CHAPTER TWELVE

Population Formation by Hybridization

The first step in a cultivar development program is to form a population with genetic variability for the characters of interest. This is done by hybridization of genetically different parents or by the use of mutagenesis. Population formation by hybridization will be the focus of this chapter.

TYPES OF POPULATIONS

The populations used for cultivar development range from a two-parent cross to a complex population involving several hundred parents. Their degree of homozygosity and homogeneity may vary considerably both within and among species. The population formed may be used for direct selection of a cultivar or may be utilized in a recurrent selection program.

Two-Parent Population

The simplest and most widely used population is formed by the mating of two parents, P1 × P2 (Fig. 12-1). The population is referred to as a two-parent cross, single cross, or two-way cross. The population may be used directly for selection or mated to other parents or populations.

Three-Parent Population

A three-parent cross is commonly referred to as a three-way cross. It is formed by mating a two-parent population to a third parent, (P1 × P2) × P3 (Fig.

Season	Procedure	Type of population
		Two-way cross (single cross, two-parent cross)
1	Cross two parents Obtain hybrid F_1 seed	Parent 1 × Parent 2 ↓
2	Grow F_1 plant Obtain F_2 seed	F_1 ↓ Self-pollination F_2 seed
		Three-way cross (three-parent cross)
1	Cross two parents Obtain hybrid F_1 seed	Parent 1 × Parent 2 ↓
2	Grow F_1 plant Cross F_1 to third parent Obtain hybrid F_1 seed	F_1 plant × Parent 3 ↓ F_1
3	Grow F_1 plants Obtain F_2 seed	↓ Self-pollination F_2 seed
		Complex population
1	Make two-way crosses between parents (first intermating) Obtain hybrid F_1 seed	Parents 1 × 2 3 × 4 5 × 6 7 × 8 ↓ ↓ ↓ ↓
2	Cross F_1 plants from different two-way crosses (second intermating) Obtain hybrid seed	F_1 × F_1 F_1 × F_1 ↘ ↙
3	Cross F_1 plants from different four-way crosses (third intermating) Obtain hybrid seed	F_1 × F_1 ↓ Hybrid seed
4	Begin selfing	↓ Plants of complex population

Figure 12-1 Development of a two-way cross, a three-way cross, and a complex population.

12-1). The plants of the two-parent population that are mated to the third parent may be in the F_1 or some later generation of self-pollination.

The three-parent cross is preferred by some breeders when one parent (P1) has a desirable character but is not adequately acceptable for other traits to be used in a successful two-parent cross. Parents P2 and P3 are desirable parents, except for the character to be obtained from P1. The three-parent cross results in a population with an average of 25 percent of the alleles from P1, 25 percent from P2, and 50 percent from P3. For example, large seed is preferred for certain uses of soybean seed. The market for large seed is minor compared with the amount of seed processed for protein and oil, for which seed size is of no consideration. The cultivars grown by farmers have relatively small seed and outyield large-seeded cultivars by over 15 percent. It would be desirable to combine the high yield of a small-seeded cultivar (P1) with the large seed weight of a lower yielding cultivar (P2). A two-parent population from the cross P1 × P2 has few, if any, segregates with adequate seed size (Bravo et al., 1981). Mating F_1 plants of the P1 × P2 cross to a third parent (P3) with large seed usually results in a population with an adequate frequency of large-seeded segregates.

Backcross Population

A backcross population is formed by mating two parents, P1 × P2, then crossing the population back to one of the two parents, (P1 × P2) × P2. The number of backcrosses can be one or more. Each backcross decreases by half the average number of alleles contributed to the population by the nonrecurrent parent: P1 × P2 = 50 percent P1, (P1 × P2) × P2 = 25 percent P1, [(P1 × P2) × P2] × P2 = 12.5 percent P1.

A backcross population is an alternative to a three-parent cross when one parent has a desirable character but is not adequately acceptable in other characters. Consider the example of seed size in soybeans that was used to illustrate a three-parent cross. An adequate frequency of large-seeded segregates in a population could be obtained from the three-parent cross (P1 × P2) × P3 or from the backcross (P1 × P2) × P2, where P1 is the high-yielding, small-seeded parent and P2 and P3 are large-seeded parents.

Four-Parent Population

A four-parent population can be formed by the mating (P1 × P2) × (P3 × P4). This mating is commonly referred to as a double cross or a four-way cross. In the mating of two single crosses, each of the parents contributes an average of 25 percent of the alleles in the final population.

A second method for the formation of a four-parent population is the mating [(P1 × P2) × P3] × P4. The genetic contribution of the parents to the final population is not the same. The parents of the initial two-way cross, P1 and P2, have an average genetic contribution of 12.5 percent each, P3 contributes 25 percent, and P4 contributes 50 percent.

Complex Population

A complex population is one that is formed by hybridization of more than four parents. Complex populations were not widely used for selection in a cultivar development program in the past. They have been more common in recent years because of the interest in population improvement by recurrent selection.

The potential advantages of a complex cross compared with a single cross are that the number of possible alleles in the population for each locus increases with the number of parents used, and the probability of heterozygosity at multiple loci is greater. The first advantage relates to individual loci with two or more alleles, one of which is more favorable than the others. With two homozygous parents, only one allele can be contributed by each. For a quantitative character controlled by multiple genes, each with small effects, the breeder does not know which allele is present in a parent. In a single cross between homozygous parents, one, both, or neither parent may have the most favorable allele possible at a locus. The probability that at least one parent has the most favorable allele at each locus increases as the number of parents in the population increases.

The second advantage of a complex cross relates to the number of loci that will be heterozygous. The probability that homozygous parents will have different alleles at two or more linked loci increases as the number of parents of the population increases. Heterozygosity for linked loci is required before effective recombination can occur between them (Chap. 3).

PRINCIPLES IN THE FORMATION OF A COMPLEX POPULATION

Selection of a procedure to form a complex population depends on the importance that the breeder places on a number of factors. These factors include (a) the need to combine alleles from all parents into the members of a population, (b) the number of parents involved, (c) the genetic contribution of each parent to the population, and (d) the amount of time available to form the population. The impact of each of these factors on the genetic makeup of the population and the efficiency of its development will be considered.

Combination of Alleles from Different Parents

The alleles obtained by the members of a population is influenced by the number of generations of intermating conducted before selection is initiated. To provide the opportunity for a segregate to possess genes from every parent of a population, two parents require a minimum of one intermating, three or four parents require two intermatings, five to eight parents require three intermatings, and so forth.

Number of parents	Number of intermating generations required
2	1
3–4	2
5–8	3
9–16	4
17–32	5

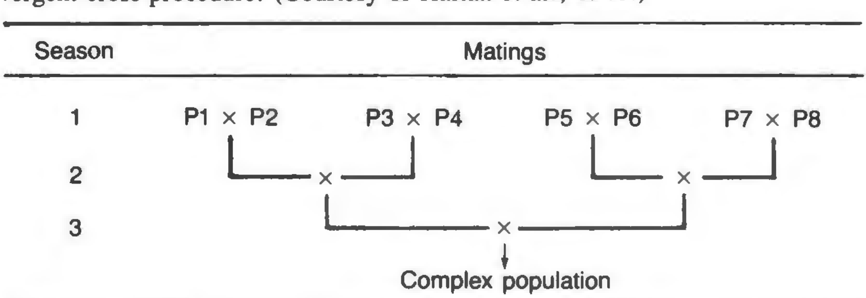
The genetic basis for the minimum number of generations of intermating required can be illustrated with alleles at four loci (A through D) for eight parents (1 through 8). The allele at a locus will be identified by the number of the parent from which it was obtained. For example, A1 is the A locus with an allele from parent 1. The mating arrangement used to illustrate the principle will be the convergent cross (Fig. 12-2).

Season 1: The matings involved and the genotype of the hybrids produced in the first generation of intermating would be

Parent 1 × parent 2 – hybrid 1–2 = A1A2B1B2C1C2D1D2
Parent 3 × parent 4 – hybrid 3–4 = A3A4B3B4C3C4D3D4
Parent 5 × parent 6 – hybrid 5–6 = A5A6B5B6C5C6D5D6
Parent 7 × parent 8 – hybrid 7–8 = A7A8B7B8C7C8D7D8

Season 2: In the second generation of intermating, the cross of hybrid 1–2 × hybrid 3–4 can produce an array of genotypes, one example of which is A1A3B2B4C1C3D2D4. The mating of hybrid 5–6 × hybrid 7–8 can produce genotypes such as A5A7B6B8C5C7D6D8. By the end of the second generation of intermating in season 2, an individual can possess alleles from four of the eight parents of the final population.

Figure 12-2 Complex population developed from eight parents by the convergent-cross procedure. (Courtesy of Harlan et al., 1940.)



Season 3: In the third generation of intermating, the cross of hybrid 1–2–3–4 × hybrid 5–6–7–8 can produce genotypes with alleles from all parents, such as A1A5B2B8C3C7D4D6.

Number of Parents Involved

The number of different alleles possible in a population theoretically increases with each additional parent used to form a population. An increase in the number of parents, therefore, enhances the possibility of increased genetic variance in the population. It often is difficult, however, to find a large number of parents that have an acceptable level of performance for the characters under selection. Out of a group of 40 genotypes available as parents, 10 may have superior performance for a character, 10 may be average, 10 may be below average, and 10 may be inferior. The breeder must decide if it is better to use the 10 superior parents to obtain a final population with a potential for high mean performance and limited genetic variability, or use more than 10 parents to increase potential genetic variability in the population with a sacrifice in its mean performance.

The number of parents used to form a population can influence the procedures available to carry out the required matings. The convergent-cross procedure can only be used with a multiple of 2^n parents: 4, 8, 16, 32, and so forth (Fig. 12-2). The choice between a complete or partial diallel design mating will be influenced by the number of parents involved relative to the resources available for hybridization. If resources were available to make 36 matings, the number of parents could not exceed six if a diallel design with reciprocal crosses and self-pollinations were preferred, could not exceed nine for a diallel design without reciprocals and selfs, and could not exceed 72 if each parent were crossed to one another in a partial diallel design.

Genetic Contribution of Each Parent

The potential advantage of mating genetically diverse parents is that each may contribute unique alleles, which when combined together may result in a superior individual. The theoretical advantage has seldom been realized for short-term improvement of cultivars for quantitative characters, such as seed yield. Populations in which highly productive cultivars contribute half the germplasm, and less productive plant introductions the other half, seldom produce a segregate that is superior to the most productive cultivar used as a parent. Breeders who are attempting to increase genetic diversity in breeding populations are faced with the choice between genetic variability and population performance. A population with broad genetic variability is of little value if the best segregates are inferior in performance to other available populations.

Some breeders choose to develop breeding populations in which certain parents contribute less than 50 percent of the germplasm. To illustrate alternative mating designs that are used, highly productive parents will be designated by the letter A and less productive plant introductions with the letters PI (Fig. 12-3).

One alternative for varying the percentage of plant introductions in a population is by the relative frequency of A and PI parents used. For a convergent cross with eight parents, the percentage of PI parentage in the final population could be varied by the fraction of PI parents used: 1 PI/7A, 2 PI/6A, 3 PI/5A, 4 PI/4A, and so forth. A disadvantage of this procedure is that a low number of different PI parents will be involved when the desired percentage of PI germplasm is low. For example, a 2 PI:6A ratio has 25 percent PI germplasm, which is contributed by only two parents.

Some form of backcrossing can be used to include a large number of PI

Figure 12-3 Two procedures for complex population formation that permit the use of multiple plant introductions (PI) as parents with a limited percentage of the germplasm of each in the final population.

Season	% A germplasm	Matings					
1	50	A1 × PI1	A2 × PI2	A3 × PI3	A4 × PI4	A5 × PI5	A6 × PI6
		↓	↓	↓	↓	↓	↓
2	75	A1 ×	A2 ×	A3 ×	A4 ×	A5 ×	A6 ×
		↓	↓	↓	↓	↓	↓
3	88	A1 ×	A2 ×	A3 ×	A4 ×	A5 ×	A6 ×
		↓	↓	↓	↓	↓	↓
4	94	A1 ×	A2 ×	A3 ×	A4 ×	A5 ×	A6 ×
5	94	Diallel of the six backcross populations					
6	94	Random mating of the hybrids from season 5					
7	94	Random mating is continued for as many generations as desired					

Season	% A germplasm	Matings					
1	50	A1 × PI1	A2 × PI2	A3 × PI3	A4 × PI4	A5 × PI5	A6 × PI6
		↓	↓	↓	↓	↓	↓
2	75	A2 ×	A3 ×	A4 ×	A5 ×	A6 ×	A1 ×
		↓	↓	↓	↓	↓	↓
3	88	A3 ×	A4 ×	A5 ×	A6 ×	A1 ×	A2 ×
		↓	↓	↓	↓	↓	↓
4	94	A4 ×	A5 ×	A6 ×	A1 ×	A2 ×	A3 ×
5	94	Diallel of the six populations					
6	94	Random mating of the hybrids from season 5					
7	94	Random mating is continued for as many generations as desired					

parents and still minimize the percentage of PI germplasm in the final population. Two of the possible procedures are illustrated in Fig. 12-3. One procedure is to mate each A parent to a PI parent, backcross to the A parents until the desired level of A parentage is achieved, then intermate the backcross populations to form a single complex population. In the second procedure, the A parents are rotated for each backcross until the desired level of A parentage is achieved, then the backcross populations are intermated to form a single population. The advantage of the second procedure is that there can be recombination of genes from the different parents during backcrossing. In the first procedure, recombination only occurs between the A and PI parent in each separate backcross program.

Amount of Time Available

The amount of genetic improvement per year in a breeding program is strongly affected by the length of time required for completion of a cycle of selection (Chap. 17). This applies to direct selection of cultivars from a population or some type of recurrent selection program. One important variable in the number of years per cycle is the length of time required to form a new population. Breeders may prefer to conduct three or more generations of intermating before selection begins, but this may add 1 or more years to the length of a cycle. The breeder must decide if more genetic improvement per year will be achieved by providing additional opportunity for recombination before selection or by conducting more cycles of selection per unit time in populations formed with minimal intercrossing.

At the present time, most breeders prefer to conduct more cycles of selection than to spend time on additional generations of intercrossing, particularly when the parents are heterozygous. It is common in recurrent selection programs involving heterozygous plants or lines to form a population by crossing parents together in a series of single-cross matings, to bulk seed from each single cross to form one population, and to initiate selection without any additional generations of intermating. As a result, the members of the new population have alleles from only two of the parents, regardless of the number of parents used to form the population. Such a population is complex in the sense that alleles from many parents are present, but it is not complex with respect to the opportunity for recombination of alleles from different parents.

PROCEDURES USED TO FORM COMPLEX POPULATIONS

Breeders can use a number of procedures to form complex populations. The following discussion will be limited to procedures reported in the literature and those commonly used by plant breeders.

Convergent Cross

The alleles obtained by an individual in a population from a set of parents is a function of the number of parents involved and the mating arrangement used to form the population. One mating procedure that provides an equal probability for alleles from each parent to be present in an individual was proposed by Harlan and colleagues (1940) as a "systematic series of bridging crosses," later referred to as a convergent cross (Fig. 12-2).

The convergent cross provides the opportunity for alleles of each parent to be present in segregates of the final population. It is seldom used, however, because of several disadvantages: (a) The number of parents is restricted to a multiple of 2^n: 4, 8, 16, 32, 64. (b) Recombination of genes from some parents is not possible until the final population is formed. For example, consider the procedure for eight parents illustrated in Fig. 12-2. Genes from parent 1 have no opportunity to recombine with those from parents 7 and 8 until the third season of intermating. (c) The number of seasons required to form the population often exceeds the length of time a breeder is willing to spend on population formation.

Diallel Mating Design with Reciprocal Crosses and Self-Pollinations

A diallel design with reciprocal crosses and self-pollinations is required to achieve Hardy–Weinberg equilibrium in a population (Chap. 3). Use of the procedure in applied breeding programs is limited to natural hybridization in certain open-pollinated populations. One example would be the mating of selected individuals in a program of recurrent phenotypic selection. Parents (plants) are selected from among male-fertile individuals before pollination, and selected parents are subjected to both self- and cross-pollination. A diallel design with reciprocals and selfs is not possible when self-incompatibility or male sterility reduces or eliminates the probability of self-pollination, or when male sterility prevents the possibility of reciprocal crosses.

The advantages of a diallel design mating with reciprocal crosses and self-pollinations are (a) the number of parents in the population is not restricted, (b) each parent has the opportunity to mate and recombine with every other parent, and (c) Hardy–Weinberg equilibrium can be achieved in the populations. However, the procedure is not considered practical for applied breeding programs that utilize artificial hybridization for several reasons. (a) Self-pollinations do not permit recombination of genes between parents. (b) Crosses in only one direction are as effective for recombination as reciprocal crosses between two parents. (c) An extensive number of matings are required (p^2), which limits the number of parents (p) that can be considered.

Diallel Mating Design without Reciprocal Crosses and Self-Pollinations

A common procedure for mating a limited number of parents to form a complex population is to use a diallel design without reciprocal crosses and self-pollinations. This type of diallel mating design provides the opportunity for genes of each parent to recombine with those of every other parent. If sufficient generations of intermating occur, individual members of a population will have the opportunity to obtain alleles from every parent. Without reciprocal crosses and self-pollinations, Hardy–Weinberg equilibrium will be approached but never achieved. This is not considered a problem for cultivar development programs.

The advantages of a diallel mating design without reciprocal crosses and self-pollinations are (a) the number of parents used to form the population is not restricted, (b) each parent has the opportunity to mate and recombine with every other parent, (c) the method applies to open-pollinated populations that involve self-incompatibility or genetic male sterility, and (d) the method involves fewer matings than necessitated by a diallel design with reciprocal crosses and self-pollinations. The primary disadvantage is that the large number of matings required, $p(p - 1)/2$, often limits the number of parents that can be included for artificial hybridization.

Partial Diallel Mating Design

The mating of each parent to some, but not all, of the other parents in a partial diallel design is commonly used in the formation of a complex population. It is used with artificial hybridization whenever the number of parents is too great to accomplish a complete diallel. A partial diallel is operative in an open-pollinated population whenever the number of seeds produced by a parent is less than the number of parents with which it must mate to achieve a diallel.

The advantages of a partial diallel design are (a) the number of parents in the population is not restricted, (b) it can be used with open-pollinated populations that involve self-incompatibility or male sterility, and (c) it involves fewer matings than necessitated by a diallel. The primary disadvantage of a partial diallel is that each parent does not have the opportunity to mate and recombine with every other parent.

Combination of the Diallel and Partial Diallel Mating Designs

Both the diallel and partial diallel designs are commonly used in the same program to form a complex population. The matings may be accomplished by artificial hybridization, open pollination, or a combination of the two. A combination of procedures is used to permit the greatest amount of recombination possible within the resources available for hybridization. Many combinations of the diallel and partial diallel mating designs are possible. A few of the more common ones are the following.

1. *Procedure 1:*

Season 1: A diallel without reciprocal crosses and self-pollinations is made to form $p(p - 1)/2$ single-cross populations.
Season 2: A partial diallel is used to intercross the single-cross populations formed in season 1.
Season 3: Hybrid seeds from season 2 are bulked and planted as one population. A partial diallel consisting of plant-to-plant crosses is used for the third generation of intermating.

2. *Procedure 2:*

Season 1: The number of parents is too large to permit a diallel; therefore, each parent is mated to one other in a partial diallel.
Season 2: There are $p/2$ single-cross populations from season 1. The number is small enough to permit a diallel mating of the populations without reciprocals and self-pollinations in season 2.
Season 3: A bulk of hybrid seed from season 2 is planted. A partial diallel consisting of plant-to-plant crosses is used for the third generation of intermating.

3. *Procedure 3:*

Season 1: A partial diallel is used to mate a large number of parents.
Season 2: Hybrid seed from season 1 is planted as a bulk in isolation. Natural hybridization is used for the second generation of intermating.
Season 3+: All subsequent generations of intermating are conducted in the same manner as described for season 2.

PLANTING ARRANGEMENTS FOR POPULATION FORMATION BY ARTIFICIAL HYBRIDIZATION

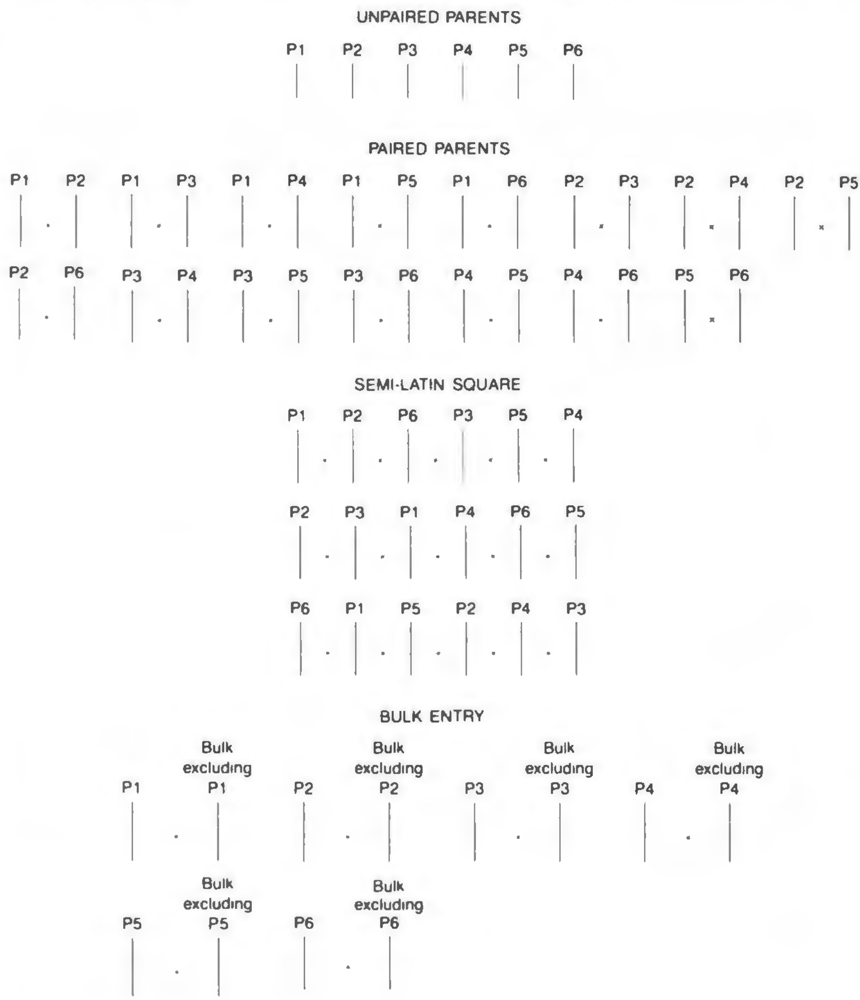
After the parents have been chosen and the mating procedure established, the breeder must select an appropriate planting arrangement when artificial hybridization is conducted in the field. Each arrangement has advantages and disadvantages, so the breeder must select the one that will be most efficient for the circumstances encountered and the resources available.

Diallel Matings

Alternative planting arrangements for artificial hybridization can best be described by first considering their use in making a diallel design of six parents without reciprocal crosses and self-pollinations. The four arrangements to be considered are unpaired parents, paired parents, a semi–latin square, and a bulk-parent method.

Unpaired Parents. A common planting arrangement for any type of mating program is the use of unpaired parents (Fig. 12-4). Each parent is planted once, regardless of the number of parents with which it will be crossed. The parents may be grown in a special area designated for crossing or may be located in different parts of a field.

Figure 12-4 Four planting arrangements that can be used for a diallel mating design of six parents without reciprocal crosses and self-pollinations.



The advantages of the unpaired-parents method are that (a) it requires the least space and parent seed of the four arrangements because there is only one row of each parent for each planting date, (b) the decision on which parents to mate can be made after planting, (c) the parents of each mating are known, and (d) it can be used with any number of parents.

One disadvantage of unpaired parents is that more time is required per mating during hybridization than for any of the other three arrangements. The availability of suitable female flowers and pollen must be determined for each pair of parents by walking from one area of the field to another. Pollen must be transported between parents. The male parent must be identified for each cross made. An inventory of number of crosses made and success of a particular mating can be time-consuming. A second disadvantage is that there is a greater chance that the wrong mating will be made accidentally, particularly by inexperienced persons.

Paired Parents. Some of the disadvantages of the unpaired-parents method can be overcome by planting adjacent to each other the pairs of parents to be mated (Fig. 12-4). When the paired parent arrangement is used, breeders employ various techniques to facilitate movement through the field during hybridization and to prevent persons from getting in the wrong place and making an incorrect mating.

1. The spacing between rows of two parents to be mated can be different than the spacing between adjacent rows of other parents. For example, the distance between rows as marked by a commercial planter may be 0.7 m. The breeder may plant parents to be mated 0.7 m apart, and skip a row on either side of the pair to provide a 1.4-m space.

P1		P2		P1		P3		P1		P4
	x				x				x	
	0.7 m		1.4 m		0.7 m		1.4 m		0.7 m	

Conversely, the breeder may choose to mate the parents on either side of the 1.4-m space, where it is easier to walk.

P1		P2		P1		P3		P1		P4
	x				x				x	
	1.4 m		0.7 m		1.4 m		0.7 m		1.4 m	

2. A string may be hung between pairs of rows that are to be mated. The string serves as a barrier to prevent persons from mating the wrong pairs of rows.
3. Stakes of the same color may be placed at the beginning of rows to be mated. Colors are alternated across the field to differentiate pairs of parents to be mated.

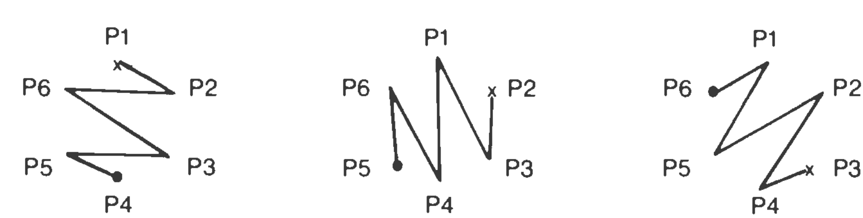
The advantages of the paired-parent arrangement are that (a) less time is required per mating during hybridization than for unpaired parents, (b) there is less chance that the wrong mating will be made than with unpaired parents, (c) the parents of each mating are known, and (d) the method can be used with any number of parents. The disadvantages are that (a) it requires the most space and parent seed of the four arrangements because the number of rows required for planting is twice the number of matings to be made and (b) the decision on parents to be mated must be made before planting.

Semi–latin Square. The disadvantage of the paired-parent arrangement for making a diallel is the large amount of space and parent seed required. To reduce the space and parent seed requirement, Fehr and Ortiz (1975) described the use of the semi–latin square to make a diallel when an even number of parents was involved. The arrangement also could be called half of a symmetric latin square or a partial latin square.

The systematic planting order for the semi–latin square is determined by the procedure illustrated in Fig. 12-5.

Figure 12-5 Arrangement of parents for a diallel design of six parents using the semi–latin square arrangement. The number of columns (mating units) utilized is equal to half the number of parents in the diallel.

P1	P2	P3
X	X	X
P2	P3	P4
X	X	X
P6	P1	P2
X	X	X
P3	P4	P5
X	X	X
P5	P6	P1
X	X	X
P4	P5	P6
Order of first column (mating unit)	Order of second column (mating unit)	Order of third column (mating unit)



x = first parent in the column.
• = last parent in the column.

1. A parent is assigned to the first position in half the number of the columns normally used in a latin square. It makes no difference which half of the parents are used to begin the columns or to which column they are assigned.
2. The order of parents in each column is determined by writing the numbers of the parents in consecutive order in a circle, one circle for each column. For a given column in the latin square, a line is drawn among the numbers in a circle beginning with the first parent in the column and ending with the last. The line proceeds from the parent heading the column to the first parent in the clockwise direction, to the first parent in the counterclockwise direction, to the second parent in the clockwise direction, to the second parent in the counterclockwise direction, to the third parent in the clockwise direction, to the third parent in the counterclockwise direction, and so on until all of the parents have been assigned a position in the column. The order of parents in the line is recorded in the appropriate column of the semi–latin square.

For planting and hybridization, each column is an independent mating unit. The mating units can be planted in separate fields if necessary without any problem. At the time of hybridization, adjacent parents in a mating unit are crossed to each other in any direction preferred. The parent at the head of the mating unit and the one at the end have only one parent adjacent to them; therefore, they are involved in only one mating. Those two parents are crossed in one of the other mating units. All other parents in a mating unit have an adjacent parent on either side and are involved in two matings.

The advantages of the semi–latin square arrangement are (a) less time is required per mating during hybridization than for unpaired parents, (b) there is less chance that the wrong mating will be made than with unpaired parents, (c) less space and parent seed are required than for paired parents, and (d) the parents of each mating are known. The disadvantages of the semi–latin square are (a) it requires more space and parent seed than unpaired-parent or bulk-parent arrangements, (b) the decision on parents to be mated must be made before planting, and (c) it can only be used with an even number of parents.

Bulk Parent. The bulk-parent arrangement can be used to form a diallel when knowledge of the parentage and frequency of each single-cross mating is not important. The arrangement also has been called the Irish and bulk-entry method (Stuber, 1980).

A row of each parent is planted adjacent to a row that is a bulk of all other parents in the diallel (Fig. 12-4). For example, if 6 parents are in the diallel, parent 1 is grown adjacent to a bulk of parents 2 to 6. All plants in the bulk row are mated to parent 1. Another row contains parent 2, adjacent to which is a bulk of parents 1 and 3 to 6. All plants in the bulk row are mated to parent 2. The plants in the bulk row are not identified; therefore, only one parent of each mating is known.

The techniques used by breeders to ensure that the correct pairs of rows are mated, as discussed for paired parents, apply also to the bulk-parent arrangement. In addition, the row of each pair that is the single parent must be differentiated from the bulk row because each plant of the bulk row must be used for crossing, if possible.

Although both parents of a mating are not known with the bulk-parent arrangement and some single-cross matings could be accidentally excluded, the arrangement provides two opportunities for each mating to occur. For example, if 20 parents are in the diallel, a row of parent 1 would be mated with a bulk containing parent 20, and a row of parent 20 would be mated with a bulk containing parent 1.

The bulk-parent arrangement used for artificial hybridization is similar in principle to the polycross method used to intermate parents by natural hybridization. In both methods, only one of the parents of a mating is known. The population, formed by bulking similar quantities of hybrid seed from each of the known parents, is assured of containing genes from every parent, but not necessarily in equal frequency.

The advantages of the bulk-parent arrangement are (a) less space and parent seed are required than for the paired-parent and semi–latin square arrangements, (b) less time is required per mating during hybridization and there is less chance of making the wrong mating than with unpaired parents, and (c) it can be used with any number of parents. The disadvantages are (a) only one parent of the hybrid seed is known, (b) the decision on parents to be mated must be made before planting, and (c) it requires more space and parent seed than unpaired parents.

Partial Diallel Design and Less Systematic Matings

Unpaired and Paired Parents. The unpaired- and paired-parent arrangements can be used to make a partial diallel with any number of matings per parent. They also can be used for any other type of mating that is desired. The advantages and disadvantages of the two planting arrangements described for the diallel also apply to their use for any other type of mating.

Circular Cross. A circular-cross arrangement can be used to form a partial diallel in which each parent is mated to two others. The procedure has also been referred to as the chain cross (Stuber, 1980). Adjacent parents are mated to each other, and the first parent in the sequence is mated to the last one. With the use of multiple circles, the number of single-cross matings for each parent can increase by multiples of two. For example, the two following arrangements each would provide two matings for each of the six parents.

×
P1 × P2 × P3 × P4 × P5 × P6

×
or P1 × P3 × P5 × P2 × P6 × P4

The advantages of the circular-cross arrangement are that (a) less time is required per mating during hybridization than for unpaired parents, (b) there is less chance that the wrong mating will be made than with unpaired parents, (c) the parents of each mating are known, (d) the method can be used with any number of parents, and (e) it requires less space and parent seed than for paired parents. The disadvantages are (a) it requires more space and parent seed than the unpaired-parents method and (b) each circle can only be used to mate each parent to two others. Additional circles would have to be used to increase the number of matings with each parent in the partial diallel.

POLYCROSS PROCEDURE

Population formation may include one or more generations of natural hybridization in species with appropriate mechanisms for open-pollination. The polycross is a widely used procedure for intercrossing parents by natural hybridization.

The polycross is a method for intercrossing parents of vegetatively propagated species with mechanisms that prevent or minimize self-pollination. Self-incompatibility in bromegrass minimizes the frequency of self-pollination. In alfalfa, a membrane over the stigma prevents self-pollination. The membrane is broken to permit cross-pollination when the flower is tripped by an appropriate insect.

The objectives of a polycross procedure are to intercross the parents as equally as possible and obtain a similar genetic contribution from each parent in the population that is formed. Several principles related to natural intercrossing must be considered.

1. Parents must be flowering at the same time for effective crossing to occur. This may restrict the range in maturity of parents that can be used in the polycross.
2. Parents adjacent to each other are most likely to be intercrossed. Replication and randomization of parents provide a reasonable assurance of random mating.
3. The method of sampling hybrid seed from the parents may influence the genetic contribution of each to the population.

Parent Arrangements

Two experimental designs normally associated with statistical analyses are used for polycrosses, the latin square and the randomized complete-block. The designs are used to arrange parents in a manner that will maximize intercrossing by placing the parents adjacent to each other as frequently as possible.

A latin square is a design in which each parent (entry) must occur in each row and column of plots in an experiment (Fig. 12-6). As a result, the number of replications of each parent is equal to the number of parents, and the number of plots is the square of the number of parents. There are four replications with four parents (16 plots), eight replications with eight parents (64 plots), and 16 replications with 16 parents (256 plots). A breeder can use two or more latin squares for a set of parents to increase replication. Four latin squares of four parents would provide 16 replications, and two latin squares of seven parents would result in 14 replications. Randomization of entries in a latin square is ensured by randomizing the order of rows, columns, or both. The advantage of the latin square arrangement compared with the randomized complete-block is that each parent occurs adjacent to every other parent somewhere in the latin square. Its disadvantage is that for a large number of parents, the number of replications required may be greater than desired.

The randomized complete-block is a design in which each parent is randomly assigned to a plot within each replication (Fig. 12-6). The advantage of the randomized complete-block design compared with the latin square is that the number of replications can be any number desired, which is particularly important for a large number of parents. Its disadvantage is that the proximity of parents to each other cannot be ensured. The difference in proximity of parents in the two designs is illustrated in Fig. 12-6.

A polycross is sometimes used to describe an unreplicated planting of parents that are open-pollinated. A group of plants in a field may be evaluated for a character and the unacceptable ones discarded. The selected ones are allowed to open-pollinate to form the new population. The advantages of an unreplicated

Figure 12-6 Latin square and randomized complete-block designs for a polycross of five parents with a total of 10 replications.

Latin square 1

P1	P2	P5	P3	P4
P3	P4	P2	P5	P1
P5	P1	P4	P2	P3
P2	P3	P1	P4	P5
P4	P5	P3	P1	P2

Latin square 2

P1	P2	P5	P4	P3
P2	P3	P1	P5	P4
P4	P5	P3	P2	P1
P5	P1	P4	P3	P2
P3	P4	P2	P1	P5

Randomized complete-block

Replication									
1	2	3	4	5	6	7	8	9	10
P5	P1	P5	P4	P2	P4	P1	P5	P2	P5
P1	P2	P1	P1	P1	P1	P4	P3	P1	P4
P3	P5	P4	P3	P5	P5	P5	P2	P3	P2
P2	P3	P2	P2	P4	P2	P3	P4	P5	P3
P4	P4	P3	P5	P3	P3	P2	P1	P4	P1

arrangement are (a) hybridization may be accomplished during the same season that selection is practiced, which reduces the number of years for population formation, and (b) much less labor is involved than with a latin square or randomized complete-block design. The disadvantage is that the mating of parents can be quite unequal, because the parents in closest proximity to each other will cross more frequently than those spaced farther apart.

Bulking Hybrid Seed

The genetic contribution of each parent to the population obtained from a polycross can be influenced by the procedure used to harvest and bulk seed from the parents. Three procedures can be used, each with inherent advantages and disadvantages.

1. The seed from all parents may be bulked without regard to the amount of seed produced by each parent. The advantage of this procedure is that it requires less labor than the following two procedures. The disadvantage is that the genetic contribution of each parent to the population may be markedly different if the amount of seed produced by each parent is different.
2. The seed from each plot (replication) of each parent may be harvested separately. The seed from plots of each parent is bulked, regardless of the amount of seed per plot. An equal amount of seed from the bulk of each parent is mixed together to form a single population. The advantage is that the genetic contribution of each parent to the final population is controlled better than with the first procedure, because the number of seeds from each female parent is the same. The disadvantages are that this procedure requires more labor than the first procedure and does not control the genetic contribution of the male parent as effectively as the third procedure.
3. The seed from each plot (replication) of each parent is harvested separately. An equal quantity of seed from each plot of a parent is bulked. An equal amount of seed from the bulk of each parent is mixed together to form a single population. The advantage is that the genetic contribution of each parent to the final population is controlled better than for the other two procedures. In each replication, the parents most adjacent to each other will cross most frequently. Sampling of an equal quantity of seed from each replication of a parent means that the genetic contribution of adjacent male parents, which differ in each replication, is similar. The disadvantage is that it requires more labor than the other two procedures.

REFERENCES

Bravo, J. A., W. R. Fehr, and S. R. Cianzio. 1981. Use of small-seeded soybean parents for the improvement of large-seeded cultivars. *Crop Sci.* 21:430–432.

Fehr, W. R., and L. B. Ortiz. 1975. Recurrent selection for yield in soybeans. *Univ. Puerto Rico J. Agri.* 59:222–232.

Harlan, H. V., M. L. Martini, and H. Stevens. 1940. A study of methods in barley breeding. *USDA Technical Bulletin 720.*

Stuber, C. W. 1980. Mating designs, field nursery layouts, and breeding records. pp. 83–104. *In* W. R. Fehr and H. H. Hadley (eds.), *Hybridization of crop plants.* American Society of Agronomy, Madison, Wis.