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# Role of Molecular Markers and their Significance

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## ABSTRACT

In this review, we have discussed the basic principles, uses, advantages and disadvantages of the most widely used molecular markers for genetic diversity studies, genetic mapping, marker-trait association studies and marker assisted selection programs. It is theoretically possible to observe and exploit genetic variation in the entire genome with different molecular markers. Most common genetic markers include allozymes, mitochondrial DNA, RFLP, RAPD, AFLP, microsatellite and SNP. Hence, the development and use of molecular markers for the detection and exploitation of DNA polymorphism is one of the most significant developments in the field of molecular genetics and medical sciences.

**Keywords:** Variation, allozymes, RFLP, RAPD, AFLP, SNP

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# INTRODUCTION

Molecular markers are used to reveal polymorphisms at the DNA level and can be applied in many genetic studies. They play a vital role in the utilization and genetic improvement of host genetic resistance. Also, known as genetic markers. Molecular markers can be found at specific locations of the genome. They are used to map the position of a particular gene. An integrated approach is provided for the use of molecular markers that generates multiple benefits and might prove attractive for application in various situations in the developing world. Various types of genetic markers are used to assess DNA polymorphism. These genetic markers are classified into morphological, biochemical and molecular markers[1]. Keys to the emergence of genomics were advances in DNA marker technology. These advances have resulted in a wealth of genetic markers including allozymes, mtDNA, RFLPs, RAPDs, AFLPs, microsatellites and SNPs with potentially wide spread utility in a variety of disease endeavors. In this review, uses, advantages and disadvantages for the various marker types are discussed. Genetic markers on the basis of their morphological traits are based on visually assessable traits like gene product including biochemical markers and relying on a DNA assay like molecular markers. The idea of using genetic markers appeared very early in literatures but the development of electrophoretic assays of isozymes and molecular markers have greatly improved our understanding in biological sciences [2-16]. Genetic markers should not be considered as normal genes, as they usually do not have any biological effect. They are identifiable DNA sequences found at specific locations of the genome and transmitted by the standard laws of inheritance from one generation to the next. The existence of various molecular techniques and methodologies need careful consideration in choosing one or more of such marker types.

**Sequence Variations**

All organisms are subject to mutations as a result of normal cellular interactions with the environment, leading to genetic polymorphism. For this variation to be useful to geneticists, it must be heritable genetic mutation distinguishable through molecular techniques. At the DNA level, types of genetic variation include base substitutions, commonly referred to as single nucleotide polymorphisms (SNPs), insertions or deletions of nucleotide sequences within a locus, inversion of a segment of DNA within a locus, and rearrangement of DNA segments around a locus of interest. Through long evolutionary pathways, many different instances of each type of mutation should exist in any given species, and the number and degree of the various types of mutations define the genetic variation within a species. DNA marker technology can be

applied to reveal these mutations. Inversions and rearrangements that involve restriction sites can be easy to detect because they disrupt the ability of a restriction enzyme to cut DNA at a given site and thus can produce relatively large changes in DNA fragment sizes. Point mutations are more difficult to detect because they do not cause changes in fragment sizes. If we consider molecular DNA markers in terms of the type of information they provide at a single locus, only three main categories can be described, in increasing degrees of interest: RAPDs (random amplification of polymorphic DNA), AFLPs (amplified fragment length polymorphism); RFLPs (restriction fragment length polymorphism), SSCPs (single stranded conformation polymorphism)and the microsatellites.

**Marker Selection**

Two main points should be considered, when using molecular markers for genetic studies. The genotyping procedure should be as simple and have as low a cost as possible, in order to generate the vast amount of genotyping data often necessary. Secondly, according to the type of analysis to be performed, a few characteristics are important, such as the dominance relationships, information content, map positions or genetic independence of markers. Molecular markers, they are classified as hybridization based markers and polymerase chain reaction PCR based markers [17, 18].

**Classification of Molecular Markers**

Molecular markers are classified into two categories: type I are markers associated with genes of known function, while type II markers are associated with anonymous genomic segments [19.] Under this classification, most RFLP markers are type I markers because they were identified during analysis of known genes. RAPD markers are type II markers because RAPD bands are amplified from anonymous genomic regions via the polymerase chain reaction (PCR). AFLP markers are type II because they are also amplified from anonymous genomic regions. Microsatellite markers are type II markers unless they are associated with genes of known function. EST markers are type I markers because they represent transcripts of genes. SNP markers are mostly type II markers unless they are developed from expressed sequences. Indels are becoming more widely used as markers since they often are discovered during genomic or transcriptomic sequencing projects, they can be either type I or type II markers depending on whether they are located in genes. Allozymes are allelic variants of proteins produced by a single gene locus, and are of interest as markers because polymorphism exists and because they represent protein products of genes with known function and are thus type I markers. Since the 1960s, starch gel electrophoresis of allozymes has been the most commonly employed molecular

method in fishery genetics [20, 21]. Allozymes were among the earliest markers used in aquaculture genetics [22-26]. Differences in the presence, absence and relative frequencies of alleles are used to quantify genetic variation and distinguish among genetic units at the levels of populations, species, and higher taxonomic designations. In few cases, correlations existed between certain allozyme markers and performance traits [27,28]. Their use in linkage mapping has been demonstrated in studies of salmonids and poeciliids [29-31].

Studies have shown that sequence divergence accumulates more rapidly in mitochondrial than in nuclear DNA [32]. This has been attributed to a faster mutation rate in mtDNA that may result from a lack of repair mechanisms during replication and smaller effective population size due to maternal inheritance of the haploid mitochondrial genome [33, 34]. RFLP markers were regarded as the first shot in the genome revolution marking the start of an entirely different era in the biological sciences [35]. RAPD procedures were first developed in 1990 using PCR to randomly amplify anonymous segments of nuclear DNA with an identical pair of primers 8–10 bp in length[36] AFLP is a PCR-based, multi-locus fingerprinting technique that combines the strengths and overcomes the weaknesses of the RFLP and RAPD methods discussed previously. The unique feature of the technique is the addition of adaptors of known sequence to DNA fragments generated by digestion of whole genomic DNA.

Microsatellites consist of multiple copies of tandemly arranged simple sequence repeats (SSRs) that range in size from 1 to 6 base pairs [37-38]. Abundant in all species studied to date, microsatellites have been estimated to occur as often as once every 10 kb in fishes[39]. Microsatellites tend to be evenly distributed in the genome on all chromosomes and all regions of the chromosome. They have been found inside gene coding regions, introns, and non-gene sequences[40]. The best known examples of microsatellites within coding regions are those causing genetic diseases in humans, such as the CAG repeats that encode polyglutamine tract, resulting in mental retardation. However, analyses using the detection of single nucleotide polymorphisms (SNP) which can cause change in one base within a DNA sequence is also gaining popularity.

**Genetic Variation**

To study genome-wide expression profiling from microarray experiments with genetic linkage analysis the technique is used to link the genotype with the phenotype and has been termed expression quantitative trait loci (QTLs). However, the approach promises to greatly improve the understanding of the underlying causes of genetic variation. The availability of the complete and validated genome architecture of chromosomes for different species is important for identifying

sequence variation underlying QTLs both within genes and other areas of the genome, which may play a role in gene regulation.

**Future applications of DNA markers**

In addition to genome mapping and various other applications, DNA markers are likely to prove useful in many aspects of genetics. The development and application of DNA marker technologies already underway in other areas such as population genetics, evolutionary biology, molecular ecology and conservation genetics. Although it may take some time to implement marker-assisted selection, the techniques of genome mapping and QTL analysis used to support MAS will eventually be used to identify and clone genes that could prove to be economically important and find applications in medicine and other bio related industries.

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