

# Enhancing Crop Resilience to Abiotic Stress through Molecular Markers: A Review

## Abstract

Plants face multiple abiotic stressors including extreme temperatures (heat or cold), drought, and salinity, which significantly diminish agricultural crop yields. These stressors often impact nutrient management and metabolic processes, leading to stunted or completely halted plant growth. From a genetic perspective, stress represents an environmental challenge that obstructs optimal genetic expression. There is an extensive array of molecular genomic markers utilized for the analysis of crops. These markers are categorized based on their application: for instance, Random Amplified Polymorphic DNA (RAPD) markers are used to identify and evaluate hybrids for salinity and drought tolerance, whereas Simple Sequence Repeat (SSR) markers are instrumental in assessing stress resilience. These markers are also vital in Quantitative Trait Loci (QTL) mapping for genes associated with stress response. Genes like dehydrins for drought and saltol for salinity play a crucial role in managing responses to these stresses. Additionally, focusing on traits through single-gene Single Nucleotide Polymorphisms (SNP) markers aids in genetic mapping and sequencing of stress-related traits in inbred lines. DNA markers are pivotal in marker-assisted breeding, enhancing tolerance to abiotic stresses through sophisticated techniques and marker enhancements.

**Keywords:** DNA, genotype, salinity, drought, temperature stress

## 1. Introduction

Plants endure various adverse climatic conditions during their growth cycles, including biotic stresses like herbivory and pathogenic infections, as well as abiotic stresses such as extremes in temperature, drought, nutrient deficiencies, high salinity, and the presence of toxic metals and metalloids (arsenic, cadmium, and aluminum) in the soil. Heat or frost, drought, and salt are the

most common environmental factors that significantly reduce agricultural crop yields, posing serious threats to food security and altering the natural geographic distribution of plants. These issues are compounded by climate change, characterized by long-term shifts in weather patterns, which serves as a significant source of abiotic stress [1,2]. In response, plants activate their basal defense systems upon stress detection. Various signaling pathways, regulated by kinase enzymes and phytohormones such as jasmonic acid, abscisic acid, ethylene, and salicylic acid, are differentially activated. These pathways lead to the generation of reactive oxygen species (ROS), which help mitigate the biological damage and are foundational to plant stress tolerance [3].

Crop production and yield are primarily affected by abiotic stresses due to adverse environmental changes [4]. In biological terms, any external factor that negatively impacts plant growth or health is labeled as a stress [5]. Plants under stress exhibit three primary response phases: the alarm phase marking the onset of stress, the resistance phase where defense systems are activated, and the exhaustion phase resulting in loss due to prolonged stress [6]. Many plants show signs of growth inhibition under stress conditions [7–10]. Salinity, in particular, limits growth and productivity by disrupting water acquisition and altering ion homeostasis [11]. Additionally, regular water scarcity, known as drought stress, significantly hampers plant survival and development. While often associated with inadequate soil water, drought can also be intensified by excessive evapotranspiration, especially under conditions of high air temperature, leading to an imbalance between water uptake and loss [12].

From a genetic viewpoint, stress is an environmental condition that prevents plants from achieving full genetic expression. Abiotic stress, not resulting from interactions with other organisms, negatively impacts organisms within an environment and is a major threat in environmental and agricultural sectors, now exacerbated by industrialization and global warming [14].

DNA markers have wide-ranging applications in enhancing a plant's genetic structure, including the identification of parental genotypes, assessment of genetic variation, and development of high-resolution genetic linkage maps. Molecular markers are extensively used for genetic analysis of crops, categorized based on usage, such as PCR-based versus non-PCR-based markers. Among them, RFLP (Restriction Fragment Length Polymorphism) markers, prevalent in

the 20th century, played a crucial role in gene mapping and other molecular biology studies. The advent of PCR by Mullis and Faloona [15] significantly advanced DNA marker technologies, reducing the time and cost associated with genetic mapping through probe hybridization. PCR is an in vitro method that amplifies DNA sequences of interest from small amounts of a single template using primers that are complementary to specific gene sequences [16]. Single nucleotide polymorphisms (SNP), sequence-characterized amplified regions (SCAR), and simple sequence repeats (SSR) are examples of PCR-based molecular markers requiring gene sequence information from samples [16]. These markers also assist in detailing the allelic positions (heterozygosis, maternal homozygosis, paternal homozygosis) of each progeny or line in a population, and are essential for the assembly of linkage maps reflecting genetic diversity based on recombination assessments by hybrid plant genotypes [16,17]. Consequently, this review aims to describe the impacts of abiotic stress on agricultural crops and the role of DNA markers in genetic control, gene mapping, and screening for stress resistance traits.

## **2. Abiotic Stress Impact on Agricultural Crops**

Among abiotic stresses, heat and drought significantly impact crop yields and productivity, leading to substantial reductions in agricultural income and benefits. For instance, a 40% reduction in water availability can decrease maize yields to 40% and wheat yields to 21% of their original levels [18]. In Africa, cowpea—a crucial agricultural crop—suffers yield reductions of 34% to 68% due to drought stress [19]. Abiotic stresses often lead to the excessive production of reactive oxygen species (ROS), which are harmful and reactive molecules that can damage carbohydrates, lipids, nucleic acids, and proteins, thereby adversely affecting plant growth [20]. Additionally, water scarcity and heat stress can impair transpiration and stomatal conductance in plant leaves [21].

Globally, 91% of the agricultural area is subjected to abiotic stresses, contributing to 50% of the production losses in agriculture. The severity and damaging effects of abiotic stress can intensify with climate changes. Enhancements in agronomic management and the development of stress-resistant genotypes through breeding programs could mitigate these effects [22]. Abiotic stresses disrupt the biochemical and physiological processes in plants. Enhancing the efficiency of light usage and photosynthetic activity could improve tolerance to these stresses. Moreover, the

activation of antioxidants and development of stress-related metabolites help protect against cellular damage, though further development of key adaptation strategies to increase stress tolerance in plants is needed [23].

Heat stress is intricately linked to temperature increases in air and soil that exceed plant tolerance levels, even for brief periods, adversely affecting crop growth and development [24]. The rising global temperatures pose a serious climatic challenge that could drastically increase crop vulnerability. Research into heat stress is crucial for understanding plant responses and developing tolerance to these conditions. Improved progeny lines with enhanced heat tolerance are necessary to sustain agriculture. Heat stress negatively affects seed germination, photosynthetic activity, and crop yields. It also disrupts reproductive cycles and the function of tapetal cells in flowers, potentially stopping pollen grain swelling and leading to the release of less viable pollen [25].

Drought stress causes numerous physiological changes such as reduced photosynthetic activity, changes in cell wall elasticity, and stomatal closure. Notably, drought and salinity stresses often overlap in their effects on plant physiology and share common tolerance mechanisms. Drought alters the nutritional status of crops by changing the ion concentration in plant tissues, and the diffusion of soil nutrients to root surfaces diminishes with moisture levels [26,27].

Changes in nutrient constituents and management, along with alterations in biosynthetic capacity, are primary factors that ultimately inhibit or halt plant growth. Developing protective systems to ensure plant survival under abiotic stress is crucial for maintaining crop growth and production levels in agriculture. These defense systems against abiotic stress are being explored through molecular genetics, with a focus on understanding and enhancing stress tolerance [28].

Abiotic stressors such as salinity, drought, nutrient scarcity, heavy metals, water/air pollution, light photoperiodicity, and intensity can induce stress individually or in combination, potentially altering plant metabolic systems and reducing productivity, development, and growth. High stress levels may become intolerable and result in plant death. Although completely avoiding stress is impossible, plants have evolved metabolic responses and produce specific molecules to survive under stressful conditions [29,30].

Abiotic stress necessitates changes in the soil and plant environment conditions, potentially leading to reduced yields of primary agricultural crops worldwide. Currently, agricultural lands in non-stressed regions account for only 10% of global crop production, with the remaining 90% facing one or more environmental stresses. Plants continue to adapt to abiotic stress biochemically, physiologically, molecularly, and phenotypically, but ongoing efforts are needed to enhance plant stress tolerance through genetic improvements, resource conservation technologies, and other strategies [14].

### **3. Utilizing DNA Markers for Enhancing Abiotic Stress Resistance in Crops**

#### **3.1. Employing RAPD Markers to Analyze Salinity and Drought Resistance**

Random Amplified Polymorphic DNAs (RAPDs) are PCR-based markers that do not require preliminary sequence data from samples, allowing the analysis of multiple loci from many individuals using limited resources. RAPDs are favored for their straightforward experimental process and are extensively utilized for the genetic screening of intra- and interspecific hybrids [31].

These markers have been pivotal in identifying genes tolerant to salinity stress in various crops. Plants possess genetically regulated mechanisms for salinity defense/tolerance, making the improvement of salinity tolerance in crops, especially in saline-affected areas, essential. RAPD markers, through PCR amplification of specific DNA sequences, facilitate the detection of salt-resistant genes. For example, a study in wheat assessed the genetic diversity among salt-resistant genotypes grown in saline fields, where these markers effectively distinguished salt-resistant from salt-sensitive genotypes. This differentiation, evidenced by polymorphic primer pairing, is crucial for breeding salt-resistant wheat varieties [32].

Salinity stress can induce DNA changes, including structural disruptions and rearrangements, often as a result of secondary stress like oxidative damage associated with ROS production (hydroxyl radicals, singlet oxygen, superoxide, and hydrogen peroxide). In saline-affected cotton seedlings treated with NaCl, RAPD markers identified genetic instabilities by displaying variations in DNA band patterns on agarose gels, such as missing bands, altered band

intensities, and the appearance of new bands. These observations confirm that RAPD analysis can effectively investigate toxicological stress [33].

Field evaluations, which are labor-intensive and time-consuming, can be complemented by molecular differences identified at the DNA level in plants developed from tissue culturing [34]. RAPD technology, being rapid and requiring only minimal DNA, is sensitive to genetic differences and allows for the efficient processing of large numbers of genomic samples under in vitro conditions. For instance, RAPD analysis in maize identified several primers linked to salt resistance, providing valuable information for breeding programs aimed at developing salt-resistant progeny through marker-assisted selection and direct genetic modifications [36].

The Genomic Template Stability (GTS) assessment, which measures DNA damage and mutations, significantly contributes to molecular marker assemblies for identifying damaged or mutant DNA in plant cells. In a study evaluating the response of various cotton genotypes to saline conditions (200 mM NaCl), RAPD profile variations were quantified using GTS percentages. The highest GTS values were observed in sensitive genotypes, while the lowest were in salt-resistant varieties, indicating a direct relationship between genetic stability and salt tolerance. Such findings underscore the usefulness of RAPD analysis in identifying DNA sequences linked to salt stress and in the early detection of salt-resistant genotypes, particularly in cotton plants [37].

Wheat quality and production are also severely impacted by drought, with losses comparable to those caused by other climatic factors, a situation exacerbated by global climate change [38,39]. Drought-induced gene expression plays a crucial role in developing drought resistance [40]. In wheat, RAPD analysis using specific primers identified genetic variations associated with drought resistance. For example, a RAPD primer amplified a specific DNA band present in drought-resistant but absent in sensitive varieties, offering insights into the genetic basis of drought resistance in wheat hybrids [41]. Rashed et al. found RAPD markers that validated the method's reliability for identifying drought-resistant wheat genotypes [42].

In horticultural plants like tomatoes, productivity decreases under high temperatures. Traits related to yield are often quantitatively inherited and heavily influenced by environmental changes, complicating the assessment of heat resistance. A study utilizing RAPD markers linked to heat-

resistant genes in tomatoes identified specific markers in recombinant inbred lines developed from a cross between heat-sensitive and heat-resistant parents. Several RAPD markers were specifically associated with heat tolerance traits, enhancing the selection of molecular markers for heat tolerance in breeding programs [43]. Further applications of DNA markers in horticultural crops focus on abiotic stress, as detailed in Table 1.

**Table 1.** Use of DNA Markers in Horticultural Crops for Abiotic Stress Management

<b>Crop</b>	<b>DNA Marker</b>	<b>Abiotic Stress</b>	<b>Objective</b>	<b>Reference</b>
<i>Petunia x atkinsiana</i>	RAPD	Salinity	<ul style="list-style-type: none"> <li>_ Genetic diversity determination in mutant clones</li> <li>_ Screening of mutants related to salt resistance</li> </ul>	[44]
D. Don cv. Prism Red. Tall fescue ( <i>Festuca arundinacea</i> )	ISSR) Sequence characterized amplified region (SCAR)	Summer stress	Development of sequence-related markers to screen summer stress-resistant plants	[45]
Perennial grass ( <i>Miscanthus sinensis</i> )	SSR	Drought	To formulate SSR markers linked to drought resistance by utilizing transcriptome sequencing	[46]
Salvia ( <i>Salvia miltiorrhiza</i> )	AFLP	Drought	<ul style="list-style-type: none"> <li>_ To segregate drought-related genes in sterile male and fertile near-isogenic lines of <i>S. Miltiorrhiza</i></li> <li>_ To evaluate the change in</li> </ul>	[47]

			fertility of plants during drought stress	
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### 3.2 Enhancing Hybrid Stress Tolerance through Marker-Assisted Selection

Marker-assisted selection is a powerful technique for enhancing abiotic stress resistance in plants. Commonly, stress-resistant genes are identified using SSR (Simple Sequence Repeat) markers. In wheat, SSR analysis, combined with bulked segregant analysis, has been instrumental in identifying markers associated with agronomic traits like grain filling and heat tolerance. Three specific markers, Xgwm132, Xgwm617, and Xgwm577, have been linked to grain filling rates under heat stress, aiding in the development of heat-resistant cultivars [48].

SSR markers have also facilitated the identification of drought-resistant tetraploid cotton hybrids. Allelic polymorphism observed with SSR primers, alongside agricultural traits, revealed significant results in "Varamin" and "Sayar 314" hybrids. The "Tabladila" hybrid exhibited extensive polymorphism with EST-SSR markers, while the drought-tolerant "Nazily" hybrid showed 53 percent polymorphism [49].

### 3.3 Identifying Genetic Diversity Under Heat and Frost Stress

Field screening for heat-resistant varieties is less desirable due to unpredictable climatic factors that affect trial repeatability and precision. Furthermore, consistent high temperatures in growing areas are not always guaranteed. Genetic evaluation of quantitative traits for adaptive responses is essential. Molecular analysis enables the use of specific genotypes in breeding strategies aimed at improving yield stability and sustainability under severe stress conditions [50].

Heat resistance involves multiple genetic components regulated by different genes across various tissues and growth stages. Sequence-Related Amplified Polymorphism (SRAP) markers, a type of PCR-based marker, are efficient at retrieving DNA fragments in a single PCR reaction. These markers are capable of amplifying many polymorphic and reproducible alleles and loci, making them suitable for DNA fingerprinting, genetic diversity assessment, and gene mapping. However, the random distribution across the plant genome limits the use of SRAP markers [51].

Another PCR-based marker, Target Region Amplified Polymorphism (TRAP), utilizes two 18-mer DNA primers—one "fixed" from an expressed sequence tag (EST), and the other paired with either a GC- or AT-rich core to target exon or intron regions [52,53].

These markers have been applied to wheat genotypes to assess genetic diversity under heat stress. A genetic analysis using SRAP and TRAP markers identified significant genetic diversity in agronomic traits under heat conditions. Although field performance data based on agronomic traits involves complex genetic interactions, marker-assisted data from SRAP and TRAP analyses provided valuable insights into genetic diversity in a more unbiased manner compared to traditional morphological evaluations [54].

Conversely, frost significantly impacts yield and survival in pea crops. A field study involving 672 diverse pea genotypes across three different locations employed trait-based marker association to evaluate frost resistance using 267 SSR markers. Among these, 16 genotypes were identified as highly winter-tolerant, surviving across all experimental sites. Population analysis revealed a structured population with two subpopulations and various combinations among the 672 genotypes. The association study pinpointed seven SSR markers consistently associated with frost resistance in at least two different environmental conditions and under two statistical models. One particular marker, EST1109 on LG VI, is believed to be linked with a gene involved in glycoprotein metabolism responsive to frost stress, suggesting a pathway for enhanced chill resistance in pea crops. These findings support the use of winter-resistant germplasms and cold-resistant linked markers in marker-assisted breeding for developing cold-resistant pea cultivars [55].

## **4. Mapping of QTL Genes Associated with Abiotic Stress in Crops**

### **4.1. Saltol**

Saltol, a major salt-resistant quantitative trait locus (QTL), has been mapped to chromosome 1 in the rice genome. This QTL includes a gene that plays a crucial role in reducing sodium (Na<sup>+</sup>) uptake and enhancing potassium (K<sup>+</sup>) absorption, leading to a low Na ratio

phenotype under conditions of high salinity [56]. Simple Sequence Repeat (SSR) markers, known for their reproducibility and precision, are effectively used to explore genetic differences in various crops, including rice. These markers facilitated a study that assessed genetic diversity among rice genotypes on chromosome 1, where saltol is located. SSR markers identified distinct salt-resistant haplotypes, using "Pokkali," a primary source of saltol expression, as a reference. The study involved eight-generation inbred hybrids (Pokkali x IR29), where 33 SSR markers were used, notably RM8094, which was critical in distinguishing genetic differences. Cluster analysis grouped the genotypes into three classes based on their salt tolerance, with the most resistant breeds clustering separately from moderately resistant and sensitive groups [57]. Marker-assisted breeding transferred the saltol gene into a widely cultivated rice variety, with SSR markers RM493 and RM3411b effectively screening for the gene's presence. This introgression from the donor parent, FL478, successfully incorporated salt resistance into the BT7 progeny genome [58].

#### **4.2. Dehydrin**

Dehydration stress triggers the production of hydrophilic proteins, including dehydrins, which play a crucial role in enhancing a plant's drought, frost, and salt tolerance. These proteins are synthesized in response to severe dehydration conditions and the presence of abscisic acid. In barley, the expression of YnSKm-type dehydrins is particularly notable, with the species expressing ten out of thirteen dehydrin genes, similar to wheat. These proteins support the protective function during cellular dehydration by maintaining enzyme activity and efficiency under limited water availability [59]. Research in barley has shown that two dehydrin-related genes, regulated by the Hv-WRKY38 gene, significantly enhance drought and salt tolerance. These genes are located near the QTL regions associated with these stress responses [60, 61]. Additionally, other genes like aquaporin and CBF also contribute to osmotic stress tolerance, with the CBF gene particularly active in salt and drought stress signaling pathways [62, 63]. In *Citrullus colocynthis*, a study utilizing ISSR markers identified drought-tolerant genes in individuals from diverse locations, with specific primers targeting genes like UB, PEPKS, Dehydrin, ACT, and P5CS, confirming their role in drought resistance [64].

### **5. QTL Mapping via DNA Markers**

Drought poses a significant threat to productivity across diverse wheat-growing regions worldwide. Leveraging DNA markers associated with specific Quantitative Trait Loci (QTLs) for drought resistance could substantially enhance drought tolerance in wheat hybrids. A study focusing on the identification of QTLs linked to grain yield genes under drought stress has yielded invaluable genetic insights. STS markers were employed for QTL mapping in inbred wheat lines exhibiting resistance to drought. Notably, a QTL for grain yield was pinpointed in the proximal region of chromosome 4AL. This locus correlates with crucial traits such as the rate of grain filling, spike density, grain yield, drought sensitivity index, and biomass production [39]. Various authors have reported stress-related QTLs in diverse agricultural crops, particularly in wheat, as documented in Tables 2 and 3.

**Table 2.** Application of DNA markers for QTL mapping in agronomic crops

Crop	Botanical Name	DNA Marker	No. of QTLs	No. of Chromosomes with QTLs Loci	Objective	Reference
Cotton	<i>Gossypium hirsutum</i>	Simple sequence repeats (SSR);  Single strand conformation polymorphic (SSCP)	14	11	_ Salt resistance trait identification _ Mapping strength assessment for QTL detection  _ Inbred line development _ resistance to drought QTL detection linked to drought resistance	65

Maize	<i>Zea mays</i>	Single nucleotide polymorphism (SNP)	29	1,3,5	Assessing salt resistance at the seedling stage using unconditional and conditional QTLs	66
Sorghum	<i>Sorghum bicolor</i>	Restriction fragment length polymorphism (RFLP)	7	1 and 2	Drought resistance and lodging resistance detection before flowering	67
Barley	<i>Hordeum vulgare</i>	Single nucleotide polymorphism (SNP)	2 (salt tolerance indices)	1,2	<ul style="list-style-type: none"> <li>_ To estimate the genetic difference of Asian barley for salt resistance</li> <li>_ To identify and screen out salt resistance traits in accessions</li> </ul>	68

**Table 3:** Utilization of DNA Markers for QTL Mapping in Wheat (*Triticum aestivum*) Crops

DNA Markers	No. of QTLs Related to Tolerance	Chromosome No. with QTL Loci	Research Objectives	References
Simple sequence repeats (SSR)	3	1,5,7	_ To classify and map QTLs for	69

and amplified fragment length polymorphism (AFLP)			heat resistance. _ To identify the DNA markers linked with QTLs	
Simple sequence repeats (SSR), diversity array technology (DarT), gene-based marker for Vrn-A1	1	5A	_ Genetic structure of drought tolerance by reproductive stage.  _ Development of drought tolerance morphological method focusing on premature microspore stage of pollen development for eliminating stress at flowering time	70
Simple sequence repeats (SSR)	8	2A	_ To detect the linkages of SSR markers with drought resistance character on chromosome 2A	71

Rice exhibits considerable genetic diversity attributed to its progenitor species and widespread distribution across vast hectares. However, numerous stress factors significantly diminish yield and overall production. Abiotic stresses alone can lead to up to 50% losses in yield. Among these stressors, salinity poses a substantial biophysical challenge to rice production in diverse regions [72]. Resistance to salinity stress is intricately governed by genetic and physiological factors. Multiple stress-related Quantitative Trait Loci (QTLs) have been pinpointed in rice, as summarized in Table 4.

**Table 4:** Utilization of DNA Markers for QTL Mapping in Rice (*Oryza sativa*)

<b>DNA Markers</b>	<b>No. of QTLs Related to Tolerance</b>	<b>No. of Chromosomes with QTLs Loci</b>	<b>Research Objectives</b>	<b>References</b>
Simple sequence repeats (SSR)	47	1–4 and 6–12	- Reliable QTLs for drought tolerance and yield performance under stress conditions.  -Marker-assisted selection in rainfed areas	72
Single feature polymorphism, simple sequence repeats (SSR)	2	4 and 10	To detect possible QTL linked with high-temperature resistance.  - Gene mapping of heat tolerance	73
Simple sequence repeats (SSR)	1	3	- Heat tolerance related QTLs identification  - Marker identification for use in marker-assisted breeding	74

Detecting quantitative trait loci (QTLs) for salinity resistance through closely linked DNA markers offers a promising alternative to traditional breeding methods, which heavily rely on morphological analysis. In rice, researchers have identified twenty QTLs distributed across chromosomes 1, 2, 4, 6, 8, 9, and 12 [75]. Notably, distinct QTLs like qSESF12.1 and qSESI12.1 stand out, holding potential for precise mapping of loci and identifying closely linked markers crucial for enhancing salt resistance.

The cultivation of *Pisum sativum*, commonly known as pea, faces challenges posed by drought stress in various climatic conditions, yet research on drought tolerance and associated genetic resources in pea remains limited. Nevertheless, a study has identified genomic regions linked to drought tolerance, leveraging indicators of drought stress and relative soil and leaf water content to assess recombinant inbred pea lines [76]. Ten quantitative trait loci, independently explaining between 9 to 33 percent of the observed morphological differences, were identified. These findings, alongside the discovery of reproducible molecular markers associated with these QTLs, hold promise for selecting individuals with desired QTLs in pea breeding programs focused on drought tolerance.

Furthermore, enhancing cold tolerance in cold-season pea varieties presents a significant agricultural challenge. Breeding efforts must consider not only freezing resistance but also seed yield and quality. Recent research targeted the genetic determinants of cold and frost resistance, utilizing a newly identified cold-tolerant source [77]. Populations of recombinant inbred lines were evaluated across various climatic conditions, resulting in the construction of a comprehensive genetic map comprising 679 molecular markers spread across seven linkage groups. Through this analysis, researchers identified 161 QTLs, explaining between 9 to 71 percent of the observed morphological differences across measured traits. These findings suggest the potential for targeted breeding to improve frost resistance and enhance seed quality and productivity in cold-season pea varieties.

## **6. Marker-Assisted Selection using SNP Markers**

Association mapping offers the advantage of assessing a greater number of alleles in large populations compared to linkage analysis, thereby revealing evolutionary recombination and mutational characteristics across various lines. This method specifically targets genes associated with phenotypic diversity, making it instrumental in identifying genes responsible for quantitative variation, such as drought resistance [78,79]. However, association mapping is less effective in identifying rare alleles within plant populations and typically involves higher costs due to the need for genotyping and sequencing numerous lines [79].

The adoption of fixed multiplex SNP chips proves to be a time-saving and cost-effective approach for association mapping and genome-wide linkage. In contrast, linkage analysis necessitates allele separation through capillary electrophoresis and the use of multi-allelic markers. Research affirms the reliability of SNP chips in delivering high-quality data, precise genotyping information, and comprehensive genetic insights. Moreover, SNPs may offer advantages over traditional DNA markers like SSRs for linkage analysis, as they are abundant and exhibit genome-wide distribution characteristics, facilitating the identification of functional genes and genetic variations [80].

For instance, Hao et al. [81] identified 27 SNPs associated with genetic variations in drought tolerance among maize lines through the detection of functional genetic differences.

## **7. Conclusions**

Effective management of abiotic stress in agricultural crops is essential for achieving higher quality and yield. Molecular genetics has revolutionized this field by providing a plethora of DNA markers that facilitate the exploration of genetic modifications, genotypic resistance, stress-tolerant lines, and genetic information associated with abiotic stresses. While early molecular marker technologies offered fundamental insights into stress resistance, advancements in marker applications now enable the identification of specific genes or gene clusters responsible for abiotic stress tolerance. Integration of DNA markers with QTL mapping reveals a discernible pattern of stress tolerance genes located at specific chromosomal loci. However, the evolving climate necessitates continuous improvements in DNA marker technology to enable even more detailed analysis of stress tolerance mechanisms. This ongoing refinement will be crucial for agricultural resilience in the face of changing environmental conditions.

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