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DNA MARKERS AND ITS USE IN CROP IMPROVEMENT

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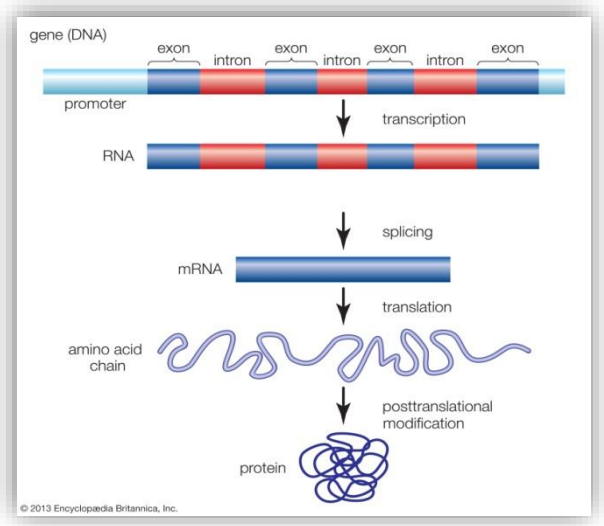
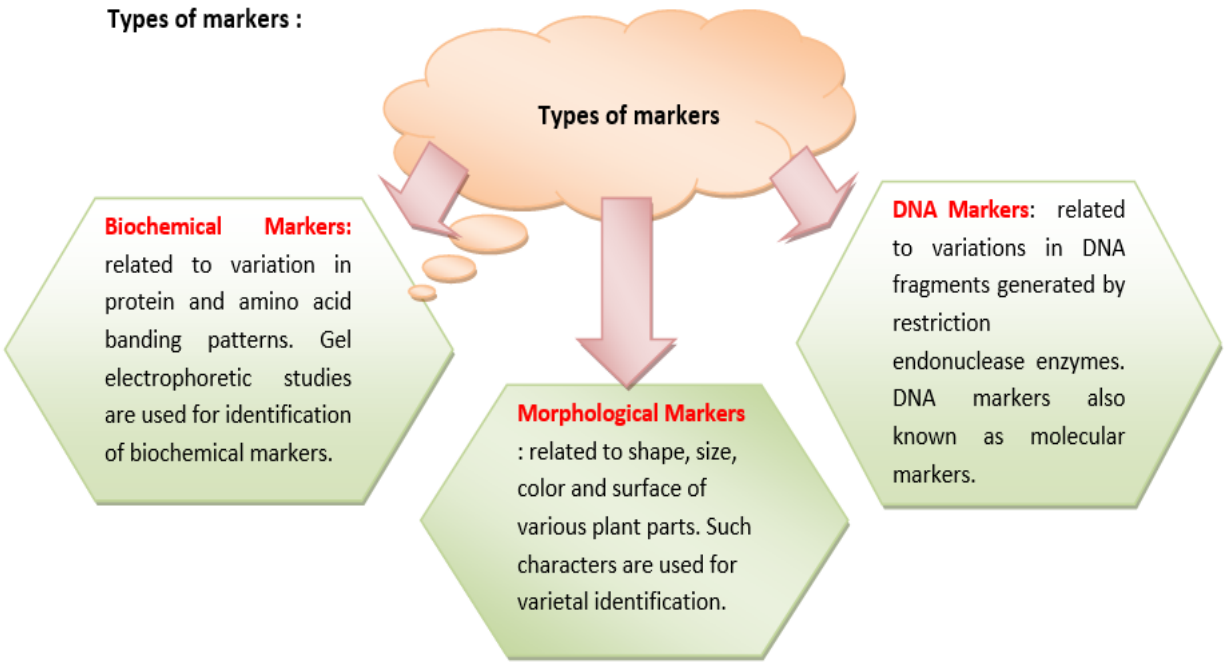
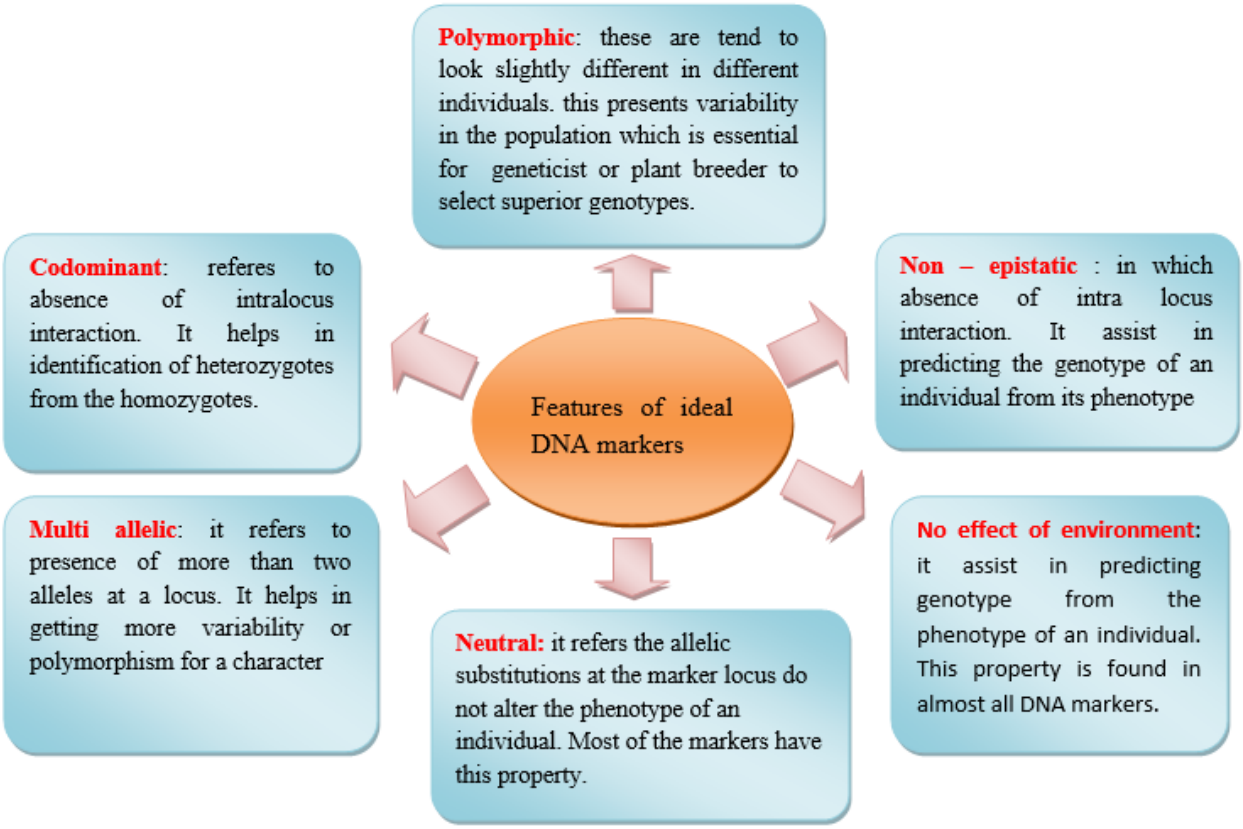
Introduction



The property of an individual which shows heritable variation is known as a character. It is a unique (DNA sequence), occurring in proximity to the gene or locus of interest. It refers to any unique DNA sequence which can be used in DNA hybridization, PCR or restriction mapping experiments to identify that sequence. DNA markers are also known as molecular markers or genetic markers. It includes morphological, physiological and biochemical properties in plant. plant characters are of two types that are qualitative (governed by one or few genes) and quantitative (governed by several genes) . To overcome problems associated with morphological markers, the DNA-based markers have been developed. those characters which can be easily identified are called as marker characters.



Features of Ideal DNA Marker



Types of DNA Markers:

1. Restriction Fragment Length Polymorphisms (RFLPs):

RFLPs known as variations found within the length of DNA fragments of a species generated by specific endonuclease. RFLPs are first type of DNA markers generated to distinguish individuals at the DNA level. RFLP technique was developed before the invention of Polymerase Chain Reaction (PCR).

2. Amplified Fragment Length Polymorphism (AFLP):

AFLPs are differences in restriction fragment lengths becuase of SNPs or INDELs that create or abolish restriction endonuclease recognition sites. AFLP assays are conducted by selectively amplifying a pool of restriction fragments with the help of PCR. The selective restriction fragment amplification (SRFA) is another name of RFLP technique.

3. Random Amplified Polymorphic DNA (RAPDs):

RAPD is known as polymorphism found within a species in the randomly amplified DNA generated by restriction endonuclease enzyme. RAPDs are PCR based DNA markers. RAPD marker assays are carried out using single DNA primer of arbitrary sequence.

4. Cleaved Amplified Polymorphic Sequences (CAPS):

CAPS polymorphisms are differences in restriction fragment lengths because of SNPs or INDELs that make or abolish restriction endonuclease recognition sites in PCR amplicons produced by locus-specific oligonucleotide primers.CAPS assays are carried out by digesting locus-specific PCR amplicons with one or more restriction enzymes and separating the digested DNA on agarose or

polyacrylamide gels.CAPS analysis is versatile and can be combined with single strand conformational polymorphim (SSCP), sequence-characterized amplified region (SCAR), or random amplified polymorphic DNA (RAPD) analysis to increase the chance of finding a DNA polymorphism. Michaels and Amasino (1998) developed a variant of the CAPS method known as dCAPS based on SNPs.

5. Simple Sequence Repeats (SSRs):

Simple sequence repeats (SSRs) or microsatellites are tandemly repeated mono-, di-, tri-, tetra-, penta-, and hexanucleotide motifs. SSR length polymorphisms are due to differences in the number of repeats. SSR locus is individually amplified by PCR using pairs of oligonucleotide primers specific to unique DNA sequences flanking the SSR sequence.

Jeffreys (1985) revealed that some restriction fragment length polymorphisms are due to VNTRs. The name "mini satellite" was coined due to the similarity of VNTRs to larger satellite DNA repeats.

6. Single Strand Conformational Polymorphisms (SSCPs):

SSCPs referred as DNA polymorphisms generated by differential folding of single-stranded DNA harboring mutations. The conformation of the folded DNA molecule is generated by intra-molecular interactions and thus it is a function of the DNA sequence. SSCP marker assays are generated by using heat-denatured DNA on non-denaturing DNA sequencing gels. Special gels (e.g., mutation detection enhancement gels) are developed to reinforce the invention of single-strand conformational polymorphisms caused by INDELs, SNPs, or SSRs.

7. Heteroduplex Analysis (HA):

It is known as DNA polymorphisms produced by separating homo-duplex from heteroduplex DNA using non-denaturing gel electrophoresis or partially denaturing high performance liquid chromatography. Single-base mismatches between genotypes results hetero-duplexes; thus, the presence of hetero-duplexes signals the presence of DNA polymorphisms. Heteroduplex analyses are often rapidly and efficiently carried out on numerous genotypes before specific alleles are sequenced, thereby greatly reducing sequencing costs in SNP discovery and SNP marker development.

8. Single Nucleotide Polymorphism (SNP):

The variations that found at one nucleotide position are referred to as single nucleotide polymorphisms or SNP. Such variation results because of substitution, deletion or insertion. This type of polymorphisms has two alleles and also known as bialleleic loci. This is the foremost common class of DNA polymorphism. It has observed in both in natural lines and after induced mutagenesis.

9. Expressed Sequence Tags (EST):

Expressed Sequence Tags (ESTs) are tiny pieces of DNA and their location and sequence on the chromosome are known. The variations that found at a single nucleotide position are known. The term Expressed Sequence Tags (ESTs) was first employed by Venter and his colleagues in 1991.

10. Sequence Tagged Sites (STS):

In genomics, a sequence tagged site (STS) is a short DNA sequence which has a single copy in a genome and whose location and base sequence are known.

Uses of DNA Markers in crop improvement:

1. Assessment of Diversity:

Molecular markers are often used for assessment of genetic diversity in cultivars, germplasm collection and advanced breeding material. This information can be used for germplasm characterization and developing varietal information system and PGR information system. This will also help in patenting plant material with special characters. The knowledge of genetic diversity will help in selection of parental lines for development of high yielding hybrids and cultivars. Crosses between distantly related parents can be made to obtain highly heterotic combinations.

2. Gene mapping:

Genetic linkage maps are important tools in plant and animal genetics. Molecular markers can be used to locate important genes or QTL in the genome. In other words, DNA markers may be used for construction of genetic linkage maps. Molecular markers are also convenient in identification of new useful alleles in the germplasm or wild species of crop plants. A tight linkage between a trait and molecular marker will assist in indirect selection of such alleles based on molecular marker. This knowledge can be used in selection programmes.

3. Marker assisted selection:

Marker assisted selection known as indirect selection for a desired plant phenotype on the basis of linked molecular marker. Selection of parents is an important step in the plant breeding. The OTL based selection helps to increased selection efficiency. Marker assisted selection is very productive for quantitative characters with low heritability. For efficient use of MAS, reliable and complete QTL products are used. The knowledge of OTLs and linked molecular markers can assist in introgressing specific segment of DNA containing gene of interest. This often achieved by marker assisted backcrossing. MAS limits the amount of linkage drag and requires less generation of backcrossing than conventional backcrossing for obtaining desired genotypes.

4. Crop evolution:

Molecular markers are convenient in the study of crop evolution. Molecular markers help in in tracing the genetic origin of crop plants. It will assist in identification of the wild species involved in the genetic evolution of different crops.

Conclusion:

DNA markers are extensively used in crop improvement because of its simplicity, reproducibility and precise location. It is not influenced by environmental effect decrease breeding cycle. Newly many DNA markers are accessible, out of this SSR, SNP are essentially used in breeding programme and other study. Application of DNA marker technologies also other areas of plant biology such as systematic, population genetics , evolutionary biology and conservation genetics, assist in genomics and identification of the wild progenitors of domestic species, the foundation of patterns of the genetic diversity