

10. Breeding Method in Segregating Generation

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Abstract:

A segregating population is a genetically heterogeneous population that results from hybridization between parents that are genetically controlled and differ in one or more traits. It is in large quantity and challenging to manage during the breeding program. Three different breeding techniques were employed to handle the segregating n F₂ and subsequent generations F₃, F₄, F₅, F₆, etc. For example, the Pedigree, Bulk, and Backcross methods. All of these methods aim to create new and pure variety. When it comes to producing new varieties, the Pedigree, bulk, and single-seed descent methods are the most commonly utilized selection techniques in segregating generation. However, F₁ and the following generation are repeatedly backcrossed to the recurrent parent to produce a new variety that is identical to the recurrent parent except for the character that was transferred. The use of the breeding method depends on the crop type.

Keywords:

Hybridization, Segregating Population, Pedigree Method, Bulk Method, Single Seed Decent Method, Backcross Method

10.1 Introduction:

Hybridization refers to crossing between different genetic makeup plants. It could be interspecific/intragenic or intergeneric. The hybridization of parents with different genetically controlled features results in thousands of plants in segregating population F₂ and subsequent generations. Since segregating generations contain Genetic diversity, we can improve crop population by employing various breeding techniques.

A crop improvement breeding program must raise and maintain the identity of parental plants and the parental seed of a large segregating population from multiple crossings. Breeding methods may differ depending on whether you are working with self-pollinated species, cross-pollinated species, or clonal species; different breeding methods may apply.

Following hybridization, there are three methods to maintain segregating generations through selection.

- Pedigree method
- Bulk method
- Backcross method

10.2 Pedigree Method:

The pedigree method is the commonly used breeding method for crop improvement self-pollinated crops. In self-pollinated crops, the Selection of highly heritable traits is done in early generations on individual plants. Yield testing is done once homozygous lines are developed. The progeny of a single best homozygote is released as a variety. Thus, a variety developed has a homozygous and homogeneous population. It is used for the development of new pure line varieties. It is mostly used in polygenic trait improvement than oligogenic traits. Also used to correct some specific weaknesses of an established variety with aims to improve the yield and quality parameters. However, in an early-generation testing procedure or a modified pedigree method, yield testing is done in early generations while within-family selection is still ongoing. The pedigree method was first outlined by Love in 1927. is maintained.

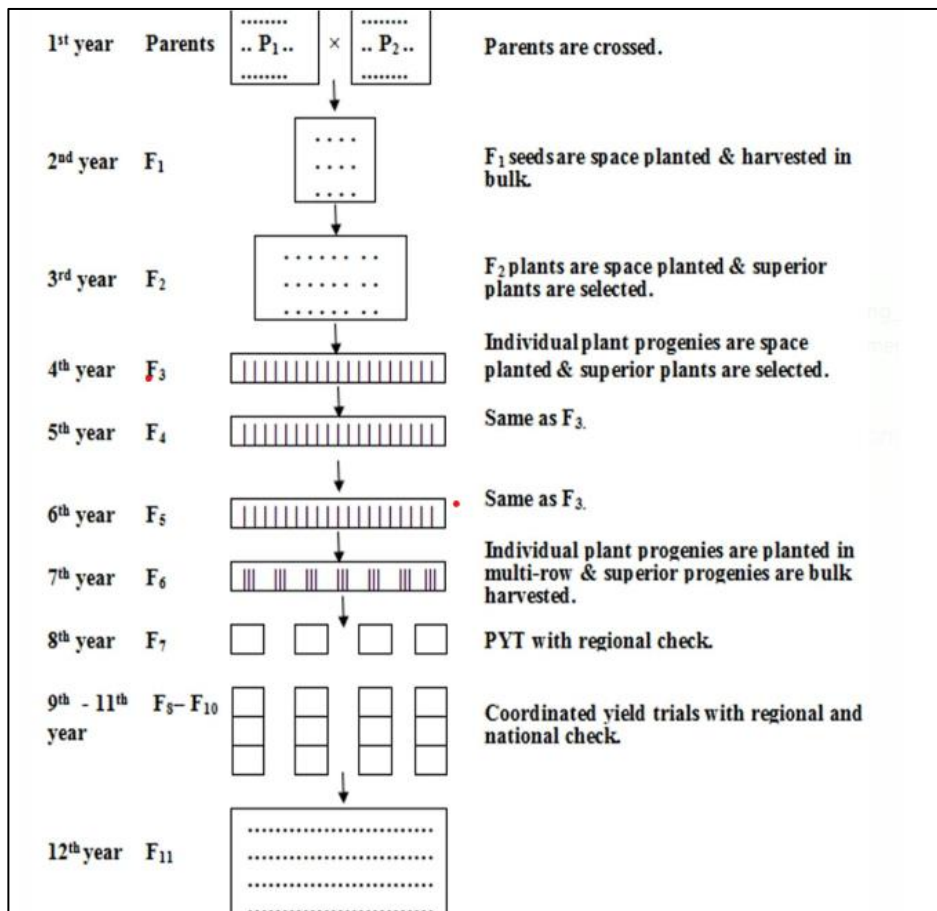


Figure 10.1: Procedure of Pedigree Method

10.2.1 Schematic Representation of Pedigree Method:

Hybridization. Selected parents Crossed to produce a progeny in the F1 generation.

Generation F₁. F1 Seeds, which are the result of hybridization, are planted with the appropriate recommended spacing between them. The bulk of Seeds of about 15-30 plants are sent to grow the F2 generation.

Generation F₂. The primary procedure used in this step is selection from F1 generation seeds to develop between 2000 to 10000 seeds in F2 generation. Using the selection method between 100-500 are selected and harvested individually for the F3 generation.

Generation F₃. Every one of the F2 generation plants produces at least thirty progenies. About 100-400 superior plants (the number should be fewer than the one chosen for the F2 generation) are selected for the F4 generation

Generation F₄. Seeds from the F₃ generation are space planted. Plants with desirable characters are selected, this number should be fewer than the ones chosen for the F4 generation.

Generation F₅. Individual plant progenies are planted in multi-row (3 or more) plots so that superior plants about 50 – 100 can be selected by comparison of their phenotypic character.

Generation F₆. Individual plant progenies are planted in multi-row (3 or more) plots. Plants are selected based on visual evaluation of phenotypic characters, and progenies showing segregation can be eliminated.

Generation F₇. In F7 generation Preliminary yield trials were conducted with a minimum of three replications and a check. Quality tests are also conducted.

Generation F₈ to F₁₀. In the F8 to F10 generation, multi-location yield trials with replications are also conducted. Tests for quality and disease resistance are conducted by providing unfavorable conditions and superior strains are produced

Generation F₁₁ to F₁₂. When a strain is to be released as a variety, the Seed of the new variety will be multiplied in the F11 generation and F12 generation distributed to the farmers.

10.2.2 Merits of Pedigree Method:

- It allows the breeder to use his skill and knowledge to select superior plants in segregating generations.
- This method is better for the improving of easily identifiable and inherited traits.
- Transgressive segregation for yield and other quantitative characters can be recovered by this method.

- In this method pedigree records are maintained. Hence Information about the inheritance of characters and the pedigree of parental lines and segregating can be obtained.
- In this method negative selection of progeny occurs. Hence Inferior plants and progenies are eliminated in early generations.
- It takes less time to develop a new variety than the bulk method.
- The pedigree record provides detailed genetic information of the cultivar.
- Pedigree record provides detailed information on ancestors; Hence the breeder can produce the progeny lines that carry the genes for the target trait.
- In this method, a high degree of genetic purity is produced in the cultivar because the selection of the cultivar is based on both phenotype and genotype traits, i.e. progeny test.

10.2.3 Demerits of Pedigree Method:

- This method depends on the breeder's skill and familiarity with the crop. So, there might be a chance that Valuable genotypes may be lost in early generations, if sufficient skill and knowledge are lacking in the breeder, at the time of selection.
- In the pedigree method, the superior genotype is manually selected, there is no chance for natural selection.
- Segregating population huge in numbers made it difficult to manage many crosses.
- Maintenance of pedigree records, selections of phenotypically superior lines, growing progeny rows, etc. are time-consuming, expensive, and laborious.

10.2.4 Achievements:

Many varieties have been developed by pedigree method in many crops.

Table 10.1: Many varieties have been developed by pedigree method in many crops

Crops	Variety Produced
Wheat	NP-52, 120,125, 700 and 800 series
Rice	ADT – 25, Jaya, Padma, Ratna, Krishna, Sarbarmati, Jaya/ Bala, Kaveri.
Rice	ADT – 25, Jaya, Padma, Ratna, Krishna, Sarbarmati, Jaya/ Bala, Kaveri.
Cotton	Lakshmi, Digvijay, LH 900, LH 1556, F846, F 1054, F1378, HS 6, Vikas, Sharda, MCU 9, MCU 11, LRA 5166
Sorghum	Co 18, RS 610, etc.
Tobacco	NP 222
Green gram	T 2, T 44,T51, Sheela.

Crops	Variety Produced
Chickpea	T1,T2,T3,T5,Radhey
Pigeon Pea	T 21, Prabhat
Pea	Pant Matar 2,,Jawahar Matat 4,Jawahar Matar 1

10.3 Bulk Method:

The bulk method allows natural selection to act and remove undesirable genotypes from the population (i.e., per cross).

The choice of growing environment will dictate what kinds of traits will be selected for or against, therefore care needs to be exercised to use environments that are suitable for realizing the objectives of the program.

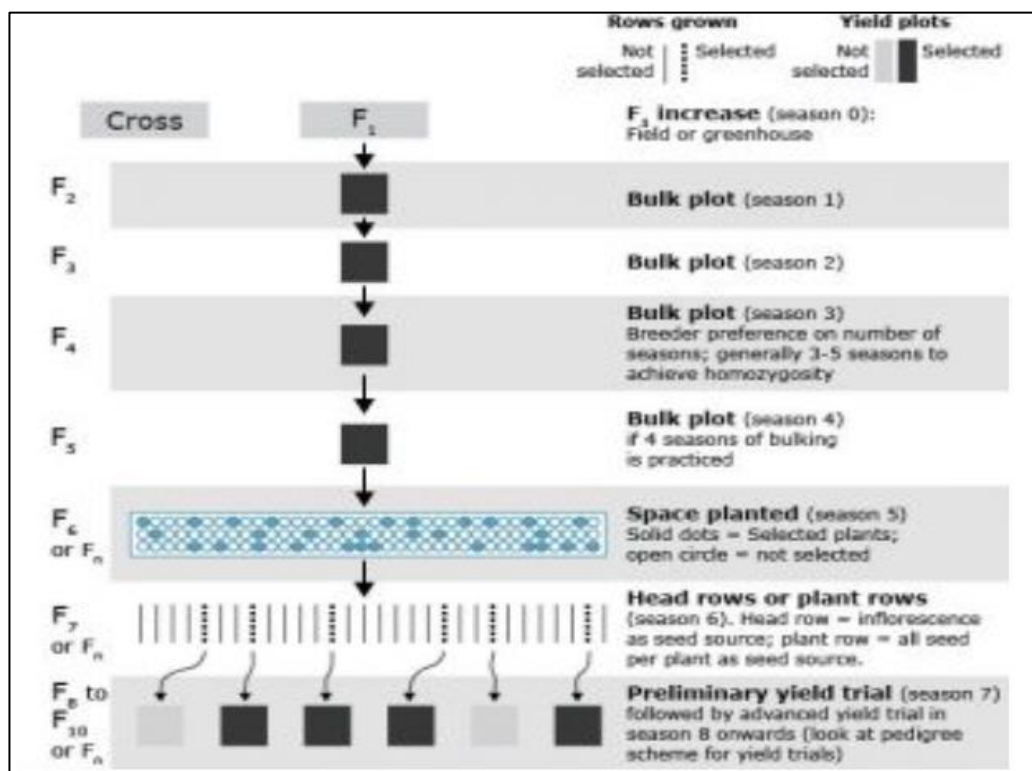


Figure 10.2: Bulk Method

10.3.1 Pictorial Representation of Bulk Method of Breeding:

In this method, F₂ and subsequent generations are harvested in bulk to grow the next generation. It is repeated up to F₆ to F₇. Selection can be made in each generation but harvested seed is bulked in each generation. This is similar to mass selection.

At the end of the bulking period i.e. after F₇ single plant selection is made and tested for yielding ability. Generations are advanced to homozygosity through bulks. Natural selection is used to remove undesirable plants. The breeder uses his skill and knowledge to select the superior plants and at the same time, there is no pedigree record which saves time and labor.

10.3.2 Merits of Bulk Method:

- Simple, convenient, less technical, and inexpensive method.
- By inducing artificial epiphytotic conditions undesirable or weaker genotypes can be eliminated.
- If the bulking period is longer natural selection operates and desirable genotypes are selected.
- pedigree record is maintained.
- Since a large population is grown there is a chance that the transgressive segregants will be superior to parents or F₂.

10.3.3 Demerits:

- Takes a much longer time to develop a new variety.
- In short-term bulk there is no chance for natural selection.
- Many progenies are to be selected in each generation which requires much labor, time, and space. We cannot get information on inheritance through this method.

10.4 Single Seed Descent (Modified Bulk Method):

It is used for segregating populations of self-pollinated species.

The progeny of a single best homozygote is released as a variety by these methods.

Thus, varieties developed by these methods are homozygous and homogeneous. single plants or inflorescence per plant are selected at each generation; while in the bulk method, plants from the entire population are harvested and seeded in the next generation and this method is called the single seed descent method.

It was developed to rapidly advance lines to homozygosity so that selection can be practiced on homozygous lines (Figure 10.3).

The original aim of this method was to maintain a large population size to mimic the genetic variation in F₂ generation for effective selection. However, this method is now used to reduce the time to develop cultivars. (Sleper and Poehlman, 2006).

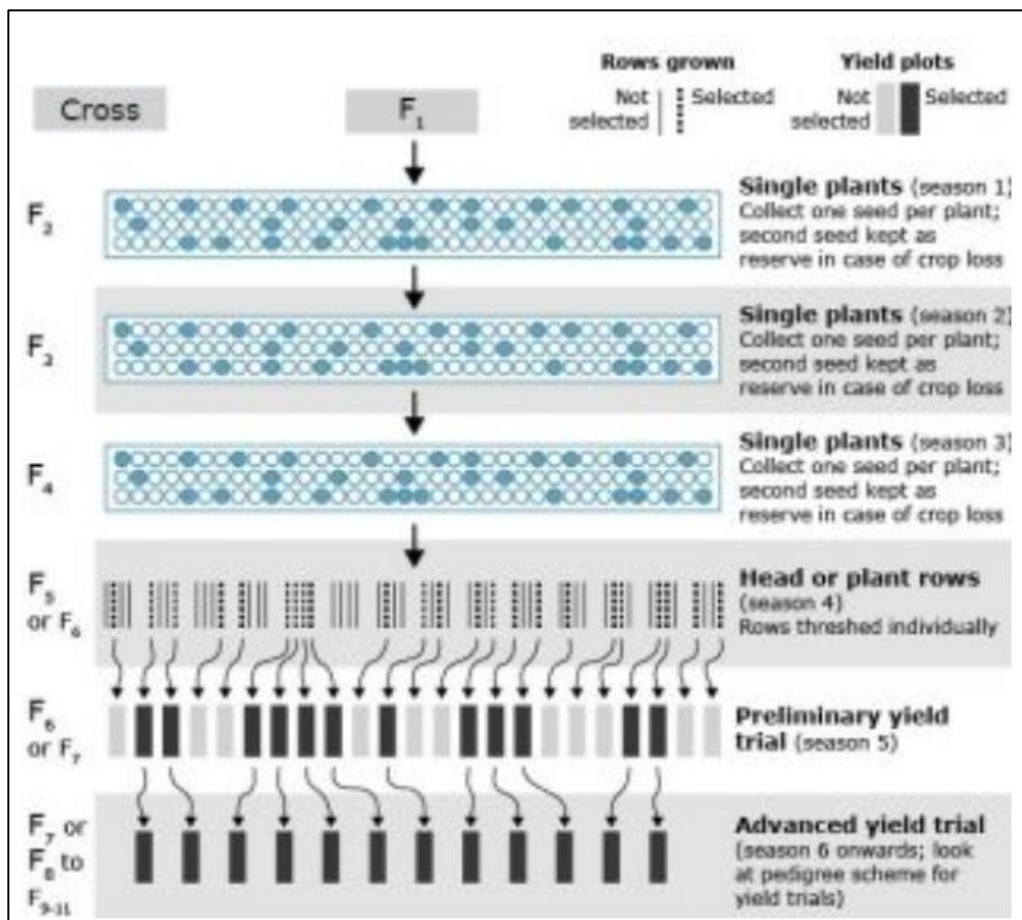


Figure 10.3: Single Seed Descent

10.4.1 Pictorial Representation of Single Seed Descent Method of Breeding:

Generations are advanced to homozygosity rapidly. In case of small grain crops (such as wheat, barley, oats), three seasons can be completed in artificial growing conditions (greenhouse etc.), and limited space is needed to keep a population size of 250-300 seed per cross. If true single seed descent is practiced (where one seed per plant is grown in successive generations, population size is reduced in each cycle due to losses due to no germination and emergence. As an alternative modified, single seed descent can be used where 2-3 seed per plant are planted in hill plots in each cycle, and 2-3 seed from each hill are collected from an inflorescence.

A. Advantages of Single Seed Descent Method:

- SSD plots can be grown in a disease or another stress condition to select for that trait.
- It is a cheaper, less technical method of breeding.
- Rapid inbreeding and homozygosity is achieved.
- No need for record keeping of individual plants while advancing through SSD.

10.5 Backcross Method:

In the backcross method of breeding, the hybrid and the progenies in subsequent generations are repeatedly backcrossed to one of the parents. As a result, the genotype of the backcross progeny becomes almost similar to that of the recurrent parent.

Self-pollinated groups of crop species are used for the transfer of oligogenic characters from a donor source to a well-adapted variety. Hence it improves one or two specific defects of a high-yielding variety.

This method is also used for the development of multilines, Isogenic lines, and transfer of male sterility. It is more effective in transferring oligogenic characters than polygenic traits.

The end product of the backcross method is similar to the parent variety except for the character which has to be transferred from the donor source.

10.5.1 Pre-Requisites for Back Cross Breeding:

- a. A suitable recurrent parent must be available which lacks in one or two traits.
- b. A suitable donor parent must be available.
- c. The character to be transferred must have high heritability and preferably it should be determined by one or two genes.
- d. The number of backcrosses should be sufficient so that the genotype of the recurrent parent is recovered in full.

10.5.2 Application of Back Cross Method:

This method is commonly used to transfer disease resistance from one variety to another. But it is also useful for the transfer of other characteristics.

- a. Intra varietal: transfer of simply inherited characters E.g., Disease resistance, seed coat color
- b. Inter varietal: transfer of quantitative characters. E.g., Plant height, Seed size, and Seed shape.
- c. Interspecific: transfer of simply inherited characters E.g., Transfer of disease resistance from related species to cultivated species. E.g., Resistance to black arm disease in cotton from wild tetraploid species into *G. hirsutum*
- d. Transfer of cytoplasm: This is employed to transfer male sterility. The female parent will have the sterile cytoplasm and the recurrent parent will be used as the male parent. E.g., *Sesamum malabariicum* x *S. indicum* Female parent Recurrent parent.
- e. Transgressive segregation: The back cross method may be modified to produce transgressive segregants. The F1 is backcrossed to the recurrent parent 2 to 3 times for getting transgressive segregants.
- f. Production of isogenic lines
- g. Germplasm conversion: E.g. Production of photo-insensitive line from photo-sensitive germplasm through backcrossing. This was done in the case of sorghum. Popularly known as a conversion program.

10.5.3 Procedure for Backcross Method:

The Plan of the backcross method would depend upon whether the gene being transferred is recessive or dominant.

A. Transfer of Dominant Gene:

First Year	Non-Recurrent		Recurrent
	Parent B	x	Parent A
	Resistant to rust		Susceptible to rust
	F1	Rr x	rr BC1
		Resistant	
	rr Rr	x	rr BC2
	rr Rr	x	rr BC3
	rr Rr	x	rr BC4
	rr Rr	x	rr BC5

Back cross up to the 6th or 7th generation. After the 7th BC rust resistant lines were self-pollinated. Harvest is done on a single-plant basis. 8th Season Individual plant progenies grown a) Homozygous plants having resistance and resembling parent A are selected harvested and bulked. 9th season: Yield trials .10th season: Seed multiplication and distribution

Steps:

- a. First Season: Hybridization: Crossing between parent B donor (Female) and Susceptible parent A recipient (male)
- b. Second Season: Raising the F1 and backcrossed to recurrent parent A.
- c. Third season: Growing the BC1F1. It will be segregating for 1 susceptible (rr): 1 resistant (Rr). Rust-resistant plants are backcrossed with recurrent parent A. This is the second backcross.
- d. Fourth Season: Raising BC2 F1 will again segregate in the ratio of 1: 1. Third backcross is done with resistant plants.
- e. Fifth Season to Eighth Season: Raising backcross F1s and crossing resistant plants with the recurrent parent is continued up to 7th backcross.
- f. Ninth season: Raising BC7F1 and observing resistant lines. The plants resembling parent A coupled with resistance is selected and harvested on a single-plant basis.
- g. Tenth Season: Growing the progeny row 8' and observing each row for resistance. Best rows are selected and harvest is done on a row basis
- h. Eleventh Season: The row bulk is raised in yield trial along with check and the best plots are selected.
- i. Twelfth season: Selected plot seeds are multiplied and released as new varieties.

B. Transfer of Recessive Gene:

I Season	Non recurrent parent B	Recurrent parent A
Hybridization	Resistant	Susceptible
	Rr	x RR
		F1 Rr
II Season	Grow the F1	Rr
III Season	Grow F2 RR “ Rr “ rr	x RR BC1
IV Season		Grow BC1F1 Rr
V Season	Grow BC2F2	RR : Rr : rr x RR BC2
VI Season	Grow BC2F1	Rr
VII Season	Grow BC2F2	RR : Rr : rr x RR BC3
VIII Season		Raise BC3F1

X Season Do as on VIII Season

XI Season does as in IX season Continue this process still 7th or 8th backcross. After studying 8th BCF2 select plants resembling parent B coupled with resistance.

Harvest them on a single plant basis. Next season raise them in progeny rows and select best progenies. Compare them in yield trial and fix the best culture, multiply it, and release it as a variety. While selecting plants artificial bombardment for disease is to be done.

Steps:

I Season: Make a cross between donor parent A and recurrent parent B and Harvest the hybrid. The donor parent A is resistant which is governed by recessive genes. The susceptibility is governed by a dominant gene in parent B.

II Season: Grow the F1 which will be susceptible – Harvest them.

III Season: Grow F2 which will be segregated in the ratio of 1:2:1 i.e. 3/4 susceptible and 1/4 resistant. The resistant lines (rr) make the first backcross with parent A having the dominant RR gene. Harvest BC1 F1

IV Season: Grow BC1F1 V season: Grow BC1 F2 which will be segregating as we saw in III season. Repeat the process of the third season. This will give BC2F1

VI Season: Grow BC2F1 VII season: Grow BC2F2 them repeat the process of

V Season. This will give BC3F1.

VIII Season: Grow BC3F1 IX Season: Grow BC3F2 and repeat the process of VII Season. Harvest BC4F1.

X season: Grow BC4F1 XI Season: Grow BC4F2 and repeat the process of IX Season. Harvest BC5F1. XII, XIII & XIV season: Repeat the process and carry out backcross upto 7 time. XV Season: While studying BC7F2 select single plants having resistance and resembling parent B.

XVI Season: Study the progenies in progeny rows and select best progenies.

XVII Season: Conduct yield trial and select best material.

XVIII Season: Multiply the seeds and distribute it as improved variety with resistance to disease.

10.5.4 Merits of Backcross Method:

- The genotype of the new variety is nearly identical to that of the recurrent parent, except for the genes transferred. Thus, the outcome of a backcross programme is known beforehand, and it can be reproduced at any time in the future.
- It is unnecessary to test the variety developed by the back cross method in extensive yield tests because the performance of the recurrent parent is already known. This may save up to 5 years and a considerable expense.
- The backcross program is not dependent upon the environment, except for that needed for the selection of the character under transfer. Therefore, off-season nurseries and greenhouses can be used to grow 2-3 generations each year. This would drastically reduce the time required for developing the new variety.
- A much smaller population is needed in the backcross method than in the case of the pedigree method.
- Defects, such as, susceptibility to disease, of a well-adapted variety can be removed without affecting its performance and adaptability. Such a variety is often preferred by the farmers and the industries to an entirely new variety because they know the recurrent variety well.
- This is the only method for interspecific gene transfers.

10.5.5 Demerits of the Backcross Method:

- The new variety is identical to the recurrent parent, except for the character that is transferred.
- Undesirable genes closely linked with the gene being transferred may also be transmitted to the new variety.

- Hybridization has to be done for each backcross. This is often difficult, time-consuming, and costly.
- By the time the backcross is over, the recurrent parent may have been replaced by other varieties superior in yielding ability and other characteristics.

Table 10.2: Achievements

Crop Name	Variety evolved	Recurrent parent	Non-recurrent parent
Cotton	V797, Digvijay, Vijalpa, and Kalyan, Vijay, Digvijay	BD8	Goghari A-28 1027 A L-F
Wheat	Pureline varieties Kharchia 65 NP852 NI5439 IWP72 HUW234 K8027 HW2004	Kharchia local NP761 NP710 Kalyan Sona HUW 12 K852 C306	EG953 EG951 REPM80 E6056 CPAN1666 HD1696
	Multiline varieties MLKS 11 KML7406 KSML3 Sonalika Multiline	Kalyan Sona Kalyan Sona Kalyan Sona Sonalika	Exotic lines resistant to leaf rust Exotic lines resistant to leaf rust Exotic lines resistant to leaf rust Six parents
Brown mustard	Narendra Rai		

Table 10.3: Comparison of Pedigree, Bulk, and Backcross Methods of Breeding

Pedigree method	Bulk method	Backcross method
Used in self and cross-pollinated crop	Used in self-pollinated crop	Used in self and cross-pollinated crop
F1 and subsequent segregating generations are allowed to self-pollinate	F1 and subsequent segregating generations are allowed to self-pollinate	F1 and subsequent segregating generations are repeatedly backcrossed to the recurrent parent
Human selection plays an important role	Both natural and human selection	Human selection plays an important role

Pedigree method	Bulk method	Backcross method
F2 population smaller than bulk method	F2 population larger than pedigree method	F2 population smaller than both bulk method and pedigree
Equally effective on both oligogenic and polygenic traits	Equally effective on both oligogenic and polygenic traits	More effective in oligogenic traits
Pedigree records are maintained	Pedigree records are not maintained	Pedigree records are not maintained
Extensive testing required before release of variety	Extensive testing required before release of variety	Extensive testing not required before release of variety
Breeding procedure is same for dominant and recessive genes	Breeding procedure is same for dominant and recessive genes	Breeding procedure is different for dominant and recessive genes
Takes 14-15 years to release new variety	Requires longer period to pedigree method	Takes less time than other two method
Variety has narrow adaptation	Variety has wide adaptation	Adaptation like parent variety
Widely used	Less popular	More popular
New variety is different from both parent	New variety is different from both parent	New variety is identical to recurrent parent except for the character transferred

10.6 Conclusion:

Hybridization is used to generate additional genetic variability for further improvement of the crop species. The methods generally used in handling segregating generations (F_2 , F_3 , F_4 , etc.) after hybridization are grouped into three types i.e., the Pedigree method, the Bulk method, and the Backcross method. The main aim of these breeding methods is to produce a Pureline, to develop a new variety, to develop an inbred line, and to improve the specific character of a well-adapted variety for which it is deficient.

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