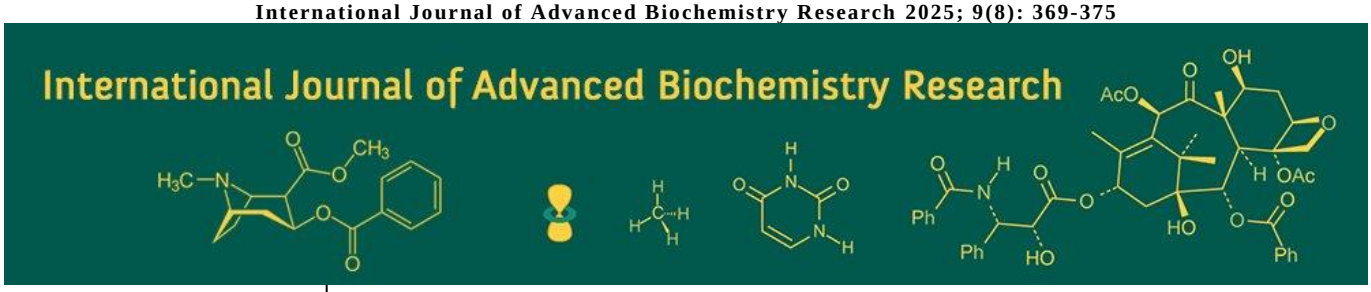

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Development of molecular markers for different traits and its application: A review

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Abstract
Crop breeding has been completely transformed by the creation of molecular markers, which make it possible to precisely identify genetic diversity and increase the effectiveness of trait selection. From first-generation RFLPs and RAPDs to more sophisticated systems like SSRs, SNPs, DArT, GBS, and KASP, this study outlines the development of marker technologies. The concepts, benefits, and drawbacks of each marker type are covered. The steps involved in developing a marker are described, including high-throughput genotyping, primer design, validation, and polymorphism area identification. The contributions of many applications, including genomic selection (GS), QTL mapping, genome-wide association studies (GWAS), genetic diversity analysis, and marker-assisted selection (MAS), to speeding up crop development are emphasized. Utilizing molecular markers in conjunction with genomic tools, machine learning, and genome editing has enormous potential for creating climate-resilient, high-performing crop varieties, despite ongoing obstacles such high costs, technical skills, and infrastructure requirements. Global food security and contemporary plant breeding continue to depend heavily on the ongoing development of marker technologies.

Keywords: Molecular markers, marker-assisted selection, SNPs, SSRs, GBS, crop breeding, CRISPR, multi-omics, genomic selection, epigenetic markers

Introduction
Crop breeding has been greatly improved by the development of molecular marker technologies, which allow for accurate identification of genetic diversity and trait selection. Marker systems began with RFLP in the 1980s and progressed to PCR-based markers such as AFLP and RAPD (Nadeem *et al*., 2017) [24]. Next came highly informative SSRs and SNPs, which were hastened by next-generation sequencing (NGS) technologies (Abbas *et al*., 2024) [1]. Breeding programs can use more precise selection thanks to functional molecular markers (FMMs), which target trait-linked genes (Kage *et al*., 2015) [17]. In order to increase productivity, disease resistance, and adaptation in important crops like maize and wheat, marker-assisted selection (MAS) has been successfully used (Prasanna *et al*., 2010; Landjeva *et al*., 2007) [27, 29]. Even with persistent infrastructural and economic issues (Kumar *et al*., 2011) [18], molecular markers are still essential tools in contemporary agriculture, and future research will likely integrate genome editing and genomic selection (Baloch *et al*., 2023) [23].

Types of Molecular Markers
First-generation markers
Restriction Fragment Length Polymorphisms (RFLPs)
Restriction Fragment Length Polymorphisms (RFLPs) were among the first DNA markers developed and are based on differences in DNA sequences that alter restriction enzyme sites, producing fragments of varying lengths upon digestion. These fragments are separated by gel electrophoresis and detected using DNA probes (Botstein *et al*., 1980) [5]. RFLPs are codominant markers, allowing clear distinction between homozygous and heterozygous genotypes. They played a key role in early genetic mapping and were used to map important genes in crops like maize, rice, and tomato (Tanksley *et al*., 1989) [33]. However, RFLPs are labour-intensive, require high-quality DNA, and have low throughput, making them less

practical than PCR-based markers.

Random Amplified Polymorphic DNA (RAPD)

Random Amplified Polymorphic DNA (RAPD) markers, introduced in the early 1990s, are PCR-based markers that use short, arbitrary primers to amplify random regions of genomic DNA without prior sequence knowledge (Williams *et al*., 1990) [39]. Polymorphisms are identified based on the presence or absence of amplification products, which reflect variations at primer binding sites. RAPDs are dominant markers, limiting their ability to distinguish between homozygous and heterozygous individuals. They have been widely used in genetic diversity, phylogenetic analysis, and germplasm characterization in crops like grapevine and sugarcane (Jones *et al*., 1997) [16]. Although cost-effective and simple, RAPDs suffer from low reproducibility due to their sensitivity to reaction conditions.

Second-Generation Markers:

Simple Sequence Repeats (SSRs)

Simple Sequence Repeats (SSRs) or microsatellites are short, tandemly repeated DNA sequences (1-6 base pairs) found throughout the genome. SSRs were classified as simple perfect, simple imperfect, compound perfect, or compound imperfect. Due to variation in the number of repeat units, SSRs are highly polymorphic and serve as codominant markers, enabling the detection of both homozygous and heterozygous genotypes (Powell *et al*., 1996) [26]. These markers are PCR-based, locus-specific, and occur in both coding and non-coding regions. SSRs are widely used in genetic diversity studies, linkage and QTL mapping, cultivar identification, and marker-assisted selection in crops such as grapevine, rice, and wheat (Gupta & Varshney, 2000) [12]. SSR are highly applicable in breeding as they are multi allele, co dominant and highly informative and occurs with high relative number, good coverage across the genome and can be experimentally reproduced (Pan, 2010; Powell *et al*., 1996; Pan *et al*., 2006) [43, 26, 42]. They have application in genetic analysis at the individual, population, cultivar, and species levels. Though highly reproducible, the main limitation of SSRs is the high cost and effort required for their initial development, particularly in non-model species.

Amplified Fragment Length Polymorphisms (AFLPs)

Amplified Fragment Length Polymorphisms (AFLPs) combine the strengths of RFLPs and PCR-based methods. AFLP involves restriction digestion of genomic DNA using specific enzymes, followed by ligation of adaptors to the sticky ends and selective amplification of fragments using primers with adaptor and selective nucleotide sequences. AFLPs are highly polymorphic, reproducible, and genome-wide, making them valuable for applications in DNA fingerprinting, genetic diversity studies, linkage mapping, and phylogenetic analysis. They are particularly useful for species with limited genomic information. Although AFLPs are dominant markers, their high multiplex ratio (ability to detect many loci in a single PCR reaction) compensates for this limitation. They are more reproducible than RAPDs and produce large amounts of data, but they require more technical skill and are costlier compared to SSRs (Vos *et al*., 1995) [36].

Third-Generation Markers

Single Nucleotide Polymorphisms (SNPs)

Single Nucleotide Polymorphisms (SNPs) are the most abundant type of genetic variation in genomes, involving a single base pair change at a specific locus. SNPs are typically biallelic, meaning they exist in two possible allelic forms, and occur on average every 100-300 base pairs in most plant and animal genomes. SNPs are highly stable, codominant, and amenable to high-throughput genotyping using platforms such as SNP arrays, KASP assays, and next-generation sequencing (NGS). Because of their abundance and genome-wide distribution, SNPs are ideal for genome-wide association studies (GWAS), high-resolution genetic mapping, genomic selection, and evolutionary studies. The development of SNP markers requires genomic sequence information and sophisticated bioinformatics tools. However, once developed, SNPs offer high automation, reproducibility, and cost-effectiveness for large-scale genotyping. SNP-based technologies have significantly advanced crop improvement programs, including in grapevine, wheat, maize, and rice (Rafalski, 2002; Edwards *et al*., 2007) [28, 10].

Diversity Arrays Technology (DART)

Diversity Arrays Technology (DART) is a microarray-based genotyping method that allows simultaneous detection of hundreds to thousands of polymorphic loci across the genome, without prior sequence information. The technique involves the digestion of genomic DNA, followed by adaptor ligation, selective amplification, and hybridization to a microarray of cloned genomic fragments. Presence or absence of hybridization signals indicates polymorphisms. DArT markers are dominant but offer a high-throughput, cost-effective, and sequence-independent approach, making them especially useful in species with large, complex, or unsequenced genomes. DArT has been widely used in genetic mapping, population structure analysis, and germplasm characterization in crops like wheat, barley, pearl millet, and grapevine (Jaccoud *et al*., 2001; Wenzl *et al*., 2004) [15, 38]. With the integration of DArT with next-generation sequencing (DArTseq), the platform now combines the advantages of SNP detection with the scalability and cost-efficiency of DArT.

Next-Generation Markers

Genotyping-by-Sequencing (GBS)

Genotyping-by-Sequencing (GBS) is a next-generation sequencing (NGS)-based technique that allows rapid, high-throughput discovery and genotyping of thousands of single nucleotide polymorphisms (SNPs) across the genome. It involves the digestion of genomic DNA with restriction enzymes, followed by adapter ligation, PCR amplification, and direct sequencing of the resulting fragments using NGS platforms. GBS is particularly effective for species with complex or unsequenced genomes, as it reduces genome complexity and does not require a reference genome, though having one improves data quality. It is widely used for genome-wide association studies (GWAS), linkage mapping, genetic diversity studies, and marker-assisted breeding. GBS offers the advantage of simultaneous marker discovery and genotyping, is cost-efficient for large populations, and generates dense marker coverage. However, it may suffer from issues like missing data and low sequencing depth per locus if not optimized. It has been

successfully applied in crops such as grapevine, maize, wheat, rice, and soybean (Elshire *et al*., 2011; Poland *et al*., 2012) [11, 25].

Kompetitive Allele Specific PCR (KASP)

Kompetitive Allele Specific PCR (KASP) is a fluorescence-based genotyping platform used primarily for SNP and small indel detection. It uses allele-specific forward primers and a common reverse primer along with fluorescently labeled reporter dyes to distinguish between alleles in a competitive PCR reaction. KASP is highly accurate, cost-effective, high-throughput, and scalable, making it ideal for marker-assisted selection, trait screening, genetic mapping, and validation of GWAS/QTL findings. The assay can be customized for single SNPs or used in multiplex formats and is compatible with both low and high sample volumes. KASP does not require gel electrophoresis, and the results are read using real-time PCR machines, providing rapid and reproducible genotyping. It has been applied successfully in crop improvement programs, including in grape, rice, wheat, and canola (Semagn *et al*., 2014) [30].

Development Process of Molecular Markers

Identification of Polymorphic Regions

The development of molecular markers begins with the identification of polymorphic genomic regions, which serve as the source of variation to be exploited for marker development. This step involves detecting differences in nucleotide sequences (e.g., SNPs, SSRs, indels) between individuals or populations. For SSR markers, tandem repeats in the genome are identified using computational tools such as MISA (MIcroSAtellite identification tool) or SSR Finder, applied on genomic or transcriptomic sequences (Thiel *et al*., 2003) [34]. For SNPs, polymorphisms are identified through whole-genome sequencing (WGS) or resequencing of multiple genotypes, followed by alignment and variant calling using software like GATK, SAMtools, or FreeBayes (McKenna *et al*., 2010) [21]. In genotyping-by-sequencing (GBS) and DArT, restriction enzyme-based genome reduction is used to target reproducible genomic regions across samples, and sequencing is used to detect polymorphic sites. These polymorphic regions are critical for designing locus-specific markers.

Primer Designing and Optimization

After polymorphic sites are identified, primers are designed to flank the region of interest for PCR amplification. For SSRs, primers are developed on sequences flanking the repeat motifs, while for SNPs, allele-specific primers may be designed. Tools like Primer3, BatchPrimer3, or Primer-BLAST are widely used for designing primers based on melting temperature (Tm), GC content, primer length, and product size. Optimization includes testing different annealing temperatures, primer concentrations, and template DNA amounts to ensure specific and efficient amplification. In the case of SNP markers, KASP primers or TaqMan probes are designed based on the SNP position and neighboring nucleotide context to allow allele discrimination.

Validation and Testing

Marker validation involves testing the designed primers on a diverse set of DNA samples to confirm:

- Polymorphism detection
- Amplification specificity
- Reproducibility

For SSRs and SNPs, validated markers should show clear and scorable banding patterns (SSR) or distinct allelic clustering (SNPs). Polymorphism Information Content (PIC), heterozygosity, and allele frequency are often calculated to assess marker informativeness (Botstein *et al*., 1980) [5]. Markers that consistently amplify the expected product and differentiate genotypes are considered validated and suitable for genetic applications.

High-Throughput Screening Methods

Validated markers are deployed in high-throughput genotyping platforms for large-scale screening. Depending on the marker type, different systems are used:

- **SSR markers:** Capillary electrophoresis using fluorescence-labeled primers on ABI genetic analyzers.
- **SNP markers:** High-throughput SNP arrays (e.g., Illumina Infinium, Affymetrix Axiom), KASP assays, or GBS pipelines using NGS.
- **DArT/DArTseq:** Use microarray hybridization or sequencing-based diversity array platforms for genome-wide genotyping.

High-throughput systems are essential for large breeding populations, genome-wide association studies (GWAS), QTL mapping, and genomic selection.

Applications of Molecular Markers in Crop Improvement

Genetic Diversity Analysis

Molecular markers are widely used to assess genetic diversity and population structure in plant germplasm. This involves screening multiple accessions or cultivars using markers such as SSRs, RAPDs, AFLPs, or SNPs to estimate parameters like:

- Allele number and frequency
- Polymorphism Information Content (PIC)
- Expected heterozygosity (He)
- Nei's genetic distance

The results are visualized using cluster analysis, Principal Coordinate Analysis (PCoA), or STRUCTURE analysis to reveal population structure, duplication, and parentage. This analysis is vital for identifying genetically diverse parents in breeding programs, conserving genetic resources, and maintaining crop adaptability.

Marker-Assisted Selection (MAS)

Marker-Assisted Selection (MAS) is the use of molecular markers linked to desirable traits for indirect selection during breeding. MAS accelerates selection for traits that are:

The process includes

Identification of trait-linked markers through mapping or association studies. Genotyping of segregating populations or breeding lines using the marker. Selection of individuals based on genotype rather than phenotype. MAS reduces breeding cycles and increases efficiency and precision in developing improved varieties. Examples include resistance gene pyramiding, early selection of dwarfing genes, and introgression of quality traits.

Quantitative Trait Loci (QTL) Mapping

QTL mapping identifies genomic regions associated with complex quantitative traits (e.g., yield, drought tolerance) using segregating populations (F2, RILs, backcross, etc.).

Procedure:

- Develop a mapping population from genetically diverse parents.
- Phenotype the population across environments.
- Genotype individuals using molecular markers (SSR, SNP, etc.).
- Construct a genetic linkage map.
- Perform QTL analysis using software like QTL Cartographer, MapQTL, or R/qtl.

Detected QTLs are characterized by LOD scores, percent phenotypic variance explained (PVE), and confidence intervals. QTLs can be used for marker-assisted selection, gene cloning, and understanding trait architecture.

Association Mapping and Genome-Wide Association Studies (GWAS)

Association mapping and GWAS use natural populations (rather than biparental mapping populations) to identify marker-trait associations by exploiting historical recombination and linkage disequilibrium (LD).

Steps in GWAS

- Phenotyping of a diverse panel of genotypes.
- Genotyping using high-density markers (e.g., SNPs from GBS or arrays).
- Population structure and kinship estimation (e.g., using STRUCTURE or PCA).
- Statistical association analysis using models like:
- GLM (General Linear Model)
- MLM (Mixed Linear Model)
- FarmCPU, BLINK (for increased power and reduced false positives)

GWAS allows fine-mapping of trait loci and is especially powerful for complex traits. It has been applied successfully in grapevine, rice, maize, and Arabidopsis.

Genomic Selection (GS)

Genomic selection (GS) is an advanced breeding approach where the genotypic data of genome-wide markers is used to predict the genetic potential (breeding value) of individuals for selection.

Steps in GS

- Develop a training population with genotypic (e.g., SNPs) and phenotypic data.
- Use statistical models (e.g., GBLUP, Bayesian models, ridge regression) to train prediction models.
- Apply the model to predict breeding values of untested individuals (test population).
- Select top individuals based on genomic estimated breeding values (GEBVs).

GS is particularly effective for

- Traits with low heritability
- Polygenic traits
- Reducing generation time in perennial crops

GS has transformed breeding programs in grapevine, wheat, maize, and forest trees.

Crop-Specific Marker Development

Cereals (e.g., Rice, Wheat, Maize)

Cereals are among the earliest crops where molecular markers were applied extensively for trait improvement. Key target traits include abiotic stress tolerance, yield components, disease resistance, and grain quality.

In rice, SSRs and SNPs have been used to map QTLs for traits such as drought tolerance, submergence resistance (e.g., Sub1), and salinity tolerance (Saltol QTL). In wheat, high-density SNP arrays (e.g., 90K and 660K chips) have facilitated genomic selection and mapping of rust resistance and quality traits. Maize has benefited from diverse marker technologies, including GBS, for genome-wide association studies (GWAS) and marker-assisted breeding. Advanced genotyping tools such as GBS, KASP, and high-density SNP arrays have significantly enhanced breeding speed and precision in cereals.

Legumes (e.g., Soybean, Chickpea, Lentil)

Molecular marker development in legumes has focused on enhancing biotic stress resistance (e.g., Fusarium wilt in chickpea, soybean cyst nematode), abiotic stress tolerance, and nutritional traits. In chickpea, EST-derived SSRs and SNPs have been developed using transcriptomics, and linked to drought and salinity tolerance traits. In soybean, large-scale SNP genotyping through Illumina arrays and GBS has enabled QTL mapping for traits like protein content, flowering time, and resistance to soybean mosaic virus Lentil marker development is supported by whole-genome resequencing, aiding in marker-assisted backcrossing for disease resistance. Legumes often require genome complexity reduction strategies due to their large genomes, making RNA-Seq and GBS effective approaches.

Fruits and Vegetables

In horticultural crops, molecular marker development is geared toward improving fruit quality traits, disease resistance, ripening behavior, and nutritional content. In grapevine, SSRs and SNPs have been employed for cultivar identification, mapping of berry traits (e.g., size, color, sugar), and resistance to powdery mildew. Tomato has been extensively studied for SNP and InDel markers linked to yield, fruit shape, and shel life traits. In apple, SSR and SNP arrays are used for QTL mapping of firmness, acidity, and scab resistance. As many fruit crops are perennial, marker-assisted breeding helps accelerate genetic gain by allowing early selection before maturity.

Cash Crops (e.g., Cotton, Sugarcane)

Marker development in cash crops is aimed at improving traits such as fiber quality, disease resistance, sugar content, and abiotic stress tolerance. In cotton, SNPs and SSRs have been used for mapping boll weight, fiber length, and Verticillium wilt resistance. The CottonSNP63K array is widely used in breeding programs. Sugarcane is a genetically complex crop; thus, GBS and DArTseq platforms are used for QTL mapping and diversity analysis related to sucrose accumulation and disease tolerance. Despite their polyploid or complex genome structure, these crops benefit from next-generation sequencing (NGS)-based

approaches that allow efficient marker discovery and deployment.

Challenges in Molecular Marker Development:

Genome Complexity and Size

Large and complex genomes pose a significant challenge in molecular marker development due to:

- High repetitive content, which makes it difficult to find unique primer binding sites.
- The presence of non-coding regions, transposons, and segmental duplications, which hinder accurate sequence alignment and marker design.
- In species with limited or no reference genomes, marker development becomes labor-intensive and time-consuming.

For instance, the wheat genome (~17 Gb) and sugarcane genome (10-12 Gb) are highly repetitive and polyploid, making marker discovery and sequence assembly challenging. Solutions include reduced-representation sequencing methods like GBS, genome skimming, and transcriptome sequencing to target informative regions.

Polyploidy and Heterozygosity

Many crop species (e.g., cotton, sugarcane, wheat, banana) are polyploids, possessing multiple sets of chromosomes. This introduces challenges such as:

- Allelic dosage complexity, where distinguishing between homozygous and heterozygous alleles is difficult.
- Paralogous sequences may confound marker specificity.
- High heterozygosity, especially in outcrossing species like grapevine and citrus, increases complexity in genotyping and marker inheritance analysis.

Polyploidy complicates linkage map construction, QTL mapping, and genomic prediction, requiring advanced analytical tools and algorithms that can model allele dosage and homeologous variation.

Cost and Accessibility of Technology

Despite technological advancements, high-throughput genotyping platforms (e.g., NGS, SNP arrays, KASP) often remain cost-prohibitive for small breeding programs and institutions in developing countries. Limitations include:

- Initial capital costs for sequencing platforms and real-time PCR systems.
- Recurring costs of reagents, enzymes, library preparation kits, and bioinformatics software licenses.
- Dependence on centralized or commercial service providers, leading to longer processing times and limited flexibility.

Additionally, unequal access to training, technical expertise, and infrastructure further exacerbates the divide between well-funded and resource-limited programs.

Bioinformatics and Data Management

- Modern molecular marker systems generate massive amounts of data that require:
- Robust bioinformatics pipelines for sequence alignment, variant calling, marker filtering, and primer design.
- Skilled personnel to manage next-generation sequencing (NGS) data and analyze large-scale marker-trait association datasets.
- Efficient data storage, curation, and sharing platforms, particularly for multi-environment trial (MET) data integration in GWAS and genomic selection.
- Challenges arise due to:
- Limited access to trained bioinformaticians.
- Lack of user-friendly tools for breeders.
- Data interoperability issues among software, databases, and institutions.

Future Perspectives in Molecular Marker Development

Integration of Multi-Omics Approaches

The integration of multi-omics approaches encompassing genomics, transcriptomics, proteomics, metabolomics, and phenomics is transforming the landscape of marker development in plant breeding (Raza *et al*., 2022) [29]. Genomics provides insights into sequence polymorphisms like SSRs and SNPs, which are foundational for molecular marker creation (Sinha *et al*., 2021) [31]. Unlike single-omics studies, multi-omics integration enables the correlation of molecular events at different biological levels, thereby uncovering functional relationships and regulatory networks that underlie traits, diseases, or responses to environmental stimuli. In crop research, this approach facilitates the identification of candidate genes, biomarkers, and pathways associated with important agronomic traits, aiding precision breeding and genetic improvement (Hasin *et al*., 2017; Chen *et al*., 2021) [13, 7]. Advanced computational tools and machine learning algorithms are increasingly being employed to manage and analyze the vast datasets generated from multi-omics platforms, enabling more accurate predictive modeling and hypothesis generation (Misra *et al*., 2019) [22]. This comprehensive strategy has proven particularly powerful in dissecting the molecular mechanisms in plants, animals, and humans, making it a cornerstone of modern biological research.

Collectively, multi-omics data allows for the identification of robust, predictive markers associated with complex traits such as stress tolerance and quality traits (Raza *et al*., 2022) [29].

Machine Learning and Artificial Intelligence in Marker Development

Machine learning (ML) and artificial intelligence (AI) are emerging as powerful tools in molecular marker discovery and trait prediction (Montesinos-López *et al*., 2018) [23]. These approaches enable the analysis of high-dimensional datasets generated from genomic and phenotypic sources to detect complex patterns and marker-trait associations (Azodi *et al*., 2020) [3]. ML algorithms such as random forest, support vector machines (SVM), and deep neural networks (DNNs) have been successfully applied to enhance the accuracy of genomic selection and QTL mapping in crops like maize and wheat (Crossa *et al*., 2017) [9]. AI also facilitates real-time data interpretation in high-throughput phenotyping platforms, enabling faster and more reliable trait evaluation (Zhang *et al*., 2017) [45]. Moreover, AI-driven tools are beginning to automate marker development pipelines, significantly reducing the time from raw sequence data to functional marker identification (Torkamani *et al*., 2018) [35].

CRISPR-Based Markers and Genome Editing

CRISPR-Cas systems have revolutionized functional genomics and have now begun to contribute to marker development through genome editing and mutation detection (Chen *et al*., 2019) [6]. CRISPR-Cas9 can create gene knockouts or insertions at target loci, which helps validate the function of QTLs or candidate genes and generate new, selectable alleles (Zhang *et al*., 2018) [46]. Recent advancements in CRISPR diagnostics using Cas12a and Cas13a have enabled the development of precise, sequence-specific detection tools that can serve as next-generation molecular markers (Aman *et al*., 2020) [44]. These CRISPR-based detection systems have been applied for rapid SNP genotyping, pathogen diagnostics, and allele differentiation in various plant species (Gootenberg *et al*., 2018;2017) [45, 46]. In addition to trait validation, CRISPR holds the potential to directly introduce beneficial alleles into elite lines, thereby merging marker development with precision breeding (Chen *et al*., 2019) [6].

Epigenetic Markers

Epigenetic modifications, including DNA methylation, histone modification (Wang *et al*.,2024) [1] and non-coding RNAs, play a crucial role in regulating gene expression without changing the underlying DNA sequence (Springer & Schmitz, 2017) [32]. These modifications can be heritable and environmentally responsive, making them valuable in identifying epigenetic markers (epimarkers) for traits like flowering time, stress memory, and heterosis (Zhang *et al*., 2018) [46]. Methylation-sensitive techniques such as MSAP (Methylation-Sensitive Amplification Polymorphism) and whole-genome bisulfite sequencing (WGBS) are commonly used to detect epigenetic polymorphisms (Alonso C *et al*., 2016) [2]. In crops like maize, rice, and Arabidopsis, epigenetic variation has been associated with phenotypic plasticity and environmental adaptability (Cortijo *et al*., 2014) [8]. The application of epimarkers in breeding programs could enhance selection for transgenerational stress tolerance, particularly under changing climatic conditions (Springer & Schmitz, 2017) [32].

Conclusion

Crop breeding has been transformed by the creation and use of molecular markers, which offer instruments for the early, accurate, and effective selection of desired traits. Every technology, from first-generation markers like RFLPs and RAPDs to next-generation platforms like GBS, KASP, and CRISPR-based systems, has made a distinct contribution to crop improvement and genetic analysis. For a variety of crops, molecular markers help with important breeding tasks such genomic selection, marker-assisted selection, QTL mapping, and genetic diversity evaluation. These techniques have made it possible to breed more quickly and precisely, despite obstacles such polyploidy, genetic complexity, and large data requirements. In the future, combining molecular markers with genome editing, multi-omics, and machine learning will improve precision breeding and aid in the creation of crop varieties that are climate-resilient and crucial for ensuring the world's food security.

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