Pedigree and Bulk method

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Abstract

Bulk and pedigree methods are frequently used in handling segregating populations in self-pollinated crops. The proportion of genotypes segregating in the population decrease by 50% every selfing generation. With the pedigree technique, line derivations begin at F_2 generation and accurate ancestry records are maintained. In the bulk technique, lines are derived in the F_3 or F_4 generation. Both methods evaluate for low heritable traits in well-designed replicated experiments in the later subsequent generations. The bulk approach is the most effective way to breed polygenic traits. Enhancing qualitative characteristics is possible with the pedigree technique. The method is used for correcting a particular defect in a preferred cultivar.

Key words: Generation advancement, homozygosity, homogeneous, line superiority

12.0 Introduction

Generation of variability and exploitation of the variation are the two major stages of conventional plant breeding. Pedigree and bulk methods are commonly used crop improvement techniques used in handling the variability released following hybridization.

Both methods were first used in the 1900s. For the bulk method, crosses were made between diverse parents, while carefully chosen parents were made to establish precise character combinations in new cultivars in the pedigree method. In the method, superior genotypes are chosen from segregating generations every generation while maintaining accurate records of the ancestry of chosen plants each generation. Emphasis is placed on selecting within and among families in elite line crosses with the main objective of retaining desirable linkage blocks intact. However, the classical pedigree method is considered labour intensive, require elaborate record keeping, and require evaluation of many families in the early generation. As a results, different modifications of the pedigree method have been established. This includes, but not limited to Modified pedigree method, Mass pedigree method, Bulk pedigree selection, Shuttle breeding system, and Backcross pedigree method

Nilsson-Ehle is considered the first to have implemented the bulk method in the improvement of yield and winter hardiness in wheat in Sweden. As part of his method, Nilsson-Ehle eliminated F_2 individual plants that had been damaged by winter or those that were prone to lodging or other defects and harvested seeds from the desirable plants in bulk. Up until the F_5 or F_6 generation, single-plant selection where continued, and the proceeds were harvested as a bulk. He reasoned that, a high percentage of suitable plants would likely increase among the winter-hardy and other desirable selections, if vast populations were grown and inferior plants were eliminated in the earliest generations. Bulk technique in its own merit allows a breeding program to handle as many segregating populations as feasible because each generation is carried over and harvested in bulk from generation to generation. Several different modifications of the bulk method have also been proposed, and some are currently being implemented in breeding programs. The techniques differ, in respect to the strictness of selection practiced in the early segregating generations. The methods share a common feature in that selection among family lines is delayed until homozygosity is high.

The genetic principle is the same for both bulk and pedigree breeding. In each generation of advancement, populations from both methods experiences a 50% increase in homozygosity and a 50% decrease in heterozygosity for the loci where the parent differs. For traits controlled by many loci, effective selection among lines are done at later generations. For example, for a trait controlled by 100 loci, 46% of the individuals at F_8 generation are expected to be fully homozygous at the loci (Table 1).

Table 1: proportion of homozygous genotypes at each selfing generation of advancement for different number of loci for which the parents differ

Loci	F_2	F_3	F_4	F_5	F_6	F_7	F_8	F_9	F_{10}	F_{11}	F_{12}	F_{13}	F_{14}	F_{15}
1	0.50	0.75	0.88	0.94	0.97	0.98	0.99	1.00	1.00	1.00	1.00	1.00	1.00	1.00
2	0.25	0.56	0.77	0.88	0.94	0.97	0.98	0.99	1.00	1.00	1.00	1.00	1.00	1.00
3	0.13	0.42	0.67	0.82	0.91	0.95	0.98	0.99	0.99	1.00	1.00	1.00	1.00	1.00
4	0.06	0.32	0.59	0.77	0.88	0.94	0.97	0.98	0.99	1.00	1.00	1.00	1.00	1.00
5	0.03	0.24	0.51	0.72	0.85	0.92	0.96	0.98	0.99	1.00	1.00	1.00	1.00	1.00
10		0.06	0.26	0.52	0.73	0.85	0.92	0.96	0.98	0.99	1.00	1.00	1.00	1.00
20			0.07	0.28	0.53	0.73	0.85	0.92	0.96	0.98	0.99	1.00	1.00	1.00
50				0.04	0.20	0.46	0.68	0.82	0.91	0.95	0.98	0.99	0.99	1.00
100					0.04	0.21	0.46	0.68	0.82	0.91	0.95	0.98	0.99	0.99
150					0.01	0.09	0.31	0.56	0.75	0.86	0.93	0.96	0.98	0.99
200						0.04	0.21	0.46	0.68	0.82	0.91	0.95	0.98	0.99
250						0.02	0.14	0.38	0.61	0.78	0.89	0.94	0.97	0.98
300						0.01	0.10	0.31	0.56	0.75	0.86	0.93	0.96	0.98

12.1.1 Parent selection

Choice of parents in a crossing block is considered the most important in a breeding program. Crosses between elite x elite, exotic x adapted, adapted x adapted, domesticated x wild relatives can be made depending on the breeding objectives. Importantly, the parents must have all the desirable genes necessary to be combined into the new variety. Either phenotypic performance, genetic diversity, or general adaptability are used in assessing for the parents' value. If more than one parent possesses the desired qualities, multiple crossing can be performed to incorporate all of these parents into the hybridization process. Combining the characters from both parents require knowledge about number of genes, their dominance, and epistatic relations, linkage, pleiotropy, effect of cytoplasm. Knowledge about the inheritance of the traits is essential in the choice of appropriate breeding method for isolating breeding lines having desirable genotype combinations in segregating generation. Most importantly, understanding the genetics of traits is crucial for determining whether one has a true F_1 hybrid or whether the plants came from seeds that were selfed.

Pedigree breeding is the most effective way to fix a specific defect in a well-known variety. The method works well for breeding for desirable qualitative traits such as disease resistance or attributes that can be easily categorized such as plant architecture or the color or shape of a plant part.

12.1.2 Pedigree records

The first selection opportunity is offered in F_2 generation. Here, maximum variability is released. However, some times, actual selection can be delayed until the F_3 or F_4 generation. In order to be able to trace parent-progeny relationships back to a specific F_2 plant in a later generation, each selection's pedigree is kept up to date using a numbering system. For instance, 27-311 is a possible designation for the selection number 311 from cross 27. An indication of 27-311-1 or 27-311-2 may be supplied if additional choices from the 27-311 family are made. The selected plants may also be designated based on location of progeny rows in the field in a given generation and in previous generation (Table 2).

Table 2: System of designating individual plant progenies by progeny row numbers of the concerned and the previous generations.

Generation	Progeny number	Description
F3	7911-7	Progeny in the 7th row in F_3 plot
F4	7911-7-4	Progeny in the 4th row in F_4 plot, selected from the progeny in 7th row of F_3 plot
F5	7911-7-4-14	Progeny in the 14th row in F_5 plot, selected from the progeny in 4th row of F_4 plot
F6	7911-7-4-14-3	Progeny in the 3rd row in F_6 plot, selected from the progeny in 14th row of F_5 plot

12.1.3 Generalized pedigree method protocol

Pedigree breeding methodology was first described by Love (1927) (Fig. 1).

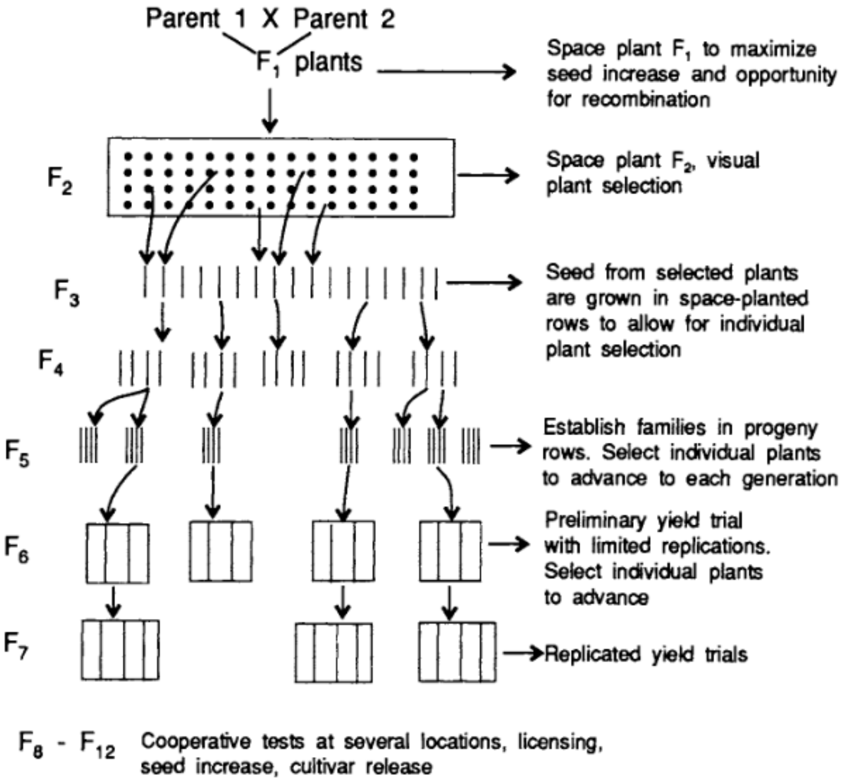


Fig. 1 Schematic presentation of pedigree method

Step-wise procedure for conducting pedigree method

In the first year, crosses between desirable parents are made, and the F_1 plants are raised to yield as much seed as is feasible. Depending on the location and the accessibility of off-season facilities, these two processes can take two years.

In the second year, depending on the seed quantity, and the available space, between 2,000 and 10,000 F_2 plants are grown in space-planted rows. Desirable plants are selected and harvested in separate packets.

In the third year, families of the particular plants that were selected in the F_2 generation are planted. For each family, 10 to 30 or more could be raised. The observations of the entire families are prioritised, and the most desirable plant is selected from each family. Some families might be completely discarded during the selection process, while in other families, more than one plant might be picked. Unless a particular cross appears to have substantial potential, the total number of selected plants shouldn't typically exceed the number of families established in the F_3 generation.

In the fourth year, by the F_5 generation, the plants will be sufficiently homozygous, enabling the commercial seeding of F_4 plant selections in the F_5 generation.

In the fifth and subsequent years, by the F_5 or F_6 generation, segregation at any given locus will have nearly reached completion, permitting yield testing. Replicated F_6 and F_7 yield trials are conducted to provide impartial benchmark for advancing better selections for additional testing. Final evaluations are carried out utilising precise on-site and regional field testing in accordance with procedures established by experience or with the objective of gathering data for licencing and recommendation reasons.

12.1.4 Concerns against pedigree breeding method

The pedigree system favours qualitative over quantitative disease resistance breeding, making it difficult to develop horizontal or broad-based disease resistance. Following the pedigree method, it is impossible to secure new sources of resistant genes quickly enough, and efforts to do so ran the possibility of depleting the accessible gene sources.

The need for resources to accommodate the space planting of several selections, record keeping, individual plant selection, and harvesting is a second issue with the pedigree system.

The third drawback of the pedigree system is the dependability of early-generation yield selection. A crucial aspect of the pedigree system is the capacity to select traits, including those that are quantitatively inherited, as early as the F_2 generation. Pedigree system breeding programmes cannot be justified by the time-consuming and laborious labour necessary if visual early-generation selection is inadequate. The majority of plant breeders agree that F_2 yield selection may not be effective for quantitatively inherited traits.

12.1.5 Modification of pedigree method

12.1.5.1 Modified pedigree method

The strategy takes into account the competitiveness of the selected plants to improve the dependability of the performance under experimentation and commercial spacing densities. The strategy was proposed in 1939 by Breakwell and I Fatten, and the procedures were later described by Kulshrestha (1989). From the F_2 generation to later generations, two programs are run concurrently. Samples of seeds from a population at F_2 generation and families at later generation are sown under high density and low density (conventional pedigree method). The performance of the populations or families under the high density trial would then help in decision on which populations and or families to focus on for single plant selection under the well-spaced conventional pedigree programme (Fig. 2).

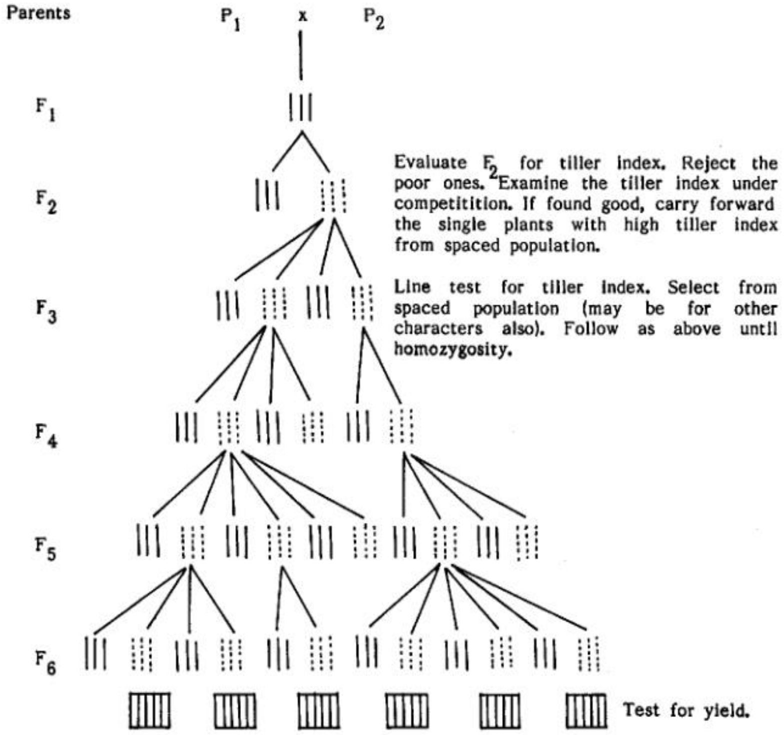


Fig. 2: Schematic representation of modified pedigree method of selection

12.1.5.2 Mass-pedigree method

The mass-pedigree method was described by Harrington (1937). Progeny rows are established using this strategy at any segregating generation where selection conditions were favourable the previous year. In unfavourable conditions, no selection is made and the population or progeny rows are harvested as a bulk. It was commonly accepted that individual plant progeny testing was conducted in F_4 after individual plant selection in F_3. However, progeny testing would be done in F_3 if significant selection was performed in F_2 due to favourable conditions. Progeny testing would be done in either F_5 or F_6 depending on whether the successful selection occurs in F_4 or F_5. To be employed as lines in F_6 or F_7, a number of progeny must be sufficiently homozygous by these generations.

The mass-pedigree approach costs less than the conventional pedigree strategy if the individual progeny testing are conducted in F_4 or F_5 as opposed to F_3. In F_2, the population is largely heterozygous, some of the desired genes are dominant or recessive, and some of the most important genes, those pertaining to quality and yield difference, often do not fully express themselves. In light of the aforementioned, it is recommended that a significant number of plants be chosen if the conditions are favourable at F_2 generation for selection in order to preserve the desired genotypes or heterozygotes that can reproduce the desired genotypes in the future generation.

But in contrast to F_2, a high proportion of the population in F_3 is homozygous, increasing the population's likelihood of having the desired genotype. As a result, at the F_3 generation, a stricter selection intensity can be employed and fewer progeny rows can be grown at the F_4 generation. If the choice to do individual progeny tests is taken after F_4, the circumstances change once more. Significantly more individuals now have homozygosity. Selecting and progeny testing consequently require less labour than F_3, where the crucial selection occurs.

12.1.5.3 Bulk-pedigree selection

The method is a modification of the mass pedigree method described by Harrington (1937). This was first implemented by Weiss (1949) in the breeding of soyabean, the methodology combines both the merits of pedigree and bulk method. In this method, bulk method is employed up to F_3 or F_4 generation, then selections are made and these selections are classified according to maturity groups. The progeny rows, selection within and among progeny rows are conducted at every generations of advancement.

12.1.5.4 Shuttle breeding system

The method is similar to bulk pedigree methods. The method is being used by CIMMYT to breed wheat for drought tolerance for semi-arid regions. The notion was initially implemented by Borlaug and colleagues at CIMMYT for wheat breeding. Borlaug and colleagues develop new wheat lines that were suited to both dry conditions (Ciudad Obrego) and humid highland environments; the breeding lines were evaluated successively in two contrasting environments in the 1940s (Toluca). This made it possible for the breeding programme to produce two generations of wheat each year and essentially cut the time required to develop a wheat cultivar in half (from 10 to 12 years to five to six years). In addition, the contrasting conditions allowed selection for a broad range of diseases and other types of stresses and also eliminate plants that were sensitive to day length (photoperiod).

In the shuttle breeding, segregating populations at F_2, F_5 and F_6 generations are "shuttled" for testing under optimum conditions of fertility and moisture and pedigree is followed based on good tillering, disease resistance, proper ear development, and grain plumpness and high leaf retention capacity. F_3 and F_4 generations are grown under reduced moisture and low fertility conditions and bulk method is followed in both the generations. The most promising lines performing well under optimum as well as under stress environmental conditions are selected and evaluated in international trials for drought resistance, yield and yield stability.

12.1.5.5 Backcross-pedigree method

The technique combines the advantages of the backcross and pedigree procedures. When using the pedigree technique, it is possible for the best lines in the various segregation generations to lack some of the desired traits of the superior parent used in the cross. To obtain more of the desired alleles from the better parent, restricted backcrossing, i.e. second- or third-cycle hybridization of the best plants in a segregating generation by the parents may be implemented. The additional hybridization cycles may also correct defects brought on by deleterious alleles from either parent.

12.7 Advantages of the pedigree method

The method gives breeders the opportunity to develop and refine their skills of selecting plants that have the most desired gene combinations to represent the new variety.

A substantial proportion of potentially inferior genotypes, particularly for undesired defects, are eliminated in the early generation before the start of expensive replicated yield evaluation experiments in the later generation.

Evaluation of family performance over a long period of time would be beneficial for selection. A reasonably accurate idea of the genetic potential of any family can be obtained from the consistent occurrence of any character in the progeny lineage through generations.

The pedigree record is utilised to bulk harvest of progenies with comparable yield potential but tracing to different F_2 plants. Despite having what appear to be uniform phenotypes, such bulk progenies almost definitely still contain substantial genetic variation and therefor do not represent traditional pure lines. Increased adaptability and longevity of the variety are two long-term benefits from such a technique.

Since the pedigree record facilitates the inclusion of lines descended from several F_2 plants in yield experiments, a broad, nay, complete spectrum of diversity of F_2 generation is recorded in homozygous form, especially up until the stage of commencing preliminary yield trials. This feature also prevents the complete elimination of some possible F_2 plants' lineages prior to entering yield trials, particularly when early yield selection is ineffective and line perpetuation is nothing more than a random process.

12.1.8 Disadvantages/Limitations of the pedigree method

The amount of breeding materials that can be handled with fixed resources is practically constrained by the fact that it is the most time- and resource-intensive method. A compromise between the number of crosses and the population size of each cross must typically be made, which does not bode well for the fundamentals of genetics.

It requires considerable data recording, which takes a lot of time and is challenging to manage over time. Breeders that rarely look back more than three years in the pedigree records are not uncommon.

The pedigree system's fundamental function is to artificially select for the best expression of economic traits, so the material can only be evaluated under ideal circumstances. Therefore, off-season nurseries are not advantageous in this method, and only one harvest each year is allowed.

The upper genetic ceiling for the potential of improved inbred lines developed utilising this technique is mainly established by the allelic composition of the foundation F_2 plants. Even the most experienced breeders cannot get eliminate off un-favourable genes that have become homozygous in the progeny being passed down. This means that the advantageous alleles at loci that unintentionally acquired homozygosity for unfavourable alleles throughout the inbreeding process cannot be recovered. However, are both experienced under bulk method as well, and can only be resolved by applying population improvement strategies.

12.1.9 Achievements of pedigree breeding method

Majority of the released cultivars of self-pollinating species have been developed through the pedigree method. For instance, in rice the following varieties were bred in India

i) Inter Varietal

- Co 37 Vaigai TN 1 × Co 29 - Short duration.
- Co 41 CuL 2410 × IR 22 - Short duration
- Co 43 Dasal × IR 20 - Medium duration.
- Co 44 ASD 5 × IR 20 - Medium duration, suitable for late planting.
- Co 45 Rathu Heenathi × IR 3403 - 207 - 1 - Medium duration, Resistant to blast, BLB and RTV.
- Ponmani (CR 1009) Pankaj × Jagannath - Long duration.

ii) Inter-Racial

- Japonica x indica cross: ADT 27 (Norin 10 × GEB 24)
- Mashuri (Taichung 65 × ME 80)

iii) Inter Specific Crosses

- Co 31: (O. perennis × GEB 24) Drought resistance.
- IR 34 Complex cross, one of the parent is *O. nivara*

12.2.0 Bulk method

The bulk method differs in basic terms from the pedigree method in the way it deals with the generations following the hybridization. Nillson-Ehle laid the first building block for this method. H. V. Haralan *et al.* (1940) laid the theoretical foundations for this method through their work on barley breeding. The seeds from selected plants from F_2 to F_5 sown as per commercial planting procedure in large plot. The plants are harvested in bulk at maturity during the generation advancement. No record of ancestry is kept. Selected plants at F_5 generation are harvested and grown as progeny rows (a head-row). Yield and quality potential are evaluated using the appropriate statistical tests in subsequent generations until the new variety is identified. Due to the extensively simplicity nature of this method, modification have been proposed to increase the likelihood of selecting favourable genotypes.

12.2.1 Selection of parents

The method is adopted when both parents are adapted to target population of environments and the targets traits are under polygenetic control.

12.2.2 General procedures for implementation of bulk method

F_1 generation: Many cross combinations are made among desirable parents to obtain F_1 seed generation. 50-100 F_1 plants of a cross is space planted and harvested in bulk. The number of F_1 plants should be large as much as possible. Markers, morphological and or molecular markers can be used for hybridity test.

F_2 to F_4 generation: 1,000 – 3,000 F_2 plants of a cross are grown and bulk-harvested. Representative sample of seed from the total harvest are sown for generation advancement to F_3 through F_4 generation. The population size is maintained through the generations. The F_2 to F_4 plots are planted in close spacing, and natural selection is allowed to occur reducing the frequency of un-adapted genotypes.

F_5 generation: 2000-5000 plants per population are space-planted for individual observation (Allard, 1960). The space planting allows for the full expression of genetic variation and effective selection.

F_6 generation: 300-500 F_5 superior individuals selected are sown as F_6 progeny rows. The progeny rows are observed for uniformity, yield potential, agronomic performance, disease resistance, quality trait etc. Undesirable plants from selected plants are rogued while undesired rows are completely discarded. Seeds from about 5 – 10 uniform progeny rows are harvested in bulk for preliminary yield testing.

F_7 generation: superior progeny rows from F_6 are sown in preliminary yield trail in large plots. Selection is based on the phenotype of the plant, grain character, disease reaction, etc.

F_8 t F_{10} generations: advanced multi-location trail is conducted for adaptability and yield performance assessment. Select the pest Line or group of similar lines are promoted for further testing for release as new variety. In each generation of yield test, appropriate design and appropriate relevant checks are included.

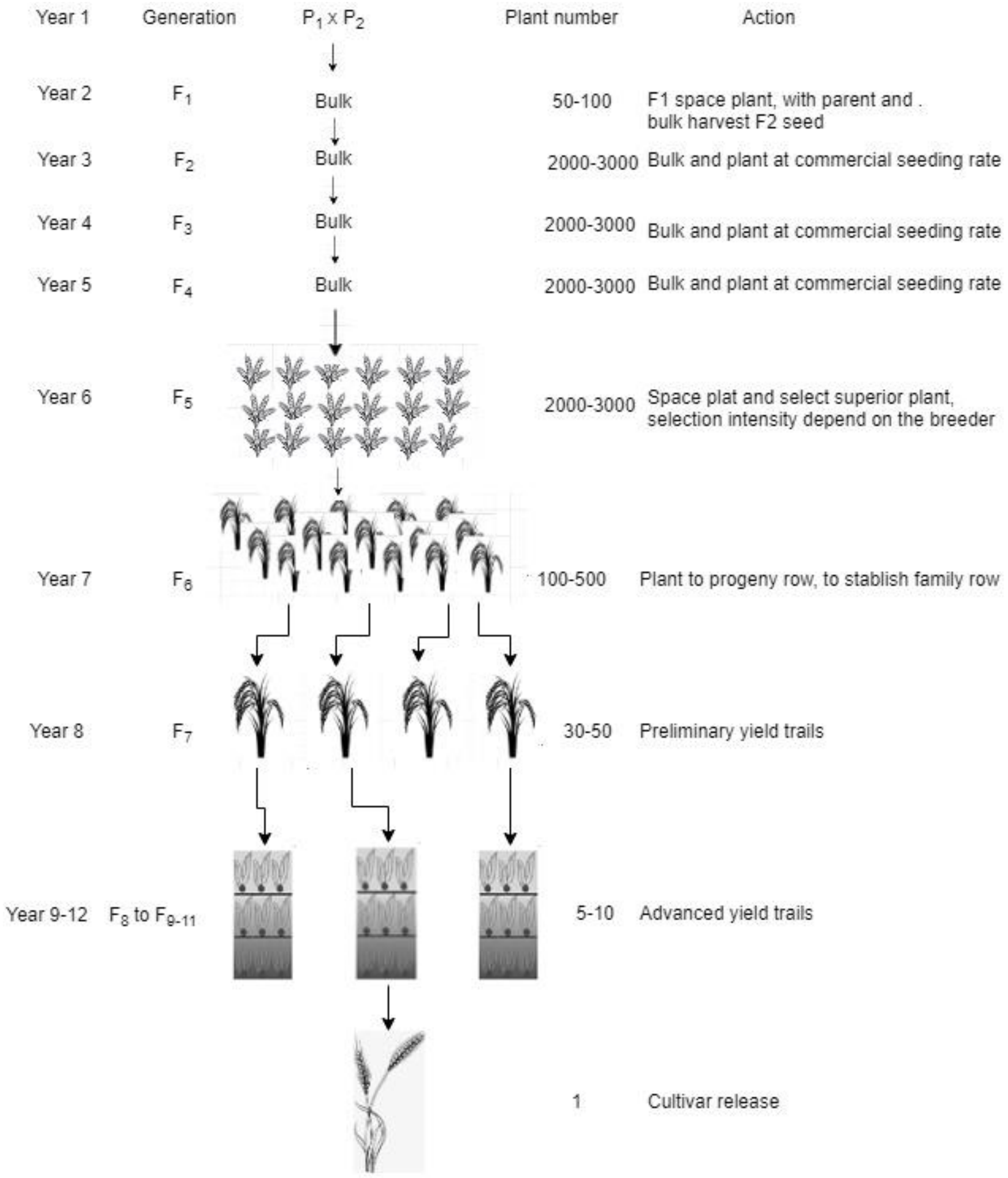


Fig. 4: Schematic representation of the bulk method for handling segregating populations.

12.2.3 Modified bulked method

Both theoretical, as well as practical modifications have been proposed for the handling of the bulk populations.

Florell (1929) recommended that artificial selection can be imposed to eliminate undesirable and weak plants in bulking generations. Even in short-term bulks, the employment of artificial selection and natural selection aids to move the population in the desired direction.

Mac Key (1962) has suggested three major modifications; bulk method in strict sense, bulk method following mass selection and bulk method with continuous mass selection approach (Fig. 4). During bulk breeding, undesirable plants are rogued, while harvests from desirable plants are bulked. Here a form of positive/ negative mass selections is applied during the bulking generations (Fig. 4).

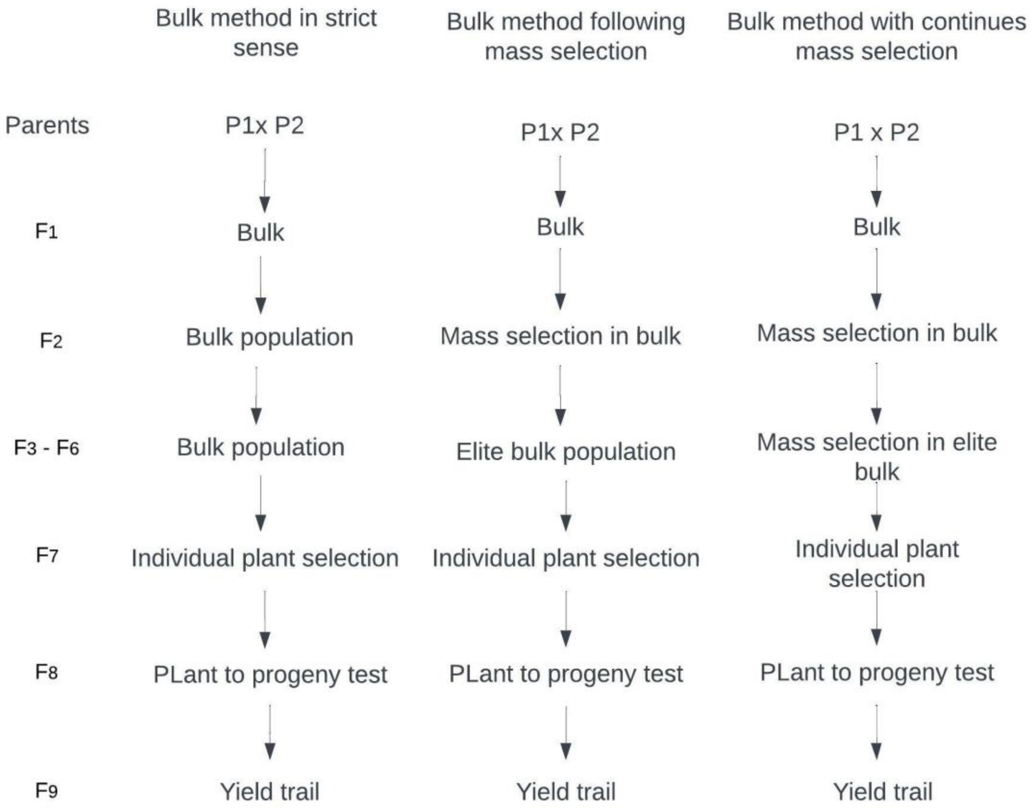


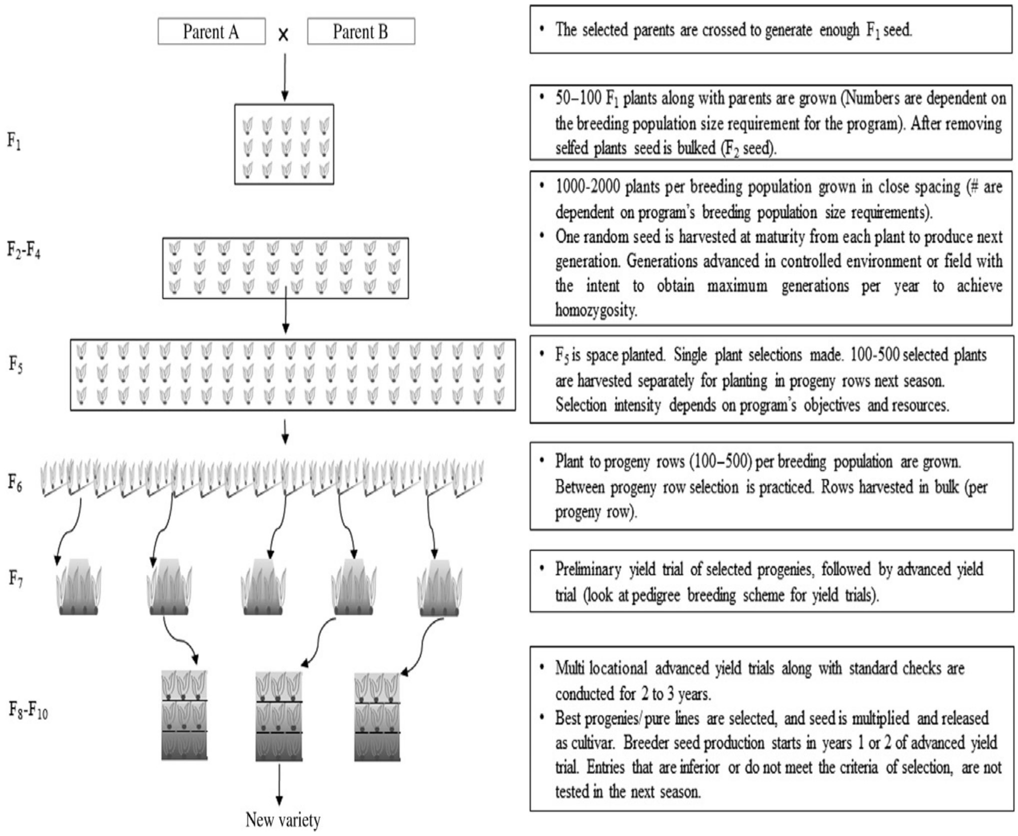
Fig. 4: Schematic representation of Modified bulk-mass selection methods as proposed by Mak Key (1962)

Harlan and Martini (1929) proposed combining progeny from multiple cross combinations to create a 'composite cross population.' This method allows for the selection of progeny from a wide range of cross combinations, followed by natural selection on the progeny.

Lupton and Whitehouse (1957) described a modified pedigree bulk that derives bulk families from selected F_2 plants and imposes selection among plants in bulk populations. The method is similar to pedigree bulk method implemented as a modified version of pedigree method.

Jensen (1988) advocated for a visual selection of desirable plants at the F_2 generation based on characteristics such plant height, disease resistance, and maturity time. F_3 progeny rows are established and evaluated visually or by yield testing, and the entire population's chosen progeny rows are harvested in one packet. Superior lines are selected from the F_4 and F_5 generation while maintaining the bulk procedures.

Goulden (1939) proposed single seed descent method as a means of reducing the loss of genetic diversity along the bulking generations. The method aids in the rapid advancement of generations even in off-season nurseries and controlled environments. It is the most commonly used method in creating recombinant inbred lines (RIL) of selected crosses. Here, rather than harvesting seeds of whole plant, single seeds from each plant at F_2 generation till F_5 or F_6 are harvested and bulked, and these are followed through the pedigree method in later generations (Fig.5).



- The selected parents are crossed to generate enough F_1 seed.

- 50–100 F_1 plants along with parents are grown (Numbers are dependent on the breeding population size requirement for the program). After removing selfed plants seed is bulked (F_2 seed).

- 1000-2000 plants per breeding population grown in close spacing (# are dependent on program's breeding population size requirements).
- One random seed is harvested at maturity from each plant to produce next generation. Generations advanced in controlled environment or field with the intent to obtain maximum generations per year to achieve homozygosity.

- F_5 is space planted. Single plant selections made. 100-500 selected plants are harvested separately for planting in progeny rows next season. Selection intensity depends on program's objectives and resources.

- Plant to progeny rows (100–500) per breeding population are grown. Between progeny row selection is practiced. Rows harvested in bulk (per progeny row).

- Preliminary yield trial of selected progenies, followed by advanced yield trial (look at pedigree breeding scheme for yield trials).

- Multi locational advanced yield trials along with standard checks are conducted for 2 to 3 years.
- Best progenies/ pure lines are selected, and seed is multiplied and released as cultivar. Breeder seed production starts in years 1 or 2 of advanced yield trial. Entries that are inferior or do not meet the criteria of selection, are not tested in the next season.

Fig. 5: Schematic representation of single seed descent method.

12.2.4 Genetic basis of bulk method

The genetic basis for bulk and pedigree method are similar. However, the end product of bulk breeding is homozygous and homogenous population. In addition, during the selfing generation, the proportions of heterozygotes decrease by half each generation of advancement. The dynamic shift in gene frequencies over generations may be observed due to the effect of natural selection over populations. The shift in the frequency is dependent on zygotic potential of heterozygotes: 1) gametic potential of the genotype for seed production, 2) survival of the genotype to produce seed in a heterogeneous population, 3) effect of Genotype by Environment interaction on seed production 4) seed sampling including maximum variation available from the population.

12.2.5 Application of bulk method

The method can be used for (1) isolation homozygous lines, (2) waiting for the opportunity for selection, and (3) to allow natural selection to change genetic makeup of the population.

12.2.5.1 Isolation of homozygous lines

The bulk method is most commonly to extract homozygous lines with minimal effort and expense. After bulking generations, plants or progeny rows selected and handled in pedigree method manner at F_6 or F_7 to later generations are nearly homozygous. Thus selected progeny rows can be effective tested at a preliminary and advanced yield trails.

2.5.2 Waiting for the opportunity for selection

Effective selection under method dependence on the occurrence conditions that favour selection, such as disease epidemics, cold, drought etc. Thus, segregated generation may be carried out in bulk up until the occurrence of such circumstances, at which point individual plants may be chosen and advanced according to the pedigree technique. The length of bulking would depend on the occurrence of the relevant environment.

12.2.5.3 Opportunity for natural selection

In the long term bulking generations, natural selection would favour maintenance of higher yielding genotypes and eliminate the poorer yielding ones. At F_2 – F_6, natural selection affects mostly heterozygous proportions, however, after F_7 natural selection starts to act on homozygous plants. As a result, superior homozygous plants are maintained.

12.2.6 Advantage of bulk method

1. The bulk technique is less expensive, straightforward, and convenient.
2. It is better suited to breeding for complicated genetic traits that are challenging to assess on single plant basis.
3. Natural selection makes it easier to isolate desirable genotypes by reducing the prevalence of un-adapted types in the population.
4. Self-fertilization in bulk from F_2 to F_4/F_5 increases homozygosity. The preselected F_5/F_6 provides homozygous phenotypes in an otherwise heterogeneous population.
5. If selection had begun in the F_2 generation, the selected F_5/F_6 plant-to-progeny rows would have far fewer negative variations.
6. No pedigree records, which saves time.

12.2.7 Disadvantage/Limitations of bulk method

1. The main drawback is that developing new varieties takes a lot longer. Natural selection is only noticeable until F_8 or F_{10}, and bulking may need to continue until F_{20} or higher.
2. Natural selection has little impact on the genetic makeup of populations in short-term bulks.
3. It gives the breeder little chance to use his or her selection skills or judgement.
4. At the end of the bulking period, a considerable number of progenies must be selected.
5. Character inheritance information, which is frequently available via the pedigree technique, cannot be obtained.
6. In some cases, at least, natural selection may act against the ergonomically desirable types.
7. Due to varied effects of natural selection may hinder populations, leading in the complete elimination of particular genes or traits like low stature, earliness, etc.

12.2.8 Achievement of bulk method

Bulk breeding method has been used in Barley crop for developing some varieties from the crosses (Allas x Vaughn), like Arival, Beecher, Glacier etc. In India, “Narendra Rai” has been developed in brown Mustard.

12.2.9 Comparison of pedigree and bulk method: Case studies

12.2.9.1 Faba Bean

In a segregating population of faba bean (Giza 429 Triple white Misr 1) x (ILB-938 Nubariah 1 Misr 1), Ali *et al*. (2020) investigated the effectiveness of pedigree and bulk methods of selection for enhancing seed yield in the F_2-F_5 generations. After three rounds of selection (using the pedigree method) or one cycle of selection (bulk method), highly significant differences among the F_5-selected families were found for the selection criterion, seed yield/plant. While all associated characteristics under study displayed lower genotypic variability using the bulk method than using the pedigree method. Pedigree selection improved seed yield/plant from the bulk sample, the best parent, and the check cultivar, respectively, by 21.82, 20.58, and 53.49% after three selection cycles. However, after a single cycle of selection, the bulk method increased seed yield/plant from the bulk sample, the best parent, and the check cultivar, respectively, by 8.43, 7.27, and 38.24%. When compared to the bulk method, the pedigree selection strategy for increasing seed production was more effective at isolating high yielding genotypes.

12.2.9.2 Soybean

Seven soybean crosses were examined by Gaurav et al. (2019) to compare the productivity and quantity of pods produced through pedigree (PM), single pod descent (SPD), single pod descent with selection (SPDS), and bulk (BM) methods of generation advancement. Analysis of variance for the mean of all lines within a cross indicated that there are significant differences in breeding methods for the studied traits across all crosses. Bulk method was the best in most of the crosses followed by pedigree method. On the basis of overall mean, bulk method was ranked first in four crosses and second in three crosses, while pedigree method was ranked first in three crosses and second in four crosses. When yield of the highest yielding line derived by different methods within each cross was compared with the mean of check, it was observed that the highest yielding line in three out of seven crosses was from the bulk method whereas in four crosses it ranked second. Performance of the pedigree method did not differ much, as it ranked first and second in two crosses each. The extent of genetic variability present in a cross had no effect on the outcome of the results.

Torrie (1958) studied six soybean crosses (Lincoln X Manchu 606, Lincoln X Flambeau, A45-251 (selection from Mukden X Richland) X Manchu 606, Lincoln X Ottawa Mandarin, A45-251 X Flambeau and Mukden X Flambeau). Torrie revealed that, there are no significant differences between the mean responses of soybean lines developed using the pedigree and bulk method in terms of plant height, lodging index, bacterial blight reaction, oil and protein content of the seed and iodine number of the oil. With two exceptions where the bulk lines excelled, mean seed yields for the two breeding method were similar.

12.2.9.3 Mustard

Singh (2022) compared unselected bulk, bulk Single-Seed-Descent and pedigree method for five characters *viz.*, number of primary branches, number of secondary branches, seed/ silique, 1000 seed weight and seed yield (plant in our crosses, *viz.*, Narenra Rai x NDR 8220, Narendra Rai x NDR 8208, Nrendra Rai x NDRE 4 and Vardan x Kranti). All the crosses showed significant differences among their progenies under all the methods for the five characters' studies. For the majority of traits, progeny from a pedigree were shown to have improved mean, range, heritability, and genetic advancement. Pedigree and SSD have shown to be better methods for identifying early progenies in different population.

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