

## Genetic Markers: An Efficient Tool

**Monika Karnawat\*, Rakesh Kumar Meena, Deepak Nagar, Rohitashav Nagar,  
Brahmanand Bairwa and Anubhav Galav**

Career Point University, Alaniya, Kota (324 005), Rajasthan, India

\*Corresponding Author E-mail: [monika.karnawat@cpur.edu.in](mailto:monika.karnawat@cpur.edu.in)

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### ABSTRACT

*Hereditary markers are one of the advances which have happened in the genomics time. Among inherited markers, sub-atomic markers as a result of their bounty, are the most by and large used them. Headway of sub-atomic markers has unfathomably changed inherited characteristics and plant raising. Inherited markers show the innate differentiations between different organs or species. A couple of assessments which were driven during the latest decade of the 20th century reported different DNA markers that have been utilized in plant reproducing programs. Beside the use of atomic markers in the advancement of linkage maps, they have different applications in plant raising, for instance, assessing the genetic assortments inside cultivars and germplasm. The most entrancing utilization of atomic markers is marker-helped assurance (MAS). Suitable DNA markers should be polymorphic in the DNA level and can be imparted in all tissues, organs, and diverse developmental stages. Differentiated and traditional raising tasks, atomic markers can construct the efficiency and feasibility of imitating programs. To research the data about sub-atomic markers, a couple of audits have been distributed over the latest thirty years; regardless, every one of these surveys were expected for investigators with bleeding edge data on sub-atomic innate characteristics. This audit is proposed to be a once-over of late upgrades in atomic markers and their applications in plant reproducing and is committed to early scientists with insufficient data on sub-atomic markers.*

**Keywords:** *Molecular marker, QTL mapping, MAS, Functional marker, Genomic selection, Genome editing.*

### INTRODUCTION

Data about the genetic assortments present inside besides, between various plant peoples and their structure and level can expect a helpful employment in the capable utilization of plants. The formative establishment, method of value stream, mating system and

people thickness are critical components used in the acknowledgment of structure and level of these assortments. To explore the different assortment and other huge credits, different kinds of agronomic and morphological boundaries have been used adequately.

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During the latest thirty years, the world has seen a quick addition in the data about the plant genome courses of action and the physiological and sub-nuclear employment of various plant characteristics, which has vexed the sub-nuclear innate characteristics and its capability in plant reproducing projects.

#### **Genetic markers:**

Nuclear markers are colossal updates in the field of plant raising. The Molecular marker is a quality or DNA movement with a realized chromosome zone controlling a specific quality or trademark (Taiz, 1991). Nuclear markers are determinedly related with the objective quality and they go most likely as sign or of course hails. Sub-nuclear markers are totally accumulated into two groupings: old style markers and DNA/atomic markers. Morphological, cytological and biochemical markers are sorts of old style markers and a few instances of DNA markers are imperative section length polymorphism (RFLP), redesigned part length polymorphism (AFLP), clear assembling goes over (SSRs), single-nucleotide polymorphism (SNP) and reasonable collection bunches advancement markers.

#### **Traditional markers**

##### **1. Morphological markers**

Morphological markers can apparently perceive attributes like seed structure, bloom concealing, improvement inclination and other critical agronomic characteristics. Morphological markers are easy to use, with no need for unequivocal instruments. They don't need a specific biochemical what's more, sub-nuclear strategy. Raisers have used such kind of markers successfully in the raising undertakings for various yields. Crucial drawbacks of morphological markers are: they are compelled in number, influenced by the plant improvement stages and distinctive characteristic factors. Since obsolete events, individuals have adequately used diverse morphological markers to investigate the assortment for use in plant raising.

##### **2. Cytological markers**

Markers that are associated with assortments present in the numbers, banding plans, size,

shape, solicitation and position of chromosomes are known as cytological markers. These assortments reveal contrasts in the allocations of euchromatin and heterochromatin (Mohammadi-Nejad et al., 2008). For example, G bunches are made by Giemsa stain, Q bunches are conveyed by quinacrine hydrochloride and R bunches are the pivoted G gatherings. These chromosome achievements can be used in the detachment of average and changed chromosomes. Such markers can similarly be used in the ID of linkage get-togethers and in actual planning.

##### **3. Biochemical markers**

Biochemical markers, or isozymes, are multi-sub-nuclear kinds of mixes which are coded by various characteristics, anyway have comparable limits. They are allelic assortments of mixes and consequently quality and genotypic frequencies can be evaluated with biochemical markers. Biochemical markers have been successfully applied in the area of innate respectable assortment, people structure, and quality stream and people improvement. They are co-overwhelming, easy to use and keen. In any case, they are less in number; they perceive less polymorphism and they are affected by various extraction draws near, plant tissues and different plant advancement stages (Boyer, 1982).

##### **4. DNA markers**

DNA markers are nucleotide groupings and can be examined through the polymorphism present between the nucleotide groupings of various people. Addition, erasure, point transformations duplication and translocation are premise of these polymorphisms; be that as it may, they do not really influence the action of qualities. A perfect DNA marker ought to be co-prevailing, uniformly dispersed all through genome, profoundly reproducible and having capacity to recognize more elevated level of polymorphism (Rejeb et al., 2014).

Classification of DNA markers are classified into various groups on the basis of:

(1) Mode of gene action (co-dominant or dominant markers)

(2) Method of detection (hybridization-based molecular markers or polymerase chain reaction (PCR) - based markers)

(3) Mode of transmission (paternal organelle inheritance, maternal organelle inheritance, bi-parental nuclear inheritance or maternal nuclear inheritance)

Various kinds of DNA atomic markers have been created and effectively applied in

hereditary qualities and rearing exercises in different farming harvests. The accompanying area gives some short data related with atomic markers dependent on their technique for location. Differences of the important characteristics of most commonly used molecular markers are described in Table 1.

**Table 1: Comparison of Characteristics in different types of markers**

S. No.	Characteristics	RFLP	RAPD	AFLP	SSR	SNP
1.	Co-dominant/Dominant	Co-dominant	dominant	Dominant	Co-dominant	Co-dominant
2.	Reproducibility	High	High	Intermediate	High	High
3.	Polymorphism level	Medium	very high	High	high	high
4.	Marker index	Low Medium	High	Medium	Medium	high
5.	Genome abundance	High	Very high	Very high	Medium	Very high
6.	Cost	High	Less	High	High	Variable
7.	Sequencing	Yes	No	No	Yes	Yes
8.	Status	Past	Past	Past	Present	Present
9.	PCR requirement	No	Yes	Yes	Yes	Yes
10.	Visualization	Radioactive	Agarose gel	Agarose gel	Agarose gel	SNP-VISTA
11.	Genomic coverage	Low copy coding region	Whole genome	Whole genome	Whole genome	Whole genome
12.	Amount of DNA required	10 µg–50	100 ng–1	100 ng–1	120 ng–50	≥ 50 ng
13.	Type of polymorphism	Single base changes, indels	Single base changes, indels	Single base changes, indels	Changes in length of repeats	Single base changes, indels
14.	Type of probes/primers	Low copy DNA cDNA clone	Usually 10 bp random nucleotides	Specific sequence	Specific sequence	Allele-specific PCR primers
15.	Proprietary rights required	No	Yes and licensed	Yes and licensed	Yes and some licensed	Yes and some licensed
16.	Suitable utility in diversity, genetics and breeding	Genetics Diversity	Diversity	All purposes	All purposes	All purposes

### **RFLP (Randomly amplified length polymorphism):**

RFLP is a distinction in homologous DNA successions that can be recognized by the nearness of sections of various lengths after absorption of the DNA tests being referred to with explicit limitation endonucleases. RFLP, as a sub-atomic marker, is explicit to a solitary clone/limitation protein blend. Most RFLP markers are co-predominant (the two alleles in heterozygous example will be identified) and profoundly locus-explicit (Mondini et al., 2009). This test is a marked DNA grouping that hybridizes with at least one pieces of the

processed DNA test after they were isolated by gel electrophoresis, in this way noteworthy an extraordinary smearing design trademark to a particular genotype at a particular locus. Short, single-or low-duplicate genomic DNA or cDNA clones are ordinarily utilized as RFLP tests (Larcher, 2003). The RFLP tests are habitually utilized in genome mapping and in variety investigation.

### **RAPD (Random access polymorphic DNA):**

This procedure was created by Williams et al. and Welsh and McClelland freely. Amplification of genomic DNA is accomplished by PCR utilizing single, short

(10 nucleotide) and arbitrary preliminary. During PCR, amplification happens when two hybridization locales are like one another and inverse way (De Leon et al., 2017). These amplified pieces are absolutely subject to the length and size of both the objective genome and the preliminary. The chose groundwork ought to have least 40% GC content, as a preliminary having under 40% GC substance will most likely not withstand the toughening temperature where DNA extension happens by DNA polymerase. For the perception, the PCR item is then isolated in agarose gel recolored with ethidium bromide. Polymorphism present either at or between preliminary restricting locales can be recognized in the electrophoresis by confirming the nearness or nonappearance of specific groups. The amount and nature of DNA, PCR cradle, magnesium chloride focus, toughening temperature and Taq DNA (sort of DNA polymerase) are some significant elements influencing the reproducibility of haphazardly amplified polymorphic DNA (RAPD) markers.

#### **AFLP (Amplified fragment length polymorphism):**

The limitations present in the RAPD and RFLP technique were overcome through the development of AFLP markers. AFLP markers combine the RFLP and PCR technology, in which digestion of DNA is done and then PCR is performed. AFLP markers are cost effective and there is no need of prior sequence information. In AFLP, both good-quality and partly degraded DNA can be used; however, this DNA should not contain any restriction enzymes or PCR inhibitors. For more information, see previous studies. In AFLP, two restriction enzymes (a frequent cutter and a rare cutter) are used for the cutting of DNA. Each end of the resulting fragments is ligated with the oligonucleotides. Oligonucleotides are short nucleic acid fragments used for the ligation in PCR. One end is specific for the rare cutter (6-bp recognition site) and the other one, for the frequent cutter (3-bp recognition site). This will lead to the amplification of only those fragments which have been cut by these cutters. For the development of primers,

known sequences of adapters are used. Adapters are actually short, enzyme specific DNA sequences generally used for fishing an unknown DNA sequence. After performing PCR, visualization is done in either agarose gel or polyacrylamide gel stained with AgNO<sub>3</sub> or by autoradiography.

#### **SSR or Microsatellites:**

Microsatellites are additionally called as SSRs; short pair rehashes and straightforward grouping length polymorphisms. SSRs are pair rehash themes of 1–6 nucleotides that are available liberally in the genome of different taxa (Mateu-Andres et al., 2005). Microsatellites can be mononucleotide (A), dinucleotide (GT), trinucleotide (ATT), tetranucleotide (ATCG), pentanucleotide (TAATC) and hexanucleotide (TGTGCA). Microsatellites are conveyed in the genome; nonetheless, they are likewise present in the chloroplast and mitochondria. Studies have likewise confirmed the nearness of SSRs in protein-coding qualities and communicated succession labels (ESTs). SSRs speak to the lesser redundancy per locus with higher polymorphism level. This high polymorphism level is because of the event of different quantities of rehashes in microsatellite locales and can be recognized effortlessly by PCR. Event of SSRs might be because of slippage of single-strand DNA, recombination of twofold strand DNA, move of versatile components (retro transposons) and crisscrosses (Liu et al., 2014). Regular themes present in SSRs are Mono: A, T; Di: AT, GA; Tri: AGG; Tetra: AAAC. For the most part the groupings which are flanking the SSRs are preserved and are utilized in the advancement of groundworks. Improvement of a genomic library and sequencing a portion of the considered genome will bring about the advancement of these ground works. The advancement of SSR markers includes the improvement of a SSR library and afterward identification of specific microsatellites. After this, the recognition of great districts for preliminary planning is done and afterward PCR is performed (Kebriyae, 2012). Translation and assessment of banding designs are performed and evaluation of PCR items is performed for examination of

polymorphism. SSR markers are viewed as a marker of decision, as they are co-predominant, with high reproducibility and more prominent genome plenitude, and they can be utilized efficiently in plant mapping considers (Younis et al., 2014).

### **Uses of molecular markers in plant sciences**

#### **Development and phylogeny:**

Before, starting investigations identified with development were absolutely subject to the land and morphological changes among the creatures. Progressions in the procedures of atomic science offer stretched out data identified with the hereditary structure. For the recreation of a hereditary guide, so as to get full data about the phylogeny and development, sub-atomic markers are being utilized for an enormous scope these days. Sub-atomic investigations identified with phylogeny are to a great extent subject to chloroplast genome grouping information because of their straightforward and stable hereditary nature, making them perfect markers in the assessment of plant phylogeny.

#### **Examination of heterosis:**

Heterosis depicts the more noteworthy exhibition of offspring (F1) over the mean of the two crossed guardians. On the off chance that the impact in F1 is more noteworthy than that in its folks, such heterosis is known as positive heterosis; while where the impact in F1 is lower than in its folks, such kind of heterosis is known as negative heterosis (Semagn et al., 2006). Different examinations have been directed by utilizing atomic markers in different yield plants, for example, wheat, maize and assault seed, to explore the hereditary assorted variety and heterosis. Sub-atomic markers like SSRs have been utilized in the examination of decent variety and heterosis in rice (Mazur et al., 2010). As of late, SSR markers were applied so as to examine the heterotic gatherings and examples in rice (Cole, 2003). A few investigations have utilized transcriptome examination to break down the qualities associated with heterosis.

#### **Identification of haploid/diploid plants and cultivars genotyping:**

Haploids are plants having a solitary arrangement of gametophytic chromosomes

and diploids are plants with two duplicates of each homologous chromosome (Bayley, 1983). These haploid/twofold haploid (DH) plants are significant on the grounds that they are utilized as a mapping populace for quantitative quality loci (QTL) mapping and in other rearing and hereditary examinations. DH plants are significant in the joining of physical and hereditary maps and in this way permit the precise recognition of up-and-comer qualities of intrigue. The R1-nj (Navajo) anthocyanin shading marker has been effectively applied for the identification of haploids. Additionally, SSR and SNP markers have been applied to distinguish DH and genotypes of isogenic lines and half breeds.

#### **Hereditary mapping**

Hereditary mapping utilizes strategies for identification of the locus of a quality just as for assurance of the separation between two qualities. Quality mapping is considered as the significant region of examination in which atomic markers are utilized today. The guideline of hereditary mapping is chromosomal recombination during meiosis which brings about the isolation of qualities. Markers present near the quality of enthusiasm on a similar chromosome are known as connected markers.

### **CONCLUSION**

The latest 30 years have seen a reliable improvement in the sub-nuclear markers development from RFLP to SNPs and an assortment of display advancement based markers. Types of progress in the sequencing headways have provoked the improvement of NGS organizes that are ease with high throughput. Despite the presence of these particularly advanced nuclear genetic methodology, we are as yet not achieving our destinations. The principal clarification for this lies in incorrect phenotyping. High-throughput phenotyping methods deal with these issues by using light, cameras, sensors, PCs and incredibly changed contraptions for the grouping of accurate phenotypic data, which is a middle need to achieving our raising goals successfully. CRISPR development has

changed the plant raising and genetic characteristics and experts are focusing in on modifying the genomes of all monetarily critical plants. The coming years are presumably going to see continued with improvements in sub-nuclear marker development to make it more precise, helpful and useful to investigate the concealed study of various qualities of premium.

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