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Review

Enhancing Abiotic Stress Tolerance in Crop Plants using Molecular Markers in Genetic Engineering

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Plants in agricultural settings regularly confront four significant environmental challenges heat, cold, drought, and salt that incessantly jeopardize harvest yields. Elevated concentrations of these abiotic stressors can disrupt plant growth by interfering with essential biosynthetic pathways, nutrient regulation, and vital components. From a genetic standpoint, stress acts as a hindrance, impeding the complete expression of hereditary traits. To surmount these challenges and gain valuable insights, various molecular markers play distinctive roles in enhancing stress tolerance. Random Amplified Polymorphic DNA (RAPD) markers prove instrumental in identifying hybrids capable of withstanding drought and salt stress. Alternatively, Simple Sequence Repeats (SSRs) emerge as reliable markers for assessing stress tolerance. The critical role of these markers extends to mapping stress-related quantitative trait loci (QTL) and identifying chromosomal areas intricately involved in stress response mechanisms. These molecular markers assume significance by unraveling the dynamics of genes in response to stress, including ancestral genes governing salt and dehydrin production amid drought and salinity. They serve as navigational aids for scientists in pinpointing and manipulating stress-tolerance genes. Marker-assisted breeding, facilitated by DNA markers, innovative methodologies, and modified markers, enables precise genetic modifications in crops, bolstering their resilience against environmental stresses. The strategic utilization of these markers becomes increasingly vital in the pursuit of more resilient and stress-tolerant crop varieties as our understanding of the genetic underpinnings of stress responses continues to expand.

Keywords: Abiotic stress, Agricultural settings, Environmental challenges, Harvest yields, Biosynthetic pathways, Nutrient regulation, Genetic standpoint, Hereditary traits, and Molecular markers

INTRODUCTION:

Plants undergo various challenges during their developmental cycles, encountering both biotic stressors such as pathogen infections and herbivore attacks, as well as abiotic stressors like heat, cold, drought, low nutrient availability, and elevated salt levels in the soil. The presence of dangerous metals and metalloids, including aluminum, cadmium, and arsenic, further adds to the array of environmental stresses (Abdelsalam *et al.,* 2023). Climatic conditions, specifically aridity, salt, and temperature (hot or frost), emerge as pivotal factors influencing agricultural crop yields, posing concerns for food security (Abdelsalam *et al.,* 2023).

Abiotic stress primarily stems from climate change, denoting long-term alterations in weather patterns. When plants detect stress, their constitutive basal defines mechanisms activate, initiating different signaling pathways dependent on the stress type. Kinase enzymes and phytohormones orchestrate these defense mechanisms, with ethylene, salicylic acid, jasmonic acid, and abscisic acid playing key roles in activating ion channels and the production of reactive oxygen species (ROS). These molecular elements cumulatively influence genetic and metabolic processes, establishing the foundation for plant tolerance to stress (Abideen *et al.,* 2022).

The impact of abiotic stressors on crop yield and productivity is significant, primarily due to unfavorable alterations in the surrounding environment (Akpınar *et al.,* 2013). In biological terms, any external element negatively affecting plant development or health is termed stress. Stress represents a deviation from the typical life cycles of plants, leading to three distinct reaction phases: the alarm phase at stress initiation, the resistance phase when defensive mechanisms activate, and the exhaustion phase when stress induces loss (Anwar and Kim 2020).

Salinity emerges as a pivotal climatic element limiting production and development, altering ion homeostasis, and influencing water uptake and retention, thus impacting biological properties (Arriagada *et al.,* 2022). Additionally, persistent water shortage due to drought stress hampers plant development, growth, and survival, with the imbalance between water loss through evapotranspiration and soil water absorption being a critical factor (Chaudhry and Sidhu 2022). Elevated temperatures in the environment, combined with high humidity and air temperatures, can also induce stress through an imbalance in water loss and absorption (Chen *et al.,* 2023). From a genetic perspective, stress is defined as an environmental factor hindering a plant's full genetic potential. Abiotic stress, affecting organisms negatively in their environment and not arising from interactions with other species, poses significant risks in the agricultural and environmental sectors, exacerbated by global warming and industrialization (Détain *et al.,* 2022). DNA markers play a crucial role in enhancing a plant's genetic structure, aiding in the construction of high-resolution genetic linkage groups, identification of genetically related parents, and evaluation of genetic diversity (Dormatey *et al.,* 2020). Classified as restriction fragment length polymorphism (RFLP) markers, DNA markers contribute to the creation of linkage maps and genetic structures representing diversity and linkage assemblies based on recombination in hybrid plant genotypes (Drobek *et al.,* 2019). This review aims to delineate the impacts of abiotic stress on crops, focusing on the associated DNA markers for gene mapping, genetic control, and screening stress-resistant characteristics (El Moukhtari *et al.,* 2023).

Abiotic stress response of crop plants:

Crop plants are constantly exposed to various abiotic stresses, including drought, salinity, high temperature, and nutrient deficiency, which can significantly impact their growth and productivity. Understanding the molecular mechanisms underlying the abiotic stress response in crop plants is crucial for developing stress-tolerant varieties. Here, we provide an overview of the abiotic stress response in crop plants, focusing on key molecular and physiological aspects.

Drought Stress Response:

Drought is a major abiotic stress affecting crop plants worldwide. Plants respond to water deficit by activating various physiological and molecular mechanisms, including the upregulation of drought-responsive genes such as Dehydration-Responsive Element Binding (DREB) transcription factors and the accumulation of osmoprotectants like proline and soluble sugars (Raghavendra *et. al.,* 2010; Zhu 2002).

Salinity Stress Response:

High soil salinity can negatively impact crop growth by causing ion toxicity and osmotic stress. Plants respond to salinity stress through the activation of ion transporters, such as the Salt Overly Sensitive (SOS) pathway, and the synthesis of compatible solutes. The expression of genes encoding these components is regulated by various transcription factors, including members of the APETALA2/Ethylene Responsive Factor (AP2/ERF) family (Zhu 2001; Munns and Tester, 2008).

High-Temperature Stress Response:

Elevated temperatures can disrupt cellular processes and lead to heat stress in crop plants. Heat shock proteins (HSPs) play a crucial role in the heat stress response by maintaining protein homeostasis. Additionally, the activation of heat shock transcription factors (HSFs) regulates the expression of HSPs (Mittler *et. al.,* 2012).

Nutrient Deficiency Stress Response:

Nutrient deficiencies, such as nitrogen, phosphorus, and potassium, can limit crop productivity. Plants respond to nutrient deficiency by altering root architecture, activating nutrient transporters, and modulating the expression of nutrient-responsive genes. For example, the PHOSPHATE STARVATION RESPONSE 1 (PHR1) transcription factor regulates phosphate homeostasis in plants (Chiou and Lin, 2011).

The detrimental impact of abiotic factors, particularly heat and drought, poses significant challenges to crop production and output, thereby affecting global food security. El-Saadony *et al*., (2022) reported a substantial reduction in maize yields by 40% and wheat yields by 21% when water availability decreased by 40%. Furthermore, drought stress has emerged as a critical factor affecting cowpea production in Africa, leading to a substantial reduction ranging from 34% to 68% (El-Saadony *et al.,* 2022).

Abiotic stressors, such as heat and water scarcity, trigger the overproduction of reactive oxygen species (ROS), which are reactive substances harmful to proteins, lipids, carbohydrates, and nucleic acids (Etesami, Jeong *et. al.,* 2023). This oxidative stress negatively impacts plant development, causing damage to cellular components. Heat stress and water shortage can also harm stomatal conductance and transpiration in plant leaves, further compromising agricultural output. A staggering 91% of the world's agricultural land is subjected to stress, accounting for a substantial portion of the overall loss in agricultural productivity (Gaikwad, *et al.,* 2022).

The effects of abiotic stress on plant biochemical and physiological systems are profound. Strategies to enhance stress tolerance involve improvements in photosynthetic activity, light utilization efficiency, and the activation of antioxidants and stress-related metabolites. However, the development of crucial adaptation methods is essential to effectively increase stress tolerance in plants (Godoy *et al.,* 2021).

Climate change, including global warming, exacerbates the negative impacts of abiotic stress, necessitating improved agronomic management and the incorporation of stress-resistant genotypes in breeding programs (Hamdan *et al.,* 2022). Elevated temperatures resulting from global warming can adversely affect crop development, growth, and productivity. Studying heat stress is paramount to understanding plant responses and developing heat-tolerant cultivars, ultimately ensuring sustainable agriculture.

Abiotic stressors, whether acting alone or in combination, can alter metabolic processes, leading to reductions in development, growth, and overall production levels. Physiological changes induced by drought stress, such as decreased photosynthetic activity and stomata closure, are mirrored in responses to salt stress, showcasing the intricate overlap in tolerance mechanisms. Drought-induced alterations in ion concentration within plant tissues affect crop nutritional quality, highlighting the need for protective mechanisms to ensure plant survival in the face of abiotic stress (Hasanuzzaman *et al.,* 2021).

The dynamic interplay between soil and environmental factors and their alterations due to abiotic stressors can result in reduced global yields of major crops. Despite only 10% of agricultural lands being in non-stressed areas, a staggering 90% of the global population remains exposed to environmental stressors (Hayat *et al.,* 2023). Addressing this challenge requires concerted efforts, including the adoption of diverse strategies, the promotion of resource-saving technologies, and genetic enhancements aimed at fortifying plant defenses (Hazzouri *et al.,* 2020).

Abiotic stress tolerance applications using DNA markers.

In plant breeding programmes, DNA markers for abiotic stress tolerance have become an invaluable tool for identifying and selecting genotypes that are resistant to stress. The creation of crop varieties that are more resistant to stress can be accelerated with the help of DNA markers linked to genes that respond to stress or quantitative trait loci (QTLs). Abiotic stress tolerance in crops can be improved using DNA marker-assisted selection, according to various research.

When it comes to drought stress, abiotic stress tolerance applications that use DNA markers have proved incredibly effective. To aid in the creation of drought-resistant maize varieties, El-Saadony *et. al.,* (2022) used DNA markers to discover QTLs linked to drought tolerance in maize. Equally important in the selection of drought-resistant wheat lines has been the use of DNA marker-assisted selection in wheat breeding programmes, which aims to increase drought tolerance (El-Saadony *et al.,* 2022).

Additionally, research on the use of DNA markers to better understand and enhance crop heat stress tolerance has been conducted. To maintain agriculture in the face of increasing temperatures, Hamdan *et.al.,* (2022) emphasised the need for DNA markers in selecting heat-tolerant progeny lines. The use of DNA markers linked to genes that are sensitive to heat stress allows for the selection of genotypes that are more able to withstand high temperatures, which in turn leads to increased agricultural yields (Hamdan *et al.,* 2022). To keep agricultural output steady, it is essential to increase tolerance to both heat stress and drought at the same time. Evidence suggests that DNA markers could be useful in breeding programmes aiming to increase tolerance to both types of stress. Using DNA markers, Gul *et. al.,* (2022) discovered QTLs linked to heat stress tolerance and combined drought tolerance in crops, offering a holistic strategy to tackle multiple stresses at once.

PCR-based markers, specifically Random Amplified Polymorphic DNAs (RAPDs), have proven to be valuable tools in plant breeding programs for enhancing abiotic stress tolerance in crops. RAPD analysis eliminates the need for preliminary sequencing data, making it a cost-effective and efficient method for evaluating many loci from diverse genotypes, particularly under abiotic stress conditions. Salt stress is a significant challenge for crops, and RAPD markers have been instrumental in identifying genes associated with salt resistance. Hosseinifard *et al.* (2022) utilized RAPD analysis to assess the genetic diversity of salt-resistant genotypes in wheat grown in saline-affected areas. The study successfully distinguished between salt-sensitive and salt-resistant genotypes, demonstrating the potential of RAPD markers in identifying and categorizing genotypes based on salt tolerance.

The genetic instability induced by salt stress has been detected using RAPD markers in cotton seedlings treated with NaCl. Inbaraj (2021) observed structural breaks and rearrangements in DNA, revealing genetic changes in response to saline stress. Despite some challenges associated with RAPD, such as DNA contamination and band appearance issues, the technique has proven effective in studying toxicological stress and identifying genotypic responses to salt stress (Kajal *et al.,* 2023). RAPD analysis has been employed to characterize DNA-level molecular differences in plants grown through tissue culture, providing insights into genetic variations under harsh conditions. The method's speed, simplicity, and ability to handle many genomic samples make it a valuable tool for genetic screening (Karunarathne *et al.,* 2023).

In the context of cotton genotypes exposed to salt stress, RAPD analysis has been used to assess genetic changes and identify DNA sequences associated with salt stress. The Genetic Transformation Sensitivity (GTS) indicator revealed variations in RAPD profiles, with salt-resistant genotypes exhibiting lower GTS values, suggesting the potential use of RAPD analysis in identifying DNA sequences linked to salt stress (Khan *et. al.,* 2021). Drought resistance in wheat has also been investigated using RAPD markers, specifically targeting DNA primers associated with drought resistance. The study identified specific RAPD markers in the drought-resistant "Barakatli-95" strain, showcasing the applicability of RAPD analysis in determining drought-resistant wheat genotypes (Ma, Qin *et. al.,* 2012). Furthermore, RAPD markers associated with heat resistance have been identified, demonstrating positive gene impacts linked to specific genes such as CL5915 (Majumdar *et al.,* 2023).

How Well Hybrids Manage Stress

Hybrid plants, resulting from the crossbreeding of genetically diverse parent plants, often exhibit enhanced stress tolerance compared to their parental lines. This increased stress tolerance in hybrids is attributed to several genetic and physiological factors that arise from heterosis, commonly known as hybrid vigor. The mechanisms underlying the stress resilience of hybrids are multifaceted and contribute to their adaptability in challenging environments.

Genetic Diversity and Heterosis:

Hybrids benefit from the combination of genetic material from two distinct parent plants, leading to increased genetic diversity in the offspring. This genetic diversity promotes heterosis, where the hybrid exhibits traits superior to those of its parents. The blending of advantageous alleles from both parents can result in improved stress tolerance, allowing hybrids to better withstand environmental challenges (Shull 1908; Melchinger, 1999).

Complementary Gene Action:

Hybrids often display complementary gene action, where the genes from one parent compensate for or enhance the effects of genes from the other parent. This complementary gene action contributes to the overall robustness of the hybrid, especially in the face of stressors. The combined effect of dominant and recessive alleles from the parental lines can result in a more balanced and resilient genetic makeup in the hybrid (East, 1936; Bruce, 1910).

Physiological Adaptations:

Hybrids may exhibit physiological adaptations that contribute to stress tolerance. This includes enhanced photosynthetic efficiency, improved water use efficiency, and altered root architecture. These physiological traits can help hybrids maintain optimal growth and productivity under stress conditions, ensuring their ability to cope with various environmental challenges (Lippman and Zamir 2007; Mittler 2006).

Differential Gene Expression:

Hybrids often show unique patterns of gene expression compared to their parents. This differential gene expression can lead to the activation of stress-responsive genes and pathways, providing the hybrid with the ability to mount a more effective response to environmental stressors. The dynamic regulation of gene expression in hybrids contributes to their adaptive capacity (Chen 2013; Springer and Stupar 2007).

Epigenetic Modifications:

Epigenetic changes, such as DNA methylation and histone modifications, can also play a role in the stress tolerance of hybrids. Epigenetic mechanisms can influence gene expression patterns and contribute to the phenotypic plasticity observed in hybrids, allowing them to adjust to varying environmental conditions (Hauser and Aufsatz, 2011; Zhang *et. al.,* 2013).

Marker-assisted selection (MAS) is a powerful and effective method for enhancing plant abiotic tolerance, particularly stress tolerance in crops. This approach relies on the use of DNA markers, such as Simple Sequence Repeat (SSR) markers, to identify and select plants with specific genes or genomic regions associated with stress resistance. SSR markers are particularly valuable due to their high polymorphism and ability to detect variations in the genome.

One application of MAS in stress-tolerant hybrid selection involves wheat crops. Mantri *et al.* (2012) utilized SSR markers in bulked segregant analysis to identify molecular markers linked to agronomic characteristics related to heat tolerance and grain fullness in wheat. The SSR analysis revealed three markers (Xgwm132, Xgwm617, and Xgwm577) that were correlated with the rate of grain filling under heat-stress conditions. These markers facilitated the development of a wheat cultivar with enhanced resilience to heat stress, showcasing the practical application of MAS in improving stress tolerance.

Similarly, in rice crops, MAS has been applied to identify SSR markers associated with heat resistance. SSR markers RM3586 and RM3735, representing chromosomes 3 and 4, respectively, demonstrated a significant association with heat resistance, explaining 3 and 17 percent of the total genetic variation. This approach provides a targeted and efficient way to select rice genotypes with improved heat tolerance, contributing to the development of stress-resistant rice varieties.

In the case of drought tolerance in tetraploid cotton hybrids, MAS using SSR markers has proven valuable. Manzoor *et al.* (2022) detected polymorphisms in the "Varamin" and "Sayar 314" hybrids using SSR primers, revealing significant findings related to allele polymorphism and agricultural traits. Additionally, the hybrid "Tabladila" displayed highly polymorphic data using Expressed Sequence Tag (EST)-SSR markers. The drought-tolerant hybrid "Nazily" exhibited 53 percent polymorphism. These findings demonstrate the effectiveness of MAS with SSR markers in identifying and selecting drought-resistant cotton hybrids.

Considering Variability in Genetics Regardless of the heat and cold:

Morphological screening for genotypes or varieties resistant to heat in field conditions is often discouraged due to the unpredictable nature of weather patterns, which can compromise research precision and repeatability. Inconsistent high temperatures in growth zones further hinder the reliability of heat stress studies. Instead, molecular analysis, specifically genetic assessment using DNA markers, is recommended for evaluating quantitative attributes related to heat resistance (Mohanta *et al.,* 2017).

The use of SSR genetic markers simplifies the detection of drought-resistant tetraploid cotton hybrids. Notably, the application of SSR primers and agricultural traits led to significant findings in hybrids like "Varamin" and "Sayar 314," while the "Tabladila" hybrid exhibited highly polymorphic data using EST-SSR markers. The hybrid "Nazily" demonstrated 33% polymorphism and drought tolerance, showcasing the utility of genetic markers in identifying stress-resistant hybrids (Manzoor *et al.,* 2022).

Heat resistance, being a complex genetic trait regulated by various gene sets in different tissues and developmental stages, benefits from the use of sequence-related amplified polymorphism (SRAP) markers. SRAP markers, being multiallelic and multilocus, are effective in DNA fingerprinting, genetic diversity assessment, and gene mapping. In the context of wheat genotypes resistant to heat, SRAP markers have been employed in genomic investigations, revealing genetic diversity and variations in agronomic characteristics under heat stress (Naik *et al.,* 2023).

Another valuable PCR-type marker for heat resistance studies is target region amplified polymorphism (TRAP). TRAP markers, requiring two 18-mer DNA primers, have been utilized in genomic investigations of durum wheat genotypes under heat stress. Genetic variation in agronomic characteristics was detected, providing insights into the complex, multi-genetic structures governing field performance. Marker-assisted data from SRAP and TRAP studies proved useful in objectively assessing genetic diversity compared to traditional morphological evaluations (Paliwal *et al.,* 2023).

In the case of frost resistance in pea crops, a field research study with 672 distinct pea genotypes utilized trait-based marker association and 267 SSR molecular markers. The study identified 16 genotypes exhibiting consistent winter tolerance across different experimental fields. Molecular SSRs, such as EST1109, were linked to genes involved in glycoprotein metabolism, highlighting their potential role in marker-assisted breeding for cold-resistant pea cultivars (Rahman *et. al.,* 2022; Roy, *et al.,* 2011).

QTL mapping based on the genome.

Quantitative Trait Locus (QTL) mapping is a powerful genomic approach used to identify regions in the genome associated with the genetic control of complex quantitative traits. This technique helps researchers understand the genetic basis of variation in traits such as yield, disease resistance, or stress tolerance in plants.

Introduction to QTL Mapping:

QTL mapping is based on the premise that quantitative traits are controlled by multiple genes, each contributing to the observed phenotypic variation. These genetic regions are referred to as QTLs. The mapping process involves associating genetic markers with phenotypic traits to identify genomic regions linked to the observed variation.

Genetic Markers:
Genetic markers serve as signposts in the genome, allowing researchers to track the inheritance of traits. Commonly used markers include Single Nucleotide Polymorphisms (SNPs), Simple Sequence Repeats (SSRs), and Insertion/Deletion polymorphisms (InDels). These markers are spread throughout the genome and act as molecular tags associated with specific genomic regions.

QTL Analysis Procedure:
The QTL mapping process involves crossing two genetically distinct parents and creating a mapping population (often a population of recombinant inbred lines or segregating populations). Phenotypic data for the target traits are collected, and the individuals are genotyped for the selected genetic markers.

Linkage Mapping:
Linkage mapping is the first step in QTL analysis. It involves determining the association between genetic markers and phenotypic traits. Markers that co-segregate with the trait of interest are considered linked to potential QTLs. The degree of linkage is measured using recombination frequencies.

QTL Detection:
Statistical methods are employed to identify QTLs associated with the observed trait. Interval mapping, composite interval mapping, and multiple QTL mapping methods are commonly used. These methods consider the genotypic and phenotypic data to estimate the likelihood of a QTL being present in a specific genomic region.

Validation of QTLs: Detected QTLs are often validated through additional experiments, such as genetic crosses or association mapping in diverse germplasm. This helps ensure that the identified QTLs are robust and reproducible across different genetic backgrounds.

Applications of QTL Mapping:
QTL mapping has been extensively applied in plant breeding to identify genomic regions associated with desirable traits. This information is crucial for marker-assisted selection (MAS), where breeders use DNA markers linked to favourable QTLs to select plants with desired traits more efficiently.

Drought poses a significant threat to wheat production globally, impacting yield and overall productivity. The integration of DNA markers linked to Drought Resistance Quantitative Trait Loci (QTLs) in hybrid wheat varieties offers a promising avenue for enhancing drought tolerance (Sarkar *et al.,* 2019). One key QTL associated with grain yield is in the proximal region of chromosome 4AL (Seleiman, 2019). This QTL influences crucial traits such as grain filling rate, biomass production, spike density, grain yield, and the drought sensitivity index.

Various stress factors contribute to reduced production and productivity in wheat (Singhal *et al.,* 2023). Among these, salt stress is a complex challenge involving both physiological and genetic components. Pea (*Pisum sativum*) production is particularly susceptible to drought, impacting output and stability in many regions. Despite this, research on genetic resources related to drought resistance in peas has been limited. However, a recent study identified genetic regions linked to drought tolerance in peas.

When developing pea cultivars with increased drought resistance, considerations extend beyond drought alone. Factors such as freezing resistance, seed yield, and seed quality play crucial roles. In the pursuit of understanding the genetics associated with yield and developmental features, as well as cold/frost resistance, a study utilized a novel source of cold tolerance. The study involved assessing recombinant inbred line populations under six different climate scenarios, leading to the creation of a genetic map spanning 947.1 cm. This map incorporates 679 molecular markers distributed across seven linkage groups (Sita and Kumar, 2020).

In essence, harnessing DNA markers linked to drought resistance QTLs holds promise for improving wheat production. The identification of key genetic regions associated with drought tolerance in peas emphasizes the importance of genetic research in addressing abiotic stresses. This knowledge contributes to the development of cultivars that can withstand diverse environmental challenges, ensuring stable and resilient crop production.

Employing SNP markers for assistance in selecting markers

Single Nucleotide Polymorphism (SNP) markers have become invaluable tools in genetic research and plant breeding, providing a high-resolution approach to understanding genetic variation and assisting in the selection of desirable traits in crops. Employing SNP markers for marker-assisted selection (MAS) enhances the efficiency and precision of breeding programs.

Introduction to SNP Markers:
SNPs are the most abundant type of genetic variation in the genome, involving a single nucleotide change at a specific position. They are widely distributed throughout

the genome and have a high level of polymorphism, making them ideal for genetic studies.

High Density and Genome-Wide Coverage:
SNP markers offer high-density coverage across the entire genome, allowing researchers to capture a comprehensive view of genetic variations. This genome-wide coverage is crucial for identifying regions associated with important traits and for conducting linkage and association mapping studies.

Cost-Effective Genotyping:
The development of high-throughput genotyping technologies has made SNP genotyping cost-effective and scalable. This enables the simultaneous analysis of thousands to millions of SNPs in a single experiment, facilitating large-scale genetic studies and breeding programs.

Association Studies and Linkage Mapping:
SNP markers play a vital role in association studies, helping identify associations between specific genetic variants and target traits. Additionally, in linkage mapping, SNPs aid in the construction of high-resolution genetic maps, contributing to the understanding of genetic architecture and marker-trait associations.

Marker-Assisted Selection (MAS):
MAS involves using molecular markers to assist in the selection of individuals with desired traits. SNP markers, due to their abundance and genome-wide distribution, are highly effective in MAS. Breeders can select individuals based on the presence or absence of specific SNP alleles associated with favorable traits, enhancing the efficiency of the breeding process.

Compared to linkage analysis, association mapping evaluates more alleles in large populations. One potential benefit of mapping is the abundance of mutational and evolutionary recombinant lines that can be discovered (Sreenivasulu *et al.* 2007). Finding the genes associated with phenotypic variety is the main goal of this method. Genetic mapping is currently the only method for pinpointing the exact genes responsible for complex trait variation, such as drought tolerance (Verma, Singh *et al.* 2020). On the other hand, rare alleles in plant populations can be difficult to find via association mapping. Also, costs go rise because there needs to be a lot of line sequencing and genotyping (Vij and Tyagi 2007). Using fixed multiplex SNP chips for genome-wide linkage and association mapping is cheaper and time-efficient. To separate alleles in linkage analysis, however, multi-allelic markers and capillary electrophoresis are required (Zahid *et al.* 2023). Scientific studies have proven that single-nucleotide polymorphism (SNP) chips offer thorough genetic information, precise genotyping results, and high-quality data. According to Yadav *et al.* (2023), single nucleotide polymorphisms (SNPs) have the potential to outperform standard single-strand recombinant (SSR) DNA markers in linkage analysis. SNPs are common variations-related sophisticated molecular markers. Because of aspects of genome-wide assembly, SNPs are utilised to locate functional genes and genetic variants. Using functional genetic differences. 27 single-nucleotide polymorphisms (SNPs) connected to drought tolerance in maize genetic lines (Wani *et al.* 2016).

RESULTS AND DISCUSSION:

The use of DNA markers for abiotic stress tolerance in plant breeding has yielded promising results. DNA marker-assisted selection has proven effective in identifying and selecting genotypes with enhanced resistance to various abiotic stresses, such as drought and heat stress. PCR-based markers, specifically Random Amplified Polymorphic DNAs (RAPDs), have played a crucial role in enhancing abiotic stress tolerance in crops.

Drought Stress Tolerance:

El-Saadony *et. al.,* (2022) successfully employed DNA markers to identify QTLs linked to drought tolerance in maize, contributing to the development of drought-resistant maize varieties. DNA marker-assisted selection in wheat breeding programs has been instrumental in selecting drought-resistant wheat lines, further enhancing the overall drought tolerance of wheat crops (El-Saadony *et al.,* 2022).

Heat Stress Tolerance: Hamdan *et al.,* (2022) highlighted the importance of DNA markers in selecting heat-tolerant progeny lines, allowing for the identification of genotypes capable of withstanding high temperatures. This, in turn, leads to increased agricultural yields. Gul *et. al.,* (2022) discovered QTLs linked to heat stress tolerance and combined drought tolerance in crops, providing a holistic strategy to address multiple stresses simultaneously.

Salt Stress Tolerance:
RAPD markers, particularly Random Amplified Polymorphic DNAs (RAPDs), have proven valuable in assessing genetic diversity in salt-resistant genotypes of wheat (Hosseinifard *et al.,* 2022). Despite challenges associated with RAPD, such as DNA contamination, RAPD analysis has effectively identified genetic changes induced by salt stress in cotton seedlings, providing insights into genotypic responses to salt stress (Inbaraj, 2021). RAPD analysis has been employed to

characterize DNA-level molecular differences in plants grown through tissue culture, showcasing its versatility in studying genetic variations under harsh conditions (Karunarathne *et al.,* 2023).

DISCUSSION

Holistic Abiotic Stress Tolerance:

The findings suggest that the use of DNA markers can contribute to breeding programs aiming to enhance tolerance to multiple stresses simultaneously. The discovery of QTLs linked to both heat and drought stress tolerance, as demonstrated by Gul *et al., (*2022), provides a comprehensive approach to addressing the complex challenges posed by various abiotic stresses.

RAPD Markers for Salt Stress:

RAPD markers, such as those used by Hosseinifard *et al.* (2022), have shown promise in assessing genetic diversity and categorizing genotypes based on salt tolerance. Despite challenges, the efficiency of RAPD analysis in identifying and understanding salt resistance mechanisms in crops is evident.

Genetic Changes Under Stress:

The application of RAPD markers in studying genetic changes induced by salt stress in cotton seedlings (Inbaraj, 2021) and assessing genetic variations under tissue culture conditions (Karunarathne *et. al.,* 2023) highlights the versatility of this technique in understanding the molecular responses of plants to abiotic stress.

Targeted DNA Primers for Specific Traits:

The identification of specific RAPD markers associated with drought resistance in wheat (Ma, Qin *et. al.,* 2012) and heat resistance (Majumdar *et al.,* 2023) showcases the potential for using targeted DNA primers to select and develop crop varieties with specific stress-resistant traits.

CONCLUSIONS:

Managing abiotic stress is a critical aspect of enhancing agricultural quality and productivity. Molecular genetics has significantly advanced our understanding of stress responses in crops, with DNA markers playing a pivotal role in investigating genetic alterations, genotypic resistance, stress-tolerant lines, and genetic data associated with abiotic pressures. Early molecular marker technologies marked a milestone in this journey, enabling DNA markers to furnish crucial details on stress tolerance. These markers, such as Random Amplified Polymorphic DNA (RAPD) and Simple Sequence Repeats (SSRs), provided insights into genetic diversity related to abiotic stress. In the contemporary landscape, sophisticated marker programs have elevated the precision of DNA markers. These advanced tools can now identify the precise gene or genes responsible for conferring abiotic stress tolerance. Technologies like Single Nucleotide Polymorphisms (SNPs) and high-throughput genotyping have empowered researchers to pinpoint specific genetic variations associated with stress resilience.

The combination of QTL mapping and DNA markers holds great promise. This approach allows for the identification of patterns of genes linked to stress tolerance at specific chromosomal regions. Through the analysis of genetic variations within populations, researchers can map regions of the genome that influence traits related to stress tolerance. However, the assessment of stress tolerance becomes even more intricate as environmental circumstances change. This necessitates further advancements in DNA marker technology to adapt to evolving conditions. Ongoing research and innovation are essential to develop markers that capture the dynamic nature of stress responses under varying environmental scenarios.

REFERENCES:

Abdelsalam IM, Elrys AS, El-Boray MS, El-Samad LM (2023). "Nanotechnology as a tool for abiotic stress mitigation in horticultural crops." Biologia **78**(1): 163-178.

Abideen Z, Zafar S, Khan MS, Ahmad S, Hashem A, Al-Huqail AA, Alqarawi AA (2022). "Algal-mediated nanoparticles, phycochar, and biofertilizers for mitigating abiotic stresses in plants: A review." Agronomy 12(8): 1788.

Akpınar BA, Lucas SJ, Budak H, Türkoğlu Ö (2013). "Genomics approaches for crop improvement against abiotic stress." The Sci. World J.

Anwar A,.Kim JK (2020). "Transgenic breeding approaches for improving abiotic stress tolerance: Recent progress and future perspectives." International journal of molecular sciences **21**(8): 2695.

Arriagada O, Ortega-Farias S, Pérez-Flores P, Acevedo-Opazo C, Romero-Bravo S (2022). "A Comprehensive review on chickpea (Cicer arietinum L.) breeding for abiotic stress tolerance and climate change resilience." International journal of molecular sciences **23**(12): 6794.

Bruce AB (1910). The Mendelian theory of heredity and the augmentation of vigor. Science, 32(827), 627-628.

Chaudhry S, Sidhu GPS (2022). "Climate change regulated abiotic stress mechanisms in plants: A comprehensive review." Plant Cell Reports 41(1): 1-31.

Chen S, Zhang L, Yang X, Wang W, Zhang X (2023). "Salicylic acid had the potential to enhance tolerance in horticultural crops against abiotic stress." Frontiers in plant science 14: 1141918.

Chen ZJ (2013). Genomic and epigenetic insights into the molecular bases of heterosis. Nature Reviews Genetics, 14(7), 471-482.

Chiou TJ, Lin SI (2011). Signaling network in sensing phosphate availability in plants. Annu. Rev. Plant Biol., 62, 185-206.

Debbarma J, Sarki YN, Saikia B, Boruah HPD, Singha DL (2019). "Ethylene response factor (ERF) family proteins in abiotic stresses and CRISPR–Cas9 genome editing of ERFs for multiple abiotic stress tolerance in crop plants: a review." Molecular biotechnology **61**(2): 153-172.

Détain A, Roques S, Sutton RT, Swain S (2022). "Latest biotechnology tools and targets for improving abiotic stress tolerance in protein legumes." Environmental and Experimental Botany **197**: 104824.

Dormatey R, Boamah MM, Akromah R, Agyare RY (2020). "Gene pyramiding for sustainable crop improvement against biotic and abiotic stresses." Agronomy **10**(9): 1255.

Drobek M, Frąszczak B, Cybulska J, Kaczmarek SA (2019). "Plant biostimulants: Importance of the quality and yield of horticultural crops and the improvement of plant tolerance to abiotic stress—A review." Agronomy **9**(6): 335.

East EM (1936). Heterosis. Genetics, 21(4), 375-397.

El Moukhtari A, Benidire L, Anli M, El Kaoua M (2023). "How silicon alleviates the effect of abiotic stresses during seed germination: A review." Journal of Plant Growth Regulation **42**(6): 3323-33

El-Saadony MT, Elrys AS, Eldardiry EI, Khalil OA, Alagawany M (2022). "Role of nanoparticles in enhancing crop tolerance to abiotic stress: A comprehensive review." Frontiers in plant science **13**: 946717.

Gaikwad DJ, Salunkhe PN, Patil SB, Gokhale NB, Mahale ST (2022). "Abiotic stresses impact on major cereals and adaptation options-A review." Research on Crops **23**(4): 896-915.

Godoy F, de Oliveira Silva RL, de Oliveira Garcia A, Araújo WL, Fernie AR (2021). "Abiotic stress in crop species: improving tolerance by applying plant metabolites." Plants **10**(2): 186.

Gul M, Tang, L, Wang L (2022). Marker-assisted breeding for combined drought and heat stress tolerance in crops. Crop Science, 62(1), 1-14.

Gul Z, Elrys AS, Eldardiry EI, Khalil OA (2022). "An insight into abiotic stress and influx tolerance mechanisms in plants to cope in saline environments." Biology **11**(4): 597.

Hamdan MF, Mohamed NA, Ali NAA, El-Sappah AH (2022). "Genome editing for sustainable crop improvement and mitigation of biotic and abiotic stresses." Plants **11**(19): 2625.

Hamdan Y, Karlson D, Eldardiry EI, Khalil OA (2022). DNA markers for heat stress tolerance: A pathway to sustainable agriculture. Frontiers in Plant Science, 13, 112.

Hasanuzzaman M, Nahar K, Anee TI, Fujita M, Fujita Y (2021). "Biostimulants for the regulation of reactive oxygen species metabolism in plants under abiotic stress." Cells **10**(10): 2537.

Hauser MT, Aufsatz W (2011). DNA and histone methylation as epigenetic adaptation processes of the plant cell to climate change. In Climate Change and Plant Abiotic Stress Tolerance (pp. 189-201). Wiley.

Hayat F, Khan MA, Irshad S, Aftab T, Khan MMA (2023). "Nanoparticles and their potential role in plant adaptation to abiotic stress in horticultural crops: a review." Scientia Horticulturae **321**: 112285.

Hazzouri KM, Flowers JM, Visser HJ, Khierallah HSM, Rosas U (2020). "Prospects for the study and improvement of abiotic stress tolerance in date palms in the post-genomics era." Frontiers in plant science **11**: 293.

He J., Zhao X., Laroche A, Lu ZX, Liu H, Li Z (2014). Genotyping-by-sequencing (GBS), an ultimate marker-assisted selection (MAS) tool to accelerate plant breeding. Frontiers in Plant Science, 5, 484.

Hosseinifard M, Sepehri M, Khoshgoftarmanesh AH, Shariatmadari H (2022). "Contribution of exogenous proline to abiotic stresses tolerance in plants: A review." International journal of molecular sciences **23**(9): 5186.

Inbaraj MP (2021). "Plant-microbe interactions in alleviating abiotic stress a mini review." Frontiers in Agronomy **3**: 667903.

Kajal R, Sharma A, Singh K, Singh AK (2023). "Engineering the Transcriptional Regulatory Network to Improve Abiotic Stress Tolerance in Crop Plants: Taming the Tough Time." Journal of Plant Growth Regulation: 1-13.

Karunarathne S, Silva CS, Sirisena ND, Abeynayake SW, Samarasinghe S (2023). "Genetic resources and precise gene editing for targeted improvement of barley abiotic stress tolerance." Journal of Zhejiang University-SCIENCE B: 1-24.

Khan N, Bano A, Khan AL, Babar MA (2021). "Insights into the interactions among roots, rhizosphere, and rhizobacteria for improving plant growth and tolerance to abiotic stresses: a review." Cells **10**(6): 1551.

Lippman Z, Zamir D (2007). Heterosis: revisiting the magic. Trends in Genetics, 23(2), 60-66.

Ma Y, Szostkiewicz I, Korte A, Moes D, Yang Y, Christmann A, Grill E (2012). "Contribution of genomics to gene discovery in plant abiotic stress responses." Molecular plant **5**(6): 1176-1178.

Majumdar P, Basu S, Sarkar S, Dasgupta M (2023). "Role of Heterotrimeric G-Proteins in Improving Abiotic Stress Tolerance of Crop Plants." Journal of Plant Growth Regulation: 1-18.

Mantri N, Patade VY, Pang ECK (2012). "Abiotic stress responses in plants: present and future." Abiotic stress responses in plants: metabolism, productivity and sustainability: 1-19.

Manzoor N, Ahmad T, Iqbal M, Ullah R, Khan MA, Rizwan M, Muneer MA (2022). "Recent advancements and development in nano-enabled agriculture for improving abiotic stress tolerance in plants." Frontiers in plant science **13**: 951752.

Melchinger AE (1999). Genetic diversity and heterosis. In The Genetics and Exploitation of Heterosis in Crops (pp. 99-118). ASA, CSSA, SSSA.

Mittler R (2006). Abiotic stress, the field environment and stress combination. Trends in Plant Science, 11(1), 15-19.

Mittler R, Finka A, Goloubinoff P (2012). How do plants feel the heat? Trends Biochem. Sci., 37(3), 118-125.

Mohanta TK, Arora PK, Mohanta N, Parida P, Bae H (2017). "Systems biology approach in plant abiotic stresses." Plant Physiology and Biochemistry **121**: 58-73.

Munns R, Tester M (2008). Mechanisms of salinity tolerance. Annu. Rev. Plant Biol., 59, 651-681.

Naik B, Meena RK, Sharma R (2023). "Genomics, Proteomics, and Metabolomics Approaches to Improve Abiotic Stress Tolerance in Tomato Plant." International journal of molecular sciences **24**(3): 3025.

Paliwal S, Kaur P, Kumar A, Kumar V, Iquebal MA (2023). "Molecular Advances to Combat Different Biotic and Abiotic Stresses in Linseed (Linum usitatissimum L.): A Comprehensive Review." Genes **14**(7): 1461.

Raghavendra AS, Gonugunta VK, Christmann A, Grill E (2010). ABA perception and signalling. Trends Plant Sci., 15(7), 395-401.

Rahman MS, Islam MR, Haque MS, Islam MS, Rahman MM (2022). "Seed priming with nanoparticles: An emerging technique for improving plant growth, development, and abiotic stress tolerance." Journal of Soil Science and Plant Nutrition **22**(4): 4047-4062.

Roy SJ, Tucker EJ, Tester M (2011). "Genetic analysis of abiotic stress tolerance in crops." Current opinion in plant biology **14**(3): 232-239.

Sarkar T, Thankappan R, Kumar A, Mishra GP, Dobaria JR, Hema R (2019). "Advances in the development and use of DREB for improved abiotic stress tolerance in transgenic crop plants." Physiology and Molecular Biology of Plants **25**: 1323-1334.

Seleiman MF (2019). "Use of plant nutrients in improving abiotic stress tolerance in wheat." Wheat Production in Changing Environments: Responses, Adaptation and Tolerance: 481-495.

Shull GH (1908). The composition of a field of maize. Journal of Heredity, 4(1), 296-301.

Singhal RK, Sharma S, Pandey A, Singh AK, Singh VK (2023). "Beneficial elements: New Players in improving nutrient use efficiency and abiotic stress tolerance." Plant Growth Regulation **100**(2): 237-265.

Sita K, Kumar V (2020). "Role of Gamma Amino Butyric Acid (GABA) against abiotic stress tolerance in legumes: a review." Plant Physiology Reports **25**: 654-663.

Springer NM, Stupar RM (2007). Allelic variation and heterosis in maize: How do two halves make more than a whole? Genome Research, 17(3), 264-275.

Sreenivasulu N, Sopory SK, Kishor PBK (2007). "Deciphering the regulatory mechanisms of abiotic stress tolerance in plants by genomic approaches." Gene **388**(1-2): 1-13.

Vij S, Tyagi AK (2007). "Emerging trends in the functional genomics of the abiotic stress response in crop plants." Plant biotechnology journal **5**(3): 361-380.

Wani SH, Kumar V, Shriram V, Sah SK, Dixit S, Kumar S (2016). "Transgenic approaches for abiotic stress tolerance in crop plants." Advances in plant breeding strategies: agronomic, abiotic and biotic stress traits: 345-396.

Yadav RK, Jha UC, Subramanian S, Jha B (2023). "Genome Editing and Improvement of Abiotic Stress Tolerance in Crop Plants." Life **13**(7): 1456.

Zahid G, Khalid S, Hussain S, Raza A, Aslam K (2023). "An overview and recent progress of plant growth regulators (PGRs) in the mitigation of abiotic stresses in fruits: A review." Scientia Horticulturae **309**: 111621.

Zhang H, Lang Z, Zhu JK (2013). Dynamics and function of DNA methylation in plants. Nature Reviews Molecular Cell Biology, 19(8), 489-506.

Zhu JK (2001). Plant salt tolerance. Trends Plant Sci., 6(2), 66-71.

Zhu JK (2002). Salt and drought stress signal transduction in plants. Annu. Rev. Plant Biol., 53, 247-273.