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# Genetics of Abiotic Stress Tolerance in Plants: Concept and Research Trends

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**ABSTRACT: Abiotic environmental stresses pose a threat to food availability in future as they may lessen farming output to only 20% or even less of the farm's inherent yields. Crop failure is mostly caused by abiotic pressures including salinity of soil, moisture stress, chilling and toxicity of heavy minerals which prevent crops from reaching their innate genetic capacity. The effect of excess salt, dryness and toxicity of heavy minerals put adverse effect on cells by upsetting their ionic and osmotic balance while chilling induces mechanical restrain to biological phospholipid membrane. Responses to abiotic pressures mediated by a variety of molecular signaling mechanisms. For crop development, it is crucial to comprehend molecular signaling networks and identify critical compounds and their specialized functions. A number of genes that code for antioxidants, enzymes that alter phospholipids of biological membrane, stress responsive transcription factors(TF$_S$), Ion (Ca$^{+2}$) homeostasis controlling cellular proteins, HSP$_S$, and enzymes that consolidate significant compounds which are responsive to stress have been identified as being responsible for endurance of abiotic pressures. Several different methods have been employed to enhance the stress resistance of various farm species, including conventional breeding techniques, shuttle breeding and mutagenesis. The present scenario of climate change demands a better level of tolerance or resistance in future varieties. Advanced breeding approaches such as GWAS, GS, Genome editing might open up new possibilities for cultivating plants that can adjust to a rapidly changing environment and still providing good yields in the face of extreme environmental stress. In this article, we will converse about the consequences of prominent abiotic pressures as well as behavior of plant to various external stresses as regards of physiology and breeding approaches used to achieve resistance or tolerance to them.**

**Keywords:** Abiotic stress, GWAS, GS, HSP$_S$, Genome editing, Mutagenesis.

## INTRODUCTION

The operation of agriculture domestication, which began over 11,000 years ago when humans began agriculture. This process of domestication led to development of high yielding cultivars of plants. Every area of human society has changed as a result of the impact of these crop varieties. These crops, however, are experiencing considerable yield losses because they are unable to adapt to the shifting environmental circumstances. Nearly 40 years ago, Boyer predicted that adverse environmental effects may lower crop productivity by roughly 70%, which would be disastrous for the globe (Boyer, 1982). Today's globe is facing significant vulnerabilities from the damaging effect of external environment pressures such as excessive temperature, moisture stress and accumulation of excessive salts which are elevated by climatic variations and global heating (Esmaeili *et al.,* 2019). The current challenge is to increase food production in changing environment condition. Therefore, in order to deal with regard to the imminent problem of food sustainability, now it is necessary to acquire stress-tolerant cultivars (Lesk *et al.,* 2016). Prior to anything else, it's critical to comprehend apprehension of stress. As plants are immobile, they are exposed to variety of pressures. Although, through a variety of molecular signaling pathways, plant counteract to numerous external stresses *viz.,* soil salinity, excessive temperature, chilling, dryness, cytotoxicity of heavy minerals. Based on nature of stress, the reaction environmental changes may be quick and may entailed either of adaptive strategies that help crop plants to endure the respective challenging circumstances or specialized growing habits to bypass the stressful situations (Esmaeili *et al.,* 2022). In actuality, plants are able to sense abiotic challenges and respond effectively by altering their inner metabolic reactions, growing and developing conditions by production of different compounds. Plants can synchronize their reactions to stressful circumstances by using signaling pathways to coordinate across cells and organs. They modulate their transcriptional activity in reaction to stress, allowing them to create particular proteins that aid in coping (Golldack *et al.,* 2014). Yet, it is challenging to fully comprehend the underlying processes of plant stress responses due to their complexity. Additionally, many stress responses are context-dependent and can vary depending on the type of stress and the plant species involved. So, more investigation is required to completely understand the plant stress response and how to control it to increase crop yields and agricultural sustainability. Although traditional breeding, molecular methods, and genetic engineering considerably contributed to the advancement of farm cultivars resistant to plenty biotic pressures (Nongpiur *et al.,* 2016), however little progress was made in tackling abiotic stresses due to the complicated genetics involved in resistance mechanisms. Popular cultivars of many crops are susceptible to drought and other abiotic stresses. Although few varieties having characteristic feature of abiotic stress tolerant have been released in commercial crops, but failed to occupy large coverage area. The present scenario of climate change demands a better level of tolerance or resistance in future varieties. Therefore, it is essential to develop a different strategy that might be used to increase crop yield and quality while improving abiotic stress resistance. Yet, recent headway in genomics, transcriptomics, proteomics, and metabolomics have given us novel strategies and tools for comprehending the genetic basis of stress tolerance and creating cultivars that are resistant to external pressures (Van Emon, 2016). Examples include the discovery of genes and genetic markers linked to agricultural stress tolerance using genome-wide association studies (GWAS) and quantitative trait loci (QTL) mapping.

**Consequences of abiotic stress.** Abiotic stress elements such as freezing, moisture scarcity, soil salinity, and accumulation of heavy minerals can have a significant impact on plant growth and farmingoutput. These stresses can occur at various times during plant growth and can have an impact on plants at multiple levels, including their morphological structure to their molecular activities (Odhong *et al.,* 2019).For instance, drought stress can result in decreased water availability, which causes plant withering, stunted development, and decreased agricultural output. It can also have an impact on a plant's physiology and metabolism, ensuing in changes in the concentration of different substances including enzymes, proteins, and carbohydrates. The physiological reactions of cultivars to stress include leaf withering, leaf abscission, shrinking of leaf dimensions, and dwindled water loss viatranspiration (Fghire*et al.,* 2015). Turgor pressure, one of the utmostintricate physiological operations that promote cell development, is lowered during drought stress. Water flow is disrupted from the xylem to the nearby elongating cells in higher plants during drought stress, which inhibits cell elongation.

Low temperatures stress has a significant mark on both plant survival and geographic dispersion. Low temperature stress in plants can lead to stunted development, chlorosis (leaf yellowing), decline in root growth, reduced seed germination and subsequent seedling growth and even mortality (Chinnusamy *et al.,* 2007). Low temperature stress can affect the activity of proteins and enzymes, chlorophyll synthesis, decrease membrane fluidity, may generate toxicity due to $H_2O_2$ and interfere with cellular signaling pathways. Cell death or damage may be the end result of these modifications (Pearce, 2001). Additionally, it inhibits metabolism, wastes energy, and leads to the formation of free radicals as a result of oxidative stress.

Increased salt content in the soil causes salinity stress. Higher salt content has two important effects on crops: ionic toxicity and osmotic stress. By impairing a plant's ability to absorb water and nutrients from the soil (preferably $K^+$ and $Ca^{2+}$), salt stress can result in decreased plant growth and output (Bassirirad, 2000). High salt concentrations can also lead to ion imbalances within the plant for example $Na^+$ and $Cl^-$ ions travel inside the cells and negatively impact the cell membrane and cytosolic metabolism along with production of Reactive oxygen species (ROS) that have an impact on the plant's overall health.

The adverse effects on farm cultivars of temperatures higher than the optimal temperature is elucidated as heat stress. Due to increased water loss through transpiration and evaporation, high temperatures can exacerbate a drought situation. High-temperature stress can seriously harm proteins, halt protein synthesis, deactivate vital enzymes, and harm membranes. The process of cell division can be significantly impacted by high temperature stress (Smertenko *et al.,* 1997). All of these negative effects can significantly impede plant growth and encourage oxidative damage. High temperatures can have an adverse effect on seed germination, plant growth, and development, as well as because an irreversible drought stresses that can be fatal (Takahashi *et al.,* 2013).

Heavy metal atmosphere pollution caused by human activity or natural processes is a common and important issue. Potentially harmful components are frequently cited as trace or Heavy metals. These metals can accumulate in plant tissues and disrupt various metabolic processes led to reduced growth, chlorosis, and even plant death in severe cases (Pourrut *et al.,* 2011). Lead, cadmium, mercury, and chromium are examples of heavy metals that are poisonous to plants and can havea number of detrimental impacts, including decreased productivity. Oxidative stress results from the production of ROS, which are highly reactive molecules that can harm biological components like DNA, proteins, and lipids. The plant cell's redox equilibrium, which is necessary for typical biological processes like photosynthesis and respiration, can be disturbed by oxidative stress (Apel and Hirt 2004). Moreover, these metals also alter metabolic pathways and interfere with enzyme activities in the plant cell, which further reduces plant output.

**How plant will counteract abiotic stress.** Abiotic stress refers to the detrimental impacts of non-living environmental conditions as drought, salt, severe temperatures, and heavy metal toxicity. Plants have developed a variety of defense mechanisms to combat abiotic stress. Under conditions of water scarcity or excess, plants may control their water uptake, transport, and loss to maintain the ideal water balance (Ashraf, 2010). For instance, they can slow down transpiration by blocking stomata, grow deep roots to access water in the soil's deeper layers, or store osmolytes like proline or carbohydrates to keep turgor pressure up (Lynch, 2011). Under stressful circumstances, plants can modify their metabolic pathways to better allocate resources and use energy. To prevent oxidative damage, for instance, they might switch from photosynthesis to respiration, make stress-specific proteins (such as chaperones, enzymes, or transporters) to repair or detoxify damaged molecules, or control the expression of genes involved in stress signaling and adaptation. Higher plants have developed a sophisticated signaling cascade that may engage in discourse in order to sense various environmental signals. A typical route concerning to excessive accumulation of salt, dryness, and cold stresses has been depicted in figure1. Apart from these, plants can reinforce their tissues and organs to withstand mechanical and osmotic stress. They may synthesis wax or trichomes to block excessive sunlight and heat absorption, for instance, or lignin, suberin, or cutin to reinforce their cell walls and decrease water loss. They may also build up calcium, boron, or silicon to improve the stability of their biological membranes and cytoskeleton (Mahajan and Tuteja 2005). To deal with stress, plants can form symbiotic or antagonistic partnerships with other living things. For instance, they can establish mycorrhizal relationships with fungi to improve their absorption of nutrients and water, draw in helpful insects or microorganisms to control infections or pests, or emit allele chemicals to fend off rival plants or herbivores (Meharg, 2003). The ability of plants to combat abiotic stress is, in general, a dynamic and complicated process that incorporates several levels of biological organization, from molecules to ecosystems. Plant breeders and genetic engineers can exploit these mechanisms to develop stress-tolerant crops and improve farming output in a changing climate (Meena *et al.,* 2016).

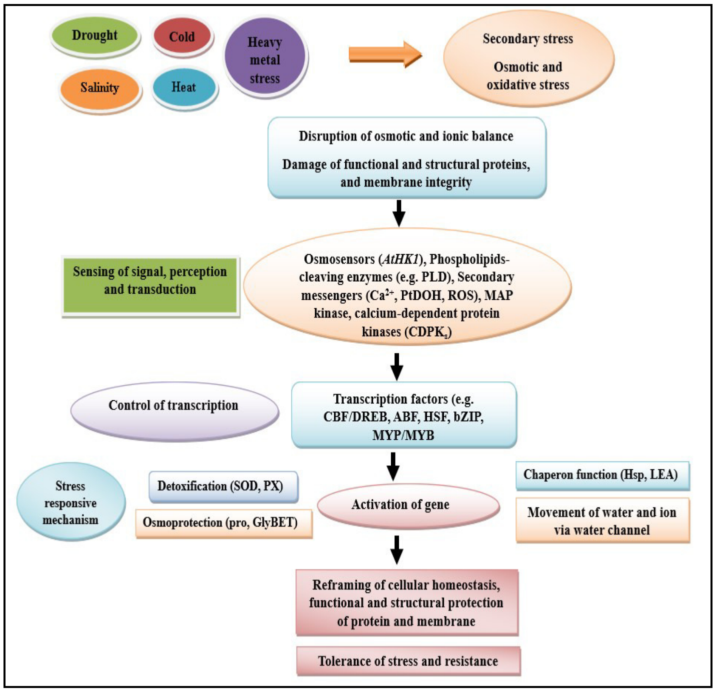

**Fig. 1.** Complexity of plant response to abiotic stress (Modified from Wang *et al.*, 2003).

**Physiology of salinity stress tolerance in plants.** A significant abiotic stress that has a global effect on vegetation development and output is salinity stress. Soil salinization is caused by both natural and anthropogenic activities. Salt buildup in soil prevents roots from absorbing water, disturbs ion homeostasis, and also causes mineral toxicity stress (Ashraf *et al.,* 2008). Here are some of the key physiological mechanisms that contribute to salinity stress tolerance in plants including ion transport and compartmentalization, Osmotic adjustment, ROS detoxification, Hormonal regulation (Shi *et al.*, 2003).The buildup of harmful ions like sodium (Na$^+$) and chloride (Cl$^-$) in plant tissues is one of the main impacts of salt stress. Plants have developed ways of avoiding these ions from entering the cytoplasm and transporting them into vacuoles or other intracellular compartments in order to prevent toxicity. A variety of transmembrane proteins, including ion channels, pumps, and antiporters, are involved in this. For instance, it has been demonstrated that the *SOS1* transporter in *Arabidopsis thaliana* is essential for Na$^+$ exclusion from root cells in saline environments (Zhu *et al.,* 1998). According to studies, over expressing a number of responsive genes linked to stress could escalate a plant's tolerance to salt by lowering the assimilation of harmful mineral ions like Sodium (Na$^+$)in the cytoplasmic matrix (Gaxiola *et al.*, 2001).Up regulation of *AtNHX1* (Arabidopsis vacuolar Na+/H+ antiporter gene 1) orthologs suchas *AtNHX5, OsNHX1, MdNHX1, TaNHX2, PgNHX1*, and*LeNHX2* also exhibited enhanced salinity tolerance in crops like *Oryza* spp. (Rice), *Solanum* spp.(eggplant, tomato), *Glycine max* (soybean), *Malus* spp. (Apple) (Li *et al.*, 2010). Salinity stress also causes a water crisis in plant tissues, which can interfere with regular metabolic functions and cause cellular damage. To combat this, plants can store suitable solutes such proline, glycine betaine, and sugars, which help toregulate turgor pressure and stabilize proteins and membranes, this process is known as osmotic adjustment and is regulated by a range of enzymes, including those responsible for the production and breakdown of suitable solutes (Munns and Tester 2008). A number of genes amenable to stress are controlled by a variety of transcription factors in order to control how plants respond to environmental challenges. *MYC, bZIP, WRKY, NAC*, and *AP2* are a few transcription factors whose roles in salt signaling pathways have been discovered (Golldack *et al*., 2011).Reactive oxygen species (ROS) including hydrogen peroxide ($H_2O_2$) and superoxide ($O_2$), which can lead to oxidative damage to lipids, proteins, and DNA, can also be produced by salinity stress. In order to prevent this, plants have developed a variety of antioxidative enzymes that scavenge and detoxify ROS, including Catalase, Peroxidase, and Superoxide dismutase (SOD). Ascorbate and glutathione, two non-enzymatic antioxidants that operate as ROS scavengers and support redox equilibrium, are also produced by plants (Gill and Tuteja, 2010).Plant hormone levels and signaling pathways may vary in response to salinity stress, which may then have an impact on a number of physiological functions. For instance, it is well known that the stress hormone abscisic acid (ABA) regulates stomatal closure and osmotic adjustment in reaction to salinity stress (Munns and Tester 2008). Other hormones such as jasmonates, ethylene, and cytokinins have also been implicated in salinity stress responses in various plant species (Deinlein *et al.,* 2014). Many transgenes have been uncovered, acquired, amplified, and reproduced in plants as possible sources of resilience to abiotic stress (Table 1).

**Table 1: Transgenes that have been shown to confer resistance to Salt Stress.**

| Transgene | Isolated from | Function | Transferred into | Protection from Abiotic stress |
|---|---|---|---|---|
| *Mtl1*D | *E. coli* | Mannitol-1-phosphate dehydrogenase | Tobacco | Salt stress |
| | | | *Arabidopsis* | Germination of seeds in high salt medium |
| P5CS | Moth bean | Pyrroline-5-carbosylate synthetase | Tobacco | Salt stress |
| *hva1* | Barley | A class-3 Lea protein | Rice | Salt stress |
| *bet*A | *E. coli* | Choline dehydrogenase | Tobacco | Salt stress |
| *cod*A | *Arthobacter globiformis* | Choline oxidase | *Arabidopsis* | Salt stress |
| *ADC* | Oat | Arginine decarboxylase | Rice | Salt stress |
| *sal1* | *Arabidopsis* sp. | Sulphur assimilation | Yeast | High salt stress |

**Physiology of drought stress tolerance in plants.** Water stress has marked effect on cellular processes, growth and development of farm plant and economic yield. Several physiological and molecular defense mechanisms, including adjustments to water relations, photosynthesis, metabolism, and hormone signaling, have been created by plants to deal with moisture stress. Performance of a crop under water stress will be distressed by the integrated effects of water stress at all the levels of plant organization (Ceccarelli and Grando 1996). Plants can reduce water loss by closing their stomata, which are pores on the leaves that regulate gas exchange and water vapor loss, leading to a reduction in carbon assimilation. Water saving species reduces transpiration mostly by closure of their stomata in response to water deficit well before wilting (stomatal sensitivity to water stress). Nevertheless, excessive stomatal closure can hinder growth and lessen carbon uptake. Research have revealed that plants with greater stomatal regulation—specifically, those that can balance carbon uptake and water loss under drought stress have a higher tolerance to drought stress (Lawson and Blatt 2014). Abscisic Acid (ABA) plays a vital role in water stress avoidance by effecting stomata closure, reducing leaf expansion and promoting root growth. Several components like ABA receptors; G-proteins, protein kinase *etc*. are inculpated in regulation of stomata closure by ABA (Fahad *et al*., 2017). Many of the stress proteins, *e.g.,* dehydrin, osmotin, Lea proteins, *etc*., are also fabricated in reaction to ABA; genes encoding such proteins are referred as *ABA-responsive (ABAR)* genes. *ABAR* genes have in their promoters an ABA response element *(ABARE),* which has ACGT as its core sequence. ABA increases the expression of the majority of genes associated with drought (such as NCED, RD22, ABREs, and RD29), and this stimulation can be up to 40 times more in the proximity of drought stress than in the proximity of usual growing circumstances (Shinozaki *et al*., 2003).The transcription factors known as drought-responsive element (DRE)-binding proteins, notably*DREB1* and *DREB2*, enchained to the promoter site of dehydration-responsive genes, like *RD29A*and trigger their activation in reaction to environmental challenges, such as drought (Shinozaki*et al*., 2007).In order to boost water uptake, plants can also alter their root systems, for instance by increasing root depth or density. Plants with deeper roots can access water from deeper soil layers, while plants with larger root systems can take up water from a larger soil volume (Lynch, 2013).Plants gather osmolytes, such as proline, carbohydrates, and amino acids, to preserve cellular water potential and avoid dehydration while they are under drought stress. Under conditions of restricted water availability, osmotic adjustment enables plants to maintain cell turgor, which is crucial for growth and development. Study by Tardieu and Simonneau (1998), revealed that maize plants that collect osmolytes are more resistant to water stress. Reactive oxygen species (ROS) build up in plant cells as a result of oxidative stress, which can cause cell death and damage. In order to scavenge ROS and stop oxidative damage, plants have developed antioxidant defense systems (Mittler, 2002).

**Physiology of heat stress tolerance in plants.** The world's patterns of rainfall and drought are impacted by shift in circling temperature that are exacerbated by atmospheric warming, which has a detrimental effect on agricultural production.The physiological effects of heat stress would impact the survival, growth & development, physiological process of the farm plants. The nature and magnitude of effects contingent mainly on temperature, species of plant and followed process (Bita and Gerats, 2013).Heat stress affects membrane composition and stability leading to enhanced permeability and leakage of ions and other important cellular constituents. Impairment of cellular integrity, a decrease in water absorption and transport, and a reduction in photosynthesis can all come from this. Grain growth, especially in cereals, is reduced by heat stress. This effect may be the result of reduced photosynthate translocation to the grain as a consequence of heat (due to a reduced sink size) (Karim *et al.,* 1999). In addition to altering enzyme activity, protein synthesis, and carbon metabolism, heat stress can also have a substantial impact on a plant's metabolism. This can end in reduced growth and development, as well as decreased photosynthetic efficiency and carbon assimilation. *HSP* (Heat Shock Protein) is a group of proteins that usually exist in cells, but their fabrication is accelerated by heat. A heat shock intensifies the magnitude of Hsp by 10 to 50-fold. Hsp seems to be essential at all the temperatures, although at elevated temperatures they are imperative in much plenty amount. Some of the Hsp may condition heat tolerance. Some Hsp, e.g., Hsp 70, function as *chaperonins,* which are a class of proteins that linked with unfolded proteins and thereby, restrict their improper folding or denaturation (Mittler *et al*., 2012). Osmoregulators like proline and glycine-betaine may have a protective role in heat stress. These osmoregulators scavenge several enzymes from heat inactivation *in vitro* (Suprasanna *et al*., 2016). Reactive oxygen species (ROS) can build up in plant cells as a result of heat stress, which can harm cellular elements such proteins, lipids, and DNA. Therefore, using genes involved in antioxidation metabolism may result in transgenic plants with improved thermo tolerance. Transgene *hsf* was transferred into Tobacco which act as heat shock factor (Transcription factor) to protect the plant against heat stress (Grover *et al*., 2013). A study by Wahid *et al.* (2007) examined how heat stress affected the physiological and biochemical processes of protein synthesis, lipid peroxidation, and antioxidant enzyme activity in wheat plants. The effects of heat stress on tomato plants were examined in another study by Hassan *et al.* (2021), who discovered that heat stress decreased photosynthetic efficiency, changed carbon metabolism, and increased oxidative stress. Also, they discovered that the use of exogenous antioxidants could lessen the detrimental effects of heat stress on plant physiology.

**Physiology of cold stress tolerance in plants.** A variety of molecular and cellular reactions to relatively low temperature are involved in the complicated physiological process known as cold stress in plants. To adapt to the challenging surroundings, plants go through alterations in their morphological, physiological, and metabolic activity during this procedure. When temperatures remain above freezing, *i.e.,*>$0^0$C, it is called *chilling.* While *freezing* narrates temperatures below-freezing, *i.e.,*<0°C. Seed germination, growth, fruit development, output, pollen fertility, and fruit quality are all ways to gauge the impact of freezing stress. It causes poor fruit set, pollen sterility, growth retardation, wilting, loss of chlorophyll, necrotic lesion, limited germination, poor seedling establishment, *etc*. (Chinnusamy *et al.,* 2007). The biological membrane's fluidity reduces at chilling temperature, which has an impact on the movement of ions, nutrients, and other molecules all across membrane. Overall, cooling causes the membrane to lose its integrity, which causes solute leakage. Plants change their lipid makeup, such as increasing unsaturated fatty acids and reducing saturated fatty acids, to preserve membrane fluidity (Singh and Usha 2003). By decreasing the effectiveness of the light responses and the Calvin cycle, chilling temperature has an impact on photosynthesis. This is brought on by the reduction in photosystem II (PSII) activity and the inhibition of important Calvin cycle enzymes like *Rubisco*. Moreover, the articulation of genes linked to the photosynthetic apparatus is impacted by chilling stress (Tuteja *et al.,* 2011). The proportion of plant hormones notably abscisic acid (ABA), proline (Pro), gibberellins (GA), and cytokinins (CK) is also impacted by chilling stress. ABA concentration elevated inreaction to chilling temperature stress which encourages stomatal closure and lowers water loss. Reduced GA and CK concentration have an impact on the accelerated plant growth and progress (Thomashow, 1999). The effectiveness of proline as a cryoprotectant has been demonstrated, and this is also one of the key elements influencing freezing tolerance. It is well recognized that the eskimo 1 (*esk1*) gene is crucial for freezing tolerance. The amount of free proline (Pro) was deemed to be 30 times progressed in esk1 mutant plants compared with wild-type plants (Xin and Browse 1998).Chilling sensitive plants may suffer from toxicity injuries as well. Chilling seems to induce, in sensitive plants, production of an inhibitor of catalase. Plant tissues normally contain $H_2O_2$, which is degraded by catalase. The degradation is restricted in chill affected plants due to induction of the catalase inhibitor. Therefore, $H_2O_2$ accumulates and acts as a source of free radical oxidants, which aggravate the chilling injury (Mahajan and Tuteja 2005). Apart from this, freezing stress causes formation of intracellular ice which is one of the major and terminal freezing impacts which is being accepted as lethal for plants. Sometimes it leads to formation of extracellular ice which in turns increases the concentration of extracellular solutes which creates water stress in the frozen tissues/plants. It also causes alternation in semi permeable properties of biological membrane, causes loss of solutes from the cells. Further, cells remain plasmolyzed even after thawing; this is often called frost plasmolysis (Guy, 2003). Transgene *ala3* was transferred into Tobacco which acts as heat antifreeze protein to protect the plant against freeze stress.

**Physiology of heavy metal stress tolerance in plants.** Toxic heavy metal emissions are so widespread that they sometimes prevent plants from adapting to their environment. Heavy metals, which can be harmful to plant growth and development, can be tolerated and detoxified by plants through a variety of methods (Jarup*,* 2003). Sequestration, chelation, subcellular localization, exclusion, and antioxidative defense systems are some of these mechanisms. The transfer of heavy metals from the cytosol into the vacuole *via* multiple transporters, including tonoplast-localized *ATPases* and antiporters, is one of the key pathways for desolation of accumulation of excess metals (Kumar and Trivedi 2016). The phytochelatin synthesis, a group of peptides that chelate heavy metals and facilitate their transport into the vacuoles, controls this process. Chelation is a different method that lessens the toxicity of heavy metals by tying them to particular substances like organic acids and amino acids. Citrate is one such; it binds to iron and aluminum ions and promotes their translocation throughout plants. Another strategy is compartmentalization, in which the metals are confined to particular organelles namely the cell wall, chloroplasts, and mitochondria (Pourrut *et al.,* 2011). Exclusion is a method for limiting heavy metal acquisition by root system.This can be done by controlling the ion channels and transporters in the roots, such as the high-affinity transporters that take up heavy metals more readily than vital nutrients (Conde *et al.,* 2011). Excessive absorption of heavy metals prompted occurrence of oxidative damage which leads to creation of enzymes like superoxide dismutase, catalase, and peroxidase that scavenge reactive oxygen species and counteract oxidative damage to the plant cells (Noctor and Foyer, 1998; Gilland Tuteja 2010).

**Breeding approaches for abiotic stress tolerance/resistance.** Crop performance is the end result of the action of thousands of genes and their interactions with external factors & cultural practices. There are several factors that limit farm output worldwide *i.e.,* no availability of inputs, soil related problems, outbreak of pests and losses due to abiotic stress. Major abiotic stress *viz.,* drought, soil salinity, chilling/freezing temperature, extreme temperature at flowering and maturity stage, chemical toxicity and oxidative stress are serious threats to agriculture and environment. By 2025, the world farmers would have to produce three billion tonnes of cereals to feed the earth population of nearly eight billion people. This means that worldwide, on average cereal mainly wheat and rice yield of four tonnes per hectare needs to be achieved and sustained (Lobell *et al.,* 2011). Modern HYV$_S$ particularly in rice and wheat has greater potential as compared to grain legumes and pulses under well managed condition. Breeding crops for specific abiotic stress tolerance is one of the core activities of varietal improvement programme.

***Selection and introduction.*** The first step in breeding for any characteristic is to assess genetic variation by gathering and analyzing the available germplasm. The introduction of the foreign germplasm might be used if a locality or species lacks the desired variability. The salt resistant varieties of rice *viz.,* Arya33, BR4-10, KR 1-24, Mo 1, Mo 2, Mo 3 were selections from locally adapted rice varieties Arya, Bhura Ratta, Kala Ratta, Chattivirippu, Kalladachampavu, Kunjathikkara in region of Maharashtra & Kerala respectively. Salt resistant variety SR26B were selected from local variety Kalambank having wider adaptability (Reddy *et al.,* 2013). When an exotic variety with abiotic stress tolerance is available, the variety can be introduced and, if tested and proven to be acceptable, released in the new area.

***Pedigree method .*** This is the process of evaluating and exploring the segregating generations which aims to isolate completely homozygous individuals surpassing parental lines of the cross. Here selection starts from F$_2$ generation from space planted plants and harvested separately. This process of selection continues up to F$_6$ until you get the reasonably homozygous lines with enough seeds. The pedigree method owes its name to the pedigree record of selected plants maintained to trace ancestral relationship among such plants (Hurd,1976). Records were maintained to ascertain the ancestral identity and genetic status of new population which still remain the most outstanding merit of pedigree method. Here, skill of breeder is one of the most important aspects of pedigree method. The pedigree selection strategy produced new lines that were highly effective at withstanding drought stress (Tammam *et al*., 2004).This method has been used to advance wheat variety KRL1-4 which was evolved from the cross Kharchia 65 x WL711 at CSSRI, Karnal. It combines high yield attribute of WL711 with salt tolerance of Kharchia 65. An advanced line, called Nesser, has been derived from the cross Jupateco 73(high yielding CIMMYT wheat) x W3918A (drought tolerant Australian variety). Nesser is considered by ICARDA to be a uniquely drought tolerant genotype, but it was bred at CIMMYT under favorable environment and identified at ICARDA as drought tolerant.

***Delayed Pedigree Approach.*** The basic tenet of this approach is to carry forward F$_2$plants as random bulk of each family till later generations where standard pedigree method can be started. The mass pedigree method of Harrington (1937) is one of the earliest modifications where single plant selection is delayed till a favorable environment for the expression of character is encountered. It has better appeal in selection for resistance against abiotic stress or threshold characters but may not be effective for improvement of farm output. The "*Veery"* wheat variety and its descendants, including the Kauz, Attila, Pastor, and Baviacora sets of lines have showna higher magnitude of abiotic tolerance to anarray of external factors (Meena *et al.,* 2016).

***Shuttle breeding.*** The notion of this method was proposed by the International Maize and Wheat Improvement Centre (CIMMYT) and popularized by father of green revolution *i.e.,* Dr.N. E. Borlaug. The wheat shuttle breeding programme at CIMMYT used two different ecological environments or locations in which varieties were raised in Obregon and Toluca during winter and summer period, respectively. In this system, an extra generation has advanced each year by using different location resulted in shorten breeding cycle. Using a shuttle breeding strategy, the salinity tolerant cultivated spp. Of Rice (Oryza spp.)Varieties CSR-27 and CSR-23 were created as part of an ICAR-IRRI collaboration project (Mishra, 1994). The flood tolerant cultivar FR13A was found to have weak combining ability but strong submergence tolerance (Reddy *et al*., 2013).

***Back cross method.*** The crossing of $F_1$ of two parents to either of the parents is referred to as backcross. It is used as a special method of breeding where an otherwise high yielding and popular cultivar is needed to be refined only for a distinctive attribute. In general, it is frequently used for stabilizing breeding where the real potential of $HYV_S$ remain unrealized due to the presence of some defect especially related to susceptibility to some external environment constrain. The incorporation of a few characters to otherwise superior genotypes through normal procedure of hybridization followed by selection leads to a major uncontrolled shake up in the genetic constitution of the parental varieties. This method is ideal for such situations as it helps to add a few genes to a variety without disturbing its basic genotype. The gene(s) to be transferred should be free from the effect of background genotype which may influence the intensity of the character when conveyed to the recipient parent. This task is replicated up to 6-8 generation of backcrosses in order to create a line with gene of interest in background of recurrent parent (Kumar *et al.,*2018).

***Mutation breeding.*** This approach is employed when there is a limited gene pool. Mutations may occur by chance or can be developed artificially. Use of induced mutants in the breeding programmes for developing superior varieties is known as mutation breeding which is not restricted to the direct release of new mutants as varieties but includes all direct & indirect use of mutations for crop improvement. It is applicable to all the crop plants but it has special application in vegetatively propagated plants. The ideal situation for mutation breeding is when the gene(s) for desired change is either not available in the germplasm or is tightly linked with undesirable genes so that recombination through hybridization is expected to be rare or impossible (Chahal and Gosal 2002). This process was used to create rice varieties *viz.,* Nucleoryza (released in Hungary), Kashmir Basmati (released in Pakistan), R.D. 15 (released in Thailand) tolerant to low temperature and drought, respectively. A mutant (Gamma ray induced) of Bermuda grass variety 'Coastcross1' (non-winter hardy) was winter hardy (Singh, 2016).

## Advance breeding approaches for abiotic stress tolerance

***Marker Assisted Selection.*** Most of the conventional breeding selections are based on resultened phenotype which relies on genetic makeup of plant along with prevailing environment. Therefore, phenotype is not always a good indicator of genotype. The phenotypic evaluation of many traits may be either cumbersome, tedious, time-consuming, destructive (e.g., for root traits, biomass), or dependent on specific threshold requirements or may require homozygous genotypes. Further, phenotypic selection for traits like yield will not be feasible in off-season nurseries/greenhouses, which are employed for rapid generation advance. This precludes the use of selectedplants for making appropriate crosses in the same generation as selection. Therefore, indirect selection for traits of interest has been a long sought after objective of plant breeders. With the discovery of molecular markers, plant breeders have long sought to achieve indirect selection of traits. In this strategy, selection is based on molecular markers linked to the desirable allele of gene or QTLs (Henkrarand Udupa 2020).Traditionally; it takes a lot of effort and money to transfer genes from a donor to well adapted popular better lines with insignificant issues in order to create isogenic lines. In contrast, marker-aided crop improvement, which takes the form of MABC, MARS, and $F_2$ enrichment, can simplify the selection processes and significantly reduce the amount of time and resources needed. Rice variety Improved Basmati-1 (Babu *et al.,* 2017), Swarna Sub-1(Neeraja *et al.,* 2007) has been developed to tolerate salinity and submergence, respectively. It has been developed by transferring genes *viz.,* Saltol, SUB-1into variety Pusa Basmati-1, Swarna.

***Genomic Selection.*** Genomic selection is one of extensively studied technique of selection or approach of breeding for abiotic stress adaptability (GS). Genomic estimated breeding values (GEBVs) are computed using data from genome-wide markers and are used to select individuals (Meuwissen *et al.*, 2001). GS captures both significant and minute effects of genes because it needs full genome markers. Therefore, GS is more advantageous than MAS sinceit sidestep the necessity to find QTL associated with the desired attributes (Nakaya and Isobe 2012). By enhancing breeding cycle intensity and selection accuracy, it has the potential to expedite genetic gain.

**Genomics based Integrated Approach.** This approach uses information and tools from several disciplines, and can be used to improve abiotic stress resistance related traits. In this, a wide range of germplasm is evaluated at several locations under different environments where the crop is cultivated. Data are collected on various traits that may contribute to the feature of interest, and on that basis of this knowledge such traits are identified. After that germplasm lines having contrasting phenotypes of the identified traits are selected. Functional genomics tools like microarrays are employed to analyze expression patterns of genes.

This will lead to identification of a group of candidate genes that are likely to be involved in the development of the desired phenotypes. In order to confirm that the identified candidate genes do actually contribute to the phenotype, either these putative genes are 'knocked out' individually or they are isolated and transformed into separate lines to study the effects of 'knock-out'/over expression of specific genes on the desired phenotype. Once the genes contributing to the desired phenotype become identified, molecular markers tightly linked to them are developed, which are then used for marker-aided selection (Singh, 2016).

***CRISPR-Cas9 gene editing technique*** With the aid of the cutting-edge gene-editing technique CRISPR-Cas9, researchers may precisely alter the DNA of living things. The technology is based on a natural defense method employed by bacteria to fend off invading viruses. A DNA sequence called CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) is present in a large number of bacteria and archaea. The Cas9 enzyme is directed by these sequences to certain regions of the genome where it can cut DNA. An RNA-guided endonuclease known as Cas9 can cleave DNA at a precise site indicated by the RNA sequence. Scientists can command the Cas9 enzyme to cut the DNA at a specific spot by adding a guide RNA that is complementary to a certain gene sequence, allowing them to edit the genome. It is a flexible and potent technique that has the ability to alter the functional properties of a gene through targeted point mutations or indels and even to reinstate a defective gene accompanying repaired exonssequence (Biswal *et al*., 2019).

## CONCLUSIONS

Ensuring food security for huge population in era of climate change is one of the biggest obstacles for agriculture scientist. Crops are being exposed to extreme weather conditions like moisture stress, excessive temperature, and salinity more frequently as the climate becomes more unpredictably unstable. These environmental stressors can drastically lower crop yields, endangering the availability of food. Plant breeders are using a variety of approaches to develop crops that can withstand these stressors. Current and rapidly developing technologies such as MAS, GWAS, GS, Genome editing will greatly accelerate the evolution of improved designer abiotic stresses-tolerant crops. The integration of these advanced approaches with each other or with conventional techniques is one way to improve the productivity in stressed condition. However, breeding for abiotic stress tolerance in plants is a complex and challenging process. The genetic basis of stress tolerance is often complex, and it can be difficult to identify the specific genes or gene networks involved. In addition, breeding for stress tolerance can sometimes result in unintended consequences, such as reduced yield or changes in plant morphology. Considering these challenges, a deeper comprehension of plant stress response and tolerance mechanisms is critically required. This comprehension study will help us to design climate-resilient crops for the future.

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