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OVERVIEW OF SSR MARKERS

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A genetic marker is a DNA sequence with known physical location on a chromosome. Genetic markers link trait of interest with the gene governing the trait. DNA segments positioned close to each other or located on same chromosome tend to inherent together. Genetic markers aid in tracking the inheritance of gene based on their relative positions. Molecular markers find their application in identifying parent lines, gene mapping, gene pyramiding, genetic diversity analysis, DNA fingerprinting etc. Molecular markers are classified as 1st generation markers which include Restriction Fragment Length Polymorphism (RFLP), 2nd generation markers which include Random Amplification Polymorphic DNAs (RAPDs), Amplified Fragment Length Polymorphisms (AFLPs), Inter Simple Sequence Repeats (ISSRs) and Simple Sequence Repeats (SSRs), 3rd generation markers include Single nucleotide polymorphisms (SNPs).

SSRs and Their Importance as a Marker System in Molecular Plant Breeding

The segments of DNA with a variable number of tandemly repeated sequences of 2-60 bp are known as variable number tandem repeats (VNTRs). Hyper variable DNA (VNTRs) include minisatellites which contain 0.2 to 2 kb long segments of 11-60 bp long tandem repeats having identical or almost identical sequences. The other category of VNTRs include microsatellites which are usually less than 100 bp long segments containing tandem repeats of 2-7 bp. Microsatellites also called as Simple sequence repeats (SSRs) or Short Tandem Repeats (STRs) or Simple Sequence Length Polymorphism and were first discovered by Litt and Luty (1989) and were applied to plants first time by Delseny *et al.* (1983).

SSR markers are gaining popularity among other marker systems due to high reproducibility, co-dominance nature, hypervariability, high abundance, chromosome specific location, amenable to automation and high throughput genotyping (Parida *et al*., 2009). On the other side SSRs suffers limitations of being expensive and time-consuming. Low degree of repetition per locus, random scattered distribution around the genome and high degree of length polymorphism are the characteristics features of SSRs. Because of the varied amount of repeats in the microsatellite sites, the high length polymorphism may be easily and reliably detected using polymerase chain reaction. Microsatellites arise due to single-stranded DNA slippage, double stranded DNA recombination mismatch/double strand break repair and retrotransposition. Slippage of DNA polymerase III (during replication) on the DNA template strand at the repeat region might cause the newly generated DNA strand to grow or contract in the repeat region. (Wang *et al*., 2009). Slip-strand errors are corrected by mismatch repair (MMR). Thus, the stablility of SSRs depends on balance between effectiveness of mismatch repair system and slippage of DNA. SSRs are classified into mono, di, tri, tetra, penta or hexanucleotides based on the number of nucleotides per repeat unit. Microsatellites are classified (Wang *et al*., 2009) as 1) Perfect repeats are the tandem arrangements of single-repeat motif 2) Imperfect repeats are the perfect repeats interrupted by non-repeat motifs 3) Two basic repeats present together in diverse arrays (compound microsatellites).

Many species include dinucleotide repeats, however they are substantially less common in coding areas than in non-coding regions. AT repeats are abundant in plants, while AC repeat is the most prevalent in animals. This appears to be the main difference between plant and animal genomes. The genomic SSRs may be mitochondrial (mtSSR), chloroplastic SSRs (cpSSR) and nuclear (nuSSR). cpSSRs have been widely utilized to analyze genomic differences in plants and gene flow in wild populations (Provan *et al*., 2001). Exons with triplet repeats are more common in many species with AAG repeats frequent in plants, while CCG repeats common in monocots. This high frequency of CCG repeats is characteristic of monocots, which can be related to their higher GC content or abundance of particular amino acids. In rice, change in the amount of GA or CT repeats in the 5' UTR of waxy gene was linked to amylose content. Another study discovered that (CCG)n in the 5' UTR of maize ribosomal protein genes was involved in regulation of fertilisation control (Dresselhaus *et al*., 1999). Although the frequency of microsatellites in plants is inversely proportional to genome size, the amount of repeated DNA in coding areas appears to be consistent, with dicots having more mononucleotide repeats and monocots having more trinucleotide repeats.

Microsatellites have become a marker of choice for an array of applications in plants due to hypervariable nature and extensive genome coverage. A large number of monogenic and polygenic loci for various traits can be easily identified and exploited for marker-assisted selection. Genome mapping is another field where microsatellites are being extensively used. Genome mapping consists of genetic mapping, comparative mapping, physical mapping and association mapping. Microsatellite markers can facilitate comparative mapping and help to identify 'linkage blocks', major gene syntenies, chromosome rearrangements and microsyntenies among species. Advent of new technologies have not affected the use of microsatellites due to their cost effectiveness and use in large scale genotyping. However, there are several potential drawbacks including the presence of stutter bands, null alleles and heterologous amplicons. Development of transferable SSR markers from sequence information being generated from major crops will provide the initial platform for evaluation, characterization and genetic map development in minor crops. Unigene derived microsatellite markers overcome the problem of redundancy in EST database and have the advantage of assaying variation in the transcribed regions of the genome with unique identity and positions. Vast amount of genetic diversity exists in plant germplasm and more microsatellite markers will become available as genomic information accumulates. Steady progress and advancement in microsatellites markers will make this marker system more attractive for molecular breeding and plant genetics and ultimately help in major crop improvement.

Conclusion

With the development of SSR markers, they have been isolated, characterized and deployed in wide range of both agriculture and horticultural crops. SSRs are found to be important in comparative mapping studies, chromosome rearrangements, major and micro gene syntenies**.** Despite a number of potential limitations, the SSR marker system has emerged as a viable tool in molecular breeding for crop improvement, thanks to advances in science and technology.

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