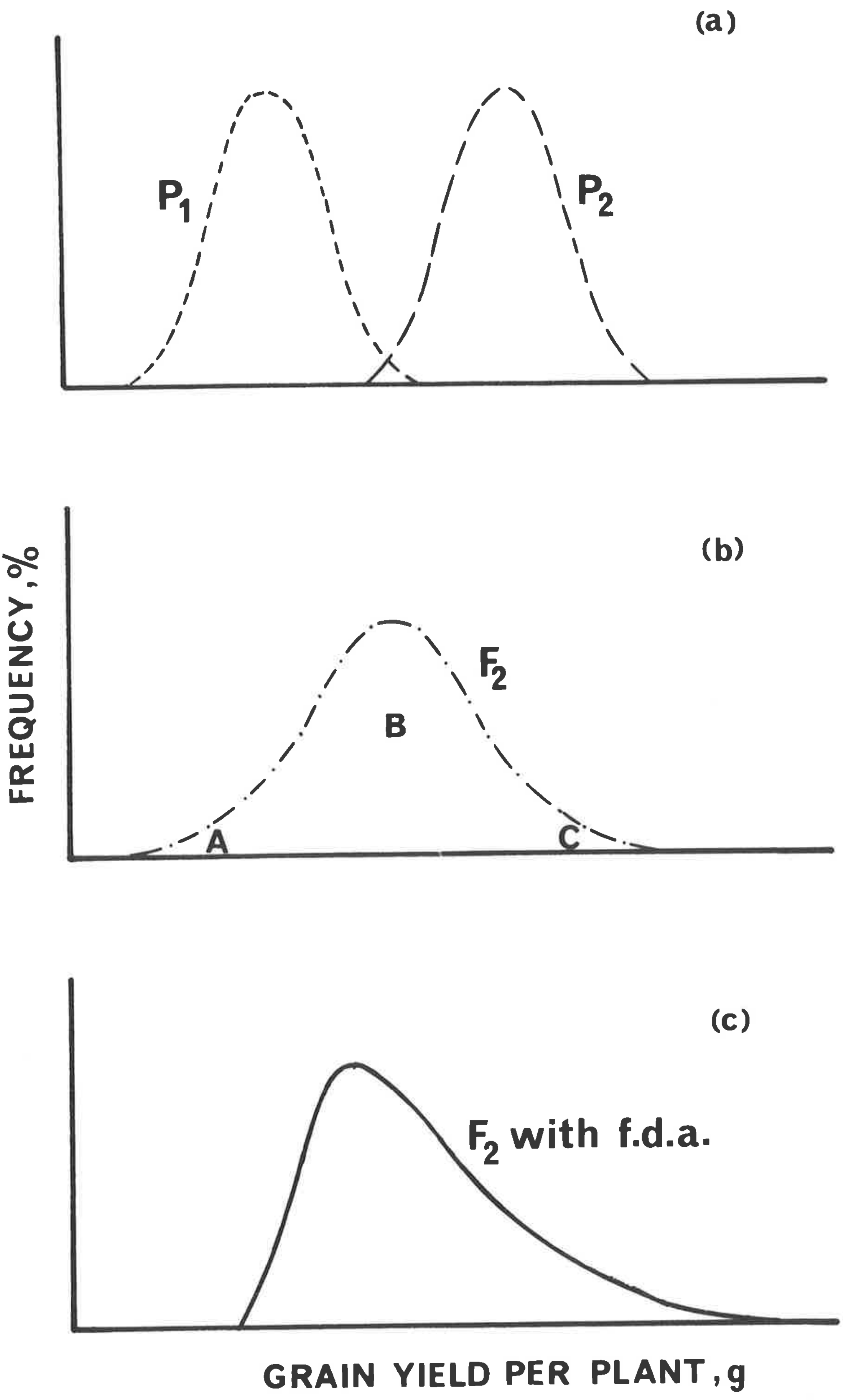

(a)
P$_1$
P$_2$
(b)
FREQUENCY, %
F$_2$
B
A
C
(c)
F$_2$ with f.d.a.
GRAIN YIELD PER PLANT, g

Table 31: Expected numbers of different genotypes containing 1 to 10 desirable alleles in an F_2 population.

		Number of desirable alleles										
		0	1	2	3	4	5	6	7	8	9	10
	1	1	2	1								
Number	2	1	4	6	4	1						
of	3	1	6	15	20	15	6	1				
	4	1	8	28	56	70	56	28	8	1		
genes	5	1	10	45	120	210	252	210	120	45	10	1

operates in such a population, plants in regions A and C are expected to increase in yield whereas those in region B would have a reduced yield. The result is that the F_2 population would have a positive skew distribution compared with the F_1 and the parents (Figure 22c). Thus, because of f.d.a., certain plants may have a higher yield than their genetic worth, and would be favoured in selection. Furthermore, it is expected that the F_2 would contain some plants which are higher yielding than the highest yielding plants in the F_1 and parental stands.

2.2 Procedures.

2.2.1 Experiment VII.

Experiments VII and VIII were grown in 1974 at Roseworthy Agricultural College. Experiment VII consisted of pure stands of three parents Warimek, Halberd and Wariquam and the F_1's and F_2's of (Warimek * Halberd) and (Wariquam * Halberd); subsequently abbreviated to (Wm*H)F_1, (Wm*H)F_2 and (Wq*H)F_1 and (Wq*H)F_2, respectively. All these populations were grown in a completely randomized block, each population occupying plots at random within the block. The whole experiment consisted of 28 plots of 100 plants, which were made up of 6 plots of Halberd, 5 plots of Warimek, Wariquam and each of the F_2's and 1 plot of each of the F_1's.

The experiment was sown on June 25. Seeds were sown by hand, using a set of boards similar to those described in experiment VI. Each board comprised of 100 holes (10x10), 6.7cm apart in both directions, and this was used to constitute a plot.

At maturity, plants were harvested singly and measurements were made on their height, number of spikes, number of grains, total weight, spike weight and grain weight.

2.2.2 Experiment VIII

Twenty five F_5 lines of the cross (Warimek * Halberd) were chosen at random from 76 F_4 lines in experiment IIA described earlier. Each line was grown at 2 frequencies, 4% and 16% in a mixture of the parents, Warimek and Halberd, to compare their f.d.a. effect. In the same experiment, each line was also grown in a pure stand and as a component in the mixtures of the 25 genotypes. Thus, in the mixture, each line also occurred at an apparent frequency of 4%.

Plants were grown in plots of 100 (10 x 10 at 6.7cm spacing on the square). The original experiment was planted on June 22-24 and consisted of 3 replicates. However, these were damaged by birds soon after sowing. Two other replicates were planted on July 22-24 and protected by netting. It was hoped to use the results from 4 replicates so 2 of the less damaged replicates of the original experiment were also harvested.

Within each replicate, there were 24 different combinations of the mixtures, consisting of 6 plots each with 4 sub-plots. Within each combination, the 25 lines were randomly arranged. In each replicate, each line was grown in a pure stand of 100 plants, and at 2 frequencies, 4% and 16%, in a mixture of the parents. The 4% frequency consisted of 2 plots, each with 4 F_5 plants in 100, giving a total of 8 F_5 plants at the low frequency. The 16% frequency consisted of a plot of 16 F_5 plants in 100, of which 8 plants were harvested at random. Figure 23 shows the arragnement for one line and an illustration of the plot containing the 25 lines.

In the mixture and at the two frequencies, 2 seeds were sown per hole and these were thinned soon after emergence to one plant. The positions of the F_5 plants at frequencies 4% and 16% were marked by small wooden sticks to enable their identification. In the pure stands, only one seed was planted per hole. Plants in the mixtures were harvested and measured singly. Plants at the two frequencies and in the pure stands were harvested in bulk and the results are expressed as means per plant and means per plot, respectively.

Since a rust (*Puccinia graminis fsp. tritici*) epidemic occurred in 1974 in South Australia, the plants were infected with stem rust as a result of late sowing, The experiment was sprayed with a solution of Diathane in late November to reduce the level of infection.

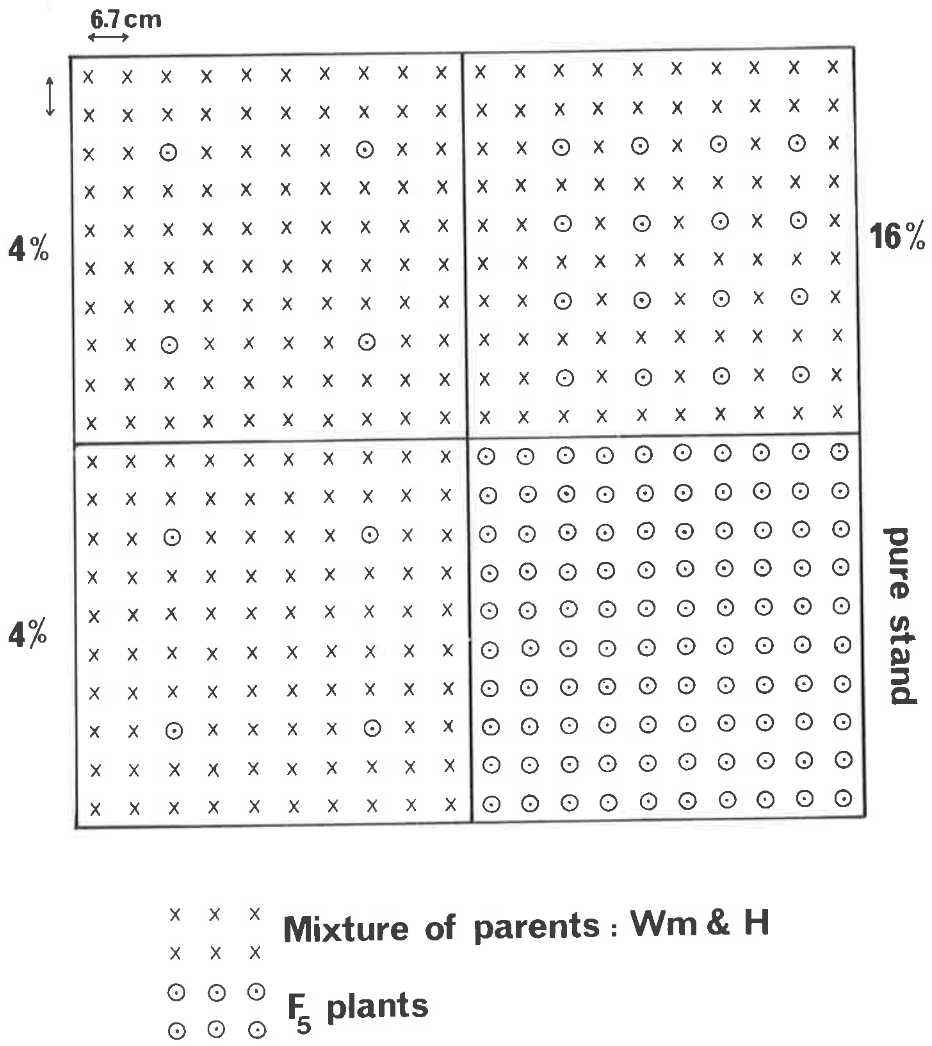
3. Results.

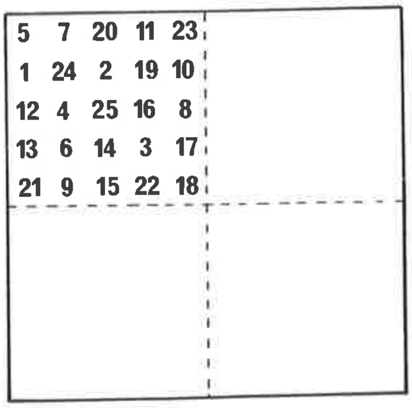
3.1 Experiment VII.

To facilitate computation, one of the Halberd stands has been

Figure 23: An arrangement of one of 25 F_5 lines of (Warimek * Halberd) at 2 frequencies and as a pure stand, and one replication of the mixture of all the lines in Experiment VIII.

a/ An arrangement of one of 25 F_5 lines of (Wm × H)





b/ One of 24 mixture combinations per replicate excluded from the analyses. This gave approximately 375 to 400 plants for each of the parents and F_2's and approximately 85 plants for each of the F_1's.

The distributions for height and grain yield per plant for each cross and the parents are shown in Figures 24, 25, 26 and 27.

The mean, variance, skewness and coefficient of variation were calculated separately for each plot of each population, using a Statscript program on a CDC6400 computer (Lamacraft, 1969). These parameters were used in the comparison of the variability between each F_2 and the parents.

In comparing the means of F_1's, F_2's and the parents, considerations must be given to the variances. An analysis of variance should be performed only when all populations have the same variance. In this experiment, since the F_1's were not replicated, they could not be included in the analysis of variance. For F_2's and the parents, there were 15 randomized plots for each cross, consisting of 5 plots of F_2's and each of the parents, an analysis of variance could take the form:

Source of variation	df
Genotypes	2
Error	12
Total	14

However, the residual variance is not the same for the three genotypes. Those of the parents should be due only to micro-environmental variation, whereas that of the F_2's should consist of the genetic variance and the environmental variance. Initially, it was decided to compare only the means of the parents. However, later attempts were

Figure 24: Distribution for height of Warimek, Halberd, (Warimek * Halberd) F_1 and (Warimek * Halberd) F_2.

Figure 25: Distribution for height of Wariquam, Halberd, (Wariquam * Halberd) F_1 and (Wariquam * Halberd) F_2.

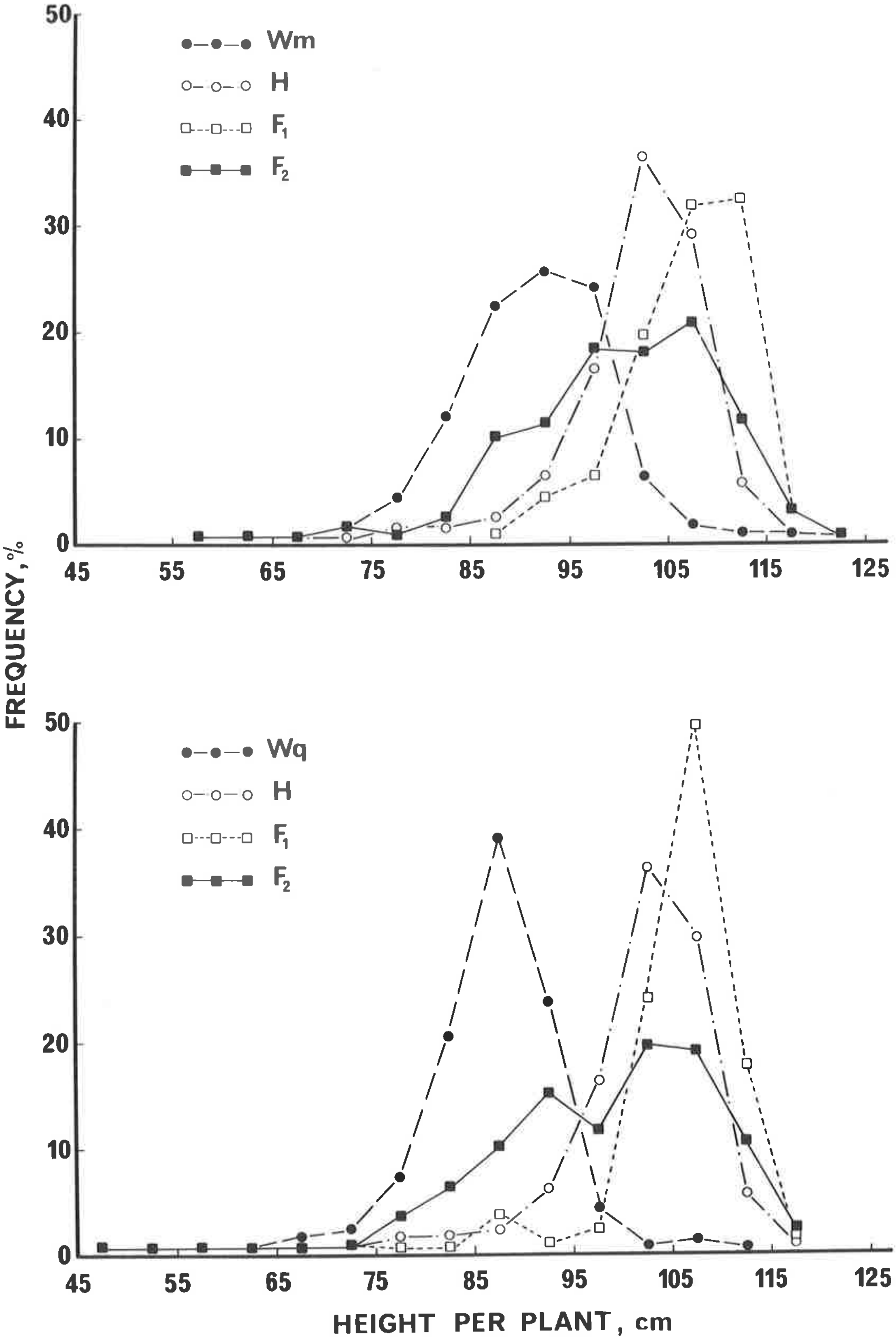
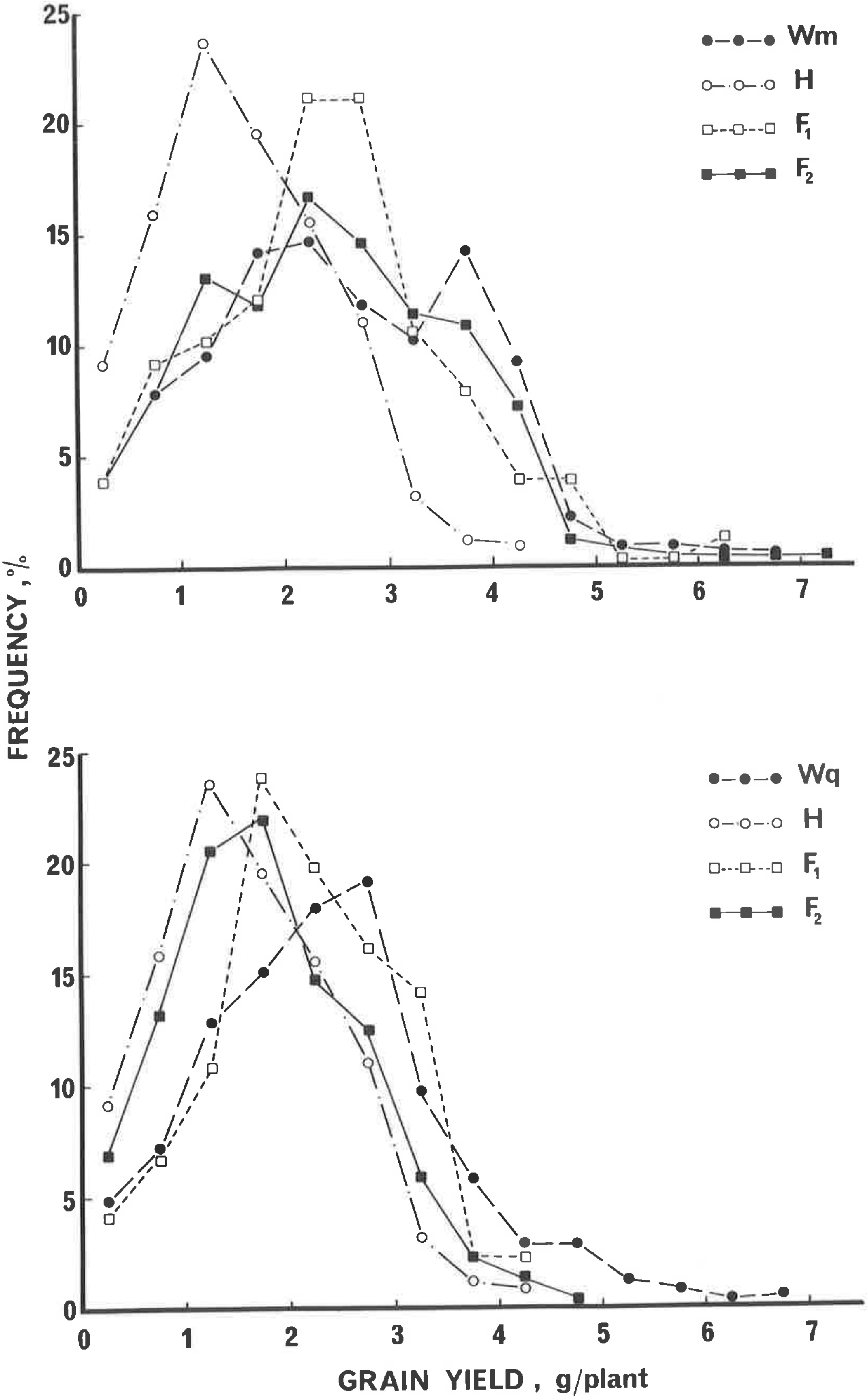

Wm
H
F1
F2
Wq
H
F1
F2
0
10
20
30
40
50
45
55
65
75
85
95
105
115
125
FREQUENCY, %
HEIGHT PER PLANT, cm

Figure 26: Distribution for grain yield per plant of Warimek, Halberd, (Warimek * Halberd) F_1 and (Warimek * Halberd) F_2.

Figure 27: Distribution for grain yield per plant of Wariquam, Halberd, (Wariquam * Halberd) F_1 and (Wariquam * Halberd) F_2.


25
20
15
10
5
0
0
1
2
3
4
5
6
7
Wm
H
F$_1$
F$_2$
FREQUENCY , %
25
20
15
10
5
0
0
1
2
3
4
5
6
7
Wq
H
F$_1$
F$_2$
GRAIN YIELD , g/plant

made to compare the means of the F_2's and the parents in each cross through an analysis of variance based on logarithmic transformed data and weighted means. These methods were aimed at inducing homogeneity of errors (Snedecor and Cochran, 1968; Yates and Cochran, 1938).

3.1.1 Means:

The means of all the characters measured of the parents, F_1 and F_2 for each cross are shown in Table 32. An analysis of variance was performed to compare the parents in each cross. Since it was found that the variances for grain weight and spike weight of Warimek and Wariquam differed significantly from those of Halberd (Table 34), the analyses of variance for these characters in Table 32 were based on logarithmic transformed data. Since there were some plants with zero grain yield, a value of 1.0 was added to the spike weight and grain weight of each plant before the log transformation.

It was found that, in each cross, Halberd was the tallest parent and Warimek or Wariquam the shortest. For grain yield, both Warimek and Wariquam were significantly higher yielding than Halberd. The higher yield of Warimek and Wariquam over Halberd was associated with more grains and more spikes per plant, respectively.

Although the F_1's were not included in the analyses of variance, it can be seen that in both crosses, they were taller than the tallest parent, Halberd, but were lower yielding than the higher yielding parents, Warimek and Wariquam.

Table 32: Means per plant of parents, F_1's and F_2's of all the characters measured.

(a) Warimek, Halberd and the F_1 and F_2 between these varieties.

Character	Warimek	Halberd	Diff. Wm & H	(Wm*H)F_1	(Wm*H)F_2
Height, cm	91.82	102.21	***	106.30	100.13
No. of spikes	2.45	2.34	NS	2.47	2.63
No. of grains	74.20	66.13	*	80.74	80.97
Total weight, g	6.37	5.80	NS	7.42	7.15
Spike weight, g	3.36	2.22	***	3.29	3.30
Grain weight, g	2.58	1.48	***	2.39	2.46

(b) Wariquam, Halberd and the F_1 and F_2 between these varieties.

Character	Wariquam	Halberd	Diff. Wq & H	(Wq*H)F_1	(Wq*H)F_2
Height, cm	86.90	102.21	***	105.81	96.06
No. of spikes	2.63	2.34	**	2.61	2.44
No. of grains	73.62	66.13	NS	75.80	68.04
Total weight, g	6.19	5.80	NS	6.85	6.04
Spike weight, g	3.13	2.22	***	2.91	2.52
Grain weight, g	2.28	1.48	***	2.05	1.73

An attempt was made to compare the means of the F_2 and parents in each cross through an alaysis of variance based on logarithmic transformed data and weighted means.

(i) Logarithmic transformed data: As can be seen in Table 34 there were differences in the variances between F_2's and parents for height, spike weight and grain weight, but not for number of spikes, number of grains and total weight per plant. Thus an analysis of variance to compare the means of F_2's and parents in each cross was based on logarithmic transformed data for height, spike weight and grain weight and on actual data for number of spikes, number of grains and total weight per plant. The results of this analysis are shown in Table 33.

It was found that there were significant differences in height between parents and F_2 in each cross. Halberd was always the tallest parent, the F_2's intermediate and Warimek and Wariquam were the shortest. In the cross, (Warimek * Halberd), there was no difference in spike weight and grain weight between Warimek and the F_2 but both were significantly higher than Halberd. In the cross, (Wariquam * Halberd), Wariquam had significantly higher spike weight and grain weight per plant than Halberd and the F_2. The high yield of Wariquam was associated with it having more spikes per plant than Halberd and the F_2. In the cross, (Warimek * Halberd), the F_2 also had significantly higher total weight per plant than Halberd.

(ii) <u>Weighted means</u>: The means of the F_2's and parents were also compared through an analysis of variance based on weighted means. The mean of each population was corrected according to its variance. Cochran (1937) suggested that the reciprocal of the error variance can be used as a measure of the appropriate weighting factor to establish homogeneity or errors. The weighting factor (w_i) for each plot of each of the F_2's and parents was calculated as follows:

$$w_i = \left(\frac{1}{s_i^2} \Big/ \sum_{1}^{15} \frac{1}{s_i^2} \right) \times 15$$

where s_i^2 = variance for plot (i)

15 = total number of plots of F_2 and parents.

Table 33: Comparisons between parents and F_2 (based on actual and logarithmic transformed data).

(a) For Warimek, Halberd and their F_2

Character	Warimek	Halberd	(Wm*H)F_2	Differences	LSD(0.05)	Comments
Height (log)	1.96	2.01	2.00	***	0.01	$H > F_2 > Wm$
No. of spikes	2.45	2.34	2.63	NS		
No. of grains	74.20	66.13	80.97	NS		
Total weight, g	6.37	5.80	7.15	*	0.99	$F_2 > H$
Spike weight (log)	0.60	0.48	0.68	*	0.12	$(F_2, Wm) > H$
Grain weight (log)	0.52	0.37	0.58	**	0.12	$(F_2, Wm) > H$

(b) For Wariquam, Halberd and their F_2

Character	Wariquam	Halberd	(Wq*H)F_2	Differences	LSD(0.05)	Comments
Height (log)	1.94	2.01	1.99	***	0.01	$H > F_2 > Wq$
No. of spikes	2.63	2.34	2.44	*	0.19	$Wq > (F_2, H)$
No. of grains	73.62	66.13	68.04	NS		
Total weight, g	6.19	5.80	6.04	NS		
Spike weight (log)	0.58	0.48	0.51	***	0.05	$Wq > (F_2, H)$
Grain weight (log)	0.48	0.37	0.41	***	0.05	$Wq > (F_2, H)$

The weighted mean of each plot (y_i) to be used in the analysis of variance was therefore:

$$y_i = x_i \cdot w_i$$

where x_i = actual mean per plot.

However, the analysis of variance based on weighted means gave unrealistic results. Since the F_2's had large variances for height, their weighted means for this character were significantly lower than those of the parents. Weighted means for height were not significantly different between Warimek and Halberd and Wariquam and Halberd. For grain yield, since Halberd had small variances, its weighted mean was increased relative to the F_2's and Warimek and Wariquam. The analysis of variance showed that there was no significant difference in weighted yield of Wariquam, Halberd and (Wq*H)F_2, but Halberd had significantly higher weighted yield than Warimek and (Wm*H)F_2. Both of these results for height and grain yield did not accord with the actual means as depicted in Figures 24, 25, 26 and 27. Thus, it was decided not to draw any statistical conclusions on the comparisons based on weighted means.

3.1.2 Variances:

The variances between the 5 plots (replicates) of each population were tested for homogeneity, using the Bartlett's test (Snedecor and Cochran, 1968). It was found that these variances did not differ significantly and hence the pooled variance was used below for each population. The pooled variances of each F_2 and the parents were then tested for homogeneity. When the chi square (with 2 degrees of freedom) was significant, the variances were compared pair-wise, using a two-tailed F test.

The pooled variances, chi squares of the Bartlett's test and variance ratios of parents and F_2's are shown in Table 34. It was found that the variances differed significantly between parents and F_2's for the characters height, spike weight and grain weight. In both crosses, the variances for height of the F_2's were approximately twice as large as those of the parents, indicating genetic segregation for this character. The parents are commonly regarded as differing by a single major gene for height (but no evidence is available to confirm this). The F_1's also had small variances for height as compared with the F_2's. The distributions for height in Figures 24 and 25 were similar for both crosses and indicate that this fitted the "ideal" genetic model. The parents differed significantly in height (Table 32) although the distributions overlapped. The F_1's showed over-dominance and the distributions of the F_2's covered the whole of the parents' ranges.

For spike weight and grain weight, the variances vary with the means. The correlation coefficients for the 5 populations between means and variances were 0.976 for spike weight and 0.992 for grain weight. Warimek and Wariquam had the largest variances, those of the F_2's were intermediate and Halberd had the smallest variances for these characters. The variances for spike weight and grain weight of the F_1's were approximately equal to those of the F_2's. Inspection of Figures 26 and 27 reveals that the distributions of Warimek, (Wm*H)F_1 and (Wm*H)F_2 were similar, but that of Halberd was appreciably smaller. In the (Wariquam * Halberd) cross, there was a gradual increase in means and variances from

Table 34: Variances of parents and F_2's, variances of F_1's, Chi squares of Bartlett's test for homogeneity and F test of variance ratios between parents and F_2's for all the plant characters measured in Expt VII.

(a) Warimek, Halberd and their F_1 and F_2

	Variances				Bartlett's		Variance ratios		
Character	Warimek	Halberd	(Wm*H)F_2	(Wm*H)F_1	Chi square		Wm/H	Wm/F_2	H/F_2
Height	58.502	48.742	98.026	35.506	49.15	***	NS	**	**
No. of spikes	0.746	0.799	0.804	0.786	0.55	NS	NS	NS	NS
No. of grains	1023.110	1018.300	1116.860	1298.100	0.98	NS	NS	NS	NS
Total weight	7.910	7.914	8.908	10.561	1.69	NS	NS	NS	NS
Spike weight	2.485	1.269	2.238	2.367	42.08	***	**	NS	**
Grain weight	1.597	0.606	1.359	1.352	83.60	***	**	NS	**

(b) Wariquam, Halberd and their F_1 and F_2

	Variances				Bartlett's		Variance ratios		
Character	Wariquam	Halberd	(Wq*H)F_2	(Wq*H)F_1	Chi square		Wq/H	Wq/F_2	H/F_2
Height	45.185	48.742	118.727	39.047	121.37	***	NS	**	**
No. of spikes	0.733	0.799	0.657	0.966	3.68	NS	NS	NS	NS
No. of grains	938.490	1018.300	840.750	1166.500	3.56	NS	NS	NS	NS
Total weight	6.490	7.914	6.889	8.983	3.89	NS	NS	NS	NS
Spike weight	1.990	1.269	1.395	1.835	22.09	***	**	**	NS
Grain weight	1.230	0.606	0.795	0.792	48.44	***	**	**	*

Halberd to (Wq*H)F_2, (Wq*H)F_1 and Wariquam in that order. Thus, there was no evidence of genetic segregation for spike weight and grain weight. If it occurred, it was over-shadowed by the large micro-environmental variation.

3.1.3 Coefficients of variation (CV's).

The CV is the standard deviation expressed as a fraction or percentage of the mean. Since, for some characters, the mean and the standard deviation tend to change together, the CV's give an estimate of the relative variation in a population. Table 35 shows the average CV's for each F_2 and its parents. These were tested for significant differences, using the Kruskal-Wallis non-parametric one-way analysis of variance by ranks (Siegel, 1956), for which the H statistics are shown. This was done by ranking the CV's in a single series. As fifteen was the total number of stands of F_2 and parents for each cross, the smallest CV was replaced by rank 1, the next to smallest by rank 2 and the largest by rank 15. The sum of the ranks in each population was found. The Kruskal-Wallis test determined whether these sums of ranks were so disparate that they were not likely to have come from samples which were all drawn from the same population.

Theoretically, it was expected that the F_2's should have the largest CV's as a result of genetic segregation. It was found that the F_2 stands had significantly larger CV's for height than those of the parents and F_1's. Even so, the CV's for height were very small, varying only between 5.6% and 11.1%. The CV's for yield and yield components were not different between F_2's and parents. However, the CV's for

Table 35: Coefficients of variation of F_2's, parents and F_1's and H statistics for the Kruskal-Wallis analysis of variance by ranks. (CV's in percentages).

(a) Warimek, Halberd and their F_1 and F_2.

Character	Warimek	Halberd	$(Wm*H)F_2$	H statistic		$(Wm*H)F_1$
Height	8.2	6.8	9.9	10.14	**	5.6
No. of spikes	35.5	38.1	34.5	2.66	NS	35.8
No. of grains	43.1	47.9	41.9	3.12	NS	44.6
Total weight	44.4	48.2	42.2	2.42	NS	43.8
Spike weight	47.0	50.7	45.6	1.46	NS	46.8
Grain weight	48.8	52.6	48.2	0.96	NS	48.6

(b) Wariquam, Halberd and their F_1 and F_2.

Character	Wariquam	Halberd	$(Wq*H)F_2$	H statistic		$(Wq*H)F_1$
Height	7.7	6.8	11.1	9.14	**	5.9
No. of spikes	32.7	38.1	33.3	5.36	NS	37.7
No. of grains	41.2	47.9	42.8	3.62	NS	45.1
Total weight	40.8	48.2	43.6	5.18	NS	43.8
Spike weight	44.4	50.7	47.0	3.92	NS	46.6
Grain weight	47.8	52.6	51.7	1.22	NS	43.3

these characters were variable with Halberd always having the largest CV's. In the cross (Warimek * Halberd), the CV for the F_2 was the smallest, whereas in the cross (Wariquam * Halberd), that of Wariquam was the smallest. Furthermore, for all populations, grain yield had the largest CV, varying from 43.3% for $(Wq*H)F_1$ to 52.6% for Halberd. In brief, there was little variation in height and some of what there was could be attributed to genetic effects. On the other hand, the variation in grain yield was large and apparently due to environmentally induced variability.

3.1.4 Skewness:

The skewness of a distribution is a measure of its departure from the symmetry of the normal distribution. It had been hypothesized that the F_2's would be more skew than the F_1's and parents, as a result of f.d.a.. The average coefficient of skewness of each F_2 and its parents is shown in Table 36. These were also compared by the Kruskal-Wallis analysis of variance by ranks. It was found that the distributions for height and number of spikes per plant were mostly negatively skewed, whereas the distributions of the remaining characters were positively skewed. Skew distributions of the same form occur commonly in plant competition, especially at high densities (Koyama and Kira, 1956). However, of all the distributions, only those for height were statistically different between F_2's and parents. In both crosses, Halberd had the largest negative skew distribution for height, followed by the F_2's and Wariquam and Warimek. In the cross (Wariquam * Halberd), the F_1 had the largest negative coefficient of skewness for height. For the remaining characters, the F_2's generally had positive coefficients of skewness similar to those of the parents.

3.1.5 The highest yielding plant in each population:

If f.d.a. and/or genetic segregation operated in the F_2 population, it was expected that this population would contain some plants which were higher yielding than the highest yielding plants in the parents and the F_1 population. Table 37 shows the performance of the highest yielding plant in each cross and their parents. In the cross

Table 36: Coefficients of skewness of parents, F_2's, F_1's and H statistics for the Kruskal-Wallis analysis of variance by ranks.

(a) Warimek, Halberd and their F_1 and F_2

Character	Warimek	Halberd	(Wm*H)F_2	H statistic		(Wm*H)F_1
Height	0.499	-1.651	-0.725	12.50	**	-0.704
No. of spikes	-0.033	-0.040	0.282	0.08	NS	-0.035
No. of grains	0.028	0.183	0.200	0.98	NS	0.071
Total weight	0.274	0.238	0.345	0.02	NS	0.028
Spike weight	0.302	0.262	0.337	0.02	NS	0.336
Grain weight	0.258	0.261	0.279	0.00	NS	0.338

(b) Wariquam, Halberd and their F_1 and F_2

Character	Wariquam	Halberd	(Wq*H)F_2	H statistic		(Wq*H)F_1
Height	-0.113	-1.651	-0.711	6.05	*	-2.595
No. of spikes	-0.086	-0.042	-0.063	0.08	NS	0.357
No. of grains	0.103	0.183	0.278	0.78	NS	0.227
Total weight	0.267	0.238	0.238	0.06	NS	0.200
Spike weight	0.321	0.262	0.347	0.42	NS	0.688
Grain weight	0.247	0.261	0.390	1.82	NS	0.026

Coefficient of skewness for significant difference from zero at the 5% probability level = 0.213 for sample size of 350 (approximate guide, for the parents & F_2's)

= 0.432 for sample size of 80 (approximate guide for the F_1's).

Table 37: Characteristics of the highest yielding plant in each population.

(a) Warimek, Halberd and their F_1 and F_2

Character	Warimek	Halberd	(Wm*H)F_1	(Wm*H)F_2
Height, cm	99.7	105.7	106.0	104.1
No. of spikes	4.0	5.0	4.0	5.0
No. of grains	164.0	165.0	182.0	188.0
Total weight, g	14.9	14.6	16.1	16.9
Spike weight, g	8.4	6.2	8.2	8.8
Grain weight, g	6.7	4.4	6.1	7.1

(b) Wariquam, Halberd and their F_1 and F_2

Character	Wariquam	Halberd	(Wq*H)F_1	(Wq*H)F_2
Height, cm	82.2	105.7	108.8	108.3
No. of spikes	6.0	5.0	6.0	4.0
No. of grains	160.0	165.0	175.0	178.0
Total weight, g	15.2	14.6	15.5	14.4
Spike weight, g	8.3	6.2	6.1	6.4
Grain weight, g	6.5	4.4	4.2	4.5

(Warimek * Halberd), the highest yielding plant was in the F_2 but this plant was only slightly higher yielding than that found in Warimek. In the cross (Wariquam * Halberd), the highest yielding plant was in Wariquam, with Halberd, F_1 and F_2 having similar maxima. It is concluded that f.d.a. had a negligible effect on the distribution of single plants in these F_2 populations.

3.2 Experiment VIII

As the original experiment (with 3 replicates) was damaged by birds, two additional replicates were planted and protected by netting.

These, and two of the less damaged replicates from the first planting, were harvested and measured. When the results were analysed, it was found that those from the first seeding were different from one another and different from the two later sown replicates. Figure 28 shows the relationship between yield in mixture and yield in pure stand for each of the four replicates. Several points worth mentioning regarding these results. Firstly, as a result of a difference of 4 weeks in the time of planting, the yields in replicates 1 and 2 were higher than those in replicates 3 and 4. Secondly, the first two replicates and particularly number 2 showed little relationship between yield in mixture and yield in pure stand whereas a positive relationship occurred in the other two. An F test of the variance ratios indicated that replicates 1 and 2 were different from one another and different from replicates 3 and 4 and this was attributed to the bird damage on the first two replicates. The variances of the two latter replicates were not different. The results from the first two replicates have been omitted from further analyses.

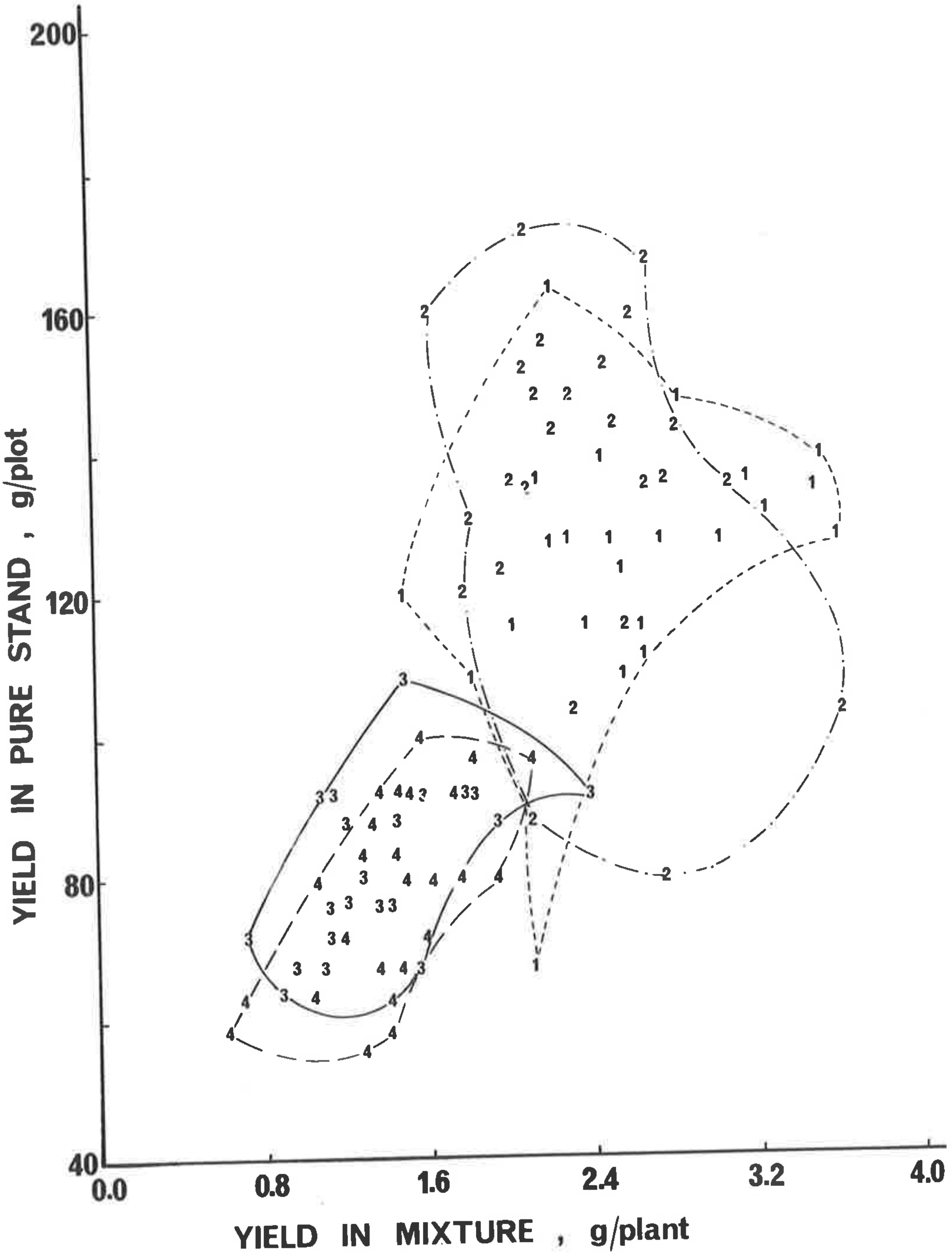
The mean yields for 25 F_5 lines are shown in Table 38. The yields at 4% and 16% frequencies and in the mixtures are the means per plant, and the yields in the pure stands are the means per plot.

Since the pure stand plots were planted with one seed per hole whereas in the other treatments two seeds were planted and the seedlings thinned, the percentage of establishment in the pure stands was lower than that in the mixtures and at the two frequencies by 23%. Hence the results in the pure stands are presented as yields per plot rather than yields per plant. As the correlation between the yield per plot and

Figure 28: Relationship between yield in mixture and yield in pure stand of 25 F_5 lines of (Warimek * Halberd) in each of the 4 replicates.

Replicates 1 and 2 were seeded in June and damaged by birds, and replicates 3 and 4 were seeded in July.

r_1, r_2, r_3 and r_4 are the correlations between yield in mixture and yield in pure stand for each of the 4 replicates, respectively.



correlations between
yields in mixture and
yields in pure stand

$r_1 =$ 0.255 NS
$r_2 =$ 0.063 NS
$r_3 =$ 0.477 *
$r_4 =$ 0.548 **

Table 38: Grain yield of 25 F_5 lines of the cross (Wm*H) when grown at frequencies 4% and 16% in the parents, in mixtures of 25 genotypes and in pure stands.

	Mean per plant, g			Mean per plot, g
Line number	4% frequency	16% frequency	Mixture	Pure stand
1	1.72	1.21	1.27	66.2
2	1.27	1.04	1.68	95.0
3	0.77	0.98	0.93	70.0
4	1.26	0.95	1.69	84.2
5	1.55	0.95	1.19	65.1
6	1.17	0.76	0.72	68.1
7	0.63	0.74	1.09	60.0
8	1.43	1.05	1.22	82.7
9	1.62	0.87	1.20	69.0
10	1.62	1.06	1.57	78.2
11	0.93	1.16	1.31	73.0
12	0.94	1.25	1.08	70.3
13	1.09	0.80	1.50	79.4
14	1.37	1.03	1.79	94.1
15	1.60	1.46	1.82	91.3
16	1.60	1.27	1.49	87.6
17	1.59	1.10	1.32	80.5
18	1.33	0.96	1.45	88.2
19	1.16	1.39	1.78	102.6
20	1.57	1.28	1.23	92.2
21	1.19	0.98	1.35	73.5
22	1.62	1.10	1.89	92.4
23	1.09	0.98	1.40	73.0
24	1.53	1.37	1.32	86.3
25	1.36	1.00	1.34	86.1
Mean	1.32	1.07	1.39	80.4

number of plants surviving to maturity of each genotype was non-significant, the difference in the pure stand yields could be attributed to genetic effects.

3.2.1 Frequency-dependent advantage of 25 lines:

Twenty of the 25 lines had higher yields at 4% frequency, whereas the other 5 lines (3,7,11,12 and 19) had higher yields at 16% frequency. For all lines, the yields per plant at 4% and 16% frequencies were higher than their yields in pure stands. The difference between the low and high frequencies was highly significant for all characters, except height (Table 39a). It is concluded that these lines again exhibited f.d.a. when grown in a stand of the parents.

However, except for height, there were no differences between lines as main effects and no interactions between frequencies and lines. The non-significance of the lines effect for grain yield was due to a significant interaction between replicates and lines (Table 39b). The mean yield of the background parents was 87 g/plot which was similar to the mean yield of the 25 lines in pure stands.

3.2.2 Yields in mixtures and yields in pure stands:

There were significant differences between the 25 lines when grown as single plants in mixtures and between the lines when they were grown in pure stands (Tables 40 and 41), indicating that there was genetic variability between these lines in both circumstances.

The yield in the mixture was higher than the yield in pure

Table 39: A summary of the analyses of variance for the comparison of 25 F_5 lines of (Wm*H) when grown in a stand of the parents at 2 frequencies, and the detailed analysis of variance for grain weight.

(a) All characters:

Character	Replicates	Lines	Frequencies	Lines x Frequencies
Height	*	*	NS	*
No. of spikes	NS	NS	***	NS
Total weight	NS	NS	***	NS
Spike weight	NS	NS	***	NS
Grain weight	NS	NS	***	NS

(b) Grain weight:

Source of variation	df	Mean square	F ratio	Probability	
Replicates	1	0.2704	1.74	0.199	NS
Lines	24	0.1665	1.07	0.429	NS
R x L (error a)	24	0.1547	2.43	0.016	*
Frequencies	1	1.5675	24.68	0.000	***
Lines x Freqs	24	0.0800	1.26	0.285	NS
R x L x F (error b)	25	0.0635			

Table 40: A summary of the analyses of variance for 25 F_5 lines of (Wm*H) when grown in mixtures of 25 genotypes.

Character	Replicates	Lines
Height	NS	*
No. of spikes	NS	NS
No. of spikelets	NS	NS
No. of grains	NS	*
Total weight	NS	NS
Spike weight	NS	*
Grain weight	NS	*

Table 41: A summary of the analyses of variance for 25 F_5 lines of (Wm*H) when grown in pure stands.

Character	Replicates	Lines
Height	NS	NS
No. of spikes	NS	NS
Total weight	NS	NS
Spike weight	NS	*
Grain weight	NS	***

stand and yields at the two frequencies. One reason for this high yield in the mixture may have been that the yield in the mixture was determined on a single plant basis. Each plant was individually measured and threshed by hand. Plants in the pure stands and at the two frequencies were harvested and processed in bulk, using a threshing machine. It was possible that there had been some slight loss of grain and that these latter yields were thereby underestimated.

As it was intended to study the relationship between the yield of F_5 plants in mixtures and the same lines in pure stands, grain yield per plant in the mixture has been plotted against grain yield per plot in pure stand (Figure 29). The correlation coefficient was 0.749 and was significant at the 0.1% level. Thus, it should have been possible to select for yield on the basis of single plants in a mixture. This is supported by the fact that 5 of the 6 highest yielding lines in mixtures were also highest yielding in pure stands.

There were highly significant correlations between grain yield in pure stands and yield and yield components in mixtures, but no correlation between height in mixtures and yield in pure stands (Table 42). The

Figure 29: Relationship between yield in mixture and yield in pure stand of 25 F_5 lines of (Warimek * Halberd).

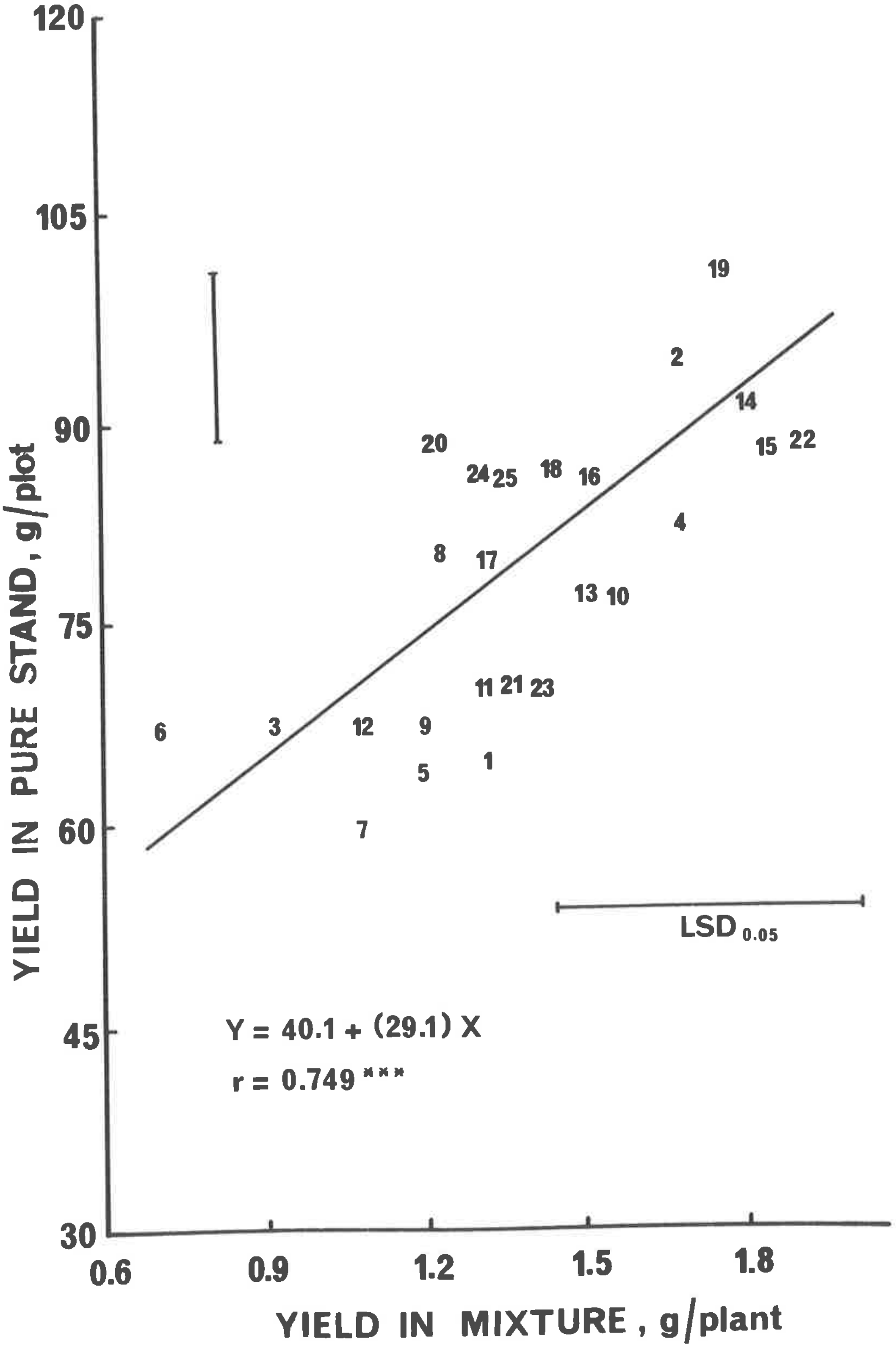

120
105
90
75
60
45
30
0.6
0.9
1.2
1.5
1.8
YIELD IN PURE STAND, g/plot
YIELD IN MIXTURE, g/plant
19
2
14
15
22
20
24
25
18
16
4
8
17
13
10
11
21
23
6
3
12
9
1
5
7
LSD 0.05
Y = 40.1 + (29.1) X
r = 0.749 ***

Table 42: Correlations between grain yield in pure stands and characters measured on single plants in mixtures.

Yield in pure stand and	Height in mixture	0.167	NS
	No. of spikes in mix.	0.579	**
	No. of spikelets in mix.	0.642	***
	No. of grains in mix.	0.627	***
	Total weight in mix.	0.668	***
	Spike weight in mix.	0.721	***
	Grain weight in mix.	0.749	***

highest correlation was 0.749, between yield in mixture and yield in pure stand. Thus, it was shown that selection for tall plants in mixtures would not result in high yielding genotypes in pure stands. Success could be achieved, however, if selection was based on yield or the yield components in the mixture.

A stepwise regression analysis was performed with yield in pure stand as the dependent variable and all the characters measured in the mixture as independent variables. The stepwise regression would indicate whether addition of any of the plant characters would increase the efficiency of selection for yield.

The result of this analysis is shown in Table 43.

Although most characters measured in the mixture had significant partial correlations with yield in pure stand, the inclusion of further characters in the equation did not reduce the error mean squares significantly. This is explained by the very high correlations between yield in the mixture and other plant characters measured in the mixture (Table 44).

Table 43: Stepwise regression of characters in mixture on yield in pure stand.

Variable entered in equation	Multiple R	Multiple R^2	Increase in R^2	Significance
Yield in mix.	0.749	0.561	0.561	***
No. of spikes in mix.	0.770	0.593	0.032	NS
Height in mix.	0.779	0.607	0.014	NS
No. of spikelets in mix.	0.782	0.612	0.005	NS
No. of grains in mix.	0.790	0.624	0.012	NS
Total weight in mix	0.792	0.627	0.003	NS
Spike weight in mix.	0.795	0.632	0.005	NS

Table 44: Correlations between grain yield in mixtures and characters measured on the same plants in mixtures.

Yield in mixture and	Height in mixture	: 0.403	*
	No. of spikes in mix.	: 0.884	***
	No. of spikelets in mix.	: 0.897	***
	No. of grains in mix.	: 0.923	***
	Total weight in mix.	: 0.954	***
	Spike weight in mix.	: 0.991	***

Thus, the variation in characters measured in mixture accounted for only 63.2% of the variation in pure stand yields. The unexplained proportion of the variation could be attributed to environmental variation within the experimental area and the difference between inter-genotypic competition in the mixture and intra-genotypic competition in the pure stand. The low proportions of the variation accounted for also reflects that there could have been some errors in obtaining the pure stand yields as these were based on only two replications.

The coefficients of variation for different characters in mixtures and pure stands give an indication of the relative variation in the two treatments (Table 45). These coefficients of variation are based on line means. For all characters, except number of spikes, the coefficient of variation in the mixture was larger than that in pure stand. The difference was largest for grain weight, indicating that yield differences between lines were increased in the mixture.

Table 45: Coefficients of variation (%) of characters measured in mixtures and pure stands.

Character	Mixture	Pure stand
Height	5.0	4.2
Total weight	15.5	10.1
Spike weight	18.8	14.1
Grain weight	20.8	13.8
No. of spikes	11.3	11.4
No. of spikelets	13.9	
No. of grains	16.3	

The correlation between grain yield in mixtures and other characters measured in mixtures showed that both height and other yield components were correlated with grain yield (Table 44). Thus, the taller plants tended to be higher yielding in the mixture. However, the correlation between height and yield was only 0.403 as compared with the correlation between yield and yield components (approximately 0.90 for most characters).

The correlations between grain yield in pure stands and other characters measured in pure stands are shown in Table 46. There were significant correlations between yield and yield components but no

Table 46: Correlations between grain yield in pure stands and characters measured on the same lines in pure stands.

Yield in pure stand and	Height in pure stand	:	0.067	NS
	No. of spikes in p.s.	:	0.528	**
	Total weight in p.s.	:	0.710	***
	Spike weight in p.s.	:	0.841	***

correlation with height. Thus, the tallest genotypes were not necessarily the highest yielding ones.

3.2.3 The relationship between f.d.a. and the yields in mixture and pure stand.

Although the 25 lines showed f.d.a. in a stand of the parents, the positive correlation between yield in mixture and yield in pure stand suggests that f.d.a. was not an important factor in the mixture. In this, and other experiments where the effect of f.d.a. was observed, the test plants were grown at low frequencies in relatively homogeneous backgrounds. In the mixtures, however, each of the 25 genotypes was occurring at a frequency of 4% and the f.d.a., if there was any, was not restricted to only a few of the genotypes. It had been suggested earlier (discussion of section B, part a) that different genotypes in a mixture may have different mechanisms for f.d.a.. Table 47 shows the correlations between grain yields in mixture and pure stand and the yields at the two frequencies and f.d.a., where f.d.a. was estimated by the difference between the yields at the low and high frequencies, for each line.

Table 47: Correlations between grain yields in mixture and pure stand and yields at 4% and 16% frequencies and f.d.a.

	Mixture		Pure stand	
Frequency 4%	0.353	NS	0.348	NS
Frequency 16%	0.386	NS	0.532	**
F.d.a. (4%-16%)	0.106	NS	0.001	NS

The correlation between the yield at 4% frequency and the yield in the mixture was non-significant, although each line also occurred at a frequency of 4% in the mixture. Nor was the correlation significant between yield in the mixture and yield at 16% frequency. The correlations between yield in pure stand and yields at the two frequencies were positive but was only significant between the yield in pure stand and the yield at 16% frequency, indicating that the yield at the high frequency was more related to the yield in pure stand than the yield at the low frequency. The very small and non-significant correlations between f.d.a. and yields in mixture and pure stand further confirmed that f.d.a. had little or no effect in a mixture of many genotypes and was unrelated to the yield in pure stand.

A stepwise regression was performed with yield in pure stand as the dependent variable and yields at 4% and 16% frequencies and f.d.a. as independent variables (Table 48). The stepwise regression would indicate whether selection for yield in pure stand could be based on a combination of the yields at low frequencies and f.d.a. in a uniform background.

It was found that the yield at the 16% frequency accounted for 28.3% of the variation of the yield in pure stand. When the yield at the 4% frequency was also considered, the proportion of the variation

Table 48: Stepwise regression of yields at 4% and 16% frequencies and f.d.a. on yield in pure stand.

Variable entered in equation	Multiple R	R^2	Increase in R^2	Significance
Yield at 16% frequency	0.532	0.283	0.283	**
Yield at 4% frequency	0.553	0.306	0.023	NS
F.d.a. (a)				

(a) F.d.a. could not be included as an additional variable in the equation since it had a very low F-level (or tolerance-level) insufficient for further computation, using the Stepwise regression program in SPSS (Nie et al., 1975).

accounted for increased by only 2.3% to 30.6%. The inclusion of f.d.a. as an additional selection criterion did not improve the efficiency of selection.

Thus it can be seen that it would be twice as efficient to select for yield in pure stand when the selection was based on yield and the yield components in mixtures rather than on yields at low frequencies and f.d.a. in homogeneous backgrounds.

4. Discussion.

Both of the present experiments were designed to investigate the effect of f.d.a. on single plant selection. The questions are whether f.d.a. would influence the choice of single plants in segregating populations and whether high yielding individuals in mixtures would also be high yielding in pure stands?

Experiment VII compared the variability between single plants in segregating populations with those in populations of genetically similar individuals. Although there were obvious genetic differences between the parents in each cross, the F_2's did not show any more variation than the F_1's and the parents, except for height. Since the F_2's had variance for grain yield intermediate between those of the parents, it was clear that there was large environmental variation among single plants in the parental stands and this variation was also very high relative to the sum of the environmental and genetic variations in the F_2 stands. Large variances in the parental stands relative to those in the F_2's have also been reported for cotton (Hutchinson, Panse and Govande, 1938) and for wheat (Palmer, 1952). This would suggest that the genetic variation within an F_2 is small relative to the environmental variation.

Of all the characters measured on single plants, only plant height showed any evidence of genetic segregation in the F_2's. Thus selection may be effectively based on this characteristic. However, the correlation between grain yield and height in the F_2 plots were small, 0.43 and 0.42 for the crosses (Wm*H) and (Wq*H), respectively. Furthermore, the results in experiment VIII indicated that there was no correlation between height in mixture and grain yield in pure stand (Table 42). Hence selection for height in the F_2 could not be used in selection for yield. Hamblin and Donald (1974) showed that there was a negative correlation between F_3 plant height and F_5 plot yield in barley, especially at high levels of nitrogen.

The use of skewness as a measure of the effect of f.d.a. in a segregating population was not successful. It had been expected that the F_2 would be more skew than the F_1 and the parents as the rare genotype would increase in yield relative to the more common ones. However, although most characters had skew distributions, the coefficients of skewness were not different between F_1's, F_2's and the parents, except for height (Table 36).

Skewness in the frequency distribution of plant weight in a population of genetically similar individuals has been reported by Koyama and Kira (1956). They found that there was a gradual change in the distribution from normal at the earliest growth stage to skewness to the right or "L" shape at a later growth stage. Their mathematical explanation of this phenomenon was based on Blackman's (1919) exponential equation of plant growth

$$w = w_o e^{rt}$$

where w is plant weight at time t, w_o is initial plant weight, and r is relative growth rate. The above equation can also be expressed in the logarithmic form

$$\log w = \log w_o + rt$$

$$r = \frac{1}{t} (\log w - \log w_o)$$

The authors postulated four possible models, with either w_o or r or both as a constant or as a normally distributed variable. Constant values of w_o can be achieved by using carefully selected uniform seeds, but normally distributed values are more usual and the value of r is influenced by a large number of factors of the environment. The two cases where w_o and r, or only r, are constant, were considered as too hypothetical, since it is unlikely that all individuals would have a constant growth rate even if they all had an equal initial weight. But when it was assumed that the r values are normally distributed and w_o is constant, then the distribution of log w must also be normal. That is, the frequency distribution of w is lognormal at any value of t. Similar results were obtained when the values of w_o are also normally distributed.

The authors concluded that the normal distribution in the seed automatically passes into the asymetric and finally into the L-shaped distribution, even when the individuals grow independently without any mutual influence.

The negative skew distribution of plant height was also reported by Koyama and Kira (1956). It can be explained that under crop conditions, plants tend to extend themselves vertically as a reaction against competition for light. A shorter plant would be less likely to survive unless it extends to reach the top of the canopy. This will eliminate the possibility of obtaining very small values for length of the main tiller, and results in a negative skew distribution of plant height.

The usual method of estimating the genetic variance of an F_2 population rests on the assumption that the environmental variance is the same in all genotypes. Falconer (1967) indicated that the environmental variance measured in one inbred line or cross is specific to this one particular genotype, and other genotypes may be more or less sensitive to environmental influence and may therefore show more or less environmental variance. This was demonstrated by the differences in the variances of the parents in each cross (Table 34). Also, Falconer (1967) pointed out that the environmental variance of a mixed population may not be the same as that measured in the genetically uniform group. In general, it was found that inbred individuals often show more environmental variation (or were less well buffered) than non-inbred individuals.

This may explain the similarity in the variances of parents, F_1's and F_2's although the latter should consist of variances attributable to genetic and environment components. In this case the large micro-environmental variation of the single plants of the parents and F_1 is approximately equal to the sum of the genetic and micro-environmental variation in the F_2.

Experiment VIII was aimed at comparing the yield of single plants in mixutres, where f.d.a. is expected to operate, and the yield of the same genotypes in pure stands. For this purpose replication was necessary and it was not possible to use F_2 seeds. The use of F_5 seeds from the same cross and extensive replications of the mixture plots is an attempt to reconstitute an F_2 population and overcome the problem of micro-environmental variation. Since the F_5 lines used in this experiment had been advanced from the F_1 by single-seed-descent (see experiment II), it was expected that they were genetically different from each other. Falconer (1967) indicated that the between-family variances increased with generations of inbreeding. The results in Tables 40 and 41 indicated that there was genetic variability between the F_5 lines in both the mixtures and pure stands.

The results on f.d.a. agreed with those found earlier for the cross (Warimek * Halberd) in experiments IA and IIA. The F_5 lines had higher yields at 4% than at 16% frequency, thus exhibiting f.d.a..

The positive correlation between yield in mixture and yield in pure stand indicated that selection in mixed population would result in high yielding genotypes in pure stands. Many authors (Allard, 1960; Bell, 1963; Shebeski, 1967) have considered that selection for yield in the

F_2 was ineffective. The main reasons usually advanced are that single plant performance is often confounded by genotype-environment interaction and inter-plant competition. The lack of success in many selection experiments could be attributed to the methods with which the experiments were conducted. For example, Hamblin (1971) selected F_3 single plants in 1967 and tested the F_5 lines in 1969, two entirely different years, climatically. McGinnis and Shebeski (1968) selected F_2 plants grown at a low density in 1966 and tested the selected lines at a commercial density in 1967. These authors concluded that single plant selection for yield was ineffective, but it is possible that their results could have been confounded by interactions between genotypes and seasons and genotypes and plant densities or a combination of these interactions. The factors limiting the efficiency of single plant selection have already been discussed in the literature review. As density, heterozygosity and genotype-environment interaction over sites and seasons were not factors in this experiment, the discussion therefore continues on the other likely contributors to the observed results.

In experiment VIII, since each genotype in the mixture was replicated 48 times, the effect of environmental variation between genotypes is minimized and the mean yield of each genotype therefore closely represents its genetic worth. The effect of micro-environmental variation in the mixture plots can be demonstrated by referring to Table 49 which shows the correlations between yield in mixture and yield in pure stand based on the yield of one plant per line, 4 plants per line (plot means), 24 plants per line (replicate means) and 48 plants per line (overall means per 2 replicates). It can be seen that the correlations increased progressively. When yields were based on single plants, the

Table 49: Correlations based on different numbers of single plants between yields in mixtures and yields in pure stands.

(a) Yields in mixtures based on single plants.

0.214	0.226	0.237	0.599 **
-0.089	0.326	-0.120	0.639 **
0.480 *	-0.198	0.258	-0.279
0.584 **	0.342	0.547 **	-0.142
0.270	0.423 *	0.255	-0.275
0.370	0.017	0.097	0.427 *
0.271	0.157	0.196	0.216
0.126	0.656 **	0.289	0.181
0.502 *	0.175	0.594 **	0.077
0.463 *	0.158	0.703 ***	0.187
0.243	0.080	-0.028	-0.067
-0.045	-0.287	0.349	0.207

Mean of 48 correlations = 0.225

(b) Yields in mixtures based on the means of 4 plants per plot.

0.395	0.320	0.291	0.606 **
0.150	0.186	0.509 **	0.362
0.501 *	0.334	0.207	0.317

Mean of 12 correlations = 0.348

(c) Yields in mixtures based on the means of 24 plants per replicate.

Replicate 1 : 0.477* Replicate 2 : 0.548 **

(d) Yields in mixtures based on the means of 48 plants in both replicates.

r = 0.749 ***

majority of the correlations was positive and some of them were statistically significant. None of the negative correlations was significant. The highest correlation was 0.749 ***, when yields in mixtures were based on the means of all 48 plants per line. However, the effect of micro-environmental variation still remains a factor since the correlation between yield in mixture and yield in pure stand only accounted for 56% of the variation. Also, there would have been fairly high errors in obtaining the yield of the pure stands and this may in part account for the low correlation above.

In this experiment, it was possible that there had been a difference between inter-genotypic competition in the mixture and intra-genotypic competition in the pure stand. Although the correlation between yield in mixture and yield in pure stand was highly significant, the unexplained proportion of the variation was still large (44%). Furthermore, the coefficient of variation for grain yield was greater in the mixture than in the pure stand and the ranking of some genotypes was different in the two treatments. Although the yields of the single plants in mixture may have been influenced by competition, it was evident that this effect was not large enough to conceal the correlation between yield in mixture and yield in pure stand. The negative correlation between competitive ability and yield in pure stand reported elsewhere could be attributed to the gross morphological differences in the components of the mixture (Jennings et al., 1968 a,b,c) which enhanced the competitive effects, or the unsatisfactory definition of competitive ability (Akihama, 1967) which includes yield in pure stand as one of its parameters.

The neighbourhood effect discussed in a previous section indicated that each plant in a mixture was competing against at least a number of neighbouring plants. Furthermore, it was suggested that f.d.a. was due to competition between plants of the same genotype growing nearby.

In experiment VIII it was found that the effect of f.d.a. was pronounced when the F_5 lines were grown at low frequencies in mixtures with the parents, but not in mixture of 25 lines. Thus, it is suggested that mixtures of many genotypes do not resemble mixtures of few genotypes, and that individuals in a multi-component mixture tend to look dissimilar from each other. This would have accounted for the lack of skewness in the distribution of F_2 plants in experiment VII and the positive correlation between yield in mixture and yield in pure stand in experiment VIII.

Although the correlation between f.d.a. and yield in pure stand was low ($r = 0.001$), the correlation between f.d.a. and mean yield at the 4% and 16% frequencies of the 25 F_5 lines was positive and significant at the 5% level ($r = 0.434$*). This is consistent with the results for the F_4 lines of (Wm*H) and (Wq*G) in experiment II. The contrast between the correlations between f.d.a. and mean yield at low frequencies and f.d.a. and yield in pure stand could be explained on the basis that genotypes with a high f.d.a. also have a large regression coefficient of yield on frequency. Hence, they tend to have a high mean yield when at low frequencies but their yield in pure stands could be lower than that of genotypes with a high yield in pure stands but low or insignificant f.d.a. at low frequencies. This is exemplified by referring to the relationship between grain yield per plant and frequency of Warimek and

Timgalen in Figure 15. Timgalen had a higher f.d.a. and a higher mean yield at low frequencies than Warimek, but its yield at a high frequency (93.75%) was similar to that of Warimek. Hence the correlation between f.d.a. and mean yield at low frequencies would be larger than that between f.d.a. and yield in pure stand.

Although 1974 was a rust epidemic year in South Australia, it was unlikely that differential responses to disease was a major factor in the mixture in experiment VIII as the susceptible genotypes would have been less affected in the mixtures than in the pure stands. If they had been, there would have been no correlation between yield in mixture and yield in pure stand or the correlation could have been negative. The small size of the pure stand plots (67cm x 67cm) and preventive spray with a solution of Diathane in November helped to reduce the effect of stem rust in the experiment.

From the results, it is concluded that micro-environmental variation was the major factor influencing the distribution of the yields of single plants in a segregating population. This would make selection in the F_2 difficult since replication is not possible in that generation. If this factor could be accounted for, it should be possible to select for high yielding genotypes based on the yield of single plants in the mixture. The positive correlation found in experiment VIII agrees with those found by Johnston (1972) for barley and supports the general conclusions of Allard (1960) and Allard and Hansche (1964) that genotypes which survive in mixed populations tend to have high yields in pure stands. F.d.a. was found to have no effect both on the distribution of single plants in F_2 populations and on the relationship between yield in mixture and yield in pure stand and was unlikely to influence single plant selection, except where differential responses to specific races of plant pathogen occurred between mixtures and pure stands.

V. COMPUTER SIMULATION OF FREQUENCY-DEPENDENT ADVANTAGE AND POPULATION CHANGES.

1. Theoretical model.

It has been suggested in the literature review that frequency-dependent selection (f.d.s.) is an important factor in maintaining genetic polymorphisms at a stable equilibrium. This section uses computer simulation to describe the changes in a polymorphic crop of a predominatly self-pollinating species resulting from frequency-dependent advantage (f.d.a.).

In recent years, a number of mathematical models describing f.d.s. have been advanced (Ayala and Campbell, 1974; a review). Some of the topics considered were the effect of f.d.s. in sexual selection, apostatic or mimetic selection, the relationship between f.d.s. and genetic loads and f.d.s. as a mechanism for the evolution of dominance. Other models have investigated the effect of niche-heterogeneity and density-dependent selection both of which often also result in frequency-dependent selective values. Of particular importance to the study here, however, are those simulations based upon the model of mixed selfing and outcrossing with selection developed by Hayman (1953). In this model, genetic changes in successive generations are described by recursion formulae which include relative viabilities of the different genotypes and relative proportions of selfing and outcrossing as the parameters.

Hayman's model considers an indefinitely large population in which yield is controlled by a single locus with 2 alleles A and a.

The genotypic frequencies of (AA, Aa, aa) in generation n are denoted by $(f_1^{(n)}, f_2^{(n)}, f_3^{(n)})$. The model supposes that the relative viabilities of the genotypes are in the ratios (x : 1 : y) and assumes that each individual has a constant probability s of selfing and $t = 1 - s$ of random outcrossing. Although the equations to describe genotypic frequencies differ depending on either the mode of selection or the stage of the life cycle at which scoring takes place (Workman and Jain, 1966), it will be assumed here that genotypic frequencies are scored just before mating and after all the selection has occurred. The recurrence relations describing genotypic frequencies in two successive generations, n and n + 1, are:

$$\left.\begin{aligned} f_1^{(n+1)} &\propto x[s(f_1^{(n)} + \tfrac{1}{4} f_2^{(n)}) + t(f_1^{(n)} + \tfrac{1}{2} f_2^{(n)})^2] \\ f_2^{(n+1)} &\propto [\tfrac{1}{2} s f_2^{(n)} + 2t(f_1^{(n)} + \tfrac{1}{2} f_2^{(n)}) (f_3^{(n)} + \tfrac{1}{2} f_2^{(n)})] \\ f_3^{(n+1)} &\propto y[s(f_3^{(n)} + \tfrac{1}{4} f_2^{(n)}) + t(f_3^{(n)} + \tfrac{1}{2} f_2^{(n)})^2] \end{aligned}\right\} \quad (1)$$

The proportionalities in equation (1) can be made equalities by dividing the terms to the right hand side of the proportionality sign for each genotype by the sum of the terms for the three genotypes.

When an independent estimate of the proportion of outcrossing, t, is available, these formulae can be used to estimate the selective values of the different genotypes (Allard and Hansche, 1964; Workman and Jain, 1966) from census data of genotypic frequencies. With these selective values, the formulae can be used to predict genotypic frequencies in successive generations (Allard and Adams, 1969b; Schutz and Usanis, 1969).

Mathematical solutions to the above equations have been developed by Hayman (1953) but they are complicated and difficult to interpret. Workman and Jain (1966) and Jain and Workman (1966) have shown that genotypic frequencies were functions of gene frequencies and the F statistic [$F = 1 - (f_2/2pq)$]. In the general case, F denotes the joint effect of inbreeding and selection and is a measure of the net deviation from Hardy-Weinberg expectations. In Wright's (1965) terminology, F is the fixation index. As such, F can vary between -1 and +1. Negative values of F indicate a higher level of heterozygosis than expected and vice versa. Using the F statistic as the fixation index, these authors showed that gene and genotypic frequencies at equilibrium can be calculated directly from relative viabilities of the three genotypes and proportions of selfing and outcrossing. However, although the use of F statistic to describe genotypic frequencies is convenient and useful, it should be noted that:

(i) $F = -1$ will arise when $x = y = 0$ regardless of the value of s.

(ii) $F = +1$ will arise when $s = 1.0$ provided that x and y are different from zero.

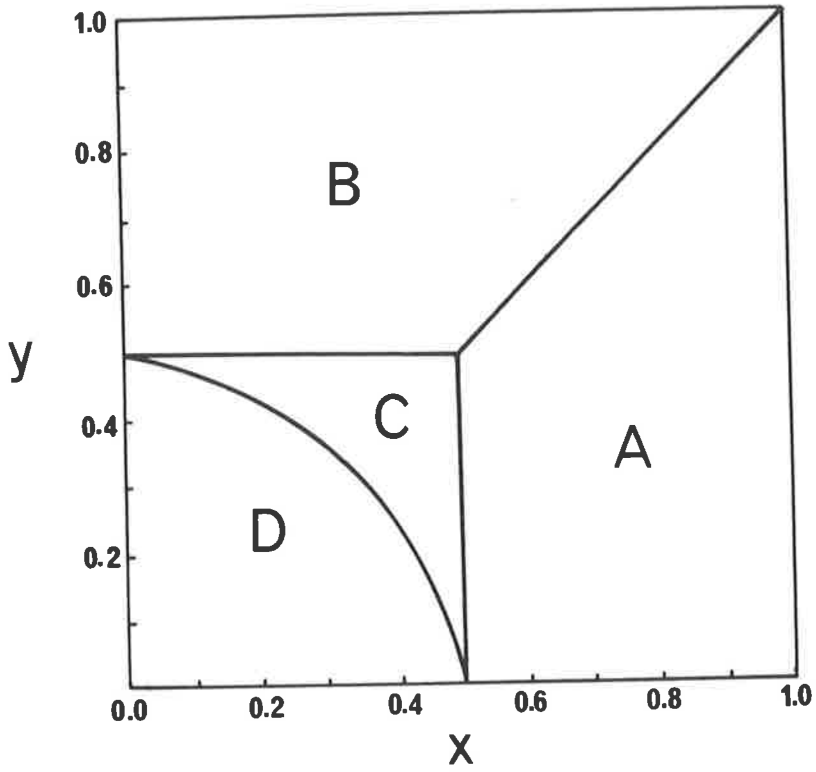
Thus, equation (1) on page 187 will be more meaningful, in general, than a reparametrisation in terms of F.

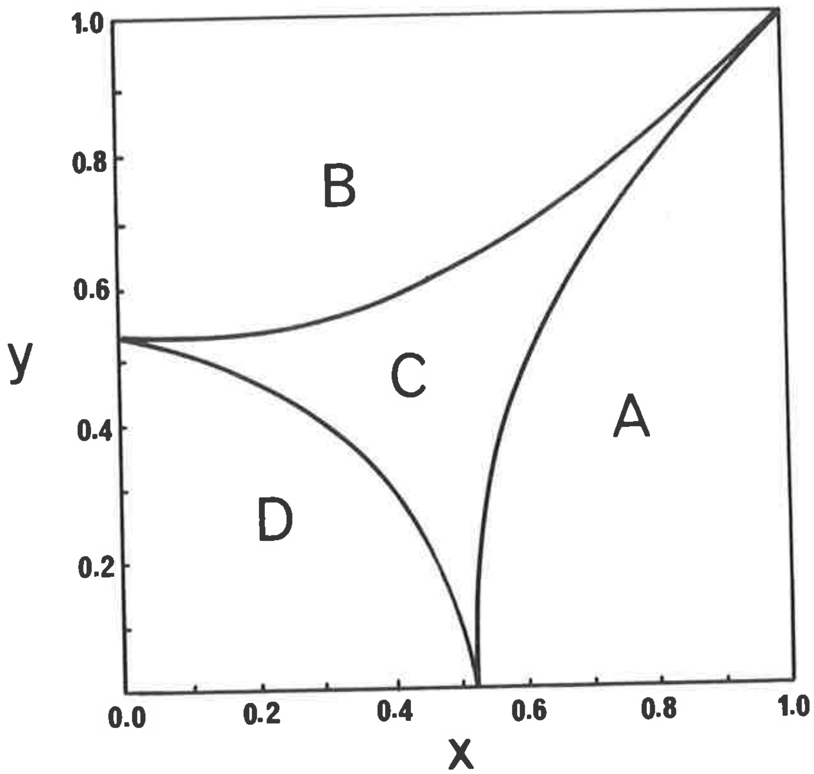
The equilibrium formulae also enable the development of phase diagrams (Hayman, 1953; Workman and Jain, 1966). Figures 30 and 31, reproduced from Workman and Jain (1966), show the types of equilibria for different magnitudes of the viabilities (x, y) of the two homozygotes for the cases of complete selfing ($s = 1.00$) and 5% outcrossing

Figures 30 and 31: (after Workman and Jain, 1966).

Phase diagrams showing the types of equilibria for different magnitudes of the relative viabilities (x, y) of the two homozygotes for the cases where there were complete selfing (Fig. 30) and 5% outcrossing (Fig. 31).

(see text for the description of areas marked A, B, C, D).


1.0
0.8
0.6
0.4
0.2
y
B
C
A
D
0.0
0.2
0.4
0.6
0.8
1.0
x


1.0
0.8
0.6
0.4
0.2
y
B
C
A
D
0.0
0.2
0.4
0.6
0.8
1.0
x

(s = 0.95), respectively.

In these diagrams, the areas marked A and B correspond to populations homozygous for AA and aa, respectively. In C the population, though containing heterozygotes, has more homozygotes than a population mating at random without selection, and in D it contains more heterozygotes. On the boundary between C and D, the effects of inbreeding and selection exactly counterbalance each other (Hayman, 1953). Several points are apparent from these diagrams:

(i) For the population to remain polymorphic, there must be heterozygote advantage.

(ii) The heterozygote advantage needed to maintain variability increases with the amount of selfing. When there is complete selfing, the fitness of the heterozygote must be at least twice those of the homozygotes for the population to remain at a non-trivial equilibrium.

(iii) F statistics and phase diagrams are useful in predicting the genotypic frequencies of the population at equilibrium, when selective values remain constant from generation to generation and are independent of genotypic frequencies.

When selective values are frequency-dependent, however, equilibrium frequencies cannot be predicted from pure stand performances. With the use of a computer, it is possible to simulate population changes resulting from frequency-dependent selective values.

Computer simulation based on Hayman's model has been studied by several authors. Allard and Adams (1969b) calculated the interaction between each pair of genotypes separately to obtain a matrix of reproductive values for computer simulation. Schutz and Usanis (1969) assumed that inter-genotypic competition is additive in nature and that the reproductive value of each genotype is determined by the sum of its competitive effects with other genotypes in the population. These authors showed that a wide range of competitive interactions (complimentary and overcompensatory with either additive, dominant or overdominant competitive effects) could maintain genotypic frequencies at a stable equilibrium. However, as their study was concerned with the effect of inter-genotypic competition, these authors expressed the reproductive value of each genotype as a function of the frequencies of other genotypes in the population.

The "inter-genotypic competition" approach did not emphasize the ability of a genotype to adjust its reproductive value as its frequency in the population changed. Thus, the role of frequency-dependent selection (or advantage) in maintaining genetic polymorphisms has not been clarified. Harding _et al_. (1966) found that, in populations of lima bean, the fitness of the heterozygotes increased as they became rare. When the frequency of the heterozygotes fell below an expected equilibrium value of 0.07, their selective value increased markedly in relation to those for the homozygotes, thus restoring the equilibrium. Frequency-dependent selection was postulated by Harding _et al_. (1966) to explain the retention of the polymorphism in these populations of lima bean. In the present study, the results from the first three experiments indicated that there was a linear relationship between the yield of each genotype and its frequency. Furthermore, as f.d.a. was apparently due to competition between plants of the same genotype, it would be more appropriate to express the reproductive value of each genotype as a linear function of its own frequency, regardless of the frequencies of other genotypes in the population.

The recursion formulae used here for the frequencies of the three genotypes in the (n + 1) generation are:

$$\left.\begin{aligned}
f_1^{(n+1)} &= [A_1 + B_1(f_1^{(n)})]\ [s(f_1^{(n)} + \tfrac{1}{4}f_2^{(n)}) + t(f_1^{(n)} + \tfrac{1}{2}f_2^{(n)})^2] \,/\, D \\
f_2^{(n+1)} &= [A_2 + B_2(f_2^{(n)})]\ [\tfrac{1}{2}sf_2^{(n)} + 2t(f_1^{(n)} + \tfrac{1}{2}f_2^{(n)})(f_3^{(n)} + \tfrac{1}{2}f_2^{(n)})] \,/\, D \\
f_3^{(n+1)} &= [A_3 + B_3(f_3^{(n)})]\ [s(f_3^{(n)} + \tfrac{1}{4}f_2^{(n)}) + t(f_3^{(n)} + \tfrac{1}{2}f_2^{(n)})^2] \,/\, D
\end{aligned}\right\}(2)$$

where $D = f_1^{(n+1)} + f_2^{(n+1)} + f_3^{(n+1)}$

The population mean fitness, W, can also be calculated. W in any generation, n, is given by:

$$W^{(n)} = \sum_{i=1}^{3} [A_i + B_i(f_i^{(n)})]\ f_i^{(n)} \qquad (3)$$

In equation (2), B_1, B_2 and B_3 are regression coefficients of reproductive values of the three genotypes on frequencies. A negative value of B indicates that the genotype has a higher reproductive value in a mixture than as a pure stand, increasing with decreasing frequencies. A value of $B = 0$ indicates that the genotype has a similar reproductive value at all frequencies. Genotypes with positive values of B suffer in competition since their reproductive values are highest in a pure stand, decreasing with lowering frequencies. The values A_1, A_2 and A_3 correspond to the selective values of the three genotypes when they occur at very low frequencies (when $f_i \simeq 0$). The pure stand reproductive value of each genotype is therefore equal to $A_i + B_i$

(when $f_i = 1.00$).

The f.d.a. of each genotype was defined as:

$$\frac{\text{yield at low frequency} - \text{yield in pure stand}}{\text{yield at low frequency}} = \frac{|B|}{A}$$

In the computer simulation, the reproductive value of the heterozygote in pure stand was set at 1.00 (i.e. $A_2 + B_2 = 1.00$). When a genotype expressed f.d.a. in a simulation, the change in its reproductive value was set at about 40%, which is comparable to that found experimentally, thus: $\frac{|B_i|}{A_i} \simeq 40\%$. The comparisons reported here were made between cases where selective values are independent of frequencies, and when selective values of 1, 2 or all 3 genotypes are frequency-dependent. Also the effect of complete selfing and partial outcrossing was compared. Outcrossing was then assumed to be 5%.

The method of simulation was deterministic iteration, with the reproductive value and frequency of each genotype in each generation being determined by its reproductive value and frequency in the previous generation. Populations were considered to have reached an equilibrium when frequencies are constant to 4 decimal places in successive generations. All populations began with F_1 plants and frequencies of homozygotes and heterozygotes were recorded for 50 generations. The mean population fitness in each generation was also recorded. Thus, in each example, the initial population was: $f_1^{(1)} = 0.00$, $f_2^{(1)} = 1.00$, $f_3^{(1)} = 0.00$ and the population fitness = 1.00 (pure stand reproductive value of the heterozygote).

2. Computer simulation.

2.1 Theoretical examples.

2.1.1 Case 1: The validity of the model was tested with the simplest case where there was no selection (equal fitness for all genotypes), no f.d.a. and no outcrossing. The parameters in equation (2) are:

$$A_1 + B_1(f_1) = A_2 + B_2(f_2) = A_3 + B_3(f_3) = 1.00 + 0.00(f_i)$$

$s = 1.00$ and $t = 0.00$

As expected, the frequency of the heterozygote decreased from 1.00 in $f^{(1)}$ to 0.50 in $f^{(2)}$, 0.25 in $f^{(3)}$ and so on. At the same time, frequency of each homozygote increased from 0.00 to 0.25 to 0.375 in the same generations (Figure 32). After 15 generations, the frequency of heterozygotes had reached 0.0000 (constant to 4 decimal places) and the population was left with the two homozygous classes each with a frequency of 0.50. The population mean fitness remained at 1.00 in all generations.

2.1.2 Case 2: The model was again tested in a case where fitnesses were the same as above (no selection), but all genotypes outcrossed at a rate of 5%. Thus $s = 0.95$ and $t = 0.05$.

The population reached equilibrium after 15 generations (Fig. 33). The proportions of the three genotypes at equilibrium were:

$$f_1^{(15+)} = 0.4762,\ f_2^{(15+)} = 0.0476,\ f_3^{(15+)} = 0.4762.$$

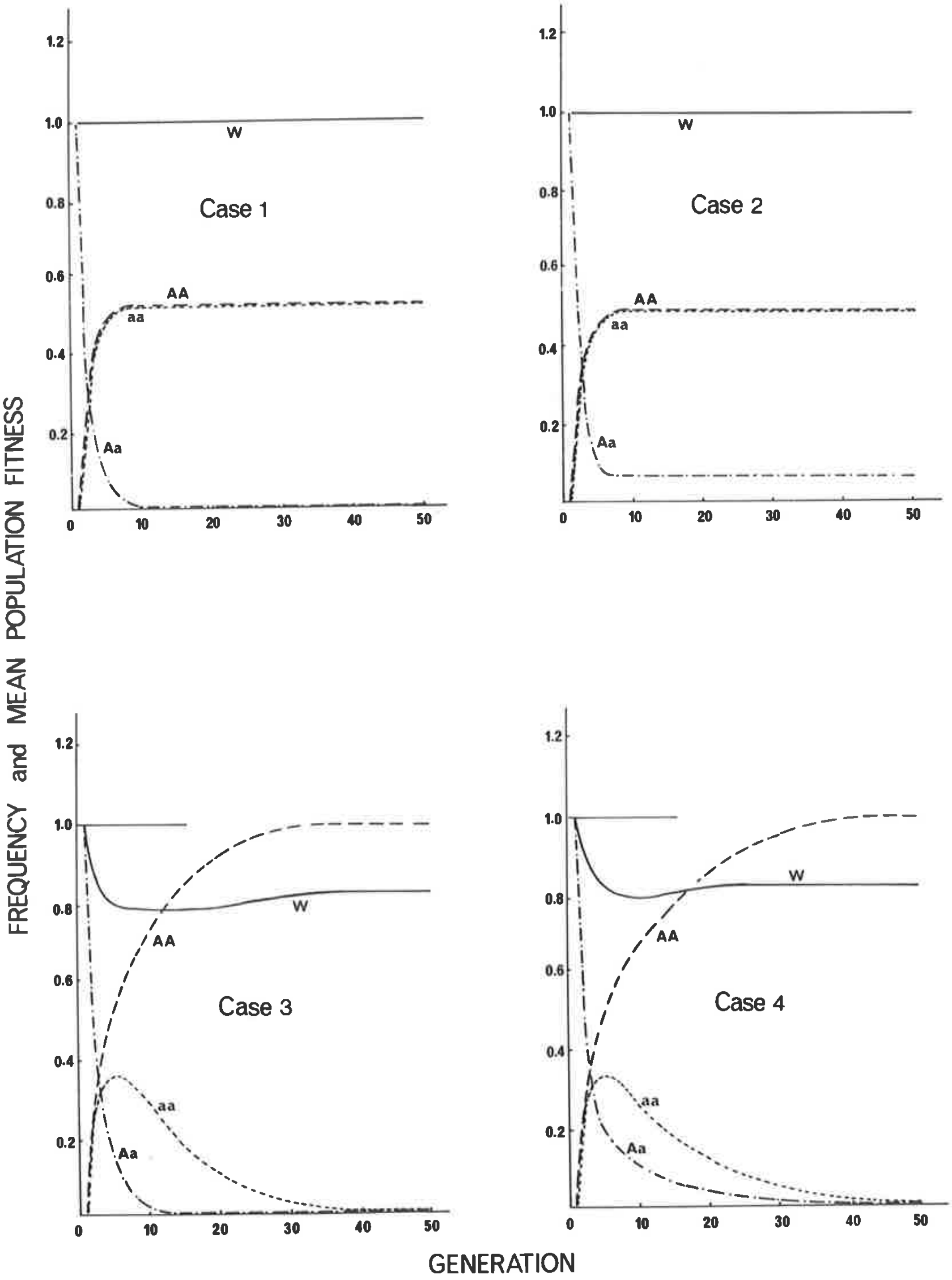
The population fitness remained at 1.00 in all generations.

Figures 32, 33, 34 and 35: Results of computer simulation of Cases 1, 2, 3 and 4.

Changes in genotypic frequencies (AA, Aa, aa) and mean population fitness (W) with generation.

Input values:

Case	A_1	B_1	A_2	B_2	A_3	B_3	t
1	1.00	0.00	1.00	0.00	1.00	0.00	0.00
2	1.00	0.00	1.00	0.00	1.00	0.00	0.05
3	0.80	0.00	1.00	0.00	0.70	0.00	0.00
4	0.80	0.00	1.00	0.00	0.70	0.00	0.05



FREQUENCY and MEAN POPULATION FITNESS
GENERATION
Case 1
Case 2
Case 3
Case 4
W
AA
aa
Aa
1.2
1.0
0.8
0.6
0.4
0.2
0
10
20
30
40
50

These results agree with those predicted. For a population with mixed selfing and outcrossing without selection, the proportions of homozygotes and heterozygotes at equilibrium are given by the equations:

$$\left.\begin{aligned} f_1 &= p^2 + pqF \\ f_2 &= 2pq\ (1 - F) \\ f_3 &= q^2 + pqF \end{aligned}\right\} \qquad (4)$$

where p and q are frequencies of the alleles A and a, respectively and F is the coefficient of inbreeding. Since there was no selection, $p = q = 0.50$ and at equilibrium $F = s/(2 - s)$ (Kempthorne, 1957). Substituting the values for p, q, and F into (4) gives the predicted genotypic frequencies at equilibrium.

2.1.3 Case 3: When there are differences in selective values ($x \neq 1 \neq y$), genotypic frequencies at equilibrium are also given by (4) but in this instance, $p \neq q$ and F is called the Fixation Index (Wright, 1965). It denotes the joint effect of inbreeding and selection. Workman and Jain (1966) showed that at equilibrium the following relations must hold:

$$p = \frac{(1 - y) - \tfrac{1}{2}\, s\ (1 - x)\ (1 + F)}{(2 - x - y)\ [\,1 - \tfrac{1}{2}\, s\ (1 + F)]} \qquad (5)$$

and $sF^2(1 - x)(1 - y) + F\ [2(xy - 1) + s\ (2 - x - y)] - 2(1 - x)(1 - y) + s\ (1 - xy) = 0$ (6)

Using the calculated values of p, q and F, genetic frequencies at equilibrium are given by:

$$\left.\begin{aligned} f_1 &= x\ [\ p^2 + \tfrac{1}{2}\ spq\ (1 + F)]\ /\ D \\ f_2 &= \ \ [\ 2pq - spq\ (1 + F)]\ /\ D \\ f_3 &= y\ [\ q^2 + \tfrac{1}{2}\ spq\ (1 + F)]\ /\ D \end{aligned}\right\} \qquad (7)$$

where D = sum of genotypic frequencies at equilibrium.

The above equations enabled the development of phase diagrams (Figures 30 and 31) which, as already mentioned, give an indication of the type of equilibrium for different selective values of the homozygotes.

In computer simulation, it was assumed that the relative viabilities were in the ratios (0.80 : 1.00 : 0.70). Phase diagrams indicated that at equilibrium, the population was homozygous for AA. Using computer simulation, it was found that with complete selfing, the heterozygotes were eliminated after 23 generations and by the 50th generation, the frequencies of AA and aa were 0.9981 and 0.0019 (Figure 34).

2.1.4 <u>Case 4</u>: When there was 5% outcrossing and the same relative viabilities as in Case 3, the proportion of the heterozygote at the 23rd generation was 0.0369 and genotypic frequencies at the 50th generation were 0.9831, 0.0053, 0.0116 respectively (Figure 35). Fixation of AA will eventually take place. The population fitness decreased from 1.00 in $f^{(1)}$ to 0.799 in $f^{(50)}$ which was approximately equal to that of AA in pure stand.

The examples thus far indicate that:

- When there was no selection, outcrossing was a powerful force in maintaining heterozygosity.

– When selection occurred, and $x > 0.5$ and $y > 0.5$, even with heterozygous advantage and outcrossing, the population eventually reached fixation. The genotype with the lower reproductive value was eventually eliminated from the mixture and hence the chance of heterozygotes being produced through outcrossing progressively diminished.

In the above examples, the B values were set at 0.00, so the fitnesses were independent of frequencies. In the following examples, the same cases were repeated with frequency-dependent selective values ($B \neq 0$). As outcrossing was necessary for the formation of heterozygotes in advanced generations, only cases where there was 5% outcrossing are reported.

2.1.5 Case 5: The hybrids and parents had similar reproductive values in pure stands but the hybrids had an advantage in mixture. The magnitude of f.d.a. of the hybrid was 37.5%. Thus input parameters were:

$$A_1 + B_1(f_1) = A_3 + B_3(f_3) = 1.00 + 0.00(f_i)$$

$$A_2 + B_2(f_2) = 1.60 - 0.60(f_2) \ ; \ \frac{|B_2|}{A_2} = 37.5\%$$

This example is comparable to case 2 above but with f.d.a. of the heterozygote. The population reached equilibrium after 21 generations (Figure 36). Genotypic frequencies at equilibrium were:

$$f_1^{(21+)} = 0.4399, \quad f_2^{(21+)} = 0.1203, \quad f_3^{(21+)} = 0.4399$$

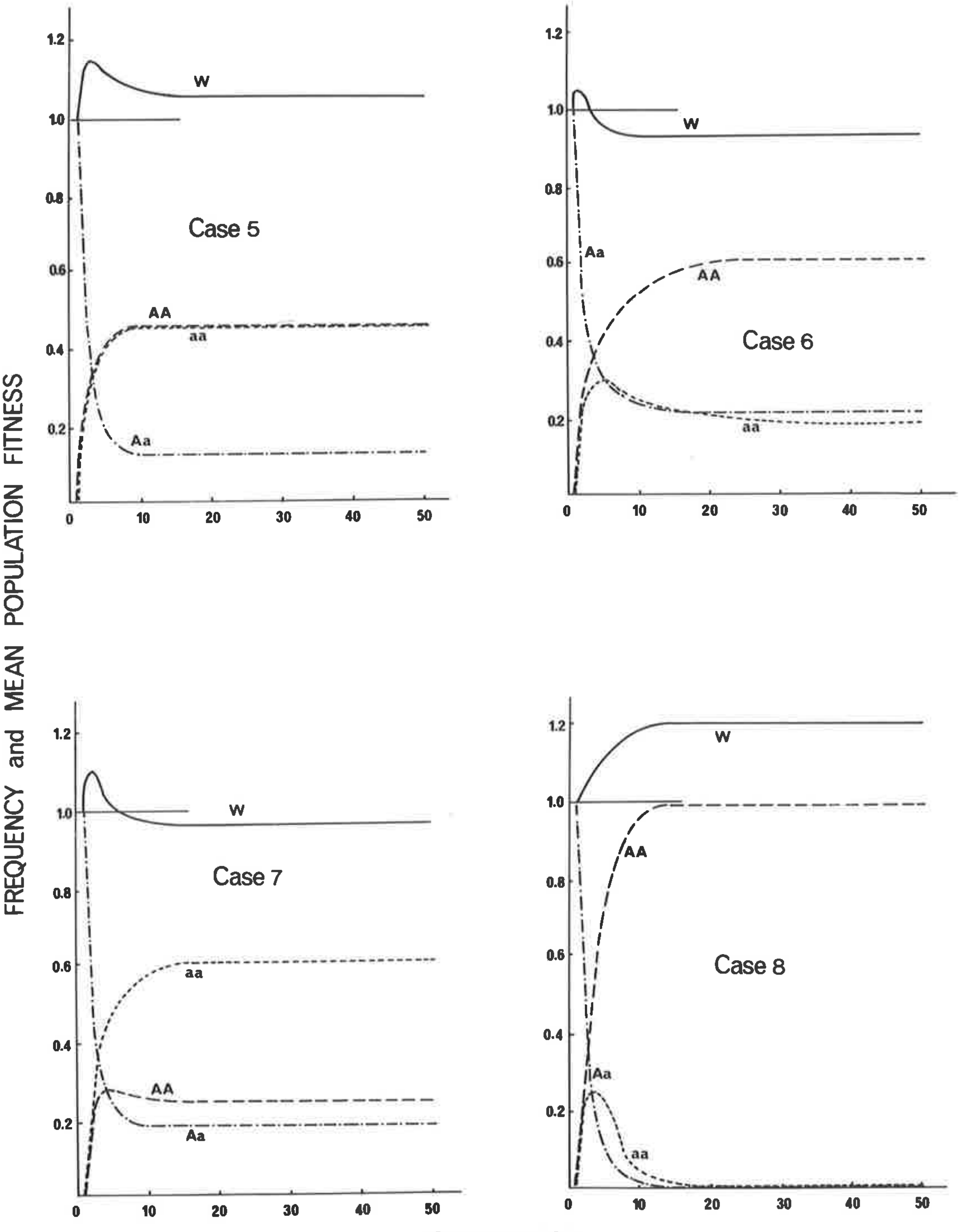
The population fitness increased from 1.00 in $f^{(1)}$ to 1.15 in $f^{(2)}$ as the heterozygote increased in its reproductive value as it decreased in frequency, but then fitness decreased as homozygotes increased in

Figures 36, 37, 38 and 39: Results of computer simulation of cases 5, 6, 7 and 8.

Changes in genotypic frequencies (AA, Aa, aa) and mean population fitness (W) with generation.

Input values

Case	A_1	B_1	A_2	B_2	A_3	B_3	t
5	1.00	0.00	1.60	-0.60	1.00	0.00	0.05
6	0.80	0.00	1.60	-0.60	0.70	0.00	0.05
7	0.80	0.00	1.60	-0.60	1.10	-0.40	0.05
8	1.20	0.00	1.00	0.00	0.80	0.00	0.05


FREQUENCY and MEAN POPULATION FITNESS
GENERATION
Case 5
W
AA
aa
Aa
Case 6
W
Aa
AA
aa
Case 7
W
aa
AA
Aa
Case 8
W
AA
Aa
aa
1.2
1.0
0.8
0.6
0.4
0.2
0
10
20
30
40
50

frequency. At equilibrium, the population fitness remained at 1.0635. Thus, when there was f.d.a. the equilibrium frequency of the heterozygote increased from 0.0476 to 0.1203 whereas those of homozygotes decreased from 0.4762 to 0.4399. The population fitness was also higher than when there was no f.d.a.. This example supports the results of Harding _et al._ (1966) which showed that f.d.a. of the heterozygote maintained its frequency at a level higher than that which could have been expected on the basis of outcrossing alone.

2.1.6 _Case 6_: Selection was now introduced together with f.d.a. of the heterozygote (c.f. case 4). The inputs were:

$$A_1 + B_1(f_1) = 0.80 + 0.00(f_1)$$

$$A_2 + B_2(f_2) = 1.60 - 0.60\ (f_2)$$

$$A_3 + B_3(f_3) = 0.70 + 0.00(f_3)$$

Although the population had not reached equilibrium by the 50th generation, there was very little change in genotypic frequencies from generation 25 onward (Figure 37). The proportions of the three genotypes at the 50th generation were 0.6088, 0.2004, and 0.1908, and the population fitness in this generation was 0.9171. Thus f.d.a. of the heterozygote was sufficient not only to increase its own frequency but also to maintain the lower yielding homozygote in the population.

2.1.7 _Case 7_: In this example, both the heterozygote and the lower yielding homozygote had an advantage in the mixture. The coefficients for the selective values were:

$$A_1 + B_1(f_1) = 0.80 + 0.00(f_1)$$

$$A_2 + B_2(f_2) = 1.60 - 0.60(f_2) \quad ; \quad \frac{|B_2|}{A_2} = 37.5\%$$

$$A_3 + B_3(f_3) = 1.10 - 0.40(f_3) \quad ; \quad \frac{|B_3|}{A_3} = 36.4\%$$

Pure stand reproductive values were still in the ratios (0.80 : 1.00 : 0.70) but there was 37.5% f.d.a. of the heterozgote and 36.4% f.d.a. of the lower yielding homozygote in the mixture. The simulation showed that there was very little change in frequencies from generation 20 onward and equilibrium was reached by the 32nd generation (Figure 38). Genotypic frequencies at that stage were 0.2393, 0.1663 and 0.5944 and the population fitness remained at 0.9535. It is noted that the frequency of the heterozygote was lower than in case 6 due to the f.d.a. of the lower yielding genotype and frequencies of the homozygotes were reversed. However, the population fitness was higher than in case 6. This example illustrates that a population may be dominated by a genotype which is low yielding in pure stand but has an advantage in mixture.

2.1.8 Case 8: In the previous exmples, the heterozygotes always had a higher selective value in pure stand than the homozygotes. In the following examples, the heterozygote was given a pure stand selective value intermediate between those of the homozygotes.

When there was no f.d.a., the selective coefficients were:

$$A_1 + B_1(f_1) = 1.20 + 0.00(f_1)$$

$$A_2 + B_2(f_2) = 1.00 + 0.00(f_2)$$

$$A_3 + B_3(f_3) = 0.80 + 0.00(f_3)$$

Even when there was 5% outcrossing, the heterozygote was eliminated by the 25th generation and by the 29th generation the population was completely dominated by AA. The population mean fitness increased steadily and remained at 1.20 (Figure 39), which is the same as the pure stand reproductive value of the higher yielding homozygote.

2.1.9 Case 9: The reproductive values of the homozygotes were the same as in case 8 but the heterozygote now had an f.d.a. of 37.5%. Thus its reproductive coefficients were: $1.60 - 0.60(f_2)$. The results showed that both elimination of the heterozygote and fixation of AA occurred by the 45th generation even with 5% outcrossing (Figure 40).

2.1.10 Case 10: In this example, both the heterozygote and the lower yielding homozygote had an advantage in the mixture. The input parameters were:

$$A_1 + B_1(f_1) = 1.20 + 0.00(f_1)$$

$$A_2 + B_2(f_2) = 1.60 - 0.60(f_2)$$

$$A_3 + B_3(f_3) = 1.30 - 0.50(f_3)$$

When there was 5% outcrossing, there was little change in genotypic frequencies after the 30th generation (Figure 41). The proportions of the three genotypes at the 50th generation were:

0.6854, 0.0669, 0.2477; population fitness = 1.2182.

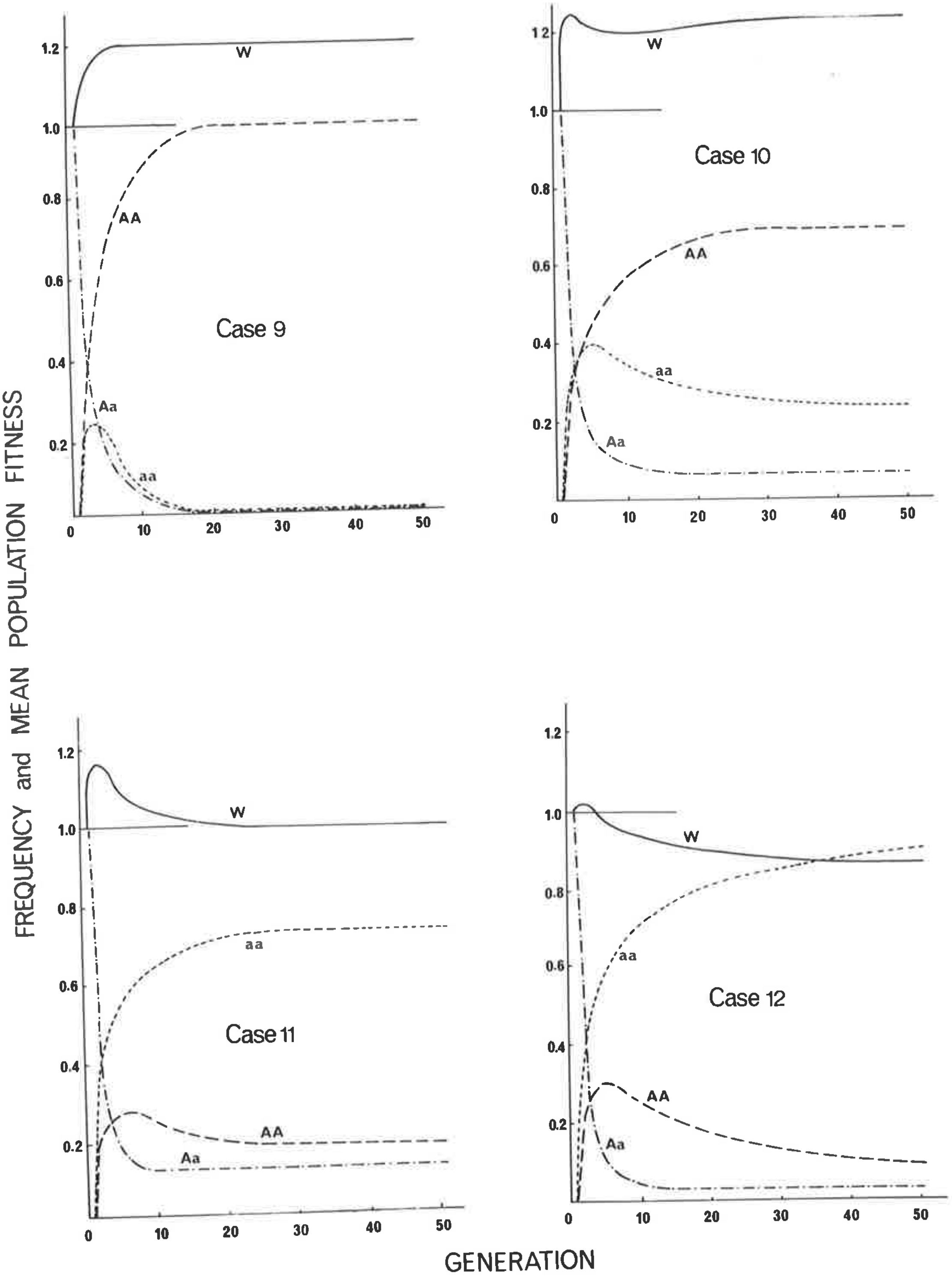
Thus, f.d.a, of the lower yielding genotype enabled it to remain in the population and contributed to the formation of heterozygotes

Figures 40, 41, 42 and 43: Results of computer simulation of Cases 9, 10, 11 and 12.

Changes in genotypic frequencies (AA, Aa, aa) and mean population fitness (W) with generation.

Input values

Case	A_1	B_1	A_2	B_2	A_3	B_3	t
9	1.20	0.00	1.60	-0.60	0.80	0.00	0.05
10	1.20	0.00	1.60	-0.60	1.30	-0.50	0.05
11	0.80	+0.40	1.60	-0.60	1.30	-0.50	0.05
12	0.80	+0.40	1.00	0.00	1.30	-0.50	0.05


FREQUENCY and MEAN POPULATION FITNESS
GENERATION
Case 9
W
AA
Aa
aa
1.2
1.0
0.8
0.6
0.4
0.2
0
10
20
30
40
50
Case 10
W
AA
aa
Aa
1.2
1.0
0.8
0.6
0.4
0.2
0
10
20
30
40
50
Case 11
W
aa
AA
Aa
1.2
1.0
0.8
0.6
0.4
0.2
0
10
20
30
40
50
Case 12
W
aa
AA
Aa
1.2
1.0
0.8
0.6
0.4
0.2
0
10
20
30
40
50

through outcrossing. The population fitness in this instance was higher than the pure stand fitness of the higher yielding homozygote.

2.1.11 Case 11: In the examples so far presented, the higher yielding genotype had been considered frequency-independent. If this genotype had an increased yield at the low frequency of the same magnitude as other genotypes, its frequency at equilibrium would have been much higher (unreported cases). There are cases, however, when the higher yielding genotype had a disadvantage when in competition (Khalifa and Qualset, 1974). The following examples consider such cases. The input parameters were:

$A_1 + B_1(f_1) = 0.80 + 0.40(f_1)$; pure stand = 1.20

$A_2 + B_2(f_2) = 1.60 - 0.60(f_2)$; pure stand = 1.00

$A_3 + B_3(f_3) = 1.30 - 0.50(f_3)$; pure stand = 0.80

With 5% outcrossing, computer simulation showed that there was little change in genotypic frequencies after the 25th generation (Figure 42). The proportions of the three genotypes in the 50th generation were:

0.1777, 0.1125, 0.7098 ; population fitness = 0.9980.

Again, a high selective value of the heterozygote in pure stand was not a necessary condition for a stable equilibrium, providing that fitnesses changed appropriately with frequency. This result is similar to that obtained by Lewontin (1958) through mathematical analysis (see Literature Review, page 25).

2.1.12 Case 12: Occasionally, it was found that f.d.a. was not associated with heterozygosity; for example, (Wm*T) F_1 in experiment IIIA. This final case is an example where the fitnesses of both homozygotes changed with frequencies but that of the heterozygote was independent of frequency. The coefficients of the three genotypes used here for simulation were:

$$A_1 + B_1(f_1) = 0.80 + 0.40(f_1) \quad ; \quad \text{pure stand} = 1.20$$

$$A_2 + B_2(f_2) = 1.00 + 0.00(f_2) \quad ; \quad \text{pure stand} = 1.00$$

$$A_3 + B_3(f_3) = 1.30 - 0.50(f_3) \quad ; \quad \text{pure stand} = 0.80$$

The population would eventually be dominated by the stronger competitor (Figure 43), but the heterozygote had not been eliminated by the 50th generation. Genotypic frequencies at that stage were:

0.1074, 0.0276, 0.8650; population fitness = 0.8685

Thus, even when the heterozygotes were intermediate between the parents in pure stand and had no advantage in mixture, they were still maintained in the population through outcrossing, providing that both parents were present.

A summary of the results for cases 1 to 12 is shown in Table 50.

2.2 Application to experimental data.

Although the previous examples had assumed genotypic frequencies arising from alleles at a single locus, similar consideration may be given to populations of parents and hybrids, particularly if they can be distinguished by simply inherited marker genes. In this

Table 50: Summary of the results of computer simulation in cases 1 to 12.

	Input Values							Results				
Case	t	A_1	B_1	A_2	B_2	A_3	B_3	n	$f_1^{(n)}$	$f_2^{(n)}$	$f_3^{(n)}$	W (n)
1	0.00	1.00	0.00	1.00	0.00	1.00	0.00	15	0.5000	0.0000	0.5000	1.0000
2	0.05	1.00	0.00	1.00	0.00	1.00	0.00	15	0.4762	0.0476	0.4762	1.0000
3	0.00	0.80	0.00	1.00	0.00	0.70	0.00	50*	0.9981	0.0000	0.0019	0.7998
4	0.05	0.08	0.00	1.00	0.00	0.70	0.00	50*	0.9831	0.0053	0.0116	0.7999
5	0.05	1.00	0.00	1.60	-0.60	1.00	0.00	21	0.4399	0.1203	0.4399	1.0635
6	0.05	0.80	0.00	1.60	-0.60	0.70	0.00	50*	0.6088	0.2004	0.1908	0.9171
7	0.05	0.80	0.00	1.60	-0.60	1.10	-0.40	32	0.2393	0.1663	0.5944	0.9535
8	0.05	1.20	0.00	1.00	0.00	0.80	0.00	29	1.0000	0.0000	0.0000	1.2000
9	0.05	1.20	0.00	1.60	-0.60	0.80	0.00	45	1.0000	0.0000	0.0000	1.2000
10	0.05	1.20	0.00	1.60	-0.60	1.30	-0.50	50*	0.6854	0.0669	0.2477	1.2182
11	0.05	0.80	+0.40	1.60	-0.60	1.30	-0.50	50*	0.1777	0.1125	0.7098	0.9980
12	0.05	0.80	+0.40	1.00	0.00	1.30	-0.50	50*	0.1074	0.0276	0.8650	0.8685

t = proportion of outcrossing

A_1 B_1, A_2 B_2, A_3 B_3 = reproductive coefficients of the three genotypes

n = generation at which equilibrium reached, 50* indicates that equilibrium had not been reached by the 50th generation.

$f_1^{(n)}$, $f_2^{(n)}$, $f_3^{(n)}$ = genotypic frequencies at equilibrium (or at the 50th generation)

$W^{(n)}$ = population mean fitness at generation n section, the changes in genotypic frequencies of Warimek, Timgalen and the F_1 were simulated using the results in experiment IIIA.

Since it was found that the (Wm*T) F_1 did not show f.d.a. in either parental stand, its pure stand yield was taken as the mean of its yield in the Warimek and Timgalen backgrounds at 2 fequencies 6.25% and 25% (Table 11). The mean yield per plant was 2.07g. Grain yields of Warimek and Timgalen were estimated from regression equations (Figure 15) which showed that pure stand yields of Warimek and Timgalen were 1.88 and 1.63 g/plant.

When the yield of the F_1 is taken to be 1.00, relative yields per plant of Warimek and Timgalen are 0.91 and 0.79, respectively.

Now, since the F_1 and Warimek did not express f.d.a., their reproductive values in mixture remained at 1.00 and 0.91, respectively, at all frequencies. However, as Timgalen was a strong competitor, its yield increased at low frequency. Its experimental f.d.a. ($\frac{|B|}{A}$) was found to be about 40% and hence the reproductive coefficients of the three genotypes were:

Warimek : $A_1 + B_1(f_{Wm}) = 0.91 + 0.00(f_{Wm})$

(Wm*T)F_1 : $A_2 + B_2(f_{F_1}) = 1.00 + 0.00(f_{F_1})$

Timgalen : $A_3 + B_3(f_T) = 1.30 - 0.51(f_T)$

pure stand of Timgalen = 0.79 and $\frac{|B_3|}{A_3} = 39.2\%$

Four cases will be presented. The population was simulated for 100 generations but as there was little change in genotypic frequencies after 50 generations, the figures only illustrate the first 50 generations.

2.2.1 Case 1: No outcrossing and no f.d.a. of Timgalen. (i.e. $A_3 + B_3 = 0.79 + 0.00$).

The F_1 was eliminated by the 19th generation and the population was completely dominated by Warimek by the 73rd generation (Figure 44). The population fitness decreased from 1.00 to 0.91 which is equal to the pure stand reproductive value of Warimek.

2.2.2 Case 2: 5% outcrossing but Timgalen did not show f.d.a.

The effect of outcrossing was to delay the stage of fixation, so the elimination of the F_1 and domination of Warimek took place in generations 79 and 92, respectively (Figure 45).

2.2.3 Case 3: No outcrossing but Timgalen had an f.d.a. of 39.2%

The F_1 was eliminated by the 17th generation but the population remained in equilibrium from generation 75 onward, when frequencies of Warimek and Timgalen were 0.2353 and 0.7647, and the population fitness at equilibrium was 0.91 (Figure 46). At these frequencies, the fitnesses of Warimek and Timgalen were equal. In this example, it is possible to predict the equilibrium frequency of Timgalen from its reproductive coefficients:

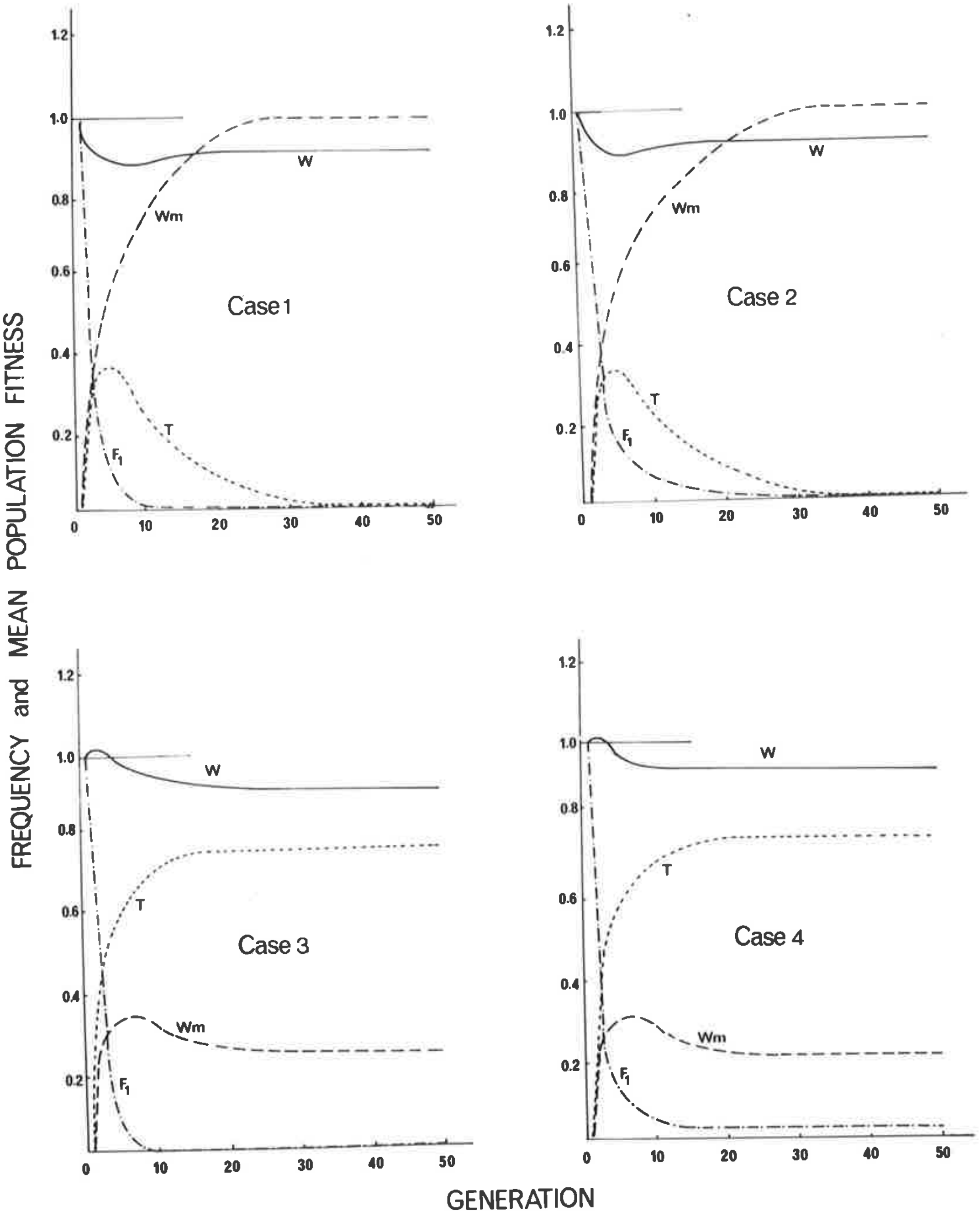
$$f_{(T)} = \frac{1.30 - 0.91}{0.51} = 0.7647$$

Figures 44, 45,46 and 47: Results of computer simulation of Warimek, Timgalen and their F_1 hybrid in Cases 1, 2, 3 and 4.

Changes in genotypic frequencies (Wm, F_1, T) and mean population fitness ($\bar{W}$) with generation.

Input values

Case	A_1	B_1	A_2	B_2	A_3	B_3	t
1	0.91	0.00	1.00	0.00	0.79	0.00	0.00
2	0.91	0.00	1.00	0.00	0.79	0.00	0.05
3	0.91	0.00	1.00	0.00	1.30	-0.51	0.00
4	0.91	0.00	1.00	0.00	1.30	-0.51	0.05


FREQUENCY and MEAN POPULATION FITNESS
GENERATION
Case 1
1.2
1.0
0.8
0.6
0.4
0.2
0
10
20
30
40
50
W
Wm
T
F1
Case 2
1.2
1.0
0.8
0.6
0.4
0.2
0
10
20
30
40
50
W
Wm
T
F1
Case 3
1.2
1.0
0.8
0.6
0.4
0.2
0
10
20
30
40
50
W
T
Wm
F1
Case 4
1.2
1.0
0.8
0.6
0.4
0.2
0
10
20
30
40
50
W
T
Wm
F1

2.2.4 Case 4: 5% outcrossing and f.d.a. of Timgalen

The population reached equilibrium by the 70th generation. Genotypic frequencies at that stage were 0.2061, 0.0395 and 0.7543, and the population mean fitness remained at 0.9175 (Fig 47).

In this example, the F_1 did not show f.d.a. but was maintained in the population due to outcrossing between Warimek and Timgalen. Thus f.d.a. of the lower yielding homozygote was sufficient to maintain that genotype in the population, but outcrossing was necessary as a source of new heterozygotes.

A summary of the results for the above four cases is shown in Table 51.

3. Discussion

Although the foregoing examples represent theoretical circumstances, they clarify the relative importance of the different factors which influence genotypic frequencies and the conditions for stable polymorphisms in a population of predominantly self-pollinating species of infinite size.

When all genotypes had an equal fitness and f.d.a. was absent, the two homozygotes always had an equal frequency (cases 1 and 2), but outcrossing was a necessary and sufficient condition to retain the heterozygote in the population (case 2). When there were differences in selective values but no f.d.a., the homozygote with a lower pure stand yield was eventually eliminated from the mixture (cases 3, 4 and 8, cases 1 and 2 of WmT), thus removing the source of new heterozygotes

Table 51: Summary of the results of computer simulation of Warimek, Timgalen and their F_1 hybrid.

	Input values							Results				
Case	t	A_1	B_1	A_2	B_2	A_3	B_3	n	$f_1^{(n)}$	$f_2^{(n)}$	$f_3^{(n)}$	$W^{(n)}$
1	0.00	0.91	0.00	1.00	0.00	0.79	0.00	19*	0.9122	0.0000	0.0878	0.8995
								73	1.0000	0.0000	0.0000	0.9100
2	0.05	0.91	0.00	1.00	0.00	0.79	0.00	79*	0.9998	0.0000	0.0002	0.9100
								92	1.0000	0.0000	0.0000	0.9100
3	0.00	0.91	0.00	1.00	0.00	1.30	-0.51	17*	0.2618	0.0000	0.7382	0.9200
								75	0.2353	0.0000	0.7647	0.9100
4	0.05	0.91	0.00	1.00	0.00	1.30	-0.51	70	0.2061	0.0395	0.7543	0.9175

t = proportion of outcrossing

A_1 B_1 and f_1 = reproductive coefficients and frequency of Warimek

A_2 B_2 and f_2 = reproductive coefficients and frequency of the hybrid

A_3 B_3 and f_3 = reproductive coefficients and frequency of Timgalen

n = generation at which heterozygote is eliminated (*), and generation of fixation of Warimek (cases 1 and 2) or equilibrium (cases 3 and 4).

$W^{(n)}$ = mean population fitness in generation n.

(cases 4 and 8, case 2 of WmT). However, if the genotype with a lower pure stand yield had a higher yield when it was grown in a mixture, its f.d.a. was sufficient to maintain the two homozygotes in equilibrium (cases 7, 10, 11 and 12, cases 3 and 4 of WmT). The population then remained polymorphic and heterozygotes were produced through intercrossing (cases 7, 10, 11 and 12, case 4 of WmT). If the magnitude of f.d.a. was appreciable, it might even enable the lower yielding genotype to dominate the mixture (cases 7, 11 and 12, cases 3 and 4 of WmT),

Assuming that the frequency of the heterozygote is reduced by inbreeding, heterozygote advantage alone was insufficient to maintain the heterozygote in the population, except the cases where the selective value of the heterozygote was more than twice those of the homozygotes (see phase diagrams, Figs 30 and 31). Furthermore, when the selective values of homozygotes approach that of the heterozygote, only a small difference in selective values of the two homozygotes was sufficient to lead to fixation (Figures 30 and 31). However, when outcrossing occurred and even without heterozygote advantage, f.d.a. was able to maintain the heterozygote in a stable equilibrium with homozygotes (cases 5, 6, 7, 10 and 11) or reduce the rate and delay the approach to fixation (case 9 c.f. case 8). This suggests that agricultural populations would tend to remain genetically variable much longer than is expected on the basis of natural crossing and segregation.

The model used in the computer simulation was based on that proposed by Hayman (1953). Workman and Jain (1966) indicated that such a model was valid if scoring was done prior to mating and soon after selection takes place, so this model was applicable to data based on seed characters (e.g. loci b, s, g, r, e in barley). These authors proposed two other models applicable to the different stage of the life cycle at which genotypic frequencies might be recorded or for differences in the mode of selection as shown here:

Stage:	zygotes $\xrightarrow{1}$	mature adults $\xrightarrow{2}$	zygotes
Generation:	n	n	n + 1

It was considered that zygotic selection could result either from differential viability at stage 1 or from differential fecundity or variation in the mating system at stage 2. Thus, Hayman's model (1953) (model I according to Workman and Jain, 1966) is applicable where both selection and the determination of genotypic frequencies occur at stage 1, and assuming that all of the selection has occurred prior to scoring. Under model II, Workman and Jain (1966) considered the case when selection occurred at either stage 1 or 2, but genotypic frequencies were scored soon after mating, based on census data of newly formed zygotes (e.g. aleurone colour loci bl_1, bl_2 in barley). At equilibrium, genotypic frequencies in this model are derived from the previous one by considering one extra generation of mixed selfing and random mating. Model III is used when selection acts on only one sex and selective differences result from differential fecundity (stage 2) determined by the maternal genotype alone and the scoring is the same as in model II.

The approach to equilibrium differs between the three models. However, at equilibrium the genotypic frequencies just prior to mating would be the same for all three models (Workman and Jain, 1966). Formulae describing population changes for the two alternative models were given by these authors but they will not be presented here. A detailed discussion on these models is outside the scope of this study.

Other factors which might influence genotypic frequencies were not considered here as they are rarely relevant to plant breeding programs. These include migration and mutation. The work reported here is applicable to populations of infinite size. When the populations are much smaller, it is more likely that the genotypes at low frequencies would be lost through sampling error. For populations of predominantly self-pollinating species, a reduction in the population size would tend to promote homozygosity.

The simulation also assumed that the rate of outcrossing was constant and that the only force which altered selective values was f.d.a.. It has been shown that the mean outcrossing rate of a population is affected by environmental and physiological factors (Fryxell, 1957). Changes in selective values can also result from genotype-environment interactions over sites and seasons. Jain and Marshall (1968) used computer simulation to study population changes in which either the rate of outcrossing or selective values of homozygotes were varied among successive generations. They concluded that fluctuation in outcrossing rate resulted in changes in the "effective" (or apparent) heterozygous advantage but left allelic frequencies unchanged. Hence, there was very little change in the bounds on selective values of the homozygotes that would yield non-trivial genotypic equilibria (boundaries between the areas marked A and C and B and C in phase diagrams). On the other hand, stochastic fluctuations in selective values markedly increased the region of stable equilibria for a polymorphism. However, Jain and Marshall (1968) used Monte Carlo procedure to simulate the model of stochastic fluctuations in selective values, so this model was applicable to finite populations of zygotes.

The literature on genetic polymorphisms also includes models where fitnesses are density-dependent which in turn may result in frequency-dependence of selective values (Anderson, 1971; Charlesworth and Giesel, 1972; Clarke, 1972; King and Anderson, 1971). In these models, genotypic fitnesses are determined by the intrinsic growth rates (density-independent), but the population size is regulated by the carrying capacity (density-dependent) of the environment for each genotype. When the population density is low (e.g. in expanding populations), selection would favour genotypes with a high reproductive rate, whereas at high population densities (e.g. stable populations), selection would favour genotypes which have a high viability in crowded conditions. The population size interacts with the carrying capacities of the environment to regulate the fecundities and viabilities of the genotypes. Genotypic fitnesses vary and until the population reaches stability, the contribution of each genotype to the population fitness will vary with its frequency.

Other models on frequency-dependent selection (e.g. sexual selection, mimetic selection) are not discussed here since they are more applicable to outcrossing species.

Thus, there are many mechanisms which determine population changes. The work reported here only points out that f.d.a., among others, contributes to the maintenance of genetic polymorphisms. However, it should be noted that frequency-dependence of reproductive values did not always result in an increase in the mean population fitness, especially when high yielding genotypes are suppressed when in competition with low yielding genotypes. This is illustrated by comparing the population fitnesses in cases 11 and 12 with those in cases 8, 9 and 10. In agricultural populations, this effect of competition is particularly important if the factor contributing to competitive success is not due to yielding ability but due to differences in morphology of the components of the mixture (Khalifa and Qualset, 1974 with wheat; Jennings and Herrera, 1968 with rice). Khalifa and Qualset (1974) compared the yields of mixtures of tall and dwarf wheat varieties grown together for 4 consecutive years without artificial selection. They showed that the relative yield (as a percentage of the bulk population) of the tall and low yielding variety, Ramona 50, increased from 50% to 78%, whereas that of the short and high yielding variety, D6301, decreased from 50% to 22%. The total grain yield of the bulk population also decreased from 6086 kg/ha to 5794 kg/ha in the same period. However, in a subsequent paper, Khalifa and Qualset (1975) reported that the bulk hybrid of the same varieties steadily increased in yield with advancing generations. Also, the increase in mean yield was accompanied by a steady increase in variance for yield among a sample of the lines selected at random. These authors drew the distinction between competition in mixtures of varieties and competition within the bulk hybrids and suggested that the greater number of genotypes in the latter may result in several kinds of competition occurring concurrently. They suggested the types of competition described by Schutz, Brim and Usanis (1968), which included overcompensation in a feedback system (similar to f.d.a.) as likely to be responsible for the increase in yield and variance of the lines in the bulk hybrid.

The retention of genetic polymorphisms for which f.d.a. may be responsible, ensures that a large number of genotypes are available for genetic recombination. This may be an important factor in the evolution of self-pollinated plants and could account for the heterogeneous nature of natural populations (Allard, 1965) or mixed agricultural populations (Harlan and Martini, 1938; Suneson, 1956). With this variability, even a small amount of outcrossing may be sufficient to produce new and highly productive genotypes. Allard and Hansche (1964) indicated that genotypes which survive in mixtures are of agricultural value. Suneson (1956) and Lohani (1976) found that high yielding genotypes were present in advanced generations of composite crosses. It is certain that agricultural populations are much more complex than the examples reported from computer simulation (where only 3 genotypes were considered). For example, Composite Cross II (Suneson and Stevens, 1953) originally consisted of 378 hybrids from the crosses of 28 varieties.

It is desirable that genotypes which are low yielding but otherwise possess useful genes such as disease resistance or high protein content be retained in natural population or bulk hybrids, as these can contribute to the improvement of the population through intercrossing with high yielding genotypes. Allard and Hansche (1965), Allard and Workman (1963), Imam and Allard (1965), Jain and Allard (1960) used census data of marker genes to show that many inferior alleles were present in natural populations of lima beans, barleys and oats. It was postulated that the persistence of heterogeneity was due to selection favouring the heterozygotes but it is probable that f.d.a. occurred to retain these alleles and allow for the possibility of genetic recombination. These previously inferior genotypes could become a source of variability and contribute to the formation of new and superior ones. The isolation of high yielding genotypes in advanced generations of composite crosses (Lohani, 1976; Suneson, 1956) indicated that superior genetic recombinations did occur in agricultural populations of self-pollinating species and f.d.a. is one of the factors which made it possible through the retention of gentic variability.

VI. GENERAL DISCUSSION AND IMPLICATIONS IN PLANT BREEDING.

A. Introduction

The main objectives of this thesis were:

(i) to establish the occurrence and to measure the magnitude of f.d.a. in wheat.

(ii) to investigate the bases of f.d.a. and the physical factors which confer the advantage.

(iii) to study the effect of f.d.a. on single plant selection.

Also, the role of f.d.a. in population dynamics was demonstrated by computer simulation, using the model of mixed selfing and random crossing developed by Hayman (1953).

The most important finding of this study was that a genotype growing at a low frequency in a mixture had a higher yield (and number of grains) per plant than when it was grown at a higher frequency in a mixture of the same components. These reductions in yield ranged from 20% to 40% and were obtained for F_1, F_4, F_5 and homozygous parental varieties. The consistent occurrence of f.d.a. in different years, when more than 150 genotypes were tested in different experimental designs ("fixed" positions in the hand planted experiments and "random" positions in the machine sown experiments) indicated that f.d.a. is likely to be widespread in wheat and to warrant attention during wheat breeding.

Under South Australian conditions, it was likely that f.d.a. was a result of interactions between plants in the soil and more specifically, between roots of the same genotype. In experiments I and IIIA, where the positions of the test plants could be accurately identified, it was found that there was a positive and significant correlation between grain yield and distance between the test plants. As the frequency of the test plants decreased, the distance between these plants increased and hence the interference between their roots is likely to have decreased. This suggests that f.d.a. was due to competition between plants of the same genotype for environmental resources in a slightly different way than that from other genotypes.

The physical basis of f.d.a. further suggested that genotypes which express f.d.a. are co-exploitative rather than competitive. This is supported by the observation that the background plants were unaffected by the enhanced growth of the test plants at low frequencies (Halberd plants in experiments IA and IB, and Warimek plants in experiment IV). Furthermore, in mixtures of strong and weak competitors (experiment IIIA), the stronger competitor, Timgalen, had an increased yield at low frequencies, but the weaker competitor, Warimek, had similar yields at all frequencies.

F.d.a. has been shown to be similar to the over-compensatory interaction reported by Schutz, Brim and Usanis (1968) for mixtures of soy bean and by Allard and Adams (1969b) for mixtures of barley genotypes. These authors considered that the over-compensatory effect might have been responsible for the maintenance of genetic polymorphisms in agricultural populations. In the study reported here, with the use of a computer, it was demonstrated that a combination of f.d.a. and natural crossing could maintain genotypic frequencies at a non-trivial equilibrium.

The effect of f.d.a. on single plant selection is discussed in the next section (B4).

B. Implications of f.d.a. in plant breeding.

The foregoing evidence on the existence and magnitude of f.d.a., and the role of f.d.a. in maintaining genetic polymorphisms, suggest that f.d.a. influences at least four aspects of plant breeding.

1. The use of Composite Crosses in breeding for higher grain yield.

The aim of the Composite Crosses (CC) is to provide a "mass reservoir" of genetic variability, which may be exploited both as a source of deriving superior recombinants and for the retention of genetic variability. Basically, the technique consists of crossing a large number of varieties in a number of combinations, mixing the hybrid seeds and propagating the composite bulk under "natural" selection - apparently indefinitely.

Preliminary considerations would suggest that the effectiveness of this method may be low as segregation will lead to homozygosity and inter-genotypic competition would then reduce such populations to homogeneity rapidly. This would restrict the likelihood of recombination. Although it has been observed that certain inferior marker genes were quickly eliminated from the CC's (Suneson and Stevens, 1953), there is evidence (Allard and Jain, 1962; review in Allard and Hansche, 1964) to show that "a vast number of different genotypes remained in the population after 18 generations of exposure to natural selection". Allard and Jain (1962) grew randomly chosen plants from the F_3, F_6, F_{13} and F_{18} generations of CCV of barley and found that the between- and within-family variances for heading time remained high in the F_{19} generation. These authors discussed the evolutionary processes which might have contributed to the observed results and suggested that "the most likely candidate was selection favouring heterozygotes".

Subsequent studies by Allard and his associates have shown that the hypothesis of heterozygote advantage was insufficient to explain the retention of heterozygosity in advanced generations. Allard and Workman (1963) used census data based on seed coat pattern of populations of lima bean to show that while there was a tendency for the selective value of the heterozygotes to increase in latter generations, their frequency at equilibrium was higher than could be expected on the basis of heterozygote advantage and outcrossing.

Frequency-dependent advantage provides an explanation for the performance of single plants which results in the persistence of heterogeneity. Harding _et al_. (1966) showed that there was a negative correlation between the fitness of the heterozygotes and their frequency in populations of lima bean. Furthermore, the fitness of the heterozygotes was largest when their frequencies were lower than that observed at equilibrium (0.07), so that at very low frequencies, they produced three times as many progeny as the homozygotes. Harding _et al_. (1966) suggested that frequency-dependent selection was "a potent force in the retention of segregational variability in this heavily inbreeding species".

The results in the experiments reported here indicated that frequency-dependent advantage was not due only to heterozygosity. The magnitude of f.d.a. was almost the same when the test genotypes were from the F_1, F_4, F_5 or were parental varieties. With the use of a computer, it was shown that f.d.a., when a genotype at low frequency produces far more than its proportionate share of offspring, enabled genotypes with a low reproductive value in pure stand to remain in the population.

Thus, a combination of f.d.a., natural crossing and heterozygous advantage is the likely mechanism maintaining heterogeneity in Composite Crosses of predominantly inbreeding species. Many of the genotypes which survive in the population through f.d.a. may not be high yielding when grown in pure stands; nevertheless, they are a source of genetic variability for hybridization and recombination. The increase in yield of the CC's in advanced generations and the isolation of high yielding genotypes from these crosses (Suneson, 1956; Lohani, 1976) indicated that useful recombination does occur. F.d.a. is important to the success of the Composite Crosses because it enables the maintenance of variability essential for continued recombination.

2. Management of multiline varieties.

Often, a multiline variety is conceived of a mixture of many genotypes, each of which occurs at a low frequency and possesses a different gene for disease resistance. The success of this method of breeding for resistance to pathogens depends on the mixture remaining genetically variable. The frequent occurrence of f.d.a. in wheat suggests that multiline varieties of this species will not need frequent re-constitution. By testing the frequency responses of a number of varieties, the breeder should be able to identify combinations of favourably interacting genotypes and predict the likely changes in the composition of potential multilines. With judicious choice, such multiline varieties would not only be more stable but could also exploit more efficiently the environmental resources than homogeneous varieties.

3. Seed certification.

There are circumstances where an f.d.a. is undesirable. In most breeding programs, selection is usually completed by the F_4 to F_6, when, on average, the lines are only about 88% to 97% homozygous. This, together with natural crossing, suggest that most varieties are likely to be a stable mixture of closely related genotypes.

It is difficult to avoid contamination from other varieties of new varieties during several generations of seed multiplication. The presence of an f.d.a. implies that the low proportion of rogues (off-type genotypes) and morphologically distinct segregants in the original seed is likely to increase during seed multiplication. In the past, it has been common for officers in charge of seed production to counter this contamination by growing hill plots from single heads. Only those typical of the variety are retained for further multiplication. This process, particularly if the number of single heads is small, and if the process is repeated frequently, could result in unconscious and perhaps deleterious selection within the variety.

In certain seed certification schemes, the requirement for uniformity specifies that a variety should not only be free from foreign materials such as weeds, but also be homozygous and homogeneous. Where there is a rigid demand for seed purity, an f.d.a. effect could considerably delay the release of a variety or render a variety unacceptable soon after it has been registered.

4. Single plant selection.

The choice of genotypes in a segregating population may be influenced by their frequencies. If a genotype performs well in a mixture because it is rare, this advantage will not persist when it is selected and propagated in a pure stand. Selection would then have been ineffective and many high yielding genotypes might have been overlooked.

However, it has been shown that the phenotypic expression of single plants depends largely on the micro-environment in which they are grown. Neither of the F_2 populations studied in experiment VII showed more variation in their yield and yield components than the parents, except in height. Also, the highest yielding plants in the F_2's were not significantly higher yielding than those in the F_1's and the parents. Since the variances for grain yield were significantly different between the parents, it was evident that the environmental effect must have been large in all populations. Thus, it was concluded that the effect of f.d.a. and genetic variability in these F_2 populations might have been masked by random variation in the micro-environment.

Experiment VIII describes an attempt to reduce the effect of micro-environmental variation between single plants by growing replicates of mixtures of F_2 derived lines. The same lines were also grown as pure stands. It was found that the correlation between yield in mixture and yield in pure stand varied from one replicate to another, but when yields in mixtures were based on the yields in all the replicates, the correlation was positive and highly significant (r = 0.749***). This implies genetic variation being expressed in the F_2 derived lines. The positive correlation agrees with that found by Johnston (1972) for barley and supports the contention of Allard and Hansche (1964) that high yielding genotypes in mixtures also tend to be high yielding in pure stands. Conversely, in the same experiment, although these lines expressed f.d.a. when grown at low frequencies in a background of the parents, the correlation between the f.d.a. of a line and its performance in a pure stand was low.

It was concluded that f.d.a. operates in mixtures of 2 or a few components, but its differential effect is small in heterogeneous populations. Results in the last two experiments indicated that f.d.a. has only a minor role in reducing the efficiency of single plant selection. Micro-environmental variation has a large influence on the phenotypic expression of single plants in a segregating population. The positive correlation between yield in mixture and yield in pure stand suggested that when the effect of micro-environmental variation could be reduced, it should be possible to select for high yielding genotypes, based on the performance of single plants in mixtures.

There are circumstances where f.d.a. might reduce the efficiency of single plant selection. Different genotypes in a mixture may react differently to specific races of a plant pathogen. Assuming that the spread of the disease is inversely related to the distance between the susceptible plants, then it may be expected that the chance of a susceptible plant being infected is less when it is at low than at high frequencies. Thus, it is possible that some high yielding plants in mixtures may be susceptible to diseases when they are grown in pure stands.

In experiment IIA, it was shown that when Halberd was grown as the test plants in a rust resistant background, Wariquam, it was relatively rust free and yielded 2.58 and 2.02 g/plant at the 6.25% and 18.75% frequencies, respectively (Figure 7). However, when Halberd was grown as the background stand (experiment IIB), the plants were heavily infected with rust and had very low yield. If differential susceptibility to diseases is the mechanism of f.d.a., then it may reduce the efficiency of single plant selection since high yielding but disease susceptible genotypes may be favoured in selection at the expense of low yielding but resistant genotypes.

C. Comparison of selection methods in breeding for yield.

As the purpose of this study is to investigate the efficiency of single plant selection for yield, it is appropriate to compare several methods of breeding pertaining to this aim. The three breeding methods used in cereals are the pedigree, the bulk population (and Composite Cross), and, more recently, the F_2 progeny method.

1. The pedigree method of breeding.

In the pedigree method of selection, desirable genotypes are selected in F_2 and successive generations on their phenotype, and a pedigree is recorded for each of the lines from a cross. The pedigree enables the breeder to decide the likely degree of similarity of two lines (e.g. relate back to a single plant in an earlier generation). He can then use this to help decide which of the families are to be retained and the ones to be discarded.

As selection begins at the generation where the frequency of the plants containing all the desirable alleles is at a maximum (Shebeski, 1967), this method gives the plant breeder the highest probability of identifying superior genotypes and has been used with success in many crops (Allard, 1960). However, there is an extensive literature to show that selection for yield in the early generations is often no more effective than random selection (e.g. Bell, 1963; De Pauw and Shebeski, 1973; Knott, 1972; McGinnis and Shebeski, 1968; Shebeski, 1967).

Shebeski (1967) surveyed techniques of breeders from many countries. He found that the number of F_2 plants grown by most breeders and the number of plants retained in each generation were too small for the chance occurrence of plants containing all the desirable alleles. Between 90% to 95% of the plants in the segregating populations were discarded each year, purely on visual inspection and morphological traits. These included disease resistance, straw strength and plant height. Yield trials were carried out only after selection for other less important but easily recognizable characters had reduced numbers. Yield, with a low heritability, was a selection criterion for only a very small proportion of the original population, so it is possible that a large number of high yielding lines may have been discarded on inadequate evidence.

Shebeski (1967) also suggested that most breeders grew the segregating populations at too few locations and in too few replications. The result is that it is difficult to separate the variances attributable to genotypes and environments and consequently, it is impossible to assess accurately the yielding ability of the selected lines.

Another limitation to effective breeding for high yield in cereals is the inability to select single plants for general adaptation based on their performance in one environment. Because of the limited amount of seed available from single plants, the procedure is to evaluate the yield potential under one environment, retaining the best and then testing them at a number of environments. Genotype-environment interactions over sites and seasons may render genotypes selected in one environment ineffective for other sites and in other seasons.

The pedigree method also allows little intercrossing between different genotypes since the segregating lines are grown in separate rows or separate plots. Because there is little genetic recombination in advanced generations, the success of this method depends on the ability of the breeder to identify high yielding genotypes in the early generations, preferably at the F_2. The breeder relies primarily on the correlation between the appearance of isolated plants and the yield of the same genotype as a crop. Townley-Smith et al. (1973) reported that nine selectors, including breeders, technicians and others, selecting at a 25% level, were not able to choose the highest yielding lines of wheat. The average of the choices of most selectors was only slightly better than the population mean. Shebeski (1967) reported the results of a selection experiment at the University of Manitoba. Four plant breeders selected a total of 440 plants which they considered to be high yielding from 11 F_2 populations each of 10,000. The progeny of each selected plant was grown alongside an unselected control from the same population. It was found that approximately 50 percent of the lines yielded more and 50 percent of the lines yielded less than their corresponding controls. It was concluded that selection for yield was ineffective in the F_2 generation.

The factors which influence the phenotypic expression of single plants have been discussed in the literature review. Several of these factors are worth mentioning as they are relevant to the present discussion. Firstly, as selection is practised when most of the plants are still heterozygous, heterozygosity and hybrid vigour may result in a false appraisal of the genetic worth of the selected lines as homozygous varieties. Secondly, F_2 plants are often grown at low densities (e.g. 45cm between plants in McGinnis and Shebeski's experiments, 1968) whereas the selected lines are grown at commercial densities which are 15 to 20 times higher than the F_2 densities. Genotype-density interactions may render single plant selection for yield ineffective. This limitation could be overcome by growing and selecting the segregating populations at a commercial density. Thirdly, replication is not possible with single plants in the F_2, so the effect of micro-environmental variation is large. This could be reduced by calculating response surfaces or moving averages as discussed previously to give an estimate of yield based on the local mean.

However, one major restriction of these methods is that they require the identification and measurement of a large number of plants, most of which are subsequently discarded. Although much of the calculation could be handled with the use of a computer, the effort to collect the data is considerable.

Other factors may limit the effectiveness of these methods. In the former, the number of terms required in the polynomial equation to represent environmental variation is arbitrary as there is no biological basis to determine the optimum number of terms. In the latter, the number of plants needed to represent the moving average must be small enough to account for irregular variation, but must be large enough to ensure that the average genetic effect of the plants approximates the population mean. Again, there is no objective criterion on which to decide how many plants should be used.

2. The bulk population and Composite Cross methods of breeding.

The bulk population method attempts to overcome some of the inadequacies of the pedigree method. In this the segregating populations are grown for a number of generations without any deliberate selection. As there is no need to keep pedigree records, a large number of crosses can be handled and there is a somewhat greater opportunity for continuing hybridization between different genotypes. The method depends primarily on "natural" selection to select out the higher yielding genotypes. Single plant selection is delayed until most of the lines are homozygous, possibly by the F_{10}, and then yield testing is undertaken in the same way as the pedigree method.

As the proportion of genotypes with desirable alleles decreases rapidly with each generation of inbreeding (Shebeski, 1967), a very large sample would need to be retained for the chance occurrence of desirable genotypes. Since only a sample of the population is retained, a large number of seeds is discarded each year at random, and it is possible that many high yielding genotypes could have been lost through sampling. Hence the chance of identifying high yielding genotypes in the bulk population method is probably much lower than in the pedigree method.

The success of the bulk method depends on the correlation between survival in mixture and yield. Studies by Harlan and Martini (1938) indicated that genotypes which dominated the mixture were often also those with high agricultural values. However, Suneson and Weibe (1942) found that the worst competitor in a mixture of four barley genotypes was the highest yielder. The bulk method is unsuited for populations originated from diverse parents. Where there are marked differences in morphological characters, e.g. tall versus dwarf, it is likely that the population will be dominated by genotypes with high competitive abilities rather than those with high yielding abilities (Jennings et al., 1968 a,b,c). Palmer (1952) found that many lines selected from the F_8 of a bulk population had small grains, so natural selection had favoured genotypes with rapid multiplication at the expense of those with heavy seeds.

The Composite Cross (CC) method is similar to the bulk population as "natural" selection is the main factor determining population changes. The success of both methods depends on the correlation between fitness and yield. The CC differs from the bulk population in that:

(i) there is often more genetic variability due to the complexity of the original cross. For example, CC II consisted of 378 hybrids from 28 varieties (Suneson and Stevens, 1953).

(ii) the population is maintained for a much longer period. Artificial selection may not be practised until the F_{20} or F_{25} whereas for a bulk population, selection can begin by the F_6 or F_7 and rarely after the F_{10}.

(iii) there is more intercrossing both due to the extra time and occasionally to the inclusion of a male sterile line.

The increase in yield of the CC's with time (Suneson, 1956; Lohani, 1970) has been attributed to the elimination of poorly adapted genotypes while allowing for continuing intercrossing between many genotypes in advanced generations. From the discussion on the implications of f.d.a. previously, it was apparent that the effect of f.d.a. is particularly important in CC's as it enables a large number of genotypes to be retained and contribute to the formation of new heterozygotes.

Both the bulk population and the CC methods are slow, especially the latter. Besides having to wait for 10 to 20 generations, many breeders are still uncertain about the correlation between success under competition and yield in pure stand. The CC's may best be used as "mass reservoirs" which allow large populations to be carried inexpensively and genetic variability remain for artificial selection (Allard and Hansche, 1964). They are often considered as a supplement to, rather than a substitute for conventional breeding methods.

3. The F_2 progeny method.

The bulk and the Composite Cross methods have not been used extensively in Australia. Much of the breeding work has been based on the pedigree method and, until recently, the backcross method. Only recently has the bulk method been investigated in New South Wales (Derera and Bhatt, 1972; Bhatt and Derera, 1973b).

In Australia, the pedigree method of breeding was successful during the early stage of wheat breeding (prior to the 1940's). During this time, improvements resulted from the selection on morphological traits closely related to yield. These included appropriate time of anthesis, shorter straw, reduced tillering and disease resistance. The breeders relied primarily on the correlation between the appearance of isolated plants and the yield of the genotype as a crop. As these characteristics were achieved, further selection for these same characteristics and the exhaustion of genetic variability resulted in these programs being less successful than those using the back-cross method.

Yield improvement due to breeding between 1940 and the mid 1960's was slow (Russell, 1973). Most of the achievement of wheat breeding during this period was in preventing serious losses of yield from diseases. The back-cross method was employed to improve pre-existing varieties by the incorporation of highly specific characteristics controlled by simple genetic systems, particularly those for resistance to specific races of a disease. With the introduction of high yielding, disease resistant Mexican lines in the early and mid 1960's, the back-cross method has been used very successfully by using these varieties as donor parents, and it seems likely that this type of program will continue to have a role, especially if even higher yielding lines with some agronomic deficiency become available.

During the last decade, the emphasis in breeding for yield has gradually been shifted to the F_2 progeny or bulk progeny method, and the Waite Agricultural Research Institute has been most committed to this program. This type of selection has promised to become of major importance in Australian breeding, for in the last Interstate Wheat Variety Trial, 9 of the top 10 unregistered lines originated from the three programs employing the new method extensively (Rathjen, per. comm.).

The F_2 progeny method was used to some extent at the Plant Breeding Institute, Cambridge, England, in the mid 1950's (Lupton and Whitehouse, 1957). This was a combination of selection for yield in early generations with selection for other desirable characters. Basically, the technique consisted of single plant selection in the F_2 generation, primarily on easily recognizable characters such as plant height, earliness and resistance to diseases. The F_3 generation was mainly for seed multiplication although there was some further selection on morphological characters. The F_4, F_5 and F_6 were grown in yield trails and tested for quality; the less desirable lines being eliminated after each year of trial. By the F_7 there would have been sufficient information on yielding ability, quality and agronomic characteristics to distinguish between lines. Finally, single plant selection was made from the most promising lines, and then multiplied for further testing.

The F_2 progeny method has a merit in that selection is based on yield data, before potentially valuable material has been discarded on inadequate evidence. However, the F_2 progeny method was later discontinued at Cambridge as it was found that the accuracy of the yield data obtained was often low as a result of the very large numbers of selections involved in each trial (Lupton and Whitehouse, 1961).

The F_2 progeny selection method used at the Waite Agricultural Research Institute is a modification of that described by Lupton and Whitehouse (1957) due to the inability to select effectively on single plants. F_2 populations are grown at a commercial density and little or no deliberate selection is practised in this generation. Only plants with obvious morphological defects such as lodging or susceptible to diseases are eliminated. Selection is actually more of a partitioning of the hybrids into closely related groups. Seeds from single spikes are then grown in rows in the F_3 for seed multiplication. Again, little selection is practised in this generation. The F_4's are grown in replicated yield trials. The mean yield of a number of plants from each line is used as an indication of the yielding ability of the F_2 line and provides a criterion for screening between lines. Subsequent generations are handled in the same way as that described by Lupton and Whitehouse (1957).

Attempts have also been made to reduce the period between successive outcrossing (Rathjen, 1973). This was done by growing the seed multiplication generations, the F_1, F_3 and F_6 during the summer and by testing the F_4, F_5, F_7, F_8 at as many locations as possible. Also, by using the F_5 or F_8 as parents, the generation interval could be shortened to three or five years.

In the study reported here, the results from experiment VIII indicated that:

(i) the effect of f.d.a. was small in a heterogeneous population and was unlikely to influence single plant selection.

(ii) micro-environmental variation was the main factor influencing the phenotypic expression of single plants.

(iii) there was a positive correlation between yield in mixture and yield in pure stand. This correlation was obtained when:

- the mixtures and pure stands were grown at the same density and planting method. Thus when there were no genotype-density and genotype-environment interactions between the two treatments.

- the yield in mixture of each genotype was based on the mean of a number of plants in replicated plots, and the effect of micro-environment was thereby minimized.

Thus, when the effect of micro-environment could be reduced, it should be possible to select for high yielding genotypes, based on the yield of single plants in mixtures. This would tend to support the F_2 progeny method in the particular form used at the Waite Agricultural Research Institute in breeding for higher grain yield.

The most important feature of this method is that selection is based on the performance of the line in a plot. Thus the correlation between the measurement on which selection is based and the performance as a crop is maximized. One of the essential requirements of the method is that the selection trials be grown in conditions closely resembling those used commercially, e.g. planting method and density. Mechanization and computerization of the breeding procedures are essential for the success of this method. They enable a large number of lines and "wider crosses" (e.g. those between European and Australian varieties) to be tested to make selection effective.

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