

## **Experiment: Handling of segregation populations: Pedigree method**

**Aim:** To know about how to handle segregating population of pedigree method

Mass selection and pure line selection cannot be applied to segregating population E.g.  $F_2$ ,  $F_3$  etc. because in these methods selection made from the existing variability present in the population. However, when there is limited genetic variability in a species than hybridization acts as an important tool to create additional genetic variability for further improvement of the species. The methods generally used for handling of segregation generation may be grouped into following three categories.

- a) Pedigree Method
- b) Bulk Method and
- c) Back Cross Method

The objectives of all these methods are to

1. Develop pureline
2. Develop new varieties.
3. Develop inbred line
4. Improve specific character of a well-adapted variety for which it is deficient.

### **Pedigree method**

In pedigree method, individual plants are selected from  $F_2$  and the subsequent generation and their progenies are tested. During the entire operation, a record of the entire parent's offspring relationship is kept, is known as pedigree record. The selection of individual plant is continued till the progenies show no segregation. At this stage, selection is done among the progenies, because there would be no genetic variation within progenies.

### **Pedigree record**

The pedigree may be defined as a description of the ancestors of an individual and it generally goes back to some distant ancestors. Thus, it describes the parents grandparents, great grandparents so on of an individual.

### **Maintenance of pedigree record**

Pedigree record may be kept in several ways, but it should be simple and accurate. Generally, each cross is given a number. The first two digits of this number refer to the year in which the cross was made, and the remaining digits denote the serial number of the cross in that year. For example, the number 7911, denotes the cross number 11 of the year 79. In the segregating generation one of the two systems of designation may be followed.

#### **i) System I**

In this system, the individual plant progenies in each generation are assigned row number, corresponding to their location in the plot. In addition each progeny in  $F_4$  and the subsequent generation is assigned the row number of the progeny in the previous generation from which it was derived.

**Table 9.1: Description of maintenance of pedigree record by System I**

Generation	Number	Description
F <sub>3</sub>	7911-7	Progeny in the 7 <sup>th</sup> row in the F <sub>3</sub> plot.
F <sub>4</sub>	7911-7-4	Progeny in the 4 <sup>th</sup> row in the F <sub>4</sub> plot, selected from the progeny in the 7 <sup>th</sup> row of the F <sub>3</sub> plot.
F <sub>5</sub>	7911-4-14	Progeny in the 14 <sup>th</sup> row in the F <sub>5</sub> plot selected from the progeny in the 4 <sup>th</sup> row of the F <sub>4</sub> plot.
F <sub>6</sub>	7911-14-3	Progeny in the 3 <sup>rd</sup> row in the F <sub>6</sub> plot selected from the progeny in the 14 <sup>th</sup> row of the F <sub>5</sub> plot.

**ii) System-II**

In this system, in each generation the selected plants are assigned serial numbers within individual progenies. Each progeny or selected plant bear the serial number of all the plants in the previous generation, related to it by direct descent. Thus, the plants selected in F<sub>2</sub> are given serial numbers of their parents (F<sub>2</sub> plants). The plants selected from a progeny in F<sub>3</sub> are given the number of that progeny and in each generation the selected plant also given a serial number.

**Table 9.2: Description of maintenance of pedigree record by System II**

Generation	Number	Description
F <sub>3</sub>	7911-7	Progeny obtained from plant number 7 selected in F <sub>2</sub>
F <sub>4</sub>	7911-7-4	Progeny from plant No.4 selected from F <sub>3</sub> progeny , derived from the plant No.7 selected in F <sub>2</sub>
F <sub>5</sub>	7911-7-4-2	Progeny from plant No.2 selected from the F <sub>4</sub> progeny derived from plant no.4 , selected from the F <sub>3</sub> progeny, obtained from the plant No.7 selected in F <sub>2</sub> .
F <sub>6</sub>	7911-7-4-2-8	Progeny from plant No 8, selected from the F <sub>5</sub> progeny, derived from the plant N0.2 selected from the F <sub>4</sub> progeny of the plant No.4 selected from F <sub>3</sub> progeny of the Plant No.7 selected in F <sub>2</sub> .

**Procedure for pedigree method****1. Hybridization**

Selection of parents to be used in a cross is the most important step in breeding program based on hybridization. The selected parents are crossed to produce a simple or a complex cross. For convenience we shall refer to the seed obtained from the width simple/complex crosses as F<sub>1</sub> seed.

**2. F<sub>1</sub> Generation**

F<sub>1</sub> seeds are space planted so that each F<sub>1</sub> plant produces the maximum number of F<sub>2</sub> seed ordinarily, 15-30 F<sub>1</sub> plants should produce enough seeds for a good F<sub>2</sub> population size.

**3. F<sub>2</sub> Generation**

In F<sub>2</sub>, 2000-10000 plants are space-planted to facilitate selection. About 100-500 plants are selected & their seeds are harvested separately. Usually the F<sub>2</sub> population size should be 10-100 times the member of F<sub>2</sub> plants that would be selected in the number may vary from 100-500 depending on the facilities available and objectives of the breeding program. When the objective of breeding is to improve yield, a relatively large no of F<sub>2</sub> plants would be selected. Selection in F<sub>2</sub> is based on characteristics that are simply inherited, e.g. plant height,

head type, seed colour, disease resistance, presence of awns etc. Selection of vigour is generally useless because vigour may be due to heterozygosis or environment or G×E interaction.

#### **4. F<sub>3</sub> Generation**

Medical plant progenies having 30 or more plants each are space-planted. Medicinal plants with desirable characteristics are selected, particularly from superior progenies may also be selected. Diseased and lodging susceptible progenies are eliminated. The no of plants selected in F<sub>3</sub> should preferably be less than that at the F<sub>3</sub> progenies. If the no of superior progenies is small, the whole cross may be rejected.

#### **5. F<sub>4</sub> Generation**

Individual plant progenies are space planted and desirable plants are selected mainly from superior progenies. The no of plants selected in F<sub>4</sub> is generally much smaller than that in F<sub>3</sub> progenies with defect and undesirable characteristics are rejected. If two or more progenies coming from the same F<sub>3</sub> progeny are similar and comparable, only on selection of desirable plants from superior progenies.

#### **6. F<sub>5</sub> Generation**

Individual plant progenies are generally planted according to the recommended commercial seed rate. Often there are more rows grown for each progeny to facilitate compression among many progenies would have in bulk. In progenies showing segregation, individual plants may be selected. The no of progenies meant be reduced to a size manageable in preliminary yield trials about 25-100 progenies.

#### **7. F<sub>7</sub> Generation**

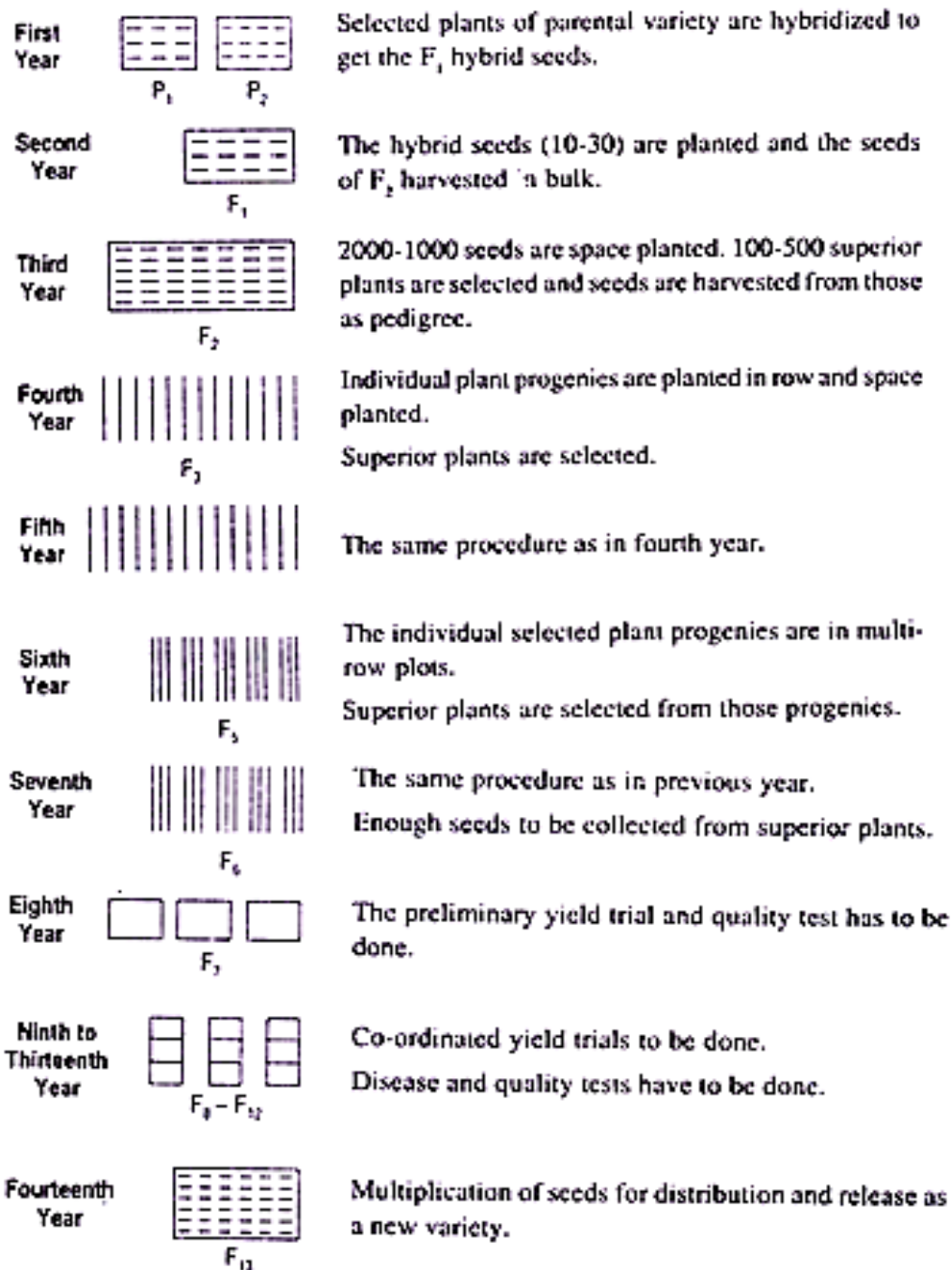
Individual plants are planted in multi row plots and evaluated visually; progenies are harvested than bulk since they would have become almost homozygous. Progeny showing outstanding, in such progenies individual plants may be selected, preliminary yields trait is usually planted in F<sub>6</sub> for those progenies, which are reasonably homozygous and have enough seed, inferior progenies are eliminated based on yield from preliminary yield trial.

#### **8. F<sub>8</sub>-F<sub>10</sub> Generation**

The superior lines are tested in replicated yield trails at several locations. These lines are evaluated for yield diseases and lodging resistance, maturity time and quality etc. A line that is superior to the test commercial variety, included in the trials as check. Yield & other characteristics will be identified for release as a new variety.

#### **9. F<sub>11</sub> Generation**

When a strain is likely to be released as a variety, the breeder usually multiplied its seed during its last year in trail. The breeders have responsibility to supply the breeder seed for production of foundation seed. Thus F<sub>11</sub> to F<sub>12</sub> the seed of the new variety will be multiplied for distribution to the farmer.



**Fig.9.1: Breeding procedure of pedigree method**

**Application of Pedigree Method:**

- 1) Selection of desirable plants from the segregating population in self- pollinated crops.
- 2) This method is commonly used to correct some specific weaknesses of an established variety (Combination breeding).
- 3) It is also used in the selection of new superior recombinant type's i.e. Transgressive breeding.
- 4) This method is suitable for improving specific characteristics such as disease resistant, plant height, maturity etc.

## **Experiment: Handling of segregation populations: Bulk method**

**Aim:** To know about how to handle segregating population of bulk method

### **Bulk population method**

Bulk population method of breeding in self –pollinated crop is also known as mass method or population method of breeding. It was first used by Nilsson Ehle in 1908. It refers to a species is grown in bulk plot (from F<sub>1</sub> to F<sub>5</sub>) with or without selection, a part of the bulk seed is used to grow the next generation and individual plant selection is practised in F<sub>6</sub> or later generation. In this method duration of bulking may vary from 6-7 to 30 generation.

### **Procedure of Bulk Population Method:**

#### **1) Hybridization:**

Parents are selected according to the objective of the breeding programmer and crossed.

#### **2) F<sub>1</sub> Generation**

The F<sub>1</sub> generation ( 10 to 25 F<sub>1</sub>) is space planted and harvested in bulk.

#### **3) F<sub>2</sub>-F<sub>6</sub> – Generation**

F<sub>2</sub> to F<sub>6</sub> generations are planted at commercial seed rate and spacing. These generations are harvested in bulk. During these generations the population size should be as possible, preferably 30 to 50 thousand plants should be grown in each generation.

#### **4) F<sub>7</sub> Generation**

About 30 to 50 thousand plants are space planted and out of this only 1000 to 5000 plants with superior phenotypes are selected and their seeds harvested separately. Selection is made on the basis of phenotypes of plants, grain characteristics etc.

#### **5) F<sub>8</sub> Generation**

Individual plant progenies are grown in single or multi row plots. Most of the progenies would be homozygous and are harvested in bulk. Weak and inferior progenies are rejected and only 100- 300 individual plant progenies with desirable characters are selected.

#### **6) F<sub>9</sub> Generation**

Preliminary yield trial is conducted along with standard variety as check. The evaluation of progeny is done for important desirable characteristics. Quality test may be conducted to reject the undesirable progenies.

#### **7) F<sub>10</sub>- F<sub>12</sub> Generation**

Replicated yield trails are conducted at several locations using standard commercial varieties as check. The lines are evaluated for important agronomic characteristics. If lines are superior to the standard check than it is released as new variety.

#### **8) F<sub>13</sub> Generation**

Seed multiplication of the newly released variety has done for distribution to the farmers.

### **Application of Bulk Population Method**

This method is suitable and most convenient for handling the segregating generation of cereals, smaller millet, grain legume and oilseeds. This may be used for three different purposes.

- 1) Isolation of homozygous lines.
- 2) Waiting for the opportunity of selection.
- 3) Opportunity for natural selection to change the composition of the population.

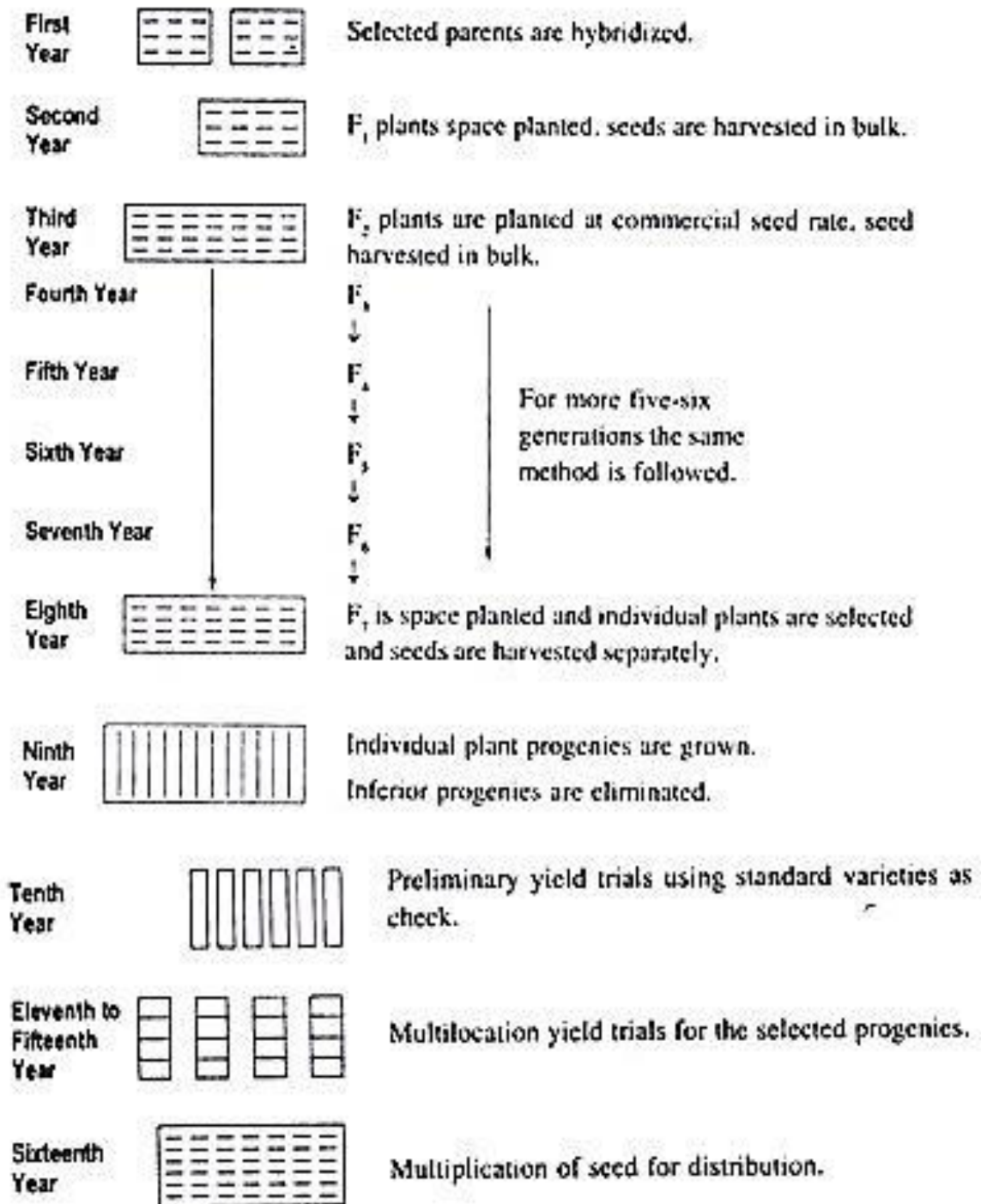


Fig.10.1: Breeding procedure of bulk population method

# **Experiment: Handling of segregation populations: Backcross method**

**Aim:** To know about how to handle segregating population of backcross method

## **Backcross method**

The plan of back cross method depend upon whether the gene being transferred is recessive or dominant. The plan for transfer of a dominant gene is quite simple than for recessive gene.

## **Requirements of a backcross programme**

1. Existence of a good recurrent parent variety which requires improvement is some qualitatively inherited character or a quantitative character with high heritability.
2. A suitable donor parent must be available possessing the character or characters to be transferred in a highly intense form.
3. High expressivity of the character under transfer through several back crosses in the genetic back ground of the recurrent parent.
4. The character to be transferred must have high heritability preferably determined by one or few genes.
5. Simple testing technique for detecting the presence of the character under transfer.
6. Recoveries of the recurrent genotype in a reasonable number of back cross generations.

## **I. Transfer of Dominant Gene**

Let us suppose that a high yielding and widely adopted variety 'A' is susceptible to stem rust ( $rr$ ) and another variety 'B' is poor yielding but resistant to stem rust ( $RR$ ) i.e. dominant to susceptibility. In this back cross programme rust resistance trait is transfer from donor parent (B) into a recurrent parent (A).

### **1) Hybridization**

Variety 'A' is crossed with variety 'B' in which variety 'A' is used as female parent which is recurrent and variety 'B' is used as donor parent.

### **2) F1 Generation**

During the second year F1 plants are backcrossed to variety 'A' since all the F1 plants will be heterozygous for rust resistance. Selection for rust resistance is not necessary.

### **3) First Back Cross Generation**

In the third year half of the plant would be resistant and remaining half would be susceptible to stem rust, rust resistant plants are selected and backcross to variety 'A'.

### **4) BC2 –BC6 Generation**

In each backcross generation, segregation would occur for rust resistance. Rust resistant plant are selected and backcrossed to the variety 'A' selection for plant type of variety 'A' may be practiced particularly in BC2 and BC3 generation.

### **5) BC6 Generation**

On an average the plant will have 98490 genes from variety A rust resistant plants are selected and selfed, their seeds are harvested separately.

### 6) BC6 F2 Generation

Individual plant progenies are grown from the selected plants. Rust resistance once plant, which are similar to variety 'A' are selected and selected plants are harvested separately.

### 7) BC5 F3 Generation

Individual plant progenies are grown homozygous progenies resistant to rust and similar to plant type of variety 'A' harvested in bulk. Several similar progenies are mixed to constitute the new variety.

### 8) Yield Test

The new variety is tested in replicated yield trials along with the variety 'A' as a check. Plant type dates of flowering date of maturity, quality, etc are critically evaluated. The new variety would be identical to variety 'A' in performance. Therefore detail yield test are not required, and the variety may be directly released for cultivation.

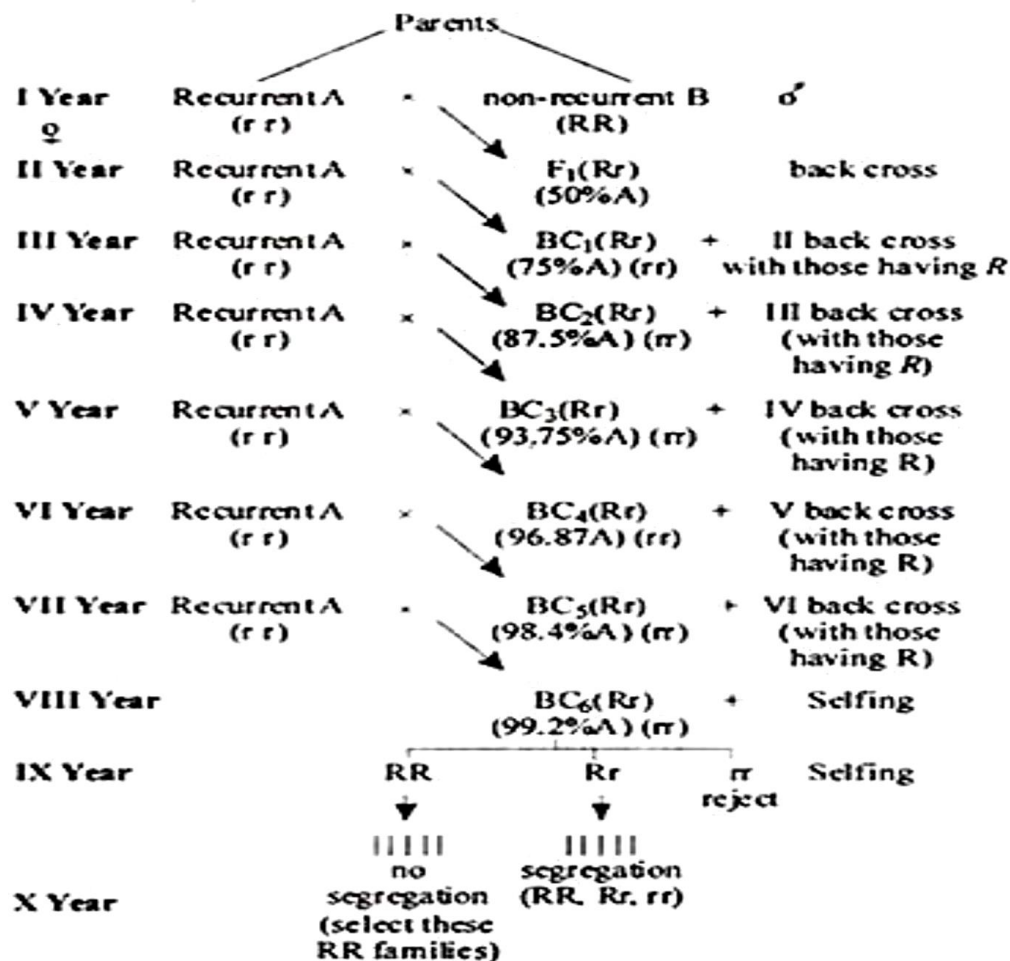


Fig. 11.1: Different steps involved in the back cross method when a dominant trait is transferred from variety B to an otherwise improved variety A



## **II. Transfer of Recessive Gene:**

When rust resistant is due to a recessive gene, all the backcross cannot make one after other. After the first backcross and after every two backcrosses  $F_2$  must be grown to identify the rust resistant plants. The  $F_1$  and the back cross progenies are not inoculated with rust because they would be susceptible to rust. Only  $F_2$  is tested for rust resistant.

### **1) Hybridization:**

The recurrent parent is crossed with rust resistant donor parent. The recurrent parent is generally used as female. i.e. ( $rr \times RR$ ).

### **2) F1 Generation:**

$F_1$  plants are backcrossed to the recurrent parent.

### **3) BC1 Generation:**

If rust resistance is recessive all the plant will be rust susceptible. Therefore, there is no test for rust resistance. All the plants are self- pollinated.

### **4) BC1 (F2) Generation:**

Rust resistance plants are selected and backcrossed with recurrent parent. i.e variety 'A'. Selection is made for the plant type and other characteristics of the variety 'A'.

### **5) BC2 Generation:**

No rust resistance test, plants are selected, which is identical to the recurrent parent (A) and backcrossed with the recurrent parent.

### **6) BC3 Generation:**

No disease resistance test. The plants are self – pollinated to raise  $F_2$ . selection is made for the plant type identical to variety 'A'.

### **7) BC3 F2 Generation:**

Plants are inoculated with stem rust. Rust resistant plant, similar to 'A' are selected and backcrossed to variety 'A'.

### **8) BC4 Generation:**

No rust resistance test plants are backcrossed to variety 'A'.

### **9) BC5 Generation:**

No rust resistance test plants are self-pollinated to raise  $F_2$  generation.

### **10) BC5 (F2) Generation:**

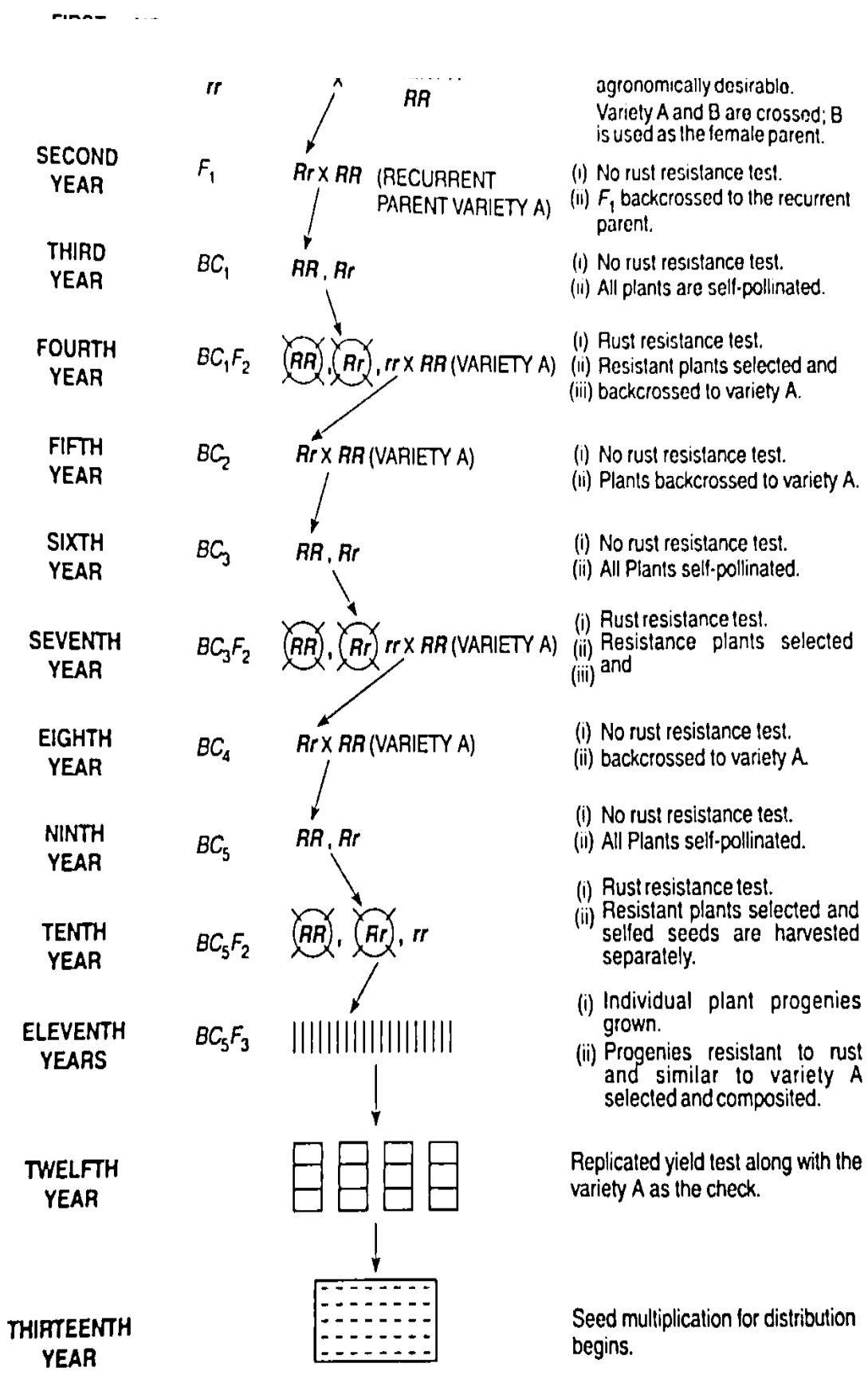
Plants are subjected to rust epidemic, resistance plant for rust and having similar characteristic of variety. 'A' is selected and self-seed are harvested separately.

### **11) BC5 (F3):**

Individual plant progenies are grown and subjected to rust epiphytotic selection is done for rust resistance and for characteristics of variety 'A' seeds from several similar rust resistant homozygous progenies are mixed to constitute new variety.

### **12) Yield Test:**

Same as in case of dominant gene transfer.



**Fig. 11.2: A generalised scheme for the transfer a recessive gene for disease resistant (here rust resistance) through the backcross method in a self-pollinated crop.**