



P-ISSN: 2349-8528

E-ISSN: 2321-4902

IJCS 2017; 5(4): 1724-1729

© 2017 IJCS

Received: 19-05-2017

Accepted: 20-06-2017

Munnesh Kumar

Department of Genetics and Plant Breeding, Post Graduate Institute, Mahatma Phule Agricultural University, Rahuri, Ahmednagar, Maharashtra, India

Sharad V Pawar

State Level Biotechnology Centre, Mahatma Phule Agricultural University, Rahuri, Maharashtra, India

Prahlad Masurkar

Department Of Mycology and Plant Pathology, Institute of Agricultural Science, Banaras Hindu University, Varanasi Uttar Pradesh, India

SM Choudhary

Department of Horticulture, Mahatma Phule Agricultural University, Rahuri, Maharashtra, India

Molecular markers: A new frontier approaches in crop improvement

Munnesh Kumar, Sharad V Pawar, Prahalad Masurkar and SM Choudhary

Abstract

Over the past few decades, functional genomics research of plants have been screened entirely rising a revolution in the area of plant biotechnology. molecular markers are a new emerging trends for estimation of genomics and another omics. Now it is a recent frontier tools in crop improvements. The unique significant of molecular markers in development in the area of molecular based genetics and plant breeding. The molecular markers classified tremendous types and differences in their principals and methodologies and applications. In this article we attempt to review most of the different molecular markers that can be regularly utilised in different aspects of dissection of plant genome such as measuring of genetic diversity, gene tagging, QTL,S analysis phylogenetic analysis, DNA fingerprinting, mapping of genome, analysis of genome crop evolution, and plant genetic resources, population genetics, plant taxonomy, plant breeding, plant disease diagnostics, forensic markers assisted selection, recurrent selection, genome wide section, gene introgression, backcrossing, gene pyramiding and estimation of molecular heterosis. In this review molecular trades increasing crop production and while same time reducing crop failure for sustainability of crops.

Keywords: Molecular marker, genome, genetic diversity, plant breeding, gene introgression, Crop Improvement, polymorphism.

Introduction

Over the last two decades, molecular markers have created new revolution in crop improvement entire field of life science and applied science. DNA marker technology is the excellent tools have been created progressive tremendous and exciting. DNA-based markers have contribute precious innovative versatile tools in different area of plant taxonomy, plant physiology, embryology, diagnosis and forensic, genetic engineering, *etc.* (Schlotterer *et al.* 2004) [32]. DNA marker technology offered another application in fingerprinting genotypes, in determining seed purity and seed viability, in systematic sampling of germplasm, and in phylogenetic analysis. gene tagging, QTLs analysis, mapping of genome, analysis of genome crop evolution, and plant genetic resources, population genetics, plant disease diagnostics, forensic, markers assisted selection, recurrent selection, genome wide section, gene introgression, backcrossing, gene pyramiding and prediction of heterosis (Kesawat *et al.*) and impact on plant and animal genome research and plant breeding. They provide to hybrid performance prediction, and to establish the distinctiveness, uniform and stability of new cultivars prior to their registration. A further study, the molecular marker widely used in dissection of root morphology and genetic architecture of heritable traits are emerging from functional genomics programs, thanks to identification of genes and DNA polymorphisms that are involved (Morgante and Salamini, 2003) [24]. Molecular markers trends offer various versatile tremendous advantages over traditional phenotype markers. They are putative, stable and manifest in all tissues regardless of growth, dissection differentiation, development, or defense status of the cell are not confounded by the environment, pleiotropic and epistatic effects.

An ideal properties of DNA marker technology should have the following criteria:

- (1) be highly polymorphic nature and random distributed throughout the genome,
- (2) highly robust, co-dominant and reproducible,
- (3) abundant in nature, easy access and reliable markers
- (4) cheap, simple, quick assay and inexpensive
- (5) required small amounts of leaves and DNA sample
- (6) Neutral markers and functional polymorphisms
- (7) null interaction among markers
- (8) have linkage to distinct phenotypes and
- (9) require no prior information about the genome

Correspondence

Munnesh Kumar

Department of Genetics and Plant Breeding, Post Graduate Institute, Mahatma Phule Agricultural University, Rahuri, Ahmednagar, Maharashtra, India

of an organism. In this review studied we investigated molecular markers are new recent emerging trends utilised in crop breeding.

Various types of molecular maker

Molecular markers are broadly divided different three groups.

(i) Hybridization based e.g. RFLPs, (ii) PCR techniques based e.g. RAPD, AFLP, microsatellites or SSRs and (iii) Single

nucleotide polymorphisms (SNPs) (iv) Other types viz:(1) Mode of inheritance (genomic inheritance and cytoplasmic inheritance). (2) Mode of gene action (dominant or co-dominant markers). (3) Method of analysis (hybridization-based or PCR-based markers) and their application for genetic diverse purpose has opened up a new opportunity in the field of molecular bioscience.

Table 1: Overview of the relevant characteristics of some important molecular markers

| Characteristics | RFLP | RAPD | AFLP | SSR | ISSR | SCAR | CAPS | SSCP | SNP | DArT | TE - Anchor |
|-------------------------|----------------------|-------------|-------------------|---------------------------|------------|-------------|-------------|-------------|-------------|-------------|-------------|
| PCR base | no | yes | yes | yes | yes | yes | yes | yes | yes | yes | yes |
| Polymorphism level | moderate | moderate | moderate | low | moderate | low | variable | moderate | moderate | variable | variable |
| Locus specificity | yes | no | no | yes | no | yes | yes | yes | yes | yes | yes |
| Genomic abundance | moderate | moderate | moderate | moderate | moderate | moderate | moderate | moderate | highest | moderate | variable |
| Co-dominance | yes | no | no | yes | no | yes | yes | | yes | no | yes |
| Reproducibility | high | low | mid-high | high | mid-high | high | high | medium | | | |
| Required DNA Quantity | high | low | medium | low | low | low | low | low | low | low | low |
| Automation | no | yes | yes | yes | yes | yes | yes | | yes | yes | yes |
| Loci per assay | 1 to few | many | many | 1 to 20 | many | single | single | | 1 to 1000 | many | single |
| Used restriction enzyme | yes | no | no | no | no | no | yes | no | no | yes | no |
| Laborious | high | low | medium | low | low | low | low-mid | low-mid | low | low | low |
| Specialised equipment | radioactive isotopes | agarose gel | polyacrami de gel | agarose/ polyacramide gel | agaroe gel | agarose gel | agarose gel | agarose gel | agarose gel | microarr ay | agarose gel |

Restriction Fragment Length Polymorphism

RFLP is the first molecular markers most extensively used, firstly used to identification DNA sequence polymorphisms for genetic mapping of a temperature-sensitive mutation of adenovirus serotypes. They were first time used for genome mapping of human (Botstein *et al.* 1980) but were later used for plant genomes. This technique is based on restriction enzymes that reveal a pattern difference between DNA fragment sizes in individual organisms. In RFLP estimation, restriction enzyme-digested plant genomic DNA is used by gel electrophoresis and then blotted on nitrocellulose membrane. Specifically bands are visualized by hybridization with labelled probe. These probes are mostly single- or multi-locus probes of about 0.5-3.0 kb in size and plant species-specific, procured from a cDNA library or a genomic library. RFLP marker is co-dominant, analysis of linkage of DNA molecules, and detected DNA sequence homologous chromosomes. The major potential of RFLP markers are their co-dominant inheritance, high reproducibility, good transferability, locus specific and provides that allow synteny analysis. Major drawback of RFLP analysis: it needs of high quantity and quality of DNA (Young *et al.* 1992) [46]. RFLPs can be used in genetic diversity and phylogenetic analyses within populations or species, to closely related species. It is most extensively used in analysis gene mapping because of their high genomic abundance due to the random distribution throughout the genome and ample availability of various restriction enzymes. Commonly, it was applied to estimation relationships of closely related taxonomy (Miller and Tanksley 1990) [22], DNA fingerprinting trends for genetic diversity analyses, and for estimates of hybridization and introgression, as well as investigation of gene flow between crops and weeds (Desplanque *et al.* 1999) [4].

PCR based molecular markers

The invention of PCR (polymerase chain reaction) was a souvenir in this progress and established to be a special

procedure that carried about a new categories of DNA amplification markers. PCR technology are used for DNA profiling of all molecular markers except RFLP marker. PCR based molecular markers classified various types viz. RAPD, AFLP, SSR, ISSR, SCAR, CAPS, SSCP, SNP, DArT, VNTR,s and INDEL and TE-Anchore.

(i) Random Amplified Polymorphic DNA (RAPD)

In RAPD marker used genomic DNA profiling through the PCR. In this techniques used short primers (10 bp) of random sequence. These oligonucleotides are used side both forward- and reverse-primers and Amplified fragments length are basically within the range of 0.5-5 kb in size are separated by agarose gel electrophoresis and polymorphisms can be estimated after stained ethidium bromide dyes, as the presence or absence of bands of particular sizes observed UV transilluminator. The major disadvantage of RAPDs is not used cloning and sequencing, low reproducibility not locus-specific, dominant markers, and similar- sized fragments may not be homologous., laborious, technical intensity is more. Usually, need purified, high molecular weight DNA, and avoid contamination from DNA samples. The application of RAPD molecular marker for assessed of phylogenetic analysis, detection of DNA polymorphism and genetic diversity.

(ii) Amplified Fragment Length Polymorphism (AFLP)

This is combined techniques of the RFLP with PCR based technology by binding primer recognition sites (adapters) to the restricted DNA. The main feature of AFLP is its capacity for "genome representation" and the simultaneous detecting of representative DNA regions distributed randomly throughout the genome. AFLPs are DNA fragments (80-500 bp) procured from digestion with restriction enzymes, followed by binding of oligonucleotide adapters to the digestion products and selective amplification by the PCR.

Generally scored as dominant markers (Vos *et al.* 1995) ^[41]. RFLP are sometimes used to refer synonymous Selective Fragment Length Amplification (SFLA) and Selective Restriction Fragment Amplification (SRFA). A variation of the AFLP technique is known as Selectively Amplified Microsatellite Polymorphic Locus (SAMPL). Don't need prior cloning and characterization (Morgante and Vogel 1994) ^[25]. SAMPL is more useful for intra-specific than inter-specific studies due to frequent null alleles. The power of AFLPs includes its high genomic abundance, considerable reproducibility, the generation of many informative bands per reaction, wide range of applications, and the fact that no sequence data for primer construction are required. Key drawback of include purified high molecular weight DNA, the dominance of alleles, and the possible non-homology of co migrating fragments belonging to different loci. AFLPs can be used in genetic diversity identity, DNA fingerprinting, clones and cultivars identification, and phylogenetic studies. highly genome abundance and generally random distribution throughout the genome make AFLPs a widely valued technology for gene mapping studies (Vos *et al.* 1995) ^[41].

(iii) Minisatellites

Minisatellite loci are also known as VNTR,s. It containing of tandem repeat units of 10-50 base motifs, flanked by conserved DNA restriction sites. mini satellite amplify of more bands, basically bands size ranged varies 4-20 kb. Minisatellite contains commonly multilocus probes capable to hybridize to different species (Jeffreys *et al.* 1985) ^[14]. More variation in the number of repeat units, due to gene conversion or unequal crossing over is considered to be the chief reason of length polymorphisms. The major potential benefits of minisatellites are high level of polymorphism, locus specificity, throughout genome sequencing and high reproducibility. The key drawback of minisatellites are similarity to RFLPs based on the methodology. In this procedure are used multilocus probes and produced highly informative profiles bands. This molecular marker mostly used in screening of genetic diversity, DNA finger printing and varietal identification (Jeffreys *et al.* 1985) ^[14].

(iv) Simple Sequence Repeats marker (SSR)

SSR marker is known as Microsatellite marker, short tandem repeats (STRs) or simple sequence length polymorphisms (SSLPs) and are the smallest class of simple repetitive DNA sequences. Microsatellites contains tandem repeats but their repeat motifs are shorter (1-6 bp). Which are highly polymorphic and variable number of repeats at a specific locus. they are extensively used through out of genome in both coding and non coding sequence. Specific primers are basically 20-25 bp can be designed to amplify the SSR by PCR. SSR and their flanking sequences can be observed by framing a small-insert genomic library. DNA Polymerase slippage during DNA replication, or slipped strand mispairing and unequal crossing over is perceived to be the chief cause of variation in the number of repeat units of a microsatellite, can be identify by gel electrophoresis (Matsuoka *et al.* 2002) ^[18]. Approximately, SSR marker displayed a high level of polymorphism, so they are very putative and informative. The power of SSR marker associated the co-dominance inheritance, highly abundance, enormous extent of allelic diversity, and the ease of assessing SSR size variation by PCR with pairs of flanking primers and random distribution throughout the genome (Morgante *et al.* 2002) ^[23]. SSR marker can be applied to disease diagnosis, forensic studies,

gene mapping, gene sequencing, gene editing, varietal identification, population genetics, molecular breeding and genetic divergence to that of closely related species (Jarne and Lagoda 1996). The most advantage of SSR markers are significantly minimised the analytical costs and the major disadvantage of microsatellites is the high development costs if able primer sequences for the species of interest are unavailable. During SSR markers analysis is the presence of stutter bands arise due to slipped-strand mispairing and unequal crossing over by DNA during PCR amplification (Freudenreich *et al.* 1997) ^[7]. Mutations are blocking amplification and occurrence null alleles and which may lead to errors in genotype scoring. The appearance of null alleles maximise with the utilise of SSR marker primers creates poor cross-species amplification ward and backward mutations, resulting in an underestimation of genetic divergence.

(v) Inter Simple Sequence Repeats (ISSR) marker

ISSR marker is the functionally combined both SSR and RFLP markers to the universality of random amplified polymorphic DNA (RAPD). Staub *et al.*, 1996 ^[39]; In this techniques used single primer in PCR processing targeting multiple genomic loci to amplify ISSR marker of different size. The primers utilised can be either unanchored (Gupta *et al.* 1994; Meyer *et al.* 1993) ^[9] or more usually anchored at 3' or 5' end with one to four degenerate bases extended into the flanking sequences (Zietkiewicz *et al.* 1994). ISSRs used longer primers (15-30 mers) as compared to RAPD primers (10 mers). In this procedure annealing temperature depend on the GC content of primers used ranged from 45 to 65 °C. In this reaction use of a high annealing temperature leading to higher stringency. PCR products are commonly ranged 200-2000 bp long and can be detected by both agarose and polyacrylamide gel electrophoresis. ISSR marker is cheap, easy, quick and randomly distributed throughout the genome and usually show high polymorphism although the level of polymorphism has been shown to vary with the detection method used. Key drawback of this markers are possibility of nonhomology of similar-sized fragments. ISSR markers most commonly used in estimation of genetic polymorphism, heterozygosity, clones and varietal identification and another systemic taxonomy sib matting spp. and mapping of gene studies (Gupta *et al.* 1994; Zietkiewicz *et al.* 1994) ^[9].

(vi) Single Nucleotide Polymorphism (SNP)

SNP molecular is the recent techniques is highly genomic abundance and throughout distributed for genome sequencing in different species of plants. These are the most functional tool for gene mapping, marker-assisted breeding, pyrosequencing and map-based cloning. SNP marker is just a single base change in the DNA sequence at which differ nucleotides arise in various individuals of populations. Over the past decade, a number of various SNP genotyping protocol have been made based on different principle of allelic discrimination and detection platforms. Base on the molecular mechanism SNP genotyping divided into four classes like oligonucleotide ligation, extension of primer, allele specific hybridization and invasive cleavage (Sobrinho *et al.* 2005) ^[38]. Mass spectrometry-based methods for SNP genotyping has continuously improved and matrix-assisted laser desorption/ionization mass spectrometry (MALDIMS) is now one of the most automated and efficient detection platforms, is price competitive, and delivers results of the highest accuracy and reliability (Tost and Gut 2002) ^[40].

(vii) Diversity Arrays Technology (DArT)

DArT is the one of the recent frontier approaches used for molecular breeding analysis including especially in paddy, wheat, barley, pigeon-pea. This techniques are most extensively used for wide dissection of genetic divergence, gene mapping. DArT is a chip and microarray hybridization-based technique.) DArT simultaneously types several thousand loci in a single assay. DArT generates whole-genome Wngerprints by scoring the presence versus absence of DNA fragments in genomic representations generated from samples of genomic DNA. The technology was originally developed for rice, a diploid crop with a small genome of 430 Mbp (Jaccoud *et al.* 2001). DArT was subsequently applied to a range of other crops. The list is expanding and currently includes 19 plant species and three fungal plant pathogens (Jaccoud *et al.* 2001 ^[11]; Lezar *et al.* 2004 ^[16]; Wenzl *et al.* 2004; Kilian *et al.* 2005; Wittenberg *et al.* 2005; Xia *et al.* 2005; Yang *et al.* 2006 ^[45]; Joseph Tohme, personal communication; DArT P/L and collaborators, unpublished data).

Advance molecular marker techniques:

This is the new emerging advance trends and genomic based analysis has increase to the enhancing molecular marker application in crop breeding and crop designing. This technology is creates of modification of various earlier techniques. Recent advance technology like organelle microsatellite, CACP marker, CAPS marker, ETS, SCAR, RAMP and another techniques.

(a) Organelle microsatellite

When the extra DNA found in cell except genomic DNA also refers to as organelle DNA like chloroplast DNA and mitochondrial DNA have promoting applied to population genetic structure and phylogenetic relationship in plants. Due to their uniparental inheritance mode of transmission, chloroplast and mitochondrial genomes exhibit various patterns of genetic differentiation compared to nuclear alleles.

(b) Plastid microsatellites

The estimation of the chloroplast organelle provides information on the population dynamics study of plants that is opposite to those obtained from the nuclear genome. Chloroplast microsatellites established as a high-resolution trends for detecting patterns of extra nuclear variation in a wide range of plant species. Chloroplast microsatellites are specially effective markers for evaluating for mating systems, gene flow via both pollen and seeds, and uniparental lineage. Chloroplast microsatellite based markers have been used for the detection of hybridization and introgression. and the analysis of the genetic diversity (Clark *et al.* 2000) ^[3] and phylogeography of plant populations (Parducci *et al.* 2001; Shaw *et al.* 2005) ^[28, 35]. One major drawback of the tool is the required of sequence data for primer construction.

(c) Mitochondrial microsatellites

Mitochondrial inheritance is governed by mtDNA in plant and animal organism. In plant mtDNA is more complex like in the maize mitochondrial genome, has been detected to be 320 MDa (Sederoff *et al.* 1981) ^[34]. In plants, mitochondrial genomes are not basically utilised for phylogenetic estimation due to a high rate of sequence reorganization (Sederoff *et al.* 1981) ^[34].

Application of molecular markers in crop improvement

Molecular markers are utilised for a widely applications in marker-aided breeding, genome mapping, genetic diversity, gene tagging, QTLs analysis phylogenetic analysis, DNA fingerprinting, mapping of genome, analysis of genome crop evolution, and plant genetic resources, population genetics, plant taxonomy, plant breeding, plant disease diagnostics, forensic markers assisted selection, recurrent selection, genome wide section, gene introgression, backcrossing, gene pyramiding and estimation of molecular heterosis. Especially, used in phylogenetic structure and crop evolution. Recently time research on molecular biology, molecular marker-aided breeding is a major achievement for scientists and breeders.

Gene mapping and gene tagging

Molecular markers are new frontier trends for marker-aided selection and gene manipulation is recent rends extensively used in in crop improvement. Molecular marker is basically used in genomics studies. The first time genome mapping analysis in plants was described in corn (Gardiner *et al.* 1993) ^[8], followed by rice (Mc Couch *et al.* 1988) ^[19], *Arabidopsis* (Nam *et al.* 1989) ^[27], etc., using RFLP markers. SSR markers, especially STMS markers, have been found to be widely applicable in mapping genome. Molecular marker broadly used into different fields like drought, salinity, chilling, disease and another nutrient supply in crop, improving qualities traits.

Estimation of phylogeny and evolution

Earlier theory of evolutionary were mainly based on the morphological and geographical variation among plant, animal and living organism. Approximately, the new emerging techniques of molecular biology becoming more andmore evident in providing detailed information about the genetic structure (Slatkin *et al.* 1987) ^[37].

Assessment diversity of exotic germplas

Molecular markers are used to estimation of genetic diversity, varietal identification, elimination of duplicates, eliminate of linkage gene, and maintain of elite genotype. Following domestication, genetic variation in crop plants has continued to narrow due to continuous selection process for specific traits, i.e. yield, thus rendering them more vulnerable to diseases and jeopardizing the potential for sustained genetic improvement over a long term. This risk was brought sharply into focus in 1970 with the outbreak of Southern corn leaf blight which drastically reduced corn yield in the USA and was attributed to the extensive use of a single, genetic male sterility factor which was unfortunately linked to the disease susceptibility.

Genotyping of genotypes

The repetitive and arbitrary DNA markers are of choice in genotyping of cultivars. Microsatellites like (CT)10, (GAA)5, (AAGG)4, (AAT)6, (GATA)4, (CAC)5 (Gupta *et al.* 1994) ^[9], and minisatellites (Ramakrishna *et al.* 1995) ^[30] have been employed in DNA fingerprinting for the detection of genetic variation, cultivar identification, and genotyping (Sant *et al.* 1999; Yang *et al.* 1994) ^[31, 44]. This information is useful for quantification of genetic diversity, characterization of accessions in plant germplasm collections, and taxonomic studies. Microsatellites have been useful for the generation of STMS markers, revealing polymorphisms within closely

related cultivars (Morjane *et al.* 1994) [26]. In plants, the first application of microsatellites was for cultivar identification and was later used to genotype unequivocally diverse materials like rice, wheat, grapevine, soybean, etc. This is important especially for protection of proprietary germplasm. Similarly microsatellite markers have also been advantageous in pedigree analysis as they represent a single locus

Other application of molecular marker

molecular markers are a new emerging trends for estimation of genomics and another omics. Now it is a recent frontier tools in crop improvements. The unique significant of molecular markers in development in the area of molecular based genetics and plant breeding. The molecular markers classified tremendous types and differences in their principals and methodologies and applications. In this article we attempt to review most of the different molecular markers that can be regularly utilised in different aspects of dissection of plant genome such as measuring of genetic diversity, gene tagging, QTLs analysis phylogenetic analysis, DNA fingerprinting, mapping of genome, analysis of genome crop evolution, and plant genetic resources, population genetics, plant taxonomy, plant breeding, plant disease diagnostics, forensic markers assisted selection, recurrent selection, genome wide section, gene introgression, backcrossing, gene pyramiding and estimation of molecular heterosis. In this review molecular trades increasing crop production and while same time reducing crop failure for sustainability of crops.

Acknowledgements

The author thank the following for support: Dr S.V. Pawar and S.M. Choudhary

References

1. Botstein D, White RL, Skolnick M, Davis RW. Construction of length polymorphisms. *Am. J. Hum. Genet.* 1980; 32:314-331.
2. Bucchi G, Anzidei M, Madaghiale A, Vendramin GG. Detection of haplotypic variation and natural hybridisation in halepensis-complex pine species using chloroplast SSR markers. *Molecular Ecology.* 1998; 7:1633-1643.
3. Chromosomes of Clark CM, Wentworth TM, Malley DMO. Genetic discontinuity revealed by chloroplast microsatellites in eastern North American *Abies* (Pinaceae). *Am J Bot.* 2000; 87:774-778.
4. Desplanque B, Boudry P, Broomberg K, Saumitou-Laprade P, Cuguen J, Van Dijk H *et al.* Genetic diversity and gene flow between wild, cultivated and weedy forms of *Beta vulgaris* L. Chenopodiaceae, assessed by RFLP and microsatellite markers. *Theor. Appl. Genet.* 1999; 98:1194-120.
5. Di Stilio VS, Kesseli RV, Mulcahy DL. A pseudoautosomal random amplified polymorphic DNA marker for the sex chromosomes of *Silene dioica*. *Genetics.* 1998; 149:2057-2062.
6. Freudenreich CH, Stavenhagen JB, Zakian VA. Stability of a CTG: CAG trinucleotide repeat in yeast is dependent on its orientation in the Genome. *Mol. Cell Biol.* 1997; 4:2090-2098.
7. Gardiner JM, Coe EH, Melia-Hancock S, Hoisington DA, Chao S. Map in maize using an immortalized F2 population. *Genetics.* 1993; 134:917-930.
8. Gupta M, Chyi YS, Romero-Severson J, Owen JL. Amplification of DNA markers from evolutionarily diverse genomes using single primers of simple-sequence repeats. *Theor. Appl. Genet.* 1994; 89:998-1006.
9. Gupta PK, Varshney RK, Prasad M. *Molecular Markers: Principles and methodology.* In: Jain SM, Ahloowalia BS and Brar DS (eds). *Molecular Techniques in Crop Improvement* Kluwer Academic Publishers. Netherlands. 2002, 9-54.
10. Jaccoud D, Peng K, Feinstein D, Kilian A. Diversity arrays: a solid state technology for sequence information independent genotyping. *Nucleic Acids Res.* 2001; 29:25.
11. Jaccoud D, Peng K, Feinstein D, Kilian A. Diversity arrays: a solid state technology for sequence information independent genotyping. *Nucleic Acids Res.* 2001; 29:25-29.
12. Jarne P, P.J.L. Lagoda. Microsatellites, from molecules to populations and back. *Trends Ecol. Evol.* 1996; 11:424-429.
13. Jeffreys AJ, Wilson V, Thein SL. Hypervariable minisatellite regions in human DNA. *Nature.* 1985; 314:67-73.
14. Kilian A, Huttner E, Wenzl P, Jaccoud D, Carling J, Caig V, *et al.* The fast and the cheap: SNP and DArT-based whole genome proWling for crop improvement. In: Tuberosa R, Phillips RL, Gale M (eds) *Proceedings of the international congress In the wake of the double helix: from the green revolution to the gene revolution*, 27-31, 2003, Avenue Media, Bologna, Italy, 2005, 443-46.
15. Lezar S, Myburg AA, Berger DK, Wing Weld MJ, Wing Weld BD. Development and assessment of microarray-based DNA Wngerprinting in *Eucalyptus grandis*. *Theor Appl Genet.* 2004; 109:1329-1336.
16. Mahipal Singh Kesawat, Basanta Kumar Das. *Molecular Markers: It's Application in Crop Improvement* J crop sci. *Biotech.* 2009; 12(4):169-181.
17. Matsuoka Y, Mitchell SE, Kresovich S, Goodman M, Doebley J. Microsatellites in *Zea*-variability, patterns of mutations, and use for evolutionary studies. *Theor. Appl. Genet.* 2002; 104:436-450.
18. McCouch SR, Kochert G, Yu ZH, Wang ZY, Khush GS, Coffina WR, *et al.* Molecular mapping of ricechromosomes. *Theor. Appl. Genet.* 1988; 76:815-829.
19. Meyer W, Mitchell TG, Freedman EZ, Vilgalys R. Hybridization probes for conventional DNA fingerprinting used as single primers in the polymerase chain reaction to distinguish strains of *Cryptococcus neoformans*. *J Clin. Microbiol.* 1993; 31:2274-2280.
20. Meyer W, Mitchell TG, Freedman EZ, Vilgalys R. Hybridization probes for conventional DNA fingerprinting used as single primers in the polymerase chain reaction to distinguish strains of *Cryptococcus neoformans*. *J Clin. Microbiol.* 1993; 31:2274-2280.
21. Miller JC, Tanksley SD. RFLP analysis of phylogenetic relationships and genetic variation in the genus *Lycopersicon*. *Theor. Appl. Genet.* 1990; 80:437-448.
22. Morgante M, Hanafey H, Powell W. Microsatellites are preferentially associated with nonrepetitive DNA in plant Genome. *Nat. Genet.* 2002; 30:194-200.
23. Morgante M, Salamini F. From plant genomics to breeding practice. *Curr Opin Biotechnol.* 2003; 14:214-219.
24. Morgante M, Vogel J. Compound microsatellite primers for the detection of genetic polymorphisms. US patent application no. 1994, 08/326456.

25. Morjane H, Geistlinger J, Harrabi M, Weising K, Kahl G. Oligonucleotide fingerprinting detects genetic diversity among *Ascochyta rabiei* isolates from a single chickpea field in Tunisia. *Curr. Genet.* 1994; 26:191-197.
26. Nam HG, Giraudat J, Den Boer B, Moonan F, Loos WDB, Hauge BM, *et al.* Restriction Fragment Length Polymorphism linkage map of *Arabidopsis thaliana*. *Plant Cell.* 1989; 1:699-705.
27. Parducci L, Szmidt AE, Madaghiele A, Anzidei M, Vendramin GG. Genetic variation at chloroplast microsatellites (cpSSRs) in *Abies nebrodensis* (Lojac.) Mattei and three neighboring *Abies* species. *Theor Appl Genet.* 2001; 102:733-740.
28. Proven J, Russell JR, Booth A, Powell W. Polymorphic chloroplast simple sequence repeat primers for systematic and population studies in the genus *Hordeum*. *Mol. Ecol.*, 1999; 8:505-511.
29. Ramakrishna W, Chowdari KV, Lagu MD, Gupta VS, Ranjekar PK. DNA fingerprinting to detect genetic variation in rice using hypervariable DNA sequences. *Theor. Appl. Genet.* 1995; 90:1000-1006.
30. Random amplified polymorphic DNA marker for the sex Sant VJ, Patankar AG, Gupta VS, Sarode ND, Mhase LB, Sainani MN, *et al.* Potential of DNA markers in detecting divergence and analysis in heterosis in Indian elite chickpea cultivars. *Theor. Appl. Genet.* 1999; 98:1217-1225.
31. Sant VJ, Patankar AG, Gupta VS, Sarode ND, Mhase LB, Sainani MN, *et al.* Potential of DNA markers in detecting divergence and analysis in heterosis in Indian elite chickpea cultivars. *Theor. A Schlotterer C.* 2004.
32. The evolution of molecular markers-just a matter of fashion? *Nat. Rev. Genet.* 5:63-69 ppl. *Genet.* 98:1217-1225.
33. Sederoff RR, Levings CS, Timothy DH, Hu WWL. Evolution of DNA sequence organization in mitochondrial genomes of *Zea*. *Proc Natl Acad Sci USA.* 1981; 78:5953-5957.
34. Shaw J, Lickey EB, Beck JT, Farmer SB, Liu W, Miller J, *et al.* The tortoise and the hare II: Relative utility of 21 noncoding chloroplast DNA sequences for phylogenetic analyses. *American Journal of Botany.* 2005; 92:142-166.
35. *Silene dioica*. *Genetics.* 149:2057-2062.
36. Slatkin M. Gene flow and population structure of natural populations. *Science.* 1987; 263:787-792.
37. Sobrino B, Briona M, Carracedoa A. SNPs in forensic genetics: a review on SNP typing methodologies. *Forensic Sci. Int.* 2005; 154:181-194.
38. Staub JE, Serquen FC, Gupta M. Genetic markers, map construction, and their application in plant breeding. *Hort Science.* 1996; 31(5):729-739.
39. Tost J, Gut IG. Genotyping single nucleotide polymorphisms by mass spectrometry. *Mass Spectrom Rev.* 2002; 21:388-418.
40. Vos P, Hogers R, Bleeker M, Reijans M, Van De Lee T, Hornes M, *et al.* AFLP: a new technique for DNA fingerprinting. *Nucleic Acids Res.* 1995; 23:4407-4414.
41. Wittenberg AHJ, van der Lee T, Cayla C, Kilian A, Visser RGF, Schouten HJ *et al.* Validation of the high-throughput marker technology DArT using the model plant *Arabidopsis thaliana*. *Mol Genet Genomics.* 2005; 274:30-39.
42. Xia L, Peng K, Yang S, Wenzl P, de Vicente C, Fregene M, *et al.* DArT for high-throughput genotyping of cassava *Manihot esculenta* and its wild relatives. *Theor Appl Genet.* 2005; 110:1092-1098.
43. Yang GP, Saghai Maroof MA, Xu CG, Zhang Q, Biyashev RM. Comparative analysis of microsatellite DNA polymorphism in landraces and cultivars of rice. *Mol Genet Genomics.* 1994; 245:187-194.
44. Yang S, Pang W, Harper J, Carling J, Wenzl P, Huttner E, *et al.* Low level of genetic diversity in cultivated pigeonpea compared to its wild relatives is revealed by diversity arrays technology (DArT). *Theor Appl Genet.* 2006; 113:585-595.
45. Young ND, Menancio-Hautea D, Fatokun CA, Danesh D. RFLP technology, crop improvement, and international agriculture. In G Thottappilly, LM Monti, DR Moham, AW Moore, eds, *Biotechnology: Enhancing research on tropical crops in Africa*. Technical Center for Agriculture and Rural Cooperation, International Institute of Tropical Agriculture, 1992, 221-230.
46. Yunbi Xu. *Molecular plant breeding*. 2009, ISBN 978-1-84593-392.
47. Zietkiewicz E, Rafalski A, Labuda D. Genome fingerprinting by simple sequence repeats anchored PCR amplification. *Genomics.* 1994; 20:176-183.