THE EFFECTS OF SELECTION AND INBREEDING ON EARLY YIELD

AND FLOWER QUALITY IN AMPHIDIPLOID DENDROBIUM

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ABSTRACT

Ten progenies involving self-, sib-, and backcross matings
and an outcross were compared to determine the effects of inbreeding
and selection in amphidiploid Dendrobium Jaquelyn Thomas. The progenies
included two sets of reciprocal matings. The measurements of yield,
size, and earliness of flowering indicated a decline due to inbreeding.
Selection increased flower size and improved the purity of flower
color. Detrimental effects of inbreeding on these characters were not
detected. Significant differences were obtained for keeping quality,
number of flowers per spray, and length of the scape of the spray.
However, it was not possible to attribute these differences to either
inbreeding or selection effects. Spray length differences were
nonsignificant. Offspring of reciprocal crosses did not differ
significantly in all characters measured.

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INTRODUCTION

Dendrobium Jaquelyn Thomas 'UH44', an important cut flower cultivar, is the seedling population produced by selfing the amphidiploid D. Jaquelyn Thomas 'Y166-1'. UH44 plants are vigorous and high-yielding, and the sprays have a long vase life and are relatively free of bud drop. Their white flowers are tinged with pink.

Although relatively uniform, the seed propagated UH44 exhibits slight variations among the offspring. By selecting the least tinged or whitest types with large flowers and by selfing or sibbing, it has been possible to improve the color and size of flowers in succeeding generations. Yield and other qualities, however, may be seriously affected by inbreeding.

In order to ascertain the effects of selection and inbreeding in amphidiploid D. Jaquelyn Thomas, ten progenies representing the first, second and third selfed generations, sib-matings, backcrosses and an outcross were produced. This study involves the evaluation of these progenies to determine whether they are affected by selection and inbreeding.

REVIEW OF LITERATURE

Inbreeding effects

Many wild species as well as cultivated varieties are naturally
self-pollinated and suffer no ill effects in terms of vigor,
productiveness, and ability to survive. However, the majority of
higher plants are endowed with devices which promote cross-pollination.
These naturally cross-pollinated plants, when artifically inbred,
display injurious effects. Most plants benefit favorably from cross-
fertilization (East and Jones, 1919).

Stebbins (1957) explained the widespread occurrence of sexual
reproduction and obligate cross-fertilization in natural populations
as the evolutionary outcome of only lines possessing mechanisms
enforcing sexuality and cross-fertilization surviving. He proposed two
explanations of self-fertilizing species. 1) Through mutation, these
species have lost their ability to cross-fertilize, and only a few
genotypes of the original heterozygous population have survived the
effects of inbreeding. These "dead-ends in evolution" will become
extinct when environmental conditions change radically. 2) Self-
fertilizing species have evolved through natural selection through some
advantages over cross-fertilizing species in surviving under certain
conditions.

In evolution heterozygosis probably played a role in the
development of mechanisms favoring cross-pollination. Variations may
have appeared that favored cross-pollination and thus such progeny
exhibited greater vigor than those of self-fertilized relatives.

Mechanisms ensuring cross-fertilization became homozygous and fixed, thereby aiding survival by increasing vigor. However, weaknesses in strains were perpetuated in cross-pollinated plants by the heterozygous condition. In self-fertilized species, the appearance of a character that weakened the individual quickly resulted in its elimination. Therefore, self-fertilized strains that have survived competition are stronger than cross-fertilized strains from which weak genotypes may be extracted (East and Hayes, 1912).

East and Jones (1919) and Jones (1925) defined inbreeding in terms of limited parentage. The manner in which individuals are mated is the basis of the idea of inbreeding. Pearl is cited by East and Jones as expressing the concept as "...a narrowing of the network of descent as a result of mating together at some point in the network of individuals genetically related to one another in some degree." Wallace (1968) defined it as "the bringing together at fertilization of two alleles that are identical by descent from some specified earlier generation."

Darwin (1900) experimented with inbreeding and crossbreeding. Ipomoea purpurea and Mimulus luteus, the two species which were inbred the longest, showed sensitivity to inbreeding. Yet in each species plants did appear that were more vigorous than the other inbred plants from the same stock and equalled or surpassed the vigor of the original cross-pollinated stock. Segregation of the inbred stock occurred and resulted in different types with different visible hereditary characters and differing in the ability to grow. The inbred plants were also observed to be more uniform in visible characters than the original cross-pollinated stock. Darwin concluded that cross-fertilization

generally had beneficial effects while self-fertilization was frequently injurious.

Shull (1908) observed rows of self-fertilized maize to differ from one another in definite characters. He concluded that these differences are not an effect of inbreeding in itself but a result of inbreeding due to an isolation of biotypes from complex hybrid combinations. In comparing cross-fertilized and self-fertilized strains of the same origin, vigor of the biotypes and their hybrids rather than the effects of the processes of inbreeding and crossbreeding are being noted. The observations of greater vigor of the cross-fertilized strains prompted Shull's suggestion that continuous hybridization rather than the isolation of pure types be the direction of the corn breeder.

Shull (1910) later modified this hypothesis to encompass the concept that although vigor in hybrids can generally be attributed to heterozygosity, in some elements the heterozygous state can be without vigor or even depressing.

East (1908) worked with two types of maize, a smooth, full kernel type and a type with a thin, peaked kernel. Crosses of plants of the same type resulted in the accentuation of type characters. Crosses between types were more vigorous and yielded more than crosses within types.

East (1908) questioned the theory of accumulation of deleterious characters being responsible for the bad effects of inbreeding. In maize the injurious effects of inbreeding were no less common when superior instead of inferior parents were involved. Also, different selfed strains from the same original stock displayed extremes of

characters, such as wide or narrow leaves and tall or short stems, both of which extremes could not be attributed to merely the self-fertilization process. Therefore, deterioration must be an indirect consequence of inbreeding.

East (1909) further argued that although there were many examples of deterioration resulting from inbreeding, there were also cases of superior inbred stock. Hence, the deterioration was made possible by the process of inbreeding but was not a direct consequence of it. Since not all species naturally cross-fertilize, inbreeding and a decrease in vigor cannot be conclusively linked as cause and effect.

Naturally crossbred species, when inbred, tend to isolate into types which are homozygous and so lack the stimulus derived from free intercrossing and appear to deteriorate. East (1909) noted that this deterioration is in no way a degeneration of hereditary characters in corn but is solely manifested in plant size and yield. Thus, this type of degeneration is a partial loss of development and decrease in cell division.

Two effects of crossbreeding are: a recombination of hereditary factors and a stimulation to development. East (1909) postulated that when two differing gametic constitutions are combined, there is an increase in stimulation of growth. Such a hypothesis accomodates the observations of decrease in vigor without the degeneration of characters. This theory also explains why this decline in vigor reaches a limit with the attainment of a completely homozygous individual.

Shull (1911) ran extensive studies comparing self-fertilized and cross-fertilized Indian corn. His major observations were: 1) progeny

of self-fertilized parents were inferior to those of cross-fertilized parents in respect to height, yield and other characters with a basis in physiological vigor, and 2) each self-fertilized family was distinguishable from other such families by particular, distinct morphological characters. Within each self-fertilized family a uniformity of these morphological characters among the individuals was apparent.

Shull (1911) also presented what he considered proof that the self-fertilized families of the same original stock were genotypically distinct and not fluctuations of the same genotype. In a population in which the mean number of ear rows was slightly above, 14 rows, selection was practiced for 12 and 14 rows. The mean number of rows in the 12-row shifted to a lower number than that selected (further generations approached 8 rows) while the 14-row family remained with a mean of 14. Since all plants were grown under nearly uniform conditions, Shull concluded that internal rather than external factors were involved. As inbreeding continued, the self-fertilized lines decreased in variability of row number.

The idea that inbreeding in itself is injurious was rejected by Shull (1911). He conceded that if such injury were real it was insignificant relative to the great vigor shown by the heterozygous condition. Further supporting evidence was that continued self-fertilization in any line did not produce the corresponding decrease in size and vigor in every generation. The decrease in the second year of self-fertilization was not as great as that observed in the first year, in the third year still less was noticed and a limit was approached as

self-fertilization continued. This supported Shull's hypothesis that when complete homozygosity is achieved no further deterioration ensues and so self-fertilization itself cannot be injurious.

In accordance with the view that the degree of vigor is due to the degree of hybridity, certain inferences were made (Shull, 1911). 1) A cross between two plants of the same self-fertilized family, or the same genotype, will show no increase in vigor over the self-fertilized plants since no new hereditary factors are introduced. 2) A cross of two individuals of different self-fertilized lines, or pure genotypes, will produce first generation hybrids exhibiting the highest degree of vigor since they are heterozygous for the characters which differentiated the parental genotypes. 3) Sib crosses among the first generation hybrids will result in progenies with the same characters, vigor, and degree of heterogeneity as progenies resulting from selfing first generation hybrids.

In Shull's (1910) experiments, yielded of F_1 hybrids of certain self-fertilized lines of maize exceeded that of the original cross-pollinated stock. The "injurious effects" of five years of inbreeding were lost through cross-fertilization. Shull attributed the high yield to the particular genetic combination of the hybrid. Thus, reciprocal crosses of two inbred strains produced equal hybrids. The same yield and crop quality of a hybrid of two inbred strains could be repeatedly obtained by remaking the cross. When F_2 hybrids were produced, they exhibited greater variability than the F_1 and this increased variability translated into a decrease in yield.

Jones (1924) as well as Shull (1911) recognized that when selection favors the most vigorous individuals of an inbred generation as progenitors of the subsequent generation, the approach to complete homozygosity is slowed. Jones emphasized that when single individuals are the progenitors of successive inbred generations, the results are dependent upon the genotypes of these individuals.

Jones (1918) found that inbreeding maize reduced the number of nodes per plant, but this decline was much less than that for height and length of ear. He observed that the number of rows per ear increased in some lines and decreased in others. He concluded that inbreeding greatly affects some characters and not others and that segregation had occurred in his plants. The extent to which variability was reduced differed among the lines.

Despite the decline in the size, general vegetative vigor and productiveness as well as greater difficulty in growing them, Jones (1918) found these inbred plants to be normal and healthy. The abnormalities commonly found in a field of maize, such as seeds found in tassels, anthers found in ears, dwarfness, sterility, mosaic and albino plants were never observed in the inbred strains. However, he was impressed by the uniformity in the size, shape, structure and position of the leaves, tassels, stalks and ears.

East and Hayes (1912) reported that normal strains with particular hereditary characters that classify them as degenerate did appear sometimes, but infrequently. They proposed that abnormalities may arise from strains lacking vigor where cell division does not occur normally.

No particular character is common to all inbred strains. The general manifestations are a loss in vigor, size and productiveness with the appearance of unfavorable characters. Such characters were never found in the same strain (East and Hayes, 1912).

East and Hayes (1912) described the developmentally weak types produced by inbreeding as those which cannot be perpetuated, are difficult to propagate and cannot complete normal development or are normal, but differ in amount of growth at maturity. After the reduction in vigor has essentially ceased, those normal, homozygous, inbred strains are comparable to self-fertilized species.

In 1939 Jones summarized 30 generations of self-fertilization in three lines of maize. Reduction in height stabilized after five generations while yield decline ceased after twenty years. Sib lines which had been separated at different points differed in some instances and not in others. Jones attributed these differences to "spontaneous transmissible variations" and not to delayed segregation. Uniformity and constancy for all visible characters were attained after twenty generations of self-fertilization as well as homozygosity for loci contributing to hybrid vigor. No variations appeared that could be construed as favorable to survival.

Inbreeding studies on alfalfa (Tysdal et al., 1942) showed a general decline in yield as the lines became more inbred. In the S_1 the average of 54 lines showed forage yield to be 68% of that of the original open-pollinated varieties while seed yield decreased to 62%. In the seventh generation of self-fertilization the forage yield was reduced to 26% where it essentially leveled off. Seed yield in the

eighth generation of inbreeding was 8% of the original open-pollinated varieties. There was great variability among the selfed lines in both seed and forage yield. In the S_1 lines forage yield ranged from 26 to 105% of the yield of the original varieties.

The inbreeding process is of value in plant improvement to eliminate abnormal, pathological, and generally unfavorable characters since when such characters appear selection can be practiced. Loss of vigor, size and productiveness results from inbreeding. However, uniform, vigorous, productive offspring are obtained when two inbred strains free of unfavorable recessives are crossed (Jones, 1918).

Heterosis

Animal breeders were first to link the effects of inbreeding with hybrid vigor, regarding hybridity to be the antidote to inbreeding effects (East and Jones, 1919).

Hybrid vigor, no doubt, was observed prior to being recorded in scientific literature. According to Zirkle (1952), Koelreuter published his work on plant hybridization from 1761 to 1766 in which hybrid vigor was first described. He observed floral mechanisms favoring cross-pollination and regarded them to be nature's design for ensuring crossbreeding.

Other botanists followed to record the effects of crossbreeding as well as to describe the mechanisms for assuring it (Zirkle, 1952). Among them were Sprengel, who in 1793 accurately detailed the structure of flowers and showed the general avoidance of self-pollination, and Knight who in 1799 attributed hybrid vigor to outcrossing and thus developed an anti-inbreeding principle. Gartner, in 1849, noted the

hardiness of many hybrids. Darwin's careful and extensive work was the forerunner of twentieth century research on hybrid vigor.

East and Jones (1919) described the manifestations of hybrid vigor as commonly being a general increase in size. This largeness is due to an increase in the size of the component parts rather than an increase in the number of parts. In maize, for example, the increase in length of the internodes is much greater than the increase in the number of internodes. Other expressions of vigor in maize include extensions in the diameter of the stalk, increased length and breadth of leaves, greater root development, larger tassels and ears, increased number of ears, and increased seed production. Jones (1918) attributed this increase in size to an increase in both size and number of cells.

East (1936) defined hybrid vigor in terms encompassing the whole organism, plant or animal. In plants its effect is likened to adding a balanced fertilizer to the soil. This vigor is not too apparent in flowers or fruits since the general vegetative stimulus is weakened by the time sexual maturity is reached. Also, reproductive processes and vegetative growth are separate phenomena. Yet preparation for reproduction involves vegetative growth and hence hybrid vigor is often shown in the profusion of flowers and fruit.

Richey (1946) defined hybrid vigor as "an excess of vigor of a hybrid over the average vigor of its parents."

The term "heterosis" was proposed by Shull (1914) to describe the increased development which may be due to heterozygosity. The term was coined for the sake of brevity and for the want of a word free from implications of Mendelian genes necessarily stimulating the cell division, growth, and other physiological processes of an organism.

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Shull (1948) later elaborated upon the scope and generality of the term. The visible and invisible phenomena resulting from the union of different gametes cannot be separated so heterosis applies to the entire process. The term also includes the differences in uniting gametes not due to analyzable Mendelian genes. Heterosis is more inclusive than hybrid vigor--all hybrid vigor can be termed heterosis but not all heterosis is hybrid vigor (as in certain groups of fungi where unlike elements are brought together by nuclear migrations and not by cross-fertilization). The phenomena of heterosis is complex and no single mechanism or cause can be presumed to apply in all instances.

Several different theories have been proposed to explain the pehnomenon of heterosis. They are not completely exclusive of each other and so more than one mechanism may be involved in a particular case of heterosis (Shull, 1948).

Bruce (1910) assumed that dominance was positively correlated to vigor and showed mathematically that crossing two different breeds resulted in the decrease of the number of homozygous recessive genotypes. Therefore, a mean vigor greater than the collective mean vigor is produced. Inbreeding a Mendelian population reduces the mean number of homozygous and heterozygous dominants and hence reduces vigor. Bruce's general treatment was the first to attribute dominant inter-action as the basis of heterosis (Richey, 1945).

Keeble and Pellew (1910) similarly explained the greater height in certain of their pea hybrids by the accumulation of dominant growth factors in the zygote, some contributed by one parent and others by the other parent.

The assumption of a dominance hypothesis is that dominant genes are favorable while the recessive counterparts are deleterious. East and Jones (1919) maintained that natural selection eliminates unfavorable dominant variations while unfavorable recessive variations tend to be perpetuated in the heterozygous state.

Two objections to the dominance hypothesis explaining heterosis have been raised. 1) Recombination should result in the appearance of an F_2 individual homozygous for all dominant factors present in the F_1. Resultant progeny of self fertilizing such an F_2 individual would all be uniform and as vigorous as the F_1. Such an individual has not been encountered. 2) If independent dominant factors are responsible for heterosis, the distribution of the F_2 characters would be skewed with the mode being above the mean. In fact, a symmetrical distribution is often obtained (Collins, 1921).

Jones (1917) believed that linkage had not been considered. Different factors are associated into linkage groups by means of distribution on chromosomes. Actions of different factors may produce the same effect. Although each variety possesses favorable as well as unfavorable characters, varieties differ in the power of development. F_1 hybrids of inbred strains of maize are quite normal and display increased vigor over parental vigor since factors lacking in one is contributed by the other and vice versa. Because of linkage, different factors exist on different chromosomes and it is practically impossible for all dominants to be combined onto the same chromosome. If the different factors are distributed on all the chromosomes, the individuals heterozygous for a certain number of factors would fall into

classes following the expansion of the binomial $(a + b)^n$ which is an illustration of the normal frequency distribution.

Collins (1921) calculated that when 10 pairs of characters are involved, more than 100,000 individuals would be needed for the laws of probability to favor the appearance of one individual homozygous for all characters. Also, as the number of characters increases, the skewness of the distribution is not as marked. Collins also calculated that in consideration of twenty characters, 1,099,514,627,776 individuals were needed to compose a representative population of 21 classes in which 99.9% of the individuals fall into the 12 classes having the greatest number of dominants. A population of 500 individuals would greatly resemble the normal distribution.

Collins (1921) criticized Jones' linkage modification of the dominance theory as being "superfluous" in accounting for heterosis. Not dismissing the probability of linkage, he argued that the objections to the dominance theory that Jones' linkage theory refuted actually had no basis in fact.

Crow (1948) claimed that the dominance hypothesis could account for little of the increased vigor of hybrids. If vigor is evaluated in terms of selective advantage, its value would merely increase by 5% when all homozygous recessive factors are replaced.

Shull (1914) credited heterosis to the "dissimilarity in the gametes" forming the organism. This heterogeneity and unbalance of differences in the germ cells result in the stimulus to increased cell division, growth, etc. Within limits the more numerous the differences between gametes, the greater is the amount of stimulation. East and

Hayes (1912) also arrived at the same hypothesis. The stimulus to development is increased by the heterozygous condition. The nature of such a stimulus may be mechanical, chemical, or electrical. By this hypothesis, inbreeding itself is not a degenerative process but instead one of Mendelian segregation (East and Jones, 1919). Unfavorable recessives hidden in the heterozygous condition are isolated in the homozygous state. A decreased power of development is due to the lack of stimulation from heterozygosity.

East (1936) confirmed that heterosis increases as the genetic differences between parental stocks increase. Hybrids between pedigreed inbred stocks display decreasing heterosis as the degree of relationship increases. Increased heterosis is also apparent when heterogamous stock is successively selfed prior to being crossed.

A. F. Shull (1912) criticized Shull's hypothesis since in accordance with this view, successive generations of inbreeding could produce a pure homozygous individual and every pure line must then reach its minimum in vigor which would be identical for all pure lines. Also, inbreeding must then always eventually reduce vigor provided random segregation and recombination occurred.

East (1910) proposed the possibility of several independently inherited allelomorphic pairs being involved in determining a particular character. The presence or absence of the dominant factor in these allelomorphic pairs would result in differing combinations, some producing the same effect on the character. The additive effects of presence or absence of the dominant factor results in quantitative variation. Hence, for a particular quantitative character, a number of

genotypes may be responsible for the same expression.

East's example was based on the hypothesis of three allelomorphic pairs determining the number of rows on ears of maize. With a basal unit of eight rows, the homozygous dominant condition contributes four rows while the heterozygous state accounts for two rows. Hence, the genotype AABBCC results in 20-rowed ears; AaBBCC, AABbCC, and AABBCc result in 18-rowed ears, etc. Therefore, since the same quantitative character may be due to differing genotypes, plants of 16-rowed ears may sometimes be obtained when crossing two plants having 12-rowed ears.

Hull (1945) assumed hybrid vigor to be a result of gene interaction. Assigning a value of 0.0 for the genotype aa and 1.0 for AA, a heterozygote with a value of 0.5 is intermediate between both parents and the locus does not contribute to hybrid vigor. As the heterozygote value approaches or exceeds 1.0, the importance of the locus in hybrid vigor is increased. Loci at which the heterozygote is superior to either homozygote contributes to hybrid vigor. The evidence of heterozygote values exceeding 1.0 is in the F_1 hybrids whose yields are in excess of the sum of the yields of two homozygous parents.

A. F. Shull (1912) recognized vigor to have its basis in metabolism. He hypothesized that when new nuclear elements encounter a cytoplasm in equilibrium as in cross-fertilization, the resulting inter-action increases metabolism and hence vigor is observed. It is not the heterozygous condition in itself, but the interaction of the heterozygous nucleus (Mm) with the cytoplasm heretofore in equilibrium with an MM or mm nucleus that produces vigor. The effect of the changed nucleus on the surrounding cytoplasm produces the stimulus to

increased cell division. Here, Shull refuted East and Hayes' stance

that the more rapid cell division determining vigor was stimulated by

the heterozygous condition.

Jones (1945) observed recessive variations in inbred lines of maize

which reduced growth but were not lethal. These variations he believed

to be degenerative changes due to single allelic modifications. Upon

crossing such mutant lines to the corresponding original inbred lines,

a great amount of heterosis resulted. Heterosis, according to Jones,

is "an accumulative effect of favorable heredity from both parents" even

when involving single allelic differences (assuming multiple effects of

genes).

Castle (1946) elaborated upon Jones' evidence. He proposed a

sensitization by a new dominant allele A, appearing in the unorganized

chromatin, on the chromatin at the opposite locus, resulting in a

recessive allele a. This sensitization is in a manner like anaphylaxis.

The two alleles establish two homozygous strains, AA in the mother

strain and aa in the mutant daughter strain. Crossing these two strains

differing in a single gene pair produces a hybrid with increased

growth energy. In cases where hybrid vigor is not apparent when two

inbred lines are crossed, the sensitized recessive allele a is absent.

Heterozygosity of the single gene pair Mama, concerning

photoperiodic response and time of floral initiation in sorghum, was

found to produce heterosis comparable in degree to commercial maize

hybrids. Quinby and Karper (1946) thus interpreted their data as

supporting the theory of interaction between unlike allelomorphs as

the plausible explanation of heterosis. The stimulation to tillering

and cell division derived from this heterozygous condition was also believed to be due to an increased capacity to utilize the available nutrient supply.

The genes determining physiological efficiency are much greater in number than genes determining morphological characters. Heterosis is mainly concerned with the speed of physiological reactions. Genes may be classified into two types--those that cause breakdowns in physiological processes and those that do not. A defective gene may be compensated for by a normal allele in the pair, and the respective processes are usually not affected. The heterosis observed when two long-inbred lines are crossed involve the "different genic isomers of the physiologically active and more or less normal genes." Non-defective intra-allelic genes, each diverging from each other in function, may have additive effects. Heterozygotes become more efficient as the component alleles diverge more greatly (East, 1936).

Homozygous strains of Drosophila melanogaster exhibit greater variance within a strain than do heterozygous strains. Decline due to inbreeding is apparent in the character of size, and heterosis is manifested in increased size and vigor as well as reduced susceptibility to environmental fluctuations. Robertson and Reeve (1952) theorized that a greater degree of heterozygosity means a greater diversity of alleles which provide "greater biochemical versatility in development." Heterosis is exhibited because of the superior ability of a highly heterozygous individual to efficiently use the available nutrients and the decrease in susceptibility to environmental fluctuations since more alternatives of overcoming such obstacles to development are available.

East (1936) emphasized that heterosis effects cannot be compared among different genera. Genetic evidence points to greater variation in some genera than others—mutation rates being higher in some. Hence, each genus requires individual consideration.

Cross- and Self-Fertilization in Orchids

The floral structures of many species of Orchidaceae were examined by Darwin (1904). He was impressed by the multitude of devices and variety in structure, all ensuring the common end of cross-fertilization. Some species of orchids are primarily or frequently self-fertilized, yet retain various structures adapted for cross-fertilization despite the fact that they are rarely if ever involved. Darwin thus concluded that such species were descended from plants cross-fertilized by insects. Under conditions of limited or no insect visitation, floral structure was gradually modified to allow for self-fertilization. Self-fertilized seeds are more advantageous to the perpetuation of the species than very few or no seeds.

Since orchid pollen must be required in a large amount to produce the great quantity of seed found in orchids and is located in anthers just above or behind the stigma, it would more safely and easily be utilized in self-fertilization than in cross-pollination where transport is necessary. Darwin, noting the beneficial effects in most cases of cross-fertilization in orchids, felt that this demonstrated that Nature "abhors perpetual self-fertilization."

MATERIALS AND METHODS

The inbred plant material used in this experiment were derived from amphidiploid D. Jaquelyn Thomas 'Y166-1', a tetraploid which arose from a diploid population. (D. Jaquelyn Thomas is a hybrid of the species D. gouldii and D. phalaenopsis.) An outcross was made to the tetraploid D. Neo Hawaii '2097 4N'.

The crosses (Table 1) were made on November 1 or 3, 1972. Seeds were set to germinate on Modified Vacin and Went Medium in 125 ml flasks on January 16, 1973. Seedlings were transflasked on April 17, 1973 to 500 ml flasks on Modified Vacin and Went Medium. On October 9, 1973, approximately 70 of the larger plants were selected from each cross and planted into community pots. Thirty-two plants per cross were further selected on May 29, 1974 (July 23, 1974 for cross 10) and individually potted into 2-inch clay pots. Selection pressure was again applied on March 3, 1975 when 20 of the most vigorous-appearing plants of each cross were repotted in a mixed rock medium in 6-inch cement pots. The plants were grown in the orchid saran house at the Upper Manoa Campus of the University of Hawaii.

A randomized block statistical design was employed. The 20 plants within a cross were ranked from 1 to 20 in decreasing order of apparent vigor. Plants of a particular rank from every cross constituted a block. On March 7 and 8, 1975, the 20 blocks were randomly arranged on two 21 feet by 4 feet benches, resulting in a 4-pot by 25-pot arrangement on each bench, buffered by a guard row of 3 or 4 pots at each end. Within each block the individual plants representing the different crosses were also randomly placed. Randomness was achieved

Table 1. Inbred and outcross progenies of amphidiploid D̲. Jaquelyn Thomas.

Progeny number	Progeny	Type of mating
1	Y166 ⊗	P selfed (S_1)
2	Y166-1 X K159-21[w]	P X S_2 (BC_2)
3	K44-5 ⊗	S_1 selfed (S_2)
4	K44-50 X K44-5[y]	S_1 sibmated
5	K44-50 X Y166-1	S_1 X P (BC_1)
6	K159-19 ⊗	S_2 selfed (S_3)
7	K159-19 X K159-21	S_2 sibmated
8	K159-21 X K159-19	S_2 sibmated
9	K159-21 X Y166-1	S_2 X P (BC_2)
10	2097 (4N)[z] X K159-21	outcross

[w]K159 is K44-50 selfed.
[y]K44 is Y166-1 selfed.
[z]2097 (4N) is a tetraploid Neo Hawaii.

through the use of a random digits table. In July, 1976, half of the plants were transferred to two adjacent benches to allow for greater spacing between the growing plants. Half a bench of plants was moved to an adjacent bench. The pots were transferred in a serpentine fashion to an arrangement with three pots across the width of the bench. The remaining plants on a bench were also rearranged in a serpentine fashion to a three-pot-wide arrangement on the same bench. Thus, the blocks were kept intact.

On March 10, 1975, height data were first recorded. The two youngest shoots were measured to the nearest half centimeter from the base of the shoot to the base of the "V" of the top leaves. The greatest measurement was taken to be the absolute height of the plant at the particular date. Measurements were continually taken at two-month intervals.

Flowering commenced on August 12, 1975, and therefore the period of one week prior to that date was designated as week 1. The following weeks were consecutively numbered and the date of flowering was recorded as the number of the week in which the first flower of a spray opened.

Sprays were harvested when 3/4 of the flowers were opened. Harvesting was done between 7:30 a.m. and 11:30 a.m. Sprays were immersed in tap water for 15 minutes and then transferred to 500 ml flasks of tap water. The water was changed three times a week at which time 1/4 to 1/2 inch of the basal part of each flower spray was cut off. The flower sprays were set in an air-conditioned laboratory where an approximate temperature of 23 degrees C. and a humidity level of 50% were normally maintained. The keeping quality of a spray was determined

as the length of time the flower spray lasted until becoming completely wilted or until half of the flowers wilted or senesced.

Flower size of a spray was established as the broadest measurement, to the nearest millimeter, of the third lowest flower on the spray.

Color of the essentially white flowers was subjectively evaluated as 1-lightly tinged, 2-moderately tinged, or 3-heavily tinged.

The length of the scape, to the nearest centimeter, was measured on the plant as the distance from the stem base to the lowest flower. Total spray length, also measured to the nearest centimeter, was later figured as the scape measurement plus the measurement of the stem from the lowest flower to the tip of the spray.

The number of flowers on a spray was recorded at harvest. Bud drop was determined as the number of flowers or buds that dropped prior to harvest.

Analysis of variance for the randomized complete block design and the Bayes Least-Significant Difference for Multiple-Comparison Testing were used to analyze the data for height, yield and date of first flowering. Due to the unequal number of flower sprays produced by individual plants, flower spray characters were analyzed according to the completely randomized design and orthogonal comparisons were made.

Height

Table 2 shows the mean progeny height at two-month intervals from the age of 26 months to 44 months.

The S_1 and S_2 plants were comparatively tall throughout the measurement period. The outcross progeny was intermediate in height during the early growth period but was relatively tall toward the latter period. S_3 plants were relatively tall during the early period but short when older. The progeny from sibmating the S_1 varied from an intermediate ranking at early growth to a high ranking at later growth while plants from S_2 sibmatings moved from an intermediate to a low position in the ranking. The backcross progeny of S_1 X P maintained a low to low-intermediate position in the comparisons, but backcross progenies of S_2 X P and P X S_2 varied within a low-intermediate to high-intermediate range.

Differences in reciprocal crosses were not statistically significant.

Yield

Mean yield of the progeny of the matings are shown in Table 3. The highest yields were obtained from the outcross, S_1 , S_1 sibbed, S_2, and the backcross S_2 X P. No significant differences were found in the mean number of sprays produced among these progenies.

The lowest yields were obtained in S_2 sibbed, the backcross S_1 X P, the S_3, and the backcross P X S_2. These yields, however, were not significantly different from those of the other S_2 sibbed, S_2, and the

Table 2. Mean height in centimeters of progeny of matings of amphidiploid $\underline{D}$. Jaquelyn Thomas at different ages.

					Age of Progeny					
Type of mating	26 months	28 months	30 months	32 months	34 months	36 months	38 months	40 months	42 months	44 months
P selfed (S_1)	14.4 a	20.1 a	28.4 a	40.2 a	42.2 ab	44.7 b	53.2 ab	69.0 ab	92.9 ab	106 ab
P X S_2 (BC_2)	12.3 c	14.9 d	22.2 de	33.7 cd	35.5 de	43.1 bc	46.3 cd	55.1 d	82.5 bcd	102 bc
S_1 selfed (S_2)	15.1 a	18.8 ab	26.8 ab	36.7 b	44.1 a	49.9 a	55.6 a	68.2 abc	89.5 ab	104 abc
S_1 sibbed	13.3 b	18.7 ab	25.0 bc	36.4 bc	40.5 bc	43.6 bc	51.8 abc	66.4 abc	86.7 bc	104 abc
S_1 X P (BC_1)	8.0 e	12.8 e	19.8 e	28.7 e	33.8 e	37.0 d	44.4 d	62.0 bcd	85.3 bc	92 cd
S_2 selfed (S_3)	13.9 b	19.6 a	26.0 abc	35.1 bcd	41.1 bc	42.8 bc	46.4 bcd	55.6 d	72.8 d	79 d
S_2 sibbed	13.4 b	18.3 abc	23.8 cd	34.8 bcd	39.2 c	42.8 bc	47.0 bcd	53.8 d	68.4 d	83 d
S_2 sibbed	10.3 d	17.4 bc	22.3 d	32.3 d	38.5 c	40.9 bc	47.5 cd	61.1 cd	75.5 cd	85 d
S_2 X P (BC_2)	11.5 c	16.8 cd	24.5 bcd	33.9 cd	38.3 cd	40.3 cd	46.0 cd	65.1 bc	87.8 ab	101 bc
Outcross	12.0 c	17.0 bc	26.8 ab	36.0 bc	38.6 c	41.8 bc	53.4 ab	74.8 a	99.1 a	116 a

[a]Means in a column followed by the same letter are not significantly different at P=0.05 by the Bayes Least-Significant Difference for Multiple-Comparison Testing. Figures are means of twenty individuals.

Table 3. Mean yield of progenies of matings of amphidiploid $\underline{D}$.
Jaquelyn Thomas to age 44 months.

Type of progeny	Mean number of sprays produced by progeny
P selfed (S_1)	5.4 a
P X S_2 (BC_2)	3.8 c
S_1 selfed (S_2)	4.4 abc
S_1 sibbed	5.2 ab
S_1 X P (BC_1)	3.7 c
S_2 selfed (S_3)	3.7 c
S_2 sibbed	3.5 c
S_2 sibbed	4.2 bc
S_2 X P (BC_2)	4.4 abc
Outcross	5.4 a

[a]Means followed by the same letter are not significantly different
at P=0.05 by the Bayes Least-Significant Difference for Multiple-
Comparison Testing. Figures are means of twenty individuals.

backcross S_2 X P.

Reciprocal matings did not produce significant differences in yield.

Date of first flowering

The mean week of earliness to flower of the progenies is found in in Table 4. The earliest flowering plants were those of the S_1, outcross, backcross P X S_2 and its reciprocal S_2 X P, S_1 sibbed, and backcross S_1 X P. The latest flowering plants were those of the sibmatings in S_2 (reciprocal crosses), the S_2, and the S_3.

Five estimated values were used in the analysis of variance, two due to flower shoots being broken off or dying in the early stages. From data on the length of time involved in the development of the flower shoot to a flowering spray, estimates were calculated based on the date of the observed flower shoot damage. These values were included in the data for crosses 7 and 8 (reciprocal sibmatings in the S_2). Three plants had not yet flowered at the time of data analysis. One was observed with two flower shoots in buds and hence, the time of expected first flowering was estimated. This plant belonged to cross 4 (sibmating in S_1). Two plants that did not exhibit signs of flowering were, in week 64, assigned the high values of week 100. These late-flowering plants belonged to mating 3 (S_2) and mating 7 (sibmating in S_2).

Keeping quality

The longer lasting quality of sprays of the outcross offspring differed significantly from sprays of the other plants (Tables 5, 6).

Table 4. Mean week of earliness to flower of progeny of matings of
amphidiploid $\underline{D}$. Jaquelyn Thomas.

Type of progeny	Mean week[z] of first flowering
P selfed (S_1)	8.1 a
P X S_2 (BC_2)	13.5 ab
S_1 selfed (S_2)	22.0 cde
S_1 sibbed	13.6 abc
S_1 X P (BC_1)	15.5 abcd
S_2 selfed (S_3)	18.8 bcde
S_2 sibbed	24.9 e
S_2 sibbed	22.3 de
S_2 X P (BC_2)	12.2 ab
outcross	9.7 a

[z]Weeks were consecutively numbered relative to the week of
August 12, 1975 (week 1).

[a]Means followed by the same letter are not significantly different
at P=0.05 by the Bayes Least-Significant Difference for Multiple-
Comparison Testing. Figures are means of twenty individuals.

Table 5. Significance of analysis of variance F values for crosses and orthogonal comparisons of the characters of keeping quality, flower size, number of flowers per spray, spray length and scape length of progeny of matings of amphidiploid $\underline{D}$. Jaquelyn Thomas.

	Keeping quality	Size of flower	Number of flowers per spray	Spray length	Scape length
Treatments (Matings)	**	**	**	ns	**
Outcross vs. others	**	**	**	ns	**
S_1 vs. S_2 and S_3	ns[z]	ns	**	ns	*
S_2 vs. S_3	**	**	**	ns	**
BC_1 vs. BC_2[y]	ns	**	ns	ns	ns
BC_2 vs. BC_2 (reciprocals)	ns	ns	ns	ns	ns
S_1 sibbed vs. S_2 sibbed	ns	**	**	ns	**
S_2 sibbed vs. S_2 sibbed (reciprocals)	ns	ns	ns	ns	ns
Residual	**	**	**	ns	ns

[y]BC_1 means S_1 backcrossed to P; BC_2 means S_2 backcrossed to P.
[z]ns means differences are nonsignificant.
*, ** One asterisk means differences are significant at P=0.05; two asterisks mean differences are significant at P=0.01.

Table 6. Mean[z] values of keeping quality, flower size, number of flowers, spray length, scape length and percentage bud drop for flower sprays of progeny of matings of amphidiploid D. Jaquelyn Thomas.

Type of mating	Keeping quality (days)	Size of flower (cm)	Number of flowers per spray	Spray length (cm)	Scape length (cm)	Percentage bud drop
P selfed (S_1)	9.5	5.81	20.2	57.9	16.1	0.9
P X S_2 (BC_2)	8.4	6.09	18.9	56.2	16.7	0.8
S_1 selfed (S_2)	11.3	5.24	18.7	57.8	17.8	2.7
S_1 sibbed	10.3	5.55	18.8	55.8	17.7	1.7
S_1 X P (BC_1)	8.8	5.84	18.5	55.2	16.6	0.2
S_2 selfed (S_3)	8.1	6.74	16.4	54.8	16.1	0.6
S_2 sibbed	9.9	6.80	14.9	51.0	16.1	0.6
S_2 sibbed	9.5	6.76	15.7	52.7	16.0	0.6
S_2 X P (BC_2)	9.0	6.22	17.8	55.1	17.0	0.6
Outcross	12.1	6.47	16.6	57.7	19.3	0.1

[z]Figures are means of twenty individuals.

Flower sprays from S_2 plants kept significantly longer than those from S_3 plants. The residual component was also significant. Reciprocal matings produced sprays that did not differ significantly in shelf life.

Flower size

The outcross significantly differed from the other matings in the size of the flower of the offspring (Tables 5, 6). Flowers of the S_3 were significantly larger than those of the S_2. The flowers produced by the backcross S_1 X P were significantly smaller than flowers produced by the backcrosses S_2 X P and P X S_2. Sibmating in the S_1 resulted in flowers significantly smaller than those resulting from sibmatings in the S_2. The residual component accounted for part of the significance of differences among matings. Flower size of reciprocal matings were not significantly different.

Number of flowers per spray

The number of flowers per spray of the outcross differed significantly from sprays of the other progenies (Tables 5, 6). S_1 sprays had significantly more flowers than sprays of the S_2 and S_3. S_2 sprays possessed a significantly greater number of flowers than the S_3 sprays. The S_1 sibbed bore sprays with significantly more flowers than sprays of S_2 sibbed. Significance also resided in the residual component. Progenies of reciprocal matings did not differ in the number of flowers per spray.

Spray length

No significant differences in spray length were found among the progenies (Tables 5, 6).

Scape length

Scape length of the outcross was significantly longer than that of
the other progenies (Tables 5, 6). S_1 plants differed at $P=0.05$ from S_2
and S_3 plants in scape length. The scape of sprays of S_2 plants was
significantly longer than the scape of S_3 sprays. Offspring of the S_1
sibbed had significantly longer scapes than offspring of S_2 sibbed.
No significant difference in scape length between offspring of
reciprocal matings was found.

Color

The outcross, S_2 selfed, and the sibmatings in the S_2 were all
classified as lightly tinged. The reciprocal backcrosses, S_2 X P and
P X S_2, produced progenies whose flowers were essentially moderately
tinged. S_1 sibbed and the backcross S_1 X P varied in flower color from
moderately tinged to heavily tinged. Flowers of the S_1 and S_2 were
essentially heavily tinged.

Bud drop

The percentage of bud drop in progenies of all matings was very
low, being 0% in the great majority of sprays. Except for the S_2 and S_1
sibbed, the average percentage of bud drop was less than 1%. In the S_2
the average bud drop percentage was 2.7% while S_1 sibbed had a 1.7%
bud drop.

DISCUSSION

Inbreeding as well as selection effects were apparent in most of the characters observed. Yield, which is of considerable importance in agricultural crop production, declined with an increased level of inbreeding in this cross-fertilized crop. The data accumulated to date represents only the early portion of the productive years of the plants. Although yield differences appear small at this point (3.5 vs. 5.4 sprays per plant), they are significant and subsequent data may show greater differences. The least inbred or non-inbred plants (the outcross and S_1) yielded significantly greater than plants inbred to a greater degree (S_2 sibmated and S_2 selfed).

Vigor is often indicated by the size of the plant. A decrease in size usually accompanies inbreeding decline. A measurement of height was used to approximate size of the plants. During the early period of growth when plants were short, shoot maturation was not synchronized. Hence, the height average of the offspring of a particular mating may have been misleading, when individual variations easily altered averages. Height averages of the latter part of the measurement period are probably more appropriate for comparisons, since the mature shoots were taller and nearer to their ceiling height. The averages obtained during this latter period revealed that the lesser inbred and noninbred progenies (the outcross, P selfed, S_1 sibbed, and S_1 selfed) were taller. Progenies of more intense inbreeding (S_2 sibbed and S_2 selfed) were comparatively shorter.

Earliness is sometimes construed as a manifestation of vigor. Hence, the week of first flowering provides a measure of earliness to determine inbreeding effects. In the two crosses where a plant had not yet flowered, and the week was estimated as a high value, the apparent lateness of these crosses may not realistically represent the degree of inbreeding. Nevertheless, some information can be gleaned from the data. The outcross and S_1 progenies showed the earliest flowering. Of the progenies of matings without a divergent individual, S_2 selfed and a sibmating in S_2 were the latest to flower.

Individuals were selected for flower size and color in the direction of larger, whiter (lower degree of the pink tinge) flowers. This selection pressure was effective in increasing the purity of flower color. A selected S_1 individual (moderately tinged K44-50) and its further selected S_2 individuals (lightly tinged K159-19 and K159-21) were involved in the matings. Sibmating the selected S_2 individuals, selfing a selected S_2 individual, and outcrossing one of the selected S_2 individuals to white Neo Hawaii resulted in progenies whose flowers were lightly tinged.

The S_1 progeny (from a selfing of the heavily tinged Y166-1) as well as the S_2 progeny (from a selfing of the heavily tinged K44-5) produced heavily tinged flowers.

Crossing selected individuals to non-selected individuals increased the degree of the pink tinge in the progeny. Reciprocal backcrosses of a selected S_2 individual (lightly tinged) to the original parent (heavily tinged) resulted in moderately tinged flowers among the progeny. The selected S_1 individual (moderately tinged) sibmated to a

non-selected S_1 individual (heavily tinged) produced offspring that varied in flower color from moderately tinged to heavily tinged. The selected S_1 individual (moderately tinged) backcrossed to the original parent (heavily tinged) also produced progeny with flowers that varied from moderately to heavily tinged.

Flower size was also clearly influenced by selection pressure. Significant differences are found between the different levels of inbreeding (S_2 vs. S_3, S_1 backcrossed to P vs. S_2 backcrossed to P, and sibmating in S_1 vs. sibmating in S_2), most likely due to the effect of selection with higher levels of inbreeding.

Accidental genetic differences rather than inbreeding effects probably account for the differences in keeping quality. Sprays of the Neo Hawaii parent involved in the outcross have a long lasting quality and this genetic tendency was contributed to the progeny. The S_2 plants with small, heavily tinged flowers, and the S_3 plants with large, lightly tinged flowers differed significantly in keeping quality. This may be related to the size of the flowers and the rate of water loss, or to the genetic tendency toward either the Ceratobium (characteristically with small flowers of long keeping quality) or the Phalaenanthe (whose cut sprays of large flowers have a short vase life) parentage.

While the spray lengths were not significantly different among the progenies, the number of flowers and the length of scape were. It is difficult to say whether inbreeding, or selection, or both are operating. In the comparisons sibmating in S_1 vs. sibmating in S_2 and S_2 vs. S_3, significantly lower values were obtained at the higher level of inbreeding. Also, the S_1 sprays had significantly more flowers than

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the S_2 and S_3 sprays. However, in the outcross and P selfed a longer scape coincided with a lower number of flowers on the spray or vice versa.

Bud drop is probably a character not influenced by inbreeding. The genetic material involved here was relatively free from bud drop. However, the progenies of the individual K44-5 had a mean bud drop percentage greater than 1%. K44-5 appears to have a tendency of transmitting this undesirable trait to its offspring.

Reciprocal crosses showed no significant differences in all characters measured. Hence, in this material, a maternal or paternal role in the mating does not influence the characters measured.

APPENDIX

Appendix A. Analysis of variance for keeping quality of progenies of matings of amphidiploid $\underline{D}$. Jaquelyn Thomas.

Source of variation	Degrees of freedom	Sum of squares	Mean square	Observed F
Total	802	12,908.531	--	--
Matings	9	1,264.611	140.512	9.569**
Outcross vs. others	1	461.653	461.653	31.441**
S_1 vs. S_2 & S_3	1	7.140	7.140	0.486 ns
S_2 vs. S_3	1	387.441	387.441	26.387**
BC_1 vs. $BC_2{}^z$	1	0.458	0.458	0.031 ns
BC_2 vs. BC_2 (reciprocals)	1	16.416	16.416	1.118 ns
S_1 sibbed vs. S_2 sibbed	1	21.706	21.706	1.478 ns
S_2 sibbed vs. S_2 sibbed (reciprocals)	1	3.669	3.669	0.249 ns
Residual	2	366.137	183.063	12.467**
Error	793	11,643.93	14.683	

$^z BC_1$ means S_1 backcrossed to P; BC_2 means S_2 backcrossed to P.

Appendix B. Analysis of variance for size of flower of progenies of matings of amphidiploid D. Jaquelyn Thomas.

Source of variation	Degrees of freedom	Sums of squares	Mean square	Observed F
Total	945	504.280	--	--
Matings	9	241.403	26.822	95.792**
Outcross vs. others	1	16.743	16.743	59.796**
S_1 vs. S_2 & S_3	1	1.008	1.008	3.600 ns
S_2 vs. S_3	1	95.326	95.326	340.450**
BC_1 vs. BC_2[z]	1	5.600	5.600	20.000**
BC_2 vs. BC_2 (reciprocals)	1	0.745	0.745	2.661 ns
S_1 sibbed vs. S_2 sibbed	1	100.125	100.125	357.589**
S_2 sibbed vs. S_2 sibbed (reciprocals)	1	0.059	0.059	0.211 ns
Residual	2	21.797	10.898	38.921**
Error	936	262.877	0.280	

[z]BC_1 means S_1 backcrossed to P; BC_2 means S_2 backcrossed to P.

Appendix C. Analysis of variance for number of flowers per spray of progenies of matings of amphidiploid $\underline{D}$. Jaquelyn Thomas.

Source of variation	Degrees of freedom	Sums of squares	Mean square	Observed F
Total	976	32,476.895	--	--
Matings	9	2,411.195	267.911	8.617**
Outcross vs. others	1	212.798	212.798	6.844**
S_1 vs. S_2 & S_3	1	465.683	465.683	14.978**
S_2 vs. S_3	1	228.970	228.970	7.364**
BC_1 vs. BC_2[z]	1	0.547	0.547	0.018 ns
BC_2 vs. BC_2 (reciprocals)	1	60.005	60.005	1.930 ns
S_1 sibbed vs. S_2 sibbed	1	846.173	846.173	27.215**
S_2 sibbed vs. S_2 sibbed (reciprocals)	1	24.077	24.077	0.774 ns
Residual	2	572.942	286.471	9.214**
Error	967	30,065.700	31.092	

[z]BC_1 means S_1 backcrossed to P; BC_2 means S_2 backcrossed to P.

Appendix D. Analysis of variance for length of spray of progenies of matings of amphidiploid $\underline{D}$. Jaquelyn Thomas.

Source of variation	Degrees of freedom	Sums of squares	Mean square	Observed F
Total	948	284,362.942	--	--
Matings	9	3,943.172	438.130	1.467 ns
Outcross vs. others	1	523.529	523.529	1.753 ns
S_1 vs. S_2 & S_3	1	137.909	137.909	0.462 ns
S_2 vs. S_3	1	358.611	358.611	1.201 ns
BC_1 vs. $BC_2{}^z$	1	10.740	10.740	0.036 ns
BC_2 vs. BC_2 (reciprocals)	1	61.234	61.234	0.205 ns
S_1 sibbed vs. S_2 sibbed	1	970.598	970.598	3.250 ns
S_2 sibbed vs. S_2 sibbed (reciprocals)	1	122.403	122.403	0.410 ns
Residual	2	1,758.148	879.074	2.944 ns
Error	939	280,419.770	298.637	

$^z BC_1$ means S_1 backcrossed to P; BC_2 means S_2 backcrossed to P.

Appendix E. Analysis of variance for length of scape of progenies of matings of amphidiploid $\underline{D}$. Jaquelyn Thomas.

Source of variation	Degrees of freedom	Sums of squares	Mean square	Observed F
Total	1000	11,391.987	--	--
Matings	9	1,124.257	124.917	12.056**
Outcross vs. others	1	732.402	732.402	70.688**
S_1 vs. S_2 & S_3	1	67.110	67.110	6.477*
S_2 vs. S_3	1	124.218	124.218	11.989**
BC_1 vs. $BC_2{}^z$	1	3.129	3.129	0.302 ns
BC_2 vs. BC_2 (reciprocals)	1	3.388	3.388	0.327 ns
S_1 sibbed vs. S_2 sibbed	1	190.200	190.200	18.357**
S_2 sibbed vs. S_2 sibbed (reciprocals)	1	0.184	0.184	0.018 ns
Residual	2	3.626	1.813	0.175 ns
Error	991	10,267.530	10.361	

$^z BC_1$ means S_1 backcrossed to P; BC_2 means S_2 backcrossed to P.

Appendix F. Analysis of variance for week of first flowering of
progenies of matings of amphidiploid D. Jaquelyn Thomas.

Source of variation	Degrees of freedom	Sums of squares	Mean square	Observed F
Total	199	41,424.380	--	--
Treatments (Matings)	9	5,835.880	648.431	1.731*
Blocks	19	5,743.380	302.283	3.715**
Error	171	29,845.120	174.532	--

LITERATURE CITED

Bruce, A. B. 1910. The Mendelian theory of heredity and the
augmentation of vigor. Science 32: 627-268.

Castle, W. E. 1946. Genes which divide species or produce hybrid
vigor. Proc. Nat. Acad. Sci. 32: 145-149.

Collins, G. N. 1921. Dominance and the vigor of first generation
hybrids. Amer. Nat. 55: 116-133.

Crow, J. R. 1948. Alternative hypotheses of hybrid vigor. Genetics
33: 477-487.

Darwin, C. R. 1900. The effects of cross and self fertilisation in
the vegetable kingdom. 2nd ed. John Murray, London.

_____. 1904. The various contrivances by which orchids are fertilised
by insects. 2nd ed. John Murray, London.

East, E. M. 1908. Inbreeding in corn. Rept. Connecticut Agric. Expt.
Sta. 1907-1908. Pp.419-428.

_____. 1909. The distinction between development and heredity in
inbreeding. Amer. Nat. 43: 173-181.

_____. 1910. A Mendelian interpretation of variation that is
apparently continuous. Amer. Nat. 44: 65-82.

_____ and H. K. Hayes. 1912. Heterozygosis in evolution and plant
breeding. U. S. D. A. Bur. Plant Indust. Bull. 243.

_____ and D. F. Jones. 1919. Inbreeding and outbreeding. J. B.
Lippincott Co., Philadelphia and London.

_____. 1936. Heterosis. Genetics 21: 375-397.

Hull, F. H. 1945. Recurrent selection for specific combining ability
in corn. J. Amer. Soc. Agron. 37: 134-145.

Jones, D. F. 1917. Dominance of linked factors as a means of
accounting for heterosis. Genetics 2: 466-479.

_____. 1918. The effects of inbreeding and crossbreeding upon
development. Connecticut Agric. Expt. Sta. Bull. 207.

_____. 1924. The attainment of homozygosity in inbred strains of
maize. Genetics 9: 405-418.

_____. 1925. Genetics in plant and animal improvement. John Wiley and Sons, Inc., New York.

_____. 1939. Continued inbreeding in maize. Genetics 24: 462-473.

_____. 1945. Heterosis resulting from degenerative changes. Genetics 30: 527-542.

Keeble, F. and C. Pellew. 1910. The mode of inheritance of stature and of time of flowering in peas (Pisum sativum). J. Genet. 1: 47-56.

Quinby, J. R. and R. E. Karper. 1946. Heterosis in sorghum resulting from their heterozygous condition of a single gene that affects duration of growth. Amer. J. Bot. 33: 716-721.

Richey, F. D. 1945. Bruce's explanation of hybrid vigor. J. Hered. 36: 243-244.

_____. 1946. Hybrid vigor and corn breeding. J. Amer. Soc. Agron. 38: 833-841.

Robertson, F. W. and E. C. R. Reeve. 1952. Heterozygosity, environmental variation and heterosis. Nature (Lond.) 170: 286.

Shull, A. F. 1912. The influence of inbreeding on vigor in Hydatina senta. Biol. Bull. 24: 1-13.

Shull, G. H. 1908. The composition of a field of maize. Amer. Breeders' Assoc. Rep. 4: 296-301.

_____. 1910. Hybridization methods of corn breeding. Amer. Breeders' Mag. 1: 98-107.

_____. 1911. The genotypes of maize. Amer. Nat. 45: 234-252.

_____. 1914. Duplicate genes for capsule form in Bursa bursa-pastoris. Zeitschr. f. ind. Abst. u. Vereb. 12: 97-149.

_____. 1948. What is "heterosis"? Genetics 33: 439-446.

Stebbins, G. L. 1957. Self-fertilization and population variability in higher plants. Amer. Nat. 91: 337-354.

Tysdal, H. N., T. A. Kiesselbach, and H. L. Westover. 1942. Alfalfa breeding. Nebraska Agric. Expt. Sta. Res. Bull. 124.

Wallace, B. 1968. Topics in population genetics. W. W. Norton and Co., Inc., New York.

Zirkle, C. 1952. Early ideas on inbreeding and crossbreeding. In Gowen, J. W. (ed.), Heterosis. Iowa State College Press, Ames, Iowa.