**Molecular plant breeding**

**Molecular breeding** may be defined as the use of molecular markers, in conjunction with linkage maps and genomics, to select plants with desirable traits on the basis of genetic assays.

Molecular breeding gained new momentum in the 1980s and has since made rapid progress, with the evolution of DNA marker technologies.

Molecular markers are used for several purposes in plant breeding.

1. **Gaining a better understanding of breeding materials and breeding system**. The success of a breeding program depends to a large extent on the materials used to initiate it. Molecular markers can be used to characterize germplasm, develop linkage maps, and identify heterotic patterns. An understanding of the breeding material will allow breeders to select the appropriate parents to use in crosses. Usually, breeders select genetically divergent parents for crossing. Molecular characterization will help to select parents that are complementary at the genetic level. Molecular markers can be especially useful in identifying markers that co-segregate with QTLs (quantitative trait loci) to facilitate the breeding of polygenic traits.
2. **Rapid introgression of simply inherited traits**. Introgression of genes into another genetic background involves several rounds of tedious backcrosses. When the source of desirable genes is a wild species, issues of linkage drag becomes more important because the dragged genes are often undesirable, requiring additional backcrosses to accomplish breeding objectives. Using markers and QTL analysis, the genome regions of the wild genotype containing the genes encoding the desirable trait can be identified more precisely, thereby reducing the fragment that needs to be introgressed, and consequently reducing linkage drag.
3. **Early generation testing**. Unlike phenotypic markers that often manifest in the adult stage, molecular markers can be assayed at an early stage in the development of the plant. Breeding for compositional traits such as high lysine and high tryptophan genes in maize can be advanced with early detection and selection of desirable segregants.
4. **Unconventional problem-solving**. The use of molecular markers can bring about novel ways of solving traditional problems, or solving problems traditional breeding could not handle. When linkage drag is recessive and tightly linked, numerous rounds of backcrosses may never detect and remove it. Disease resistance is often a recessive trait. When the could be difficult to remove by traditional backcross procedures. Marker analysis can help to solve the problem, as was done by J. P. A. Jansen when he introgressed resistance to the aphid *Nasonovia ribisnigi* from a wild lettuce *Lactuca virosa* by repeated backcrosses. The result of the breeding was a lettuce plant of highly undesirable quality. The recessive linkage drag was removed by using DNA markers flanking the introgression to preselect for individuals that were recombinant in the vicinity of the gene. The lifespan of new cultivars can be extended through the technique of **gene pyramiding** (i.e., transferring multiple disease-resistance genes into one genotype) for breeding disease-resistant cultivars. Marker-assisted backcross can be used to achieve this rapidly, especially for genes with indistinguishable phenotypes.
5. **Plant cultivar identification**. Molecular markers are effective in cultivar identification for protecting proprietary rights as well as authenticating plant cultivars. The types of molecular markers are discussed next.

**Molecular markers**

Plant breeders use **genetic markers** (or simply markers) to study genomic organization, locate genes of interest, and facilitate the plant breeding process.

**Concept of markers**

Genetic markers are simply landmarks on chromosomes that serve as reference points to the location of other genes of interest when a genetic map is constructed. Breeders are interested in knowing the association (linkage) of markers to genes controlling the traits they are trying to manipulate. The rationale of markers is that an easy-to-observe trait (marker) is tightly linked to a more difficult-to-observe and desirable trait. Hence, breeders select for the trait of interest by indirectly selecting for the marker (that is readily assayed or detected or observed). When a marker is observed or detected, it signals that the trait of interest is present (by association).

Genetic markers can be detected at both the morphological level and the molecular or cellular level – the basis for classification of markers into two general categories as **morphological markers** and **molecular markers**. Morphological markers are manifested on the outside of the organism as a product of the interaction of genes and the environment (i.e., an adult phenotype). On the other hand, molecular markers are detected at the subcellular level and can be assayed before the adult stage in the life cycle of the organism. Molecular markers of necessity are assayed by chemical procedures and are of two basic types – **protein** and **DNA markers**. Markers are indispensable in genetic engineering, being used in selection stages to identify successful transformation events.

**Types of markers**

1. **Morphological markers**
   * Seed color e.g. Kernel color in maize
   * Function based e.g. Plant height associated with salt tolerance in rice

**Limitations**

* 1. Most phenotypic markers are undesirable in the final product (Yellow color in maize).
  2. Dominance of the markers: homozygotes/ heterozygotes not distinguishable
  3. Sometimes dependent on the environment for expression e.g. Height of plants

1. **Molecular markers**
   * Non-DNA such as isozyme markers: Restricted due limited number of enzyme systems available.
   * DNA based markers: Markers based on the differences in the DNA profiles of individuals.

Some molecular markers are pieces of DNA that have no know function or impact on plant performance (Linked Markers):

* Detected via mapping.
* Linked markers are near the gene of interest and are not part of the DNA of the gene.

Other markers may involve the gene of interest itself (Direct Markers):

* Based on part of the gene of interest.
* Hard to get but great once you have it.

**Requirements for a useful molecular marker**

1. Molecular markers must be tightly linked to a target gene. The linkage must be really tight such that the presence of the marker will reliably predict the presence of the target gene.
2. The marker should be able to predict the presence of the target gene in most if not all genetic backgrounds.

**Marker-assisted breeding**

Molecular markers may be used in several ways to make the plant breeding process more efficient. The adoption of a **marker-assisted selection** (MAS) or **marker-aided selection** in a breeding program hinges on the availability of useful molecular markers. Fortunately, this resource is becoming increasingly available to many species, thanks to the advances in biotechnology. This breeding approach is applicable to improving both simple and complex traits, as a means of evaluation of a trait that is difficult or expensive to evaluate by conventional methods. The basic requirement is to identify a marker that co-segregates with a major gene of the target trait. MAS is more beneficial to breeding quantitative traits with low heritability.

**Conditions under which MAS is valuable**

1. Low heritability traits
2. Traits too expensive to score: Soybean Cyst Nematode (SCN) resistance. Young (1999)
3. Recessive genes: Pyramiding of dominant and recessive genes conferring resistance to important crop diseases which would otherwise be very difficult
4. Multiple genes (Quantitative traits): QTLs underlying phenotypic and physiological traits can be traced using markers. Although QTL mapping is tedious, markers once identified can be used fast and accurately to detect the QTLs of interest.
5. Quarantine: No need to grow plants to screen for viral diseases that can not be visually detected, and small tissues can be used for DNA typing.

**Advantages of MAS**

1. Improvement of response to selection (Rs)
2. Assays require small amount of tissue, therefore no destructive sampling.
3. Use of codominant markers allows accurate identification of individuals for scoring without ambiguity
4. Multiple sampling for various QTLs is possible from same DNA prep
5. Can assay for traits before they are expressed, e.g. before flowering
6. Time saving.

**Limitations of MAS**

1. Cost of equipment, reagents and personnel.
2. Data collected in the field is assumed to be normally distributed, but usually is not.
3. Integration of the DNA information into existing systems is difficult.
4. Linkage drag. As the marker distance from the target gene increases, more of the donor DNA is retained in the desired background resulting in need for more backcrosses.