

Marker-assisted selection in plant breeding

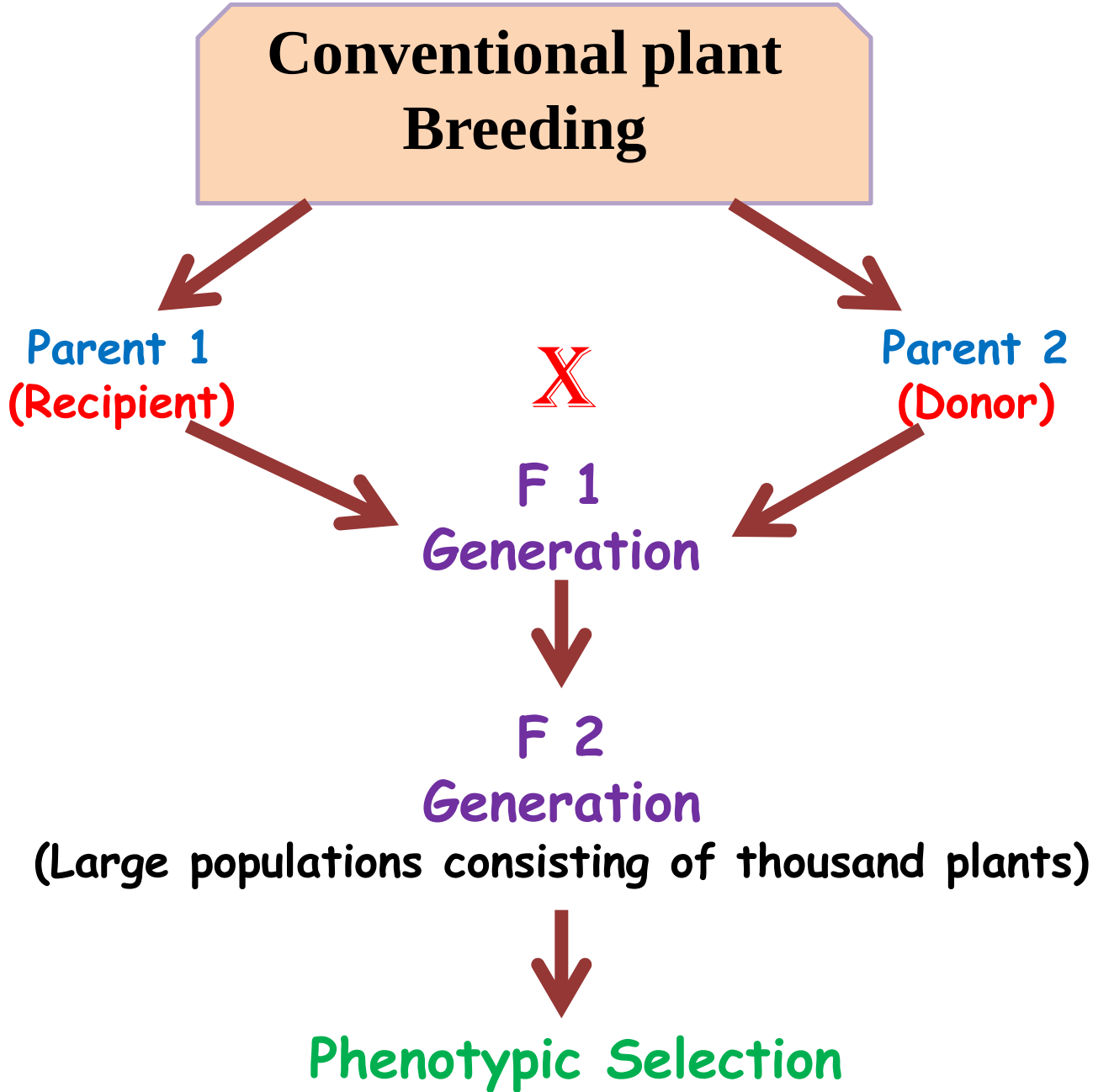


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Marker Assisted Selection

- Marker-assisted selection refers to the manipulation of genomic regions that are involved in the expression of traits through DNA markers.
- The principal concept on which Marker Assisted Selection (MAS) works on is that the presence of a gene from the presence of a marker must be tightly linked to the gene of interest.
- If the marker and the gene are located far apart then the possibility of transmitting together to the progeny individuals will be reduced.
- The important traits that are most looked for are tolerance to abiotic stresses, resistance to pathogen, insects and nematodes, quality parameters and quantitative traits.

Conventional plant Breeding



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graph TD; A[Conventional plant Breeding] --> B[Parent 1  
(Recipient)]; A --> C[Parent 2  
(Donor)]; B --> D[F 1  
Generation]; C --> D; D --> E[F 2  
Generation  
(Large populations consisting of thousand plants)]; E --> F[Phenotypic Selection];
```

Parent 1
(Recipient)

X

Parent 2
(Donor)

F 1
Generation

F 2
Generation

(Large populations consisting of thousand plants)

Phenotypic Selection

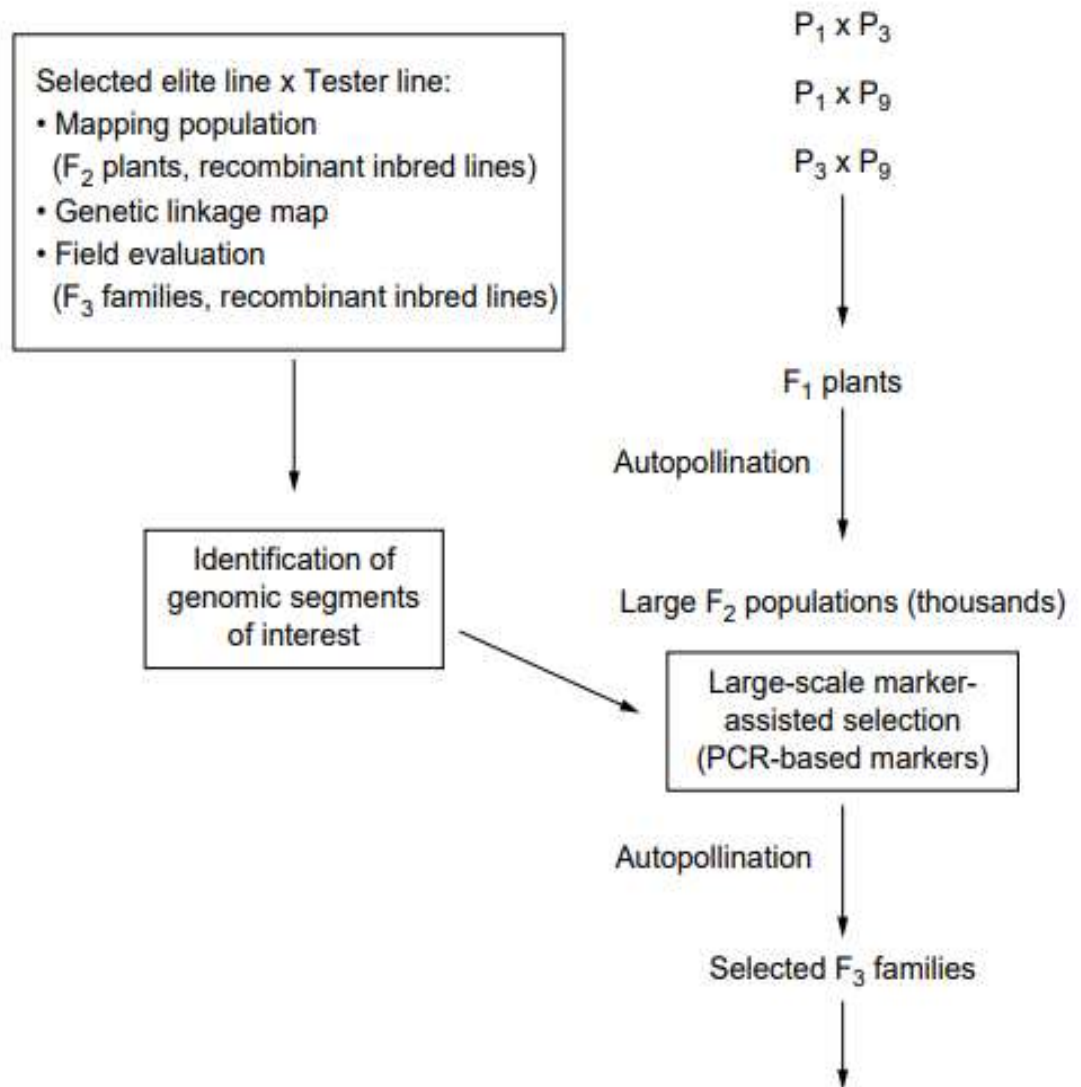
Phase I

Elite parental line selection ($P_1, P_2 \dots P_n$):

- Genetic experimental design (e.g. diallel)
- Fingerprinting

Phase II

Crosses from selected elite lines

**Phase III**

Population and line development

population development
parental selection and hybridization



QTL mapping
linkage map construction
phenotypic evaluation for trait(s)
QTL analysis



QTL validation
confirmation of position and effect of QTLs
verification of QTLs in independent
populations and testing in different genetic
backgrounds
fine mapping



marker validation
testing of markers in important breeding
material
identification of 'toolbox' of polymorphic
markers



marker-assisted selection

- **Pre-Requisites:**

There are certain pre-requisites for marker assisted selection such as:

- (i) a tight linkage between molecular marker and gene of interest (1cM or less)
- (ii) high heritability of the gene of interest.
- (iii) An efficient means of screening large populations for molecular markers should be available.
- (iv) High reproducibility across laboratories.
- (v) It should be cost effective.

- **Markers Used:**

MAS makes use of various types of molecular markers. The most commonly used molecular markers include:

- amplified fragment length polymorphisms (AFLP),
- restriction fragment length polymorphisms (RFLP),
- random amplified polymorphic DNA (RAPD),
- simple sequence repeats (SSR) or micro satellites,
- single nucleotide polymorphisms (SNP), etc.

The use of molecular markers differs from species to species also.

Advantages of MAS

- Accuracy,
- Rapid Method,
- Non-transgenic Product,
- Identification of Recessive Alleles,
- Early Detection of Traits,
- Screening of Difficult Traits,
- Highly Reproducible,
- Small Sample for Testing,
- Gene Pyramiding
- Permits QTL Mapping

Table 1. Examples of marker-assisted backcrossing in cereals.

species	trait(s)	gene/QTLs	foreground selection	background selection	reference
barley	barley yellow dwarf virus	<i>Yd2</i>	STS	not performed	Jefferies <i>et al.</i> (2003)
barley	leaf rust	<i>Rphq6</i>	AFLP	AFLP	van Berloo <i>et al.</i> (2001)
barley	stripe rust	QTLs on 4H and 5H	RFLP	not performed	Toojinda <i>et al.</i> (1998)
barley	yield	QTLs on 2HL and 3HL	RFLP	RFLP	Schmierer <i>et al.</i> (2004)
maize	corn borer resistance	QTLs on chromosomes 7, 9 and 10	RFLP	RFLP	Willcox <i>et al.</i> (2002)
maize	earliness and yield	QTLs on chromosomes 5, 8 and 10	RFLP	RFLP	Bouchez <i>et al.</i> (2002)
rice	bacterial blight	<i>Xa21</i>	STS ^a	RFLP	Chen <i>et al.</i> (2000)
rice	bacterial blight	<i>Xa21</i>	STS ^a	AFLP	Chen <i>et al.</i> (2001)
rice	bacterial blight	<i>xa5</i> , <i>xa13</i> and <i>Xa21</i>	STS, CAPS	not performed	Sanchez <i>et al.</i> (2000)
rice	bacterial blight	<i>xa5</i> , <i>xa13</i> and <i>Xa21</i>	STS	not performed	Singh <i>et al.</i> (2001)
rice	bacterial blight + quality	<i>xa13</i> , <i>Xa21</i>	STS and SSR	AFLP	Joseph <i>et al.</i> (2004)
rice	blast	<i>Pi1</i>	SSR	ISSR ^b	Liu <i>et al.</i> (2003)
rice	deep roots	QTLs on chromosomes 1, 2, 7 and 9	RFLP and SSR	SSR	Shen <i>et al.</i> (2001)
rice	quality	waxy	RFLP ^a	AFLP	Zhou <i>et al.</i> (2003a)
rice	root traits and aroma	QTLs on chromosomes 2, 7, 8, 9 and 11	RFLP and SSR	RFLP and SSR	Steele <i>et al.</i> (2006)
rice	submergence tolerance	<i>Sub1</i> QTL	phenotyping and SSR ^a	SSR	Mackill <i>et al.</i> (2006)
rice	submergence tolerance, disease resistance, quality	<i>Subchr9</i> QTL, <i>Xa21</i> , <i>Bph</i> and blast QTLs and quality loci	SSR and STS	not performed	Toojinda <i>et al.</i> (2005)
wheat	powdery mildew	22 <i>Pm</i> genes	phenotyping	AFLP	Zhou <i>et al.</i> (2005)

^a Indicates recombinant selection performed to minimize linkage drag around target locus.^b ISSR and inter SSRs.

Few Papers On Marker Assisted Selection

PHILOSOPHICAL
TRANSACTIONS
OF
THE ROYAL
SOCIETY **B**

Phil. Trans. R. Soc. B (2008) **363**, 557–572
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Marker-assisted selection: an approach for precision plant breeding in the twenty-first century

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trends in plant science
update

technical focus

**Marker-assisted
selection: new
tools and
strategies**

Journal of Experimental Botany, Vol. 58, No. 2, pp. 351–360, 2007
*Integrated Approaches to Sustain and Improve Plant Production under
Drought Stress Special Issue*
doi:10.1093/jxb/erl214 Advance Access publication 6 December, 2006

 Journal of
Experimental
Botany

SPECIAL ISSUE PAPER

Marker-assisted selection to improve drought adaptation in maize: the backcross approach, perspectives, limitations, and alternatives

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Markers and marker techniques

Allele mining	Identification and access to allelic variation of a sequenced gene that affects the plant phenotype. The DNA sequence or parts of it are used for searching new allelic variants of the same (resistance) gene.
Diagnostic marker	Markers that could be used in multiple genetic backgrounds, ideally the marker-trait association is valid in all germplasm.
Genomic selection	Selection based on genomic breeding values that are calculated as the sum of the effects of dense genetic markers, or haplotypes of these markers, across the entire genome, thereby potentially capturing all QTL that contribute to variation in a trait and similarly to multiple traits.
MABC: marker-assisted backcrossing	A molecular marker is used for indirect selection during backcrossing of the gene/QTL of interest into elite material.
Mapping	Assigning markers, genes, and/or QTL in order, indicating the relative distances among them, and assigning them to their linkage groups on the basis of their recombination values from all pairwise combinations.
Molecular marker	Specific fragments of DNA with a known location on the chromosome that can be identified within the whole genome and reveal neutral sites of variation. Markers that are in close linkage with a particular gene/QTL of interest can be used for MAS.
MAS: marker-assisted selection	A marker (morphological, biochemical, or one based on DNA/RNA variation) is used for indirect selection of a gene/QTL.
Perfect marker	A marker without recombination to the gene of interest, ideally drawn directly from the gene sequence.
QTL: quantitative trait locus/loci	Location of a gene that affects a trait measured on a quantitative (linear) scale. QTL are identified via statistical procedures that integrate genotypic and phenotypic data.
SNP: single nucleotide polymorphism	DNA sequence variation occurring when a single nucleotide in a shared sequence differs between members of a biological species or paired chromosomes in an individual.
SSR: single sequence repeat	Also called microsatellite. It consists of a specific sequence of DNA nucleotides that contain tandem repeats, e.g., GTGTGTGTGTGT would be referred to as (GT) ₆ . Alleles at a specific locus can differ in the number of repeats.

Limitations of MAS

- MAS is not cost effective as it requires well equipped laboratory.
- MAS requires well trained manpower for handling of sophisticated equipment.
- The detection of various linked DNA markers (AFLP, RFLP, RAPD, SSR, SNP etc.) is a difficult, laborious and time consuming task.
- MAS sometimes involves use of radioactive isotopes in labeling of DNA, which may lead to serious health hazards.
- It has been reported that MAS may become less efficient than phenotypic selection in the long term.