



Molecular Markers: An Advanced Technique for Crop Improvement

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Molecular markers are fragments of nuclear, mitochondrial or chloroplast DNA, which are linked with the gene of interest and hence acting as representative of the gene. Molecular marker analysis based on polymorphism in DNA, can be considered as objective measures of genetic variations and have catalyzed research in a variety of disciplines such as phylogeny, taxonomy, ecology, genetics and plant breeding. Different types of molecular markers with different properties exist, each with its own advantages and disadvantages. Application of molecular markers as a novel tool for crop improvement will definitely improve the ability of capturing desirable characters in new progenies of plants.

Introduction

Molecular markers are small segments of DNA that flag the presence or absence of particular traits or character. The application of molecular markers is based on naturally occurrence of variation in DNA. The molecular marker system can be efficiently used in the breeding programme for the development of high yielding varieties. Different types of molecular markers with different properties exist, each with its own advantages and disadvantages (Karp *et al.*, 1997). There is a long list of molecular marker system that has been developed so far. The most important among them are: RFLP, AFLP, SSRs, ISSRs, SNPs and ESTs etc. (Chhabra and Somveer, 2002). Suitable marker system should have following desirable properties:

1. Easy, fast and cheap to use and analysis,
2. It should require small amount of DNA,
3. Co-dominance,
4. It should have repeatability/reproducibility of results,
5. High level of polymorphism and
6. Occurrence and uniform distribution throughout the genome

In the marker system variation in DNA or polymorphism for a specific region of DNA sequence can be identified based on the product, such as band size and mobility. Markers are identifying the variation in cellular level so called as molecular markers also known as DNA markers. There is no any single molecular marker which gave all these characters, for that a wide range of molecular techniques are available to detect the variation in the DNA level.

Types of Markers

Non PCR based: Marker system that does not require PCR amplification of genomic DNA. Those are also based on DNA-DNA hybridisation between the DNA or RNA probe and total genomic DNA, e.g. Restriction Fragment Length Polymorphism (RFLP).

PCR based: Those based on the PCR amplification of genomic DNA fragments; these techniques are Random Amplified Polymorphic DNA (RAPD), Amplified Fragment Length Polymorphism (AFLP), Simple Sequence Repeats (SSR), Inter Simple Sequence Repeats (ISSR), Arbitrarily Primed PCR (AP-PCR), Single Nucleotide Polymorphism (SNP), Sequence Characterised Amplified Region (SCAR), etc.

The selection and use of DNA markers in research and agricultural breeding is still a challenge for plant scientists. The comparative features, advantages and disadvantages of DNA markers are given in **Table 1**. A breeder has to make a decision for choice of a good molecular marker that meets the bets requirements according to the conditions and resources available for the breeding programme (Semagn *et al.*, 2006a).

Table 1. Comparison of most commonly used DNA marker systems in plants taken from Collard *et al.* (2005), Semagn *et al.* (2006a), Xu (2010) and others.

Feature and Description	RFLP	RAPD	AFLP	SSR	SNP
Genomic abundance	High	High	High	Moderate to High	Very High
Genomic coverage	Low copy coding region	Whole Genome	Whole Genome	Whole Genome	Whole Genome
Expression/inheritance	Co-dominant	Dominant	Co-dominant/Dominant	Co-dominant	Co-dominant
Number of loci	Small (<1000)	Small (<1000)	Moderate (<1000)	High (1000-10000)	Very High (>100000)
Level of polymorphism	Moderate	High	High	High	High
Type of polymorphism	Single base changes, indels	Single base changes, indels	Single base changes, indels	Changes in length of repeats	Single base changes, indels
Cloning and/or Sequencing	Yes	No	No	Yes	Yes
PCR-based	Usually no	Yes	Yes	Yes	Yes
Radioactive detection	Usually yes	No	Yes or No	Usually No	No
Reproducibility/Reliability	High	Low	High	High	High
Genotyping Throughput	Low	Low	High	High	High
Quality of DNA Required	High	Moderate	High	Moderate to High	High
Cost per analysis	High	Low	Moderate	Low	Low
Primary application	Genetics	Diversity	Diversity and Genetics	All purposes	All purposes

Applications of Molecular Marker in the Agriculture

Most important applications of molecular markers are in the marker assisted breeding which include:

Genetic diversity analysis: Genetic diversity analysis is a powerful tool for breeders to identify different heterotic groups and to increase the efficiency of finding crosses with good specific combinability (SCA). The result are visualize in the form of dendrogram (Karp *et al.* 1997).

Genotyping, variety identification and seed purity analysis: Genotyping using DNA markers can be considered as the most reliable method for the identification of lines and varieties. Therefore, the DNA fingerprinting methods can be used to analyze the purity of seed lots (Chhabra, 2001).

Isolation of markers tightly linked to the specific gene i.e. gene tagging: Identification of markers for disease resistance, high yield, increased aroma, pesticide, herbicide resistant gene is now possible through molecular marker system (Michelmore *et al.*, 1991).

Marker assisted back cross breeding (MABC): MABC aims to transfer one or few genes or quantitative trait regions from wide germplasm in to superior cultivar to increase the performance (Semagn *et al.*, 2006b).

Conclusion

To fulfill the need of food for the growing world population there is need to develop the high yielding hybrids, varieties and superior population of food crops. This can be achieved through the use of molecular marker system. Molecular markers are independent on the environment so it offers important tool for breeders to directly select for the genotype. The efficiency of molecular marker depends upon the linkage relationship of marker with target gene and resolving power of co-segregating molecular marker. However, the Marker Assisted Selection technology should be economically affordable and easy to practice in laboratory.

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