

SPECIAL ISSUE ARTICLE

Drought: Sensing, signalling, effects and tolerance in higher plants

Mohammad Mukarram^{1,2}  | Sadaf Choudhary¹ | Daniel Kurjak² | Anja Petek² | M. Masroor A. Khan¹

¹Advance Plant Physiology Section, Department of Botany, Aligarh Muslim University, Aligarh, India

²Department of Integrated Forest and Landscape Protection, Faculty of Forestry, Technical University in Zvolen, Zvolen, Slovakia

Correspondence

Mohammad Mukarram, Advance Plant Physiology Section, Department of Botany, Aligarh Muslim University, Aligarh 202002, India.

Email: mdmukarram007@gmail.com

Edited by: M. Ahanger

Abstract

Drought can be considered as a cocktail of multiple stressful conditions that contribute to osmotic and ionic imbalance in plants. Considering that water is vital for plant life, the very survival of the plant becomes questionable during drought conditions. Water deficit affects a wide spectrum of morpho-physiological phenomena restricting overall plant growth, development and productivity. To evade such complications and ameliorate drought-induced effects, plants have a battery of various defence mechanisms. These mechanisms can vary from stomatal adjustments to osmotic adjustments and antioxidant metabolism to ion regulations. In this review, we critically evaluate how drought is perceived and signalled through the whole plant via abscisic acid mediated pathways. Additionally, the impact of drought on photosynthesis, gas exchange variables and reactive oxygen species pathway was also reviewed, along with the reversal of these induced effects through associated morpho-physiological counter mechanisms.

1 | INTRODUCTION

Although drought threatens plant and animal kingdom alike, there is no unanimous scale that classify drought (Ault, 2020; Gupta et al., 2020). It is a shared understanding that organisms strive hard to meet their water demand during drought. This struggle could be fatal depending on the duration and severity of the drought. Considering plants cannot migrate from such environments, like their animal counterparts, they tend to take a heavier blow from the drought. Depending on the stage of the plant life cycle, drought could cause substantial damage to plant growth and productivity through various processes.

Plants close their stomatal aperture as the first response at the onset of drought to maintain the leaf water potential (Laxa et al., 2019). Drought creates an ionic imbalance among K^+ , Cl^- and H^+ in the stomatal guard cells. This ionic imbalance dominates the turgor pressure in the guard cells, which subsequently controls the closing of the stomata. The strict regulation of stomatal conductance under water deficiency enables plants to minimise water loss through evaporation or transpiration. However, this survival trick cost plants their productivity as CO_2 uptake is downregulated with stomatal

closure, and thus, in turn, the rate of photosynthesis also declines. During normal circumstances, plants synthesise reactive oxygen species (ROS) as the by-product of different vital physiological phenomena such as photosynthesis and photorespiration (Corpas et al., 2020). These reactive species could act as signalling molecules for different metabolic pathways, including seed germination, senescence, plant growth and development (González-Gordo et al., 2020). However, with reduced stomatal conductance and CO_2 uptake, drought overproduces such potential toxic reactive species due to over-reduction of the electron transport system and the carbon starvation of the cellular organelles (Laxa et al., 2019). This, subsequently, provokes oxidative damage of various vital macromolecules limiting plant growth and development (Corpas et al., 2020).

Plants have multiple physiological and biochemical counter measures to minimise drought-induced oxidative damage. These include antioxidant metabolism, osmotic adjustment (OA) and ion homeostasis. Almost all subcellular organelles have an antioxidant battery to counteract and scavenge ROS to palliate oxidative stress. OA is covered by a range of osmotically active compounds such as amino acids, soluble sugars, polyamines and ammonium compounds. These osmolytes work

towards creating a more hydrating internal environment for the plant through cell hydration and osmoregulation. Whereas ionic homeostasis is dominated by several antiporters and channels of Ca^{2+} , Na^+ , H^+ and Cl^- and abscisic acid (ABA) mediated signalling to ensure proper ion uptake and transfer to adjacent cells and organelles (Chen & Jiang, 2010).

In this regard, the present study renders a timely account of how drought is sensed and signalled via ABA-mediated pathways, its interaction with plant physiology, and the counter mechanisms higher plants adopt for drought tolerance.

2 | SENSING AND SIGNALLING DROUGHT

Plants have developed several signalling mechanisms to recognise the approaching stress and regulate their physiological responses and metabolism in order to cope (Zhang et al., 2006). Such signalling pathways are interconnected at several checkpoints and form a complex defence network. This provides 'cross-resistance' against multiple biotic and abiotic stresses, or several pathways can work together against one particular stress (Chaves et al., 2003).

Plants, primarily roots, perceives drought through chemical and hydraulic signalling. Any significant change in soil conditions such as water and oxygen contents, bulk density or nutrients composition triggers chemical signalling from the root towards the shoot to initiate necessary molecular responses. These responses induce morpho-physiological changes, including stomatal behaviour and prepare the plant for the upcoming drought (Christmann et al., 2007; Janiak et al., 2016). Through stomatal conductance, leaf growth and leaf transpiration are also well regulated by this signalling. Chemical signals from the roots travel through the xylem and probably dominate the early stress stages before hydraulic signals are produced. However, the identity of these signals in the xylem sap is controversial (Schachtman & Goodger, 2008).

Plants have a complex signalling system to regulate stomatal opening mediated by the uptake and intracellular synthesis of solutes that reduces the water potential in guard cell and creates a driving force for water uptake (Zhao et al., 2018). In general, guard cells close the stomatal aperture in the absence of photosynthesis to prevent water loss, however, stomatal closure could be triggered during daytime if the plant encounters water deficit owing to low soil moisture, low air humidity or some other conditions (Luan, 2002). Dynamic regulation of stomatal aperture is crucial to prevent excessive transpirational water loss and minimise the drought impact. Stomatal behaviour plays a critical role during drought sensing and signalling, yet its modus operandi and crosstalk with other plant phenomena are complex and debatable (Gollack et al., 2014; Lawson & Matthews, 2020).

3 | ABSCISIC ACID MEDIATED SIGNAL TRANSDUCTION

Specific receptors can intercept the stress signals and initiate (or suppress) a signalling cascade as a molecular response and further

convey the stress message through a complex signal transducing pathway (Chaves et al., 2003). ABA is said to have a pivotal role during drought owing to its potential to act as a primary chemical messenger. The ABA-signal transduction system consists of PYR/PYL/RCAR type ABA receptors, group A 2C-type protein phosphatase (PP2C) and SNF-1 (sucrose non-fermenting 1) related protein kinase 2 (SnRK2) family of proteins, which suppress ABA signalling. Several cellular parts, including the cytosol, nucleus, plasma membrane and chloroplast envelope, reportedly have ABA receptor elements that are involved in ABA signal transduction (Mahmood et al., 2020). There are at least four independent regulatory systems that have been reported for gene expression in response to water stress, out of which two are ABA-independent, while the others are ABA-dependent. In Arabidopsis, the cis- and trans-acting elements that function in ABA-independent and ABA-responsive gene expression have been studied to understand the molecular mechanism regulating gene expression in response to water deficit (Huang et al., 2012).

The ABA-dependent pathway regulates the expression of genes in two ways based on whether new protein synthesis is required or not. An ABA-responsive element ABRE (a major cis acting element PyACGTGGC, where Py indicates pyrimidine base, C or T) is present in the promoter domain of all ABA-responsive genes, where protein synthesis is not required. The ABRE can lead to ABA-induced gene expression, once bound with its corresponding bZip family of transcription factors such as EmBP-1 (Zhang et al., 2006). ABA accumulates and binds to PYR/RCARs in drought conditions, which in turn inactivates PP2C. Since PP2C is a suppressor for SnRK2 activity, its inactivation restores SnRK2 activity. The activated SnRK2 induces stomatal closure and OA through downstream regulation of ion channels and transcription factors (AREB). (Yu et al., 2019). The activation of SnRK2 by osmotic stress and ABA is mediated by a distinct mechanism, and it has been confirmed by the elucidation of variance in their C-terminal sequences. However, they all required an 'SnRK2' box for activation under osmotic stress and an 'ABA' box for the ABA response (Wu et al., 2019).

Whereas, de novo synthesis of proteins is the prerequisite when new proteins are required for ABA-induced gene expression. The ABA-responsive element of such genes combines with the MYC family transcription factor (master regulatory of cell cycle entry and proliferative metabolism) and contains no ABREs like elements. The synthesis and the activation of the genes of the MYC family transcription factors should lead up to ABA-initiated genes activation (Zhang et al., 2006).

Stomatal aperture regulation is mediated by the SnRK2 protein kinase OST1 (SnRK2 OPEN STOMATA 1), which connects the guard cell movement to the ABA transduction pathway. It regulates inward K^+ channels, Ca^{2+} permeable channels, NADPH oxidase, and the slow anion channel SLAS1 in guard cells. Moreover, the SnRK2 protein kinase OST1 also targets the R-QUAC-type voltage dependent quickly activating anion channels in guard cells. These findings suggest systemised control of QUAC1-mediated transport of malate and SLAS1-mediated transport of chloride and nitrate in the same ABA signalling pathway (Gollack et al., 2014).

The expression of ABA-independent stress-responsive genes is regulated by DREB, that is, dehydration responsive element binding (plant-specific transcription factor) proteins that bind to DRE (dehydration responsive element) cis-elements. The DREBs induce several abiotic stress related genes and transmit immunity to plant systems during different stress, including drought. They consist of two subclasses, DREB1/CBF and DREB2, and they have different binding specificities, hence, upregulate a distinct set of abiotic stress related genes. However, the activation mechanism of DREB2-type genes is not well understood, and it was presumed that some post translation modification, that is, phosphorylation and/or dephosphorylation phenomenon, might play a role in the activation of stress-responsive genes (Agarwal & Jha, 2010).

4 | DROUGHT-INDUCED EFFECTS IN HIGHER PLANTS

Scarcity of water generates a wide range of plant responses constraining plant growth, development and productivity. Such morphological and physiological responses are visible at various phenological stages. Drought retards the expansion of roots, shoots and leaves, division, expansion and differentiation of cells, germination and seedling growth, resulting in reduced plant growth and biomass (Farooq et al., 2012; Iqbal et al., 2020). These vegetative complications are expected to confer productivity setbacks in the stressed plant. Along with yield reduction, drought can induce plant mortality depending on its severity, duration and developmental stage of the plant (Akram, 2011; Farooq, Wahid, et al., 2009b). Furthermore, higher plants, including trees, are more susceptible to drought stress as potentially greater rootstock is unable to compensate for the greater water demand (McGregor et al., 2020). There are two main mechanisms discussed as a possible reason for drought-related mortality: carbon starvation and hydraulic failure (McDowell et al., 2008). Even though the first hypothesis has just a few direct pieces of evidence (Rowland et al., 2015), hindered photosynthesis influence basically all physiological processes. The main role of stomatal closure is to inhibit water loss, thereby stimulate drought tolerance under conditions of water deficit stress. Stomatal opening and closure affect different kinds of physiological processes and properties, such as photosynthesis and water status (Koyro et al., 2012). Under normal conditions, the opening and closing of the stomata (in most plants) are connected to the light–dark cycle in order to maximise the efficiency of light utilisation for photosynthesis. Plants react to light by opening the stomata for CO₂ uptake. During the dark period, stomata gradually close to prevent water loss in the absence of light (Luan, 2002). Stomatal regulation is a key process involved in maintaining the photosynthetic capacity in plants under stress conditions (Koyro et al., 2012). If the plants undergo water deficit stress, stomatal closure is triggered (Schroeder et al., 2001), and plants prioritise survival over productivity (Luan, 2002). This quick response, defined as the active closure mechanism, is controlled by a diverse network of signalling pathways, in which the major and the key player ABA, performs together with

jasmonates, ethylene, auxins and cytokinins (Nemhauser et al., 2006). The complexity of the reaction is mainly dependent on the initial threshold of stress and the individual plant's stress history.

During a period of drought, with increased leaf evaporation and lower soil water potential, xylem tension increases, leading to increasing cavitation events, drastically reducing hydraulic conductance (Cochard, 2006). At short time-scales, plants control water-loss to avoid a water potential causing significant damage to their water-transport system due to embolism (Brodrribb et al., 2003). Cavitation occurs within the xylem of vascular plants when water tension becomes so high that dissolved air within water expands to fill either vessel or tracheids. Under water stress, when cavitation appears, the small initial gas-bubble spreads to fill the entire cell, which is therefore no longer functional, thus reducing the plant's overall capacity to feed water to its aerial organs (Cochard, 2006).

Moreover, most of the environmental stresses lead to increased ROS production. Drought-induced ROS accumulation has been reported in various plant species growing in different environments. Under normal or non-stressed conditions, most of the plants maintain a steady homeostatic condition. Upon the occurrence of stress, increased ROS level is significant. The ROS production beyond the plant's quenching capacity is usually defined as a disruption of redox signalling and redox control, which can cause oxidative stress by damaging membrane lipids, proteins, photosynthetic pigments and nucleic acids (Corpas et al., 2020; Mukarram, Khan & Corpas, 2021a). Besides its main effect on photosynthesis, the closure of the stomata generates most of the oxidative stress during drought. The decrease in CO₂ availability for photosynthesis leads to a misdirection of electrons in the photosystems, resulting in the overproduction of ROS. Superoxide radicals (O₂⁻), single oxygen (O₂), hydrogen peroxide (H₂O₂), alkoxy radicals (RO) and hydroxyl radicals (OH⁻) are among the major ROS generated in plants subjected to drought stress (Aroca, 2012). ROS are highly reactive and impair normal plant metabolism through lipid peroxidation or protein denaturation (Mukarram, Khan, Uddin, et al., 2021b). In chloroplasts, PSI and PSII and in mitochondria, complex I, ubiquinone and complex III of electron transport chain are the primary production locations of ROS (Gill & Tuteja, 2010). Hence, reduced function of essential enzymes such as ribulose-1,5-bisphosphate carboxylase/oxygenase (Rubisco), phosphoenolpyruvate carboxylase, and ATP synthesis due to oxidative damage may restrict photosynthetic and respiratory activities. Downregulation and inhibition of metabolic processes result in reduced ribulose-1,5-bisphosphate (RuBP), which then dominates limitation under severe drought, and therefore inhibits photosynthetic CO₂ assimilation (Aroca, 2012). The transformations of RuBP to 3-phosphoglyceric acid decreases with decreasing leaf relative water content suggesting a negative impact of drought on the regeneration of RuBP. Therefore, declining RuBP contents and its regeneration are responsible for the additional rapid decline in photosynthesis under drought (Reddy et al., 2004). Figure 1 depicts the stomatal behaviour under drought environment as well as traces the drought-induced effects on the major cellular organelles, including chloroplast, mitochondria and peroxisomes.

5 | DROUGHT TOLERANCE

5.1 | Through antioxidant metabolism

Plants possess a battery of enzymatic and non-enzymatic antioxidants to offer a primary defence line against drought-induced oxidative stress. These antioxidants are present in almost all the major ROS producing sites, including chloroplast, mitochondria, peroxisomes, nucleus, endoplasmic reticulum, along with the cytosol. Categorically, they can be one of two types: enzymatic and non-enzymatic. Catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GPX), glutathione sulfo-transferase (GST), peroxidase (POX), ascorbate peroxidase (APX), glutathione reductase (GR), monodehydroascorbate reductase (MDAR) and dehydroascorbate reductase (DHAR) are some of the principle enzymatic antioxidants (Lukatkin & Anjum, 2014). While non-enzymatic antioxidants include glutathione (GSH), glutathione disulphide (GSSG), ascorbic acid (AsA), dehydroascorbate (DHA), carotenoids, tocopherols and phenolics (Lukatkin & Anjum, 2014). As drought poses an imminent oxidative threat, plants upregulate their antioxidants synthesis to maintain ionic and osmotic homeostasis.

This upregulation can be transcriptional or post-transcriptional however, its degree might depend on multiple factors, including the degree of sensing and signalling of drought as well as the metabolic and developmental state of the plant (Laxa et al., 2019).

Overproduction of O_2^- during drought can be considered as the first significant step in plant oxidative damage. The increased activity of SOD isoforms, that is, Mn-SOD, Fe-SOD, Cu, Zn-SOD during drought, depicts SOD as an efficient tool against O_2^- overproduction in plants (Zandalinas et al., 2017). SOD (EC. 1.15.1.1) provides protection against oxidative damage through the dismutation of O_2^- to the less damaging H_2O_2 . However, H_2O_2 itself is fairly toxic, and its accumulation can still cause substantial membrane and organelle damage through lipid peroxidation or by interfering with other metabolic pathways. This necessitates the removal of H_2O_2 . Regarding this, a large machinery of antioxidants dedicated for H_2O_2 detoxification is found in the plants.

CAT (EC. 1.11.1.6) is the primary enzymatic antioxidant for H_2O_2 removal in plants (Laxa et al., 2019). It reduces H_2O_2 produced through mitochondrial electron transport, photorespiratory oxidation and β -oxidation of fatty acids to water molecules (Quan et al., 2008).

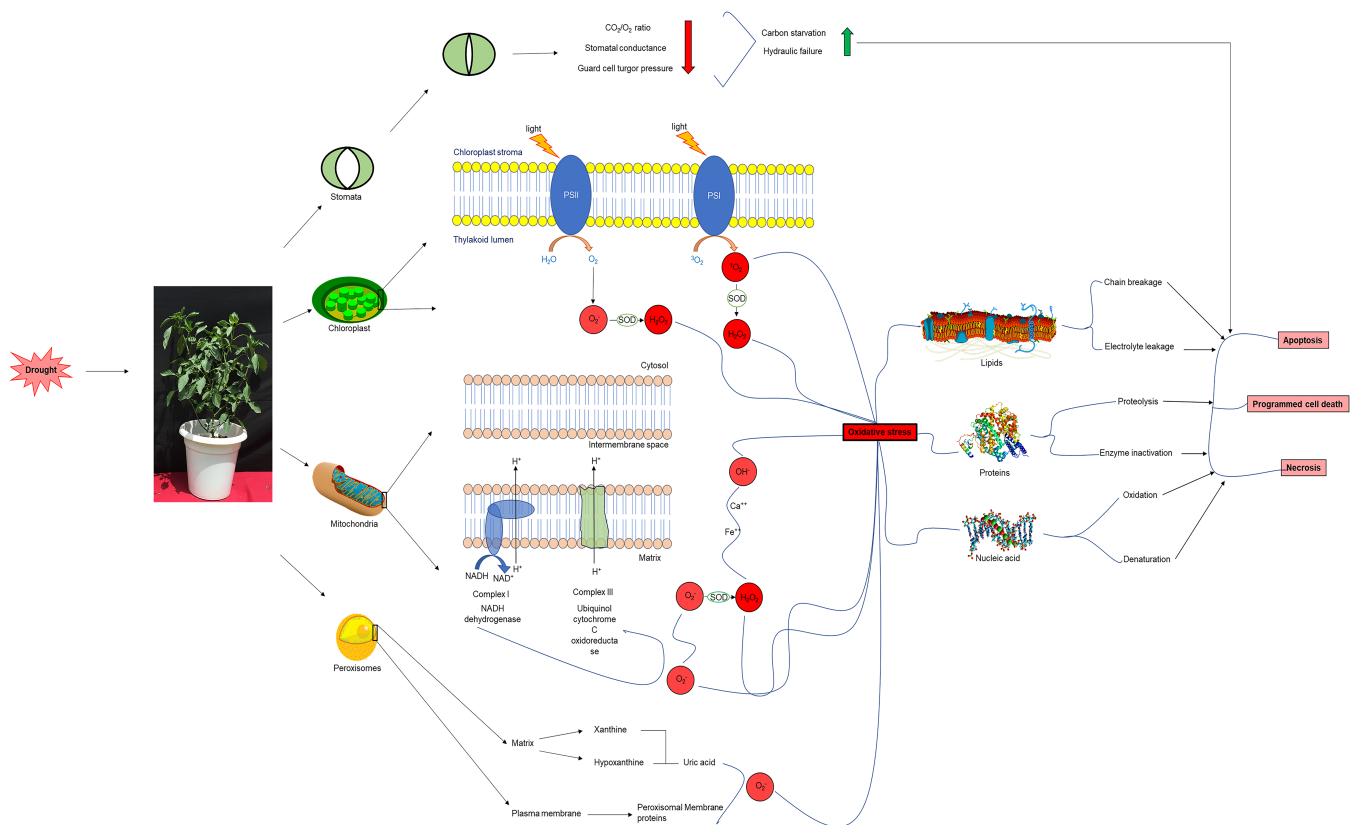


FIGURE 1 Interaction of drought with various cellular organelles and their subsequent effect on overall plant growth and development. At the physiological level, drought can significantly ($p \leq 0.05$) reduce guard cell turgor pressure and regulate stomatal behaviour. Subsequent hydraulic failure and carbon starvation might accompany. Additionally, major cellular organelles experience overproduction in reactive oxygen species causing an oxidative stress. Depending on the intensity of oxidative stress, substantial damage can be observed in the plant including complete termination in severe cases. 1O_2 , singlet oxygen; 3O_2 , triplet oxygen; Ca^{++} , calcium ion; CO_2 carbon dioxide; Fe^{++} , iron ion; H^+ , hydrogen ion; H_2O_2 , hydrogen peroxide; NAD^+ , nicotinamide adenine dinucleotide ion; $NADH$, nicotinamide adenine dinucleotide hydride; O_2 , Oxygen molecule; O_2^- , superoxide radical; OH^- , hydroxyl ion; PS, photosystem; SOD, superoxide dismutase

Interestingly despite sufficient CAT in the peroxisomes, it is not the primary scavenger for low concentrated peroxisomal H₂O₂. This suggests that CAT works better against higher H₂O₂ concentrations (Quan et al., 2008). Alternatively, H₂O₂ can also be converted into water by the APX (EC. 1.11.1.11). However, in addition to H₂O₂ scavenging, APX also plays a crucial role in the ascorbate-glutathione (AsA-GSH) cycle. APX catalyses the conversion of AsA to monodehydroascorbate (MDHA) during AsA-GSH cycle.

In addition to APX, the AsA-GSH cycle is comprised of the MDHR, GR, DHAR enzymes for efficient H₂O₂ detoxification and operates in the chloroplast, mitochondria, peroxisomes and cytosol. Various studies have reported an upsurge in the expression level of these enzymes during drought, suggesting their crucial role in drought tolerance (Laxa et al., 2019). Besides carrying out the conversion between certain non-enzymatic antioxidants (MDHA to DHA), MDAR (E.C. 1.6.5.4) can also reduce MDHA directly to AsA using electrons derived from the electron transport chains. GR (EC.1.6.4.2) is another important antioxidative enzyme, which provides AsA-GSH cycle mediated drought tolerance given its upregulated activity under water stress (Laxa et al., 2019). It catalyses reactions that convert GSSG to GSH in an NADPH-dependent reaction and ensures high amounts of GSH and AsA for an efficient AsA-GSH cycle (Koca et al., 2007).

Contradictory to this, GPX and DHAR are two enzymes that oxidise glutathione from GSH to GSSG. The cytosol is the prime action site for GPX and DHAR mediated scavenging of H₂O₂. Given that H₂O₂ can be transported through the membranes to other organelles (chloroplast, peroxisomes or mitochondria) and to the cytosol itself, a complex antioxidant system is present in the cytosol as well. In addition to scavenging H₂O₂, GPX (EC. 1.11.1.12) also converts reduced glutathione (GSH) to oxidised glutathione (GSSG). Besides GSH and GSSG, AsA is another key non-enzymatic antioxidant that participates in the AsA-GSH cycle for H₂O₂ removal from the chloroplast, mitochondria, peroxisomes and from the cytosol itself.

Apart from the enzymes of the AsA-GSH cycle, POX (E.C. 1.11.1.7) can minimise drought-induced cellular damages due to its ability to lignify and cross-link structural proteins in the cell wall (Quan et al., 2008). This complex antioxidant metabolism comprises a biochemical defence line and ensures better drought tolerance in plants (Laxa et al., 2019; Quan et al., 2008; Zandalinas et al., 2017).

5.2 | Through osmotic adjustment

The regulation of favourable water status is crucial for plant survival under adverse environmental condition, such as drought. The accumulation of cellular compatible solutes, also known as OA, is generally accepted as a most common reaction to overcome the negative effects of water deficit (Blum, 2017; Serraj & Sinclair, 2002). The OA leads to a net increase of the osmotically active solutes in the cell while decreasing the osmotic potential, which consequently improves the level of cell hydration and maintains turgor in leaf and other metabolically active cells. Therefore, OA is recognised as a mechanism for drought tolerance rather than just a drought response (Sanders & Arndt, 2012).

Osmotically active solutes involved in OA include amino acids (e.g., proline), ammonium compounds (e.g., glycine betaine), soluble sugars (e.g., fructans, sucrose) and polyamines (e.g., spermidine, spermine) (Farooq et al. 2008; Farooq, Aziz, et al., 2009a; Farooq, Wahid, et al. 2009b). All these solutes have numerous hydroxyl (—OH) groups that assist in facilitating hydrogen bonds with the water molecules in the cytoplasm. Thus, the solutes not only contribute to osmoregulation but as well, protection of the enzymes and macromolecules in cells from the damaging effects of ROS (Farooq, Aziz, et al., 2009a), hence operate as antioxidants (Ashraf & Foolad, 2007).

Proline is a significant compatible solute that accumulates in the cytoplasm of various plants in response to water deficit. Elevated levels of free proline enable plants to sustain low water potentials (Manivannan et al., 2007) and obtain water from the surrounding environment. In higher plants, two pathways of proline biosynthesis are known, the glutamate and the ornithine pathway. The role of the glutamate pathway in proline synthesis is well documented and has been recognised as the primary pathway in response to osmotic stress (Kishor et al., 2005). While in the ornithine pathway, the ornithine- δ -aminotransferase has been found to be involved in proline synthesis. Moreover, proline can stabilise the structure of proteins and membranes, scavenge free radicals and maintain cytoplasmic pH under stress (Hayat et al., 2012).

Another well-known osmoprotectant against various stresses, including drought, is glycine betaine (GB) (Ashraf & Foolad, 2007). GB, a quaternary ammonium compound, synthesised in the chloroplast, which consists of a two-step oxidation of choline to betaine through betaine aldehyde (Ahanger et al., 2014). These two biosynthetic steps are catalysed by choline monooxygenase and betaine aldehyde dehydrogenase. GB has been shown to protect functional proteins, enzymes (e.g., Rubisco) and lipids of the photosynthetic apparatus, and to maintain electron flow through the thylakoid membranes. Furthermore, intracellular accumulation of GB allows for water retention in the cell and prevents its dehydration, hence it has a positive correlation with stress tolerance (Ahanger et al., 2014). GB is an effective metabolite for alleviating stress effects due to its small size, solubility in water and non-interference with other metabolites within the cell.

Plants also react to drought stress by the accumulation of soluble sugars. Sugars are easily available organic osmolytes in the cell. Under water deficit, soluble sugars operate in two ways which are inseparable: as osmotic agents and as osmoprotectants (Ingram & Bartels, 1996). As an osmotic agent, the main function of sugars is connected with turgor maintenance and OA. As osmoprotectants, they protect the structure of proteins and membranes, potentially by substituting the water in the formation of hydrogen bonds with polypeptide polar residue and phospholipid phosphate groups (Strauss & Hauser, 1986). When subjected to osmotic stress, soluble sugars are generated by hydrolysis of common carbohydrates (Levitt, 1980). The most frequent soluble sugars in the cell are sucrose, glucose, and fructose (Taiz & Zeiger, 2010). During periods of drought, besides these simple sugars, various plants accumulate trehalose. Drought tolerant plants can accumulate trehalose up to 1% of their dry weight under non-stress conditions. Even though trehalose is the most effective

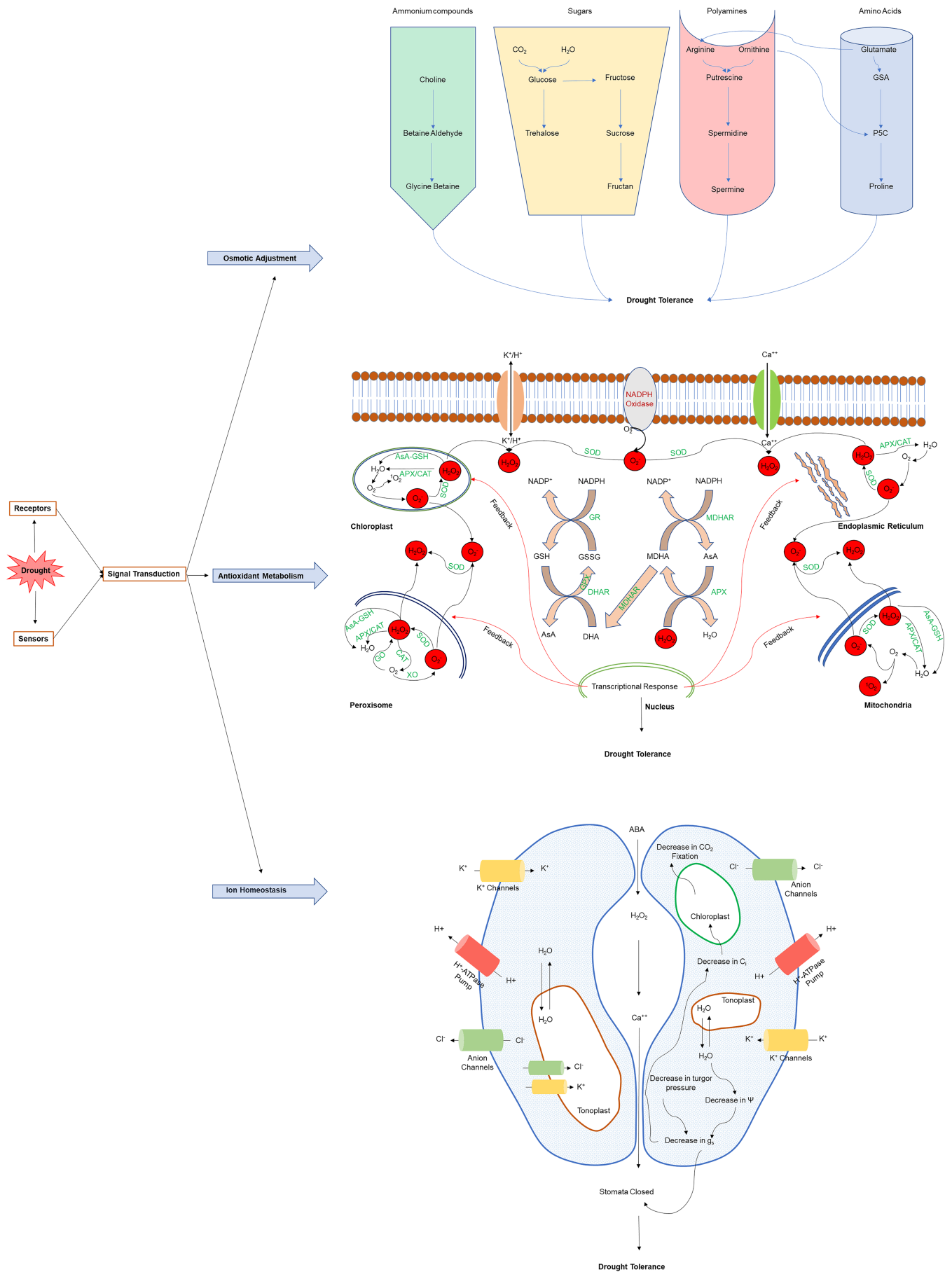


FIGURE 2 Legend on next page.

osmoprotective sugar, its concentration in most plants is often very low (Ingram & Bartels, 1996). If trehalose accumulates abundantly in a plant, it can act as an osmolyte and a vital osmoprotectant (Djilianov et al., 2005). Additionally, trehalose is a significant source of carbon and energy, and it can stabilise membranes and proteins in cells exposed to osmotic stress (Djilianov et al., 2005).

The diamine putrescine, the triamine spermidine and the tetra-amine spermine have been identified as the main group of the polyamines (PAs) present in all living organisms, including plants. PAs are aliphatic nitrogen compounds which appear in plants as free and conjugated structure (Hussain et al., 2011; Martin-Tanguy, 2001). Due to the positively charged form of the PA molecules, they can interact with negatively charged macromolecules like DNA and RNA, proteins and phospholipids, hence they are involved in the regulation of physical and chemical properties of membranes, nucleic acids structure and functions and modulation of enzyme activities (Hussain et al., 2011). Moreover, more studies have shown the important roles of PAs in, for example, cell growth and development (Fariduddin et al., 2013; Kusano et al., 2008). However, the most interest invokes PAs' function as a stress messenger in plant responses to abiotic stresses (Yamaguchi et al., 2007). Plants exposed to environmental stresses have enhanced content of the PAs (Hussain et al., 2011; Kasukabe et al., 2004), and usually, plants with a high concentration of polyamines are more stress tolerant than those with low one (Kasukabe et al., 2004).

5.3 | Through ion homeostasis

As drought causes an impaired ionic balance, several inorganic ions, mainly K^+ , Na^+ , Cl^- and Ca^{2+} could help achieve drought tolerance in higher plants through ion homeostasis. A large number of ion antiporters and ion channels, including the Na^+/H^+ antiporter, the Na^+/Ca^{2+} antiporter, the K^+/Na^+ antiporter, Ca^{2+} channel and Cl^- channel, mediate the ion transport process that regulates ion uptake, flux and transfer in diverse parts of plants (Chen & Jiang, 2010). K^+ and Cl^- regulates the turgor pressure in guard cells by dominating the water transport system of the plasma membrane and tonoplast. K^+/Cl^- ion homeostasis thus subsequently controls stomatal behaviour by regulating guard cells turgidity (Singh et al., 2015).

The H^+ coupled H^+ -ATPases is a crucial enzyme that induces a negative electrical potential gradient and causes a hyperpolarisation of the plasma membrane. The hyperpolarisation opens voltage regulated inward K^+ channels and results in stomatal opening (Osakabe et al., 2014). However, under drought, the activity of H^+ -ATPases is inhibited in order to restrict excessive water loss by inducing stomatal closing. This ABA-mediated regulation reduces the phosphorylation level of H^+ -ATPases, which is essential to maintain membrane depolarisation (Singh et al., 2015). Thus, transportation of ABA to guard cells from leaf vascular tissues and subsequent reduction in H^+ -ATPases activity makes the plant more tolerant during drought conditions by inducing stomatal closure and minimising transpiration or evaporation.

Calcium (Ca^{2+}) plays major structural and physiological roles in plants and maintains the stability of cell walls, membranes and membrane-bound proteins through bridging chemical residues among these structures. Several physiological and molecular processes are mediated by Ca^{2+} in plants, including cell division, cell differentiation, cell polarity, photomorphogenesis, as well as stress-responses and plant defence (Hadi & Karimi, 2012). Calcium has unique properties and the universal ability to transmit diverse signal, hence, serves as a critical messenger. It triggers primary physiological functions in cells upon exposure to an environmental-stimuli (e.g., stress factors). The three main Ca^{2+} channels located at the plasma membrane and organellar membranes, namely calcium ion-permeable channels, Ca^{2+}/K^+ antiporters, and Ca^{2+} -ATPases, respond to drought stress signals and elevate cytosolic Ca^{2+} concentration, that transduce messages and initiate a signal cascade of biochemical processes (Majid et al., 2007). These elevated Ca^{2+} levels are decoded and transmitted by calcium sensor proteins like calmodulin (CaM), calcineurin B-like proteins (CBLs) and calcium-dependent protein kinases (CDPKs). CBLs interact with a specific group of protein kinases designated CBL-interacting protein kinases (CIPKs), and perform essential roles in numerous Ca^{2+} -dependent processes in plants (Cheong et al., 2010). Moreover, various studies have confirmed the role of Ca^{2+} in inhibiting the synthesis of activating oxides, protecting the structure of cellular plasma membranes and boosting the overall plant drought resistance (Song et al., 2008).

Figure 2 summarises the principal physiological and molecular approaches, including osmotic and ionic adjustments that plants adapt as the counter mechanisms against the drought.

FIGURE 2 Physiological and molecular regulations for drought tolerance in higher plants. Physiological adjustment of stomatal opening and closing during drought curtails excessive water loss from the plant. This adjustment is strictly regulated by the ABA and Ca^{++} . In addition to physiological regulations, a complex system of enzymes and osmolytes minimises the drought-induced oxidative damage by readily scavenging overproduced reactive species. 1O_2 , singlet oxygen; ABA, abscisic acid; APX, ascorbate peroxidase; AsA, ascorbic acid; AsA-GSH, ascorbate-glutathione cycle; ATPase, adenosine triphosphatase; Ca^{++} , calcium ion; CAT, catalase; C_i , internal carbon dioxide concentration; Cl^- , chloride ion; CO_2 , carbon dioxide; DHA, dehydroascorbate; DHAR, dehydroascorbate reductase; GO, glutamate oxidase; GPX, glutathione peroxidase; GR, glutathione reductase; g_s , stomatal conductance; GSA, glutamate semialdehyde; GSH, glutathione; GSSG, glutathione disulphide; H^+ , hydrogen ion; H_2O , water; H_2O_2 , hydrogen peroxide; K^+ , potassium ion; MDHA, monodehydroascorbate; MDHAR, monodehydroascorbate reductase; NAD^+ , nicotinamide adenine dinucleotide ion; NADH, nicotinamide adenine dinucleotide hydride; O_2 , oxygen molecule; O_2^- , superoxide radical; P5C, pyrrolidine-5-carboxylate; SOD, superoxide dismutase; XO, xanthine oxidase; Ψ , leaf water potential

6 | CONCLUSIONS AND FUTURE PERSPECTIVES

It can be concluded that drought imposes oxidative and ionic imbalance in plants through more than one pathway. The

morphological and physiological complications in the plant depend on the severity of the stress and the sensitivity of the plant species. Drought is sensed and signalled through plants via multiple pathways, including ABA dependent and ABA independent pathways. Due to locomotive restrictions, plants possess a complex defence system to counter drought-induced damage. The primary defence line against drought is the change in stomatal behaviour. However, longer acclimatisation requires more than just stomatal adjustment. This protection is provided through biochemical and physiological modulations, that is, OAs, antioxidant metabolism and ion homeostasis. The efficiency of these adjustments, along with other factors, could determine the extent of drought tolerance among various plant species.

While the population is growing exponentially and covering more land than ever, the demand for water has also seen an unprecedented spike. With lesser water available for irrigation, the agriculture sector has suffered drastically and considering the potential impact of these patterns, a serious concern over drought on a global scale has risen recently. Therefore, to ensure global food security, future trends might include:

1. Genetic manipulations to develop more tolerant plant species.
2. Development of plant species with minimised water requirement.
3. Sustainable implication of certain nontoxic growth regulators to provoke OA and antioxidant metabolism in plants under drought conditions.

AUTHOR CONTRIBUTIONS

All authors equally contributed in writing this review and approved the final version.

DATA AVAILABILITY STATEMENT

Data sharing is not applicable to this article as no datasets were generated during this study.

ORCID

Mohammad Mukarram  <https://orcid.org/0000-0002-9034-9366>

REFERENCES

- Agarwal, P.K. & Jha, B. (2010) Transcription factors in plants and ABA dependent and independent abiotic stress signalling. *Biologia Plantarum*, 54, 201–212.
- Ahanger, M.A., Tyagi, S.R., Wani, M.R. & Ahmad, P. (2014) Drought tolerance: role of organic osmolytes, growth regulators, and mineral nutrients. In: Ahmad, P. & Wani, M.R. (Eds.) *Physiological mechanisms and adaptation strategies in plants under changing environment*. New York: Springer, pp. 25–55.
- Akram, M. (2011) Growth and yield components of wheat under water stress of different growth stages. *Bangladesh Journal of Agricultural Research*, 36, 455–468.
- Aroca, R. (2012) *Plant responses to drought stress*. Berlin, Heidelberg: Springer-Verlag GmbH.
- Ashraf, M. & Foolad, M.R. (2007) Roles of glycine betaine and proline in improving plant abiotic stress resistance. *Environmental and Experimental Botany*, 59, 206–216.
- Ault, T.R. (2020) On the essentials of drought in a changing climate. *Science*, 368, 256–260.
- Blum, A. (2017) Osmotic adjustment is a prime drought stress adaptive engine in support of plant production: osmotic adjustment and plant production. *Plant, Cell & Environment*, 40, 4–10.
- Brodribb, T.J., Holbrook, N.M., Edwards, E.J. & Gutierrez, M.V. (2003) Relations between stomatal closure, leaf turgor and xylem vulnerability in eight tropical dry forest trees. *Plant, Cell & Environment*, 26, 443–450.
- Chaves, M.M., Maroco, J.P. & Pereira, J.S. (2003) Understanding plant responses to drought—from genes to the whole plant. *Functional Plant Biology*, 30, 239–264.
- Chen, H. & Jiang, J.G. (2010) Osmotic adjustment and plant adaptation to environmental changes related to drought and salinity. *Environmental Reviews*, 18, 309–319.
- Cheong, Y.H., Sung, S.J., Kim, B.G., Pandey, G.K., Cho, J.S., Kim, K.N. et al. (2010) Constitutive overexpression of the calcium sensor CBL5 confers osmotic or drought stress tolerance in Arabidopsis. *Molecules and Cells*, 29, 159–165.
- Christmann, A., Weiler, E.W., Steudle, E. & Grill, E. (2007) A hydraulic signal in root-to-shoot signalling of water shortage. *The Plant Journal*, 52, 167–174.
- Cochard, H. (2006) Cavitation in trees. *Comptes Rendus Physique*, 7, 1018–1026.
- Corpas, F.J., González-Gordo, S. & Palma, J.M. (2020) Plant peroxisomes: a factory of reactive species. *Frontiers in Plant Science*, 11, 853–864.
- Djilianov, D., Georgieva, T., Moyankova, D., Atanassov, A., Shinozaki, K., Smeeken, S.C.M. et al. (2005) Improved abiotic stress tolerance in plants by accumulation of osmoprotectants—gene transfer approach. *Biotechnology and Biotechnological Equipment*, 19, 63–71.
- Fariduddin, Q., Varshney, P., Yusuf, M. & Ahmad, A. (2013) Polyamines: potent modulators of plant responses to stress. *Journal of Plant Interactions*, 8, 1–16.
- Farooq, M., Basra, S.M.A., Wahid, A., Cheema, Z.A., Cheema, M.A. & Khaliq, A. (2008) Physiological role of exogenously applied glycinebetaine to improve drought tolerance in fine grain aromatic rice (*Oryza sativa* L.). *Journal of Agronomy and Crop Science*, 194, 325–333.
- Farooq, M., Aziz, T., Wahid, A., Lee, D.J. & Siddique, K.H.M. (2009a) Chilling tolerance in maize: agronomic and physiological approaches. *Crop & Pasture Science*, 60, 501.
- Farooq, M., Wahid, A., Kobayashi, N., Fujita, D. & Basra, S.M.A. (2009b) Plant drought stress: effects, mechanisms and management. *Agronomy for Sustainable Development*, 29, 185–212.
- Farooq, M., Hussain, M., Wahid, A. & Siddique, K.H.M. (2012) Drought stress in plants: an overview. In: Aroca, R. (Ed.) *Plant responses to drought stress*. Berlin, Heidelberg: Springer.
- Gill, S.S. & Tuteja, N. (2010) Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. *Plant Physiology and Biochemistry*, 48, 909–909, 930.
- Golldack, D., Li, C., Mohan, H. & Probst, N. (2014) Tolerance to drought and salt stress in plants: unraveling the signaling networks. *Frontiers in Plant Science*, 5, 151.
- González-Gordo, S., Rodríguez-Ruiz, M., Palma, J.M. & Corpas, F.J. (2020) Superoxide radical metabolism in sweet pepper (*Capsicum annuum* L.) fruits is regulated by ripening and by a NO-enriched environment. *Frontiers in Plant Science*, 11, 485.
- Gupta, A., Rico-Medina, A. & Caño-Delgado, A.I. (2020) The physiology of plant responses to drought. *Science*, 368, 266–269.
- Hadi, M.R. & Karimi, N. (2012) The role of calcium in plants' salt tolerance. *Journal of Plant Nutrition*, 35, 2037–2054.

- Hayat, S., Hayat, Q., Alyemeni, M.N., Wani, A.S., Pichtel, J. & Ahmad, A. (2012) Role of proline under changing environments: a review. *Plant Signaling & Behavior*, 7, 1456–1466.
- Huang, G.T., Ma, S.L., Bai, L.P., Zhang, L., Ma, H., Jia, P. et al. (2012) Signal transduction during cold, salt, and drought stresses in plants. *Molecular Biology Reports*, 39, 969–987.
- Hussain, S.S., Ali, M., Ahmad, M. & Siddique, K.H.M. (2011) Polyamines: natural and engineered abiotic and biotic stress tolerance in plants. *Biotechnology Advances*, 29, 300–311.
- Ingram, J. & Bartels, D. (1996) The molecular basis of dehydration tolerance in plants. *Annual Review of Plant Physiology and Plant Molecular Biology*, 47, 377–403.
- Iqbal, M.S., Singh, A.K. & Ansari, M.I. (2020) Effect of drought stress on crop production. In: Rakshit, A., Singh, H., Singh, A., Singh, U. & Fraceto, L. (Eds.) *New Frontiers in stress management for durable agriculture*. Singapore: Springer.
- Janiak, A., Kwaśniewski, M. & Szarejko, I. (2016) Gene expression regulation in roots under drought. *Journal of Experimental Botany*, 67, 1003–1014.
- Kasukabe, Y., He, L., Nada, K., Misawa, S., Ihara, I. & Tachibana, S. (2004) Overexpression of spermidine synthase enhances tolerance to multiple environmental stresses and up-regulates the expression of various stress-regulated genes in transgenic *Arabidopsis thaliana*. *Plant & Cell Physiology*, 45, 712–722.
- Kishor, P.B.K., Sangam, S., Amrutha, R.N., Laxmi, P.S., Naidu, K.R., Rao, K.R. S.S. et al. (2005) Regulation of proline biosynthesis, degradation, uptake and transport in higher plants: its implications in plant growth and abiotic stress tolerance. *Current Science*, 88, 424–438.
- Koca, H., Bor, M., Özdemir, F. & Türkan, İ. (2007) The effect of salt stress on lipid peroxidation, antioxidative enzymes and proline content of sesame cultivars. *Environmental and Experimental Botany*, 60, 344–351.
- Koyro, H.W., Ahmad, P. & Geissler, N. (2012) Abiotic stress responses in plants: an overview. In: Ahmad, P. & Prasad, M.N.V. (Eds.) *Environmental adaptations and stress tolerance of plants in the era of climate change*. New York: Springer New York, pp. 1–28.
- Kusano, T., Berberich, T., Tateda, C. & Takahashi, Y. (2008) Polyamines: essential factors for growth and survival. *Planta*, 228, 367–381.
- Lawson, T. & Matthews, J. (2020) Guard cell metabolism and stomatal function. *Annual Review of Plant Biology*, 71, 273–302.
- Laxa, M., Liebthal, M., Telman, W., Chibani, K. & Dietz, K.J. (2019) The role of the plant antioxidant system in drought tolerance. *Antioxidants*, 8, 94.
- Levitt, J. (1980) *Responses of plants to environmental stress*, (Vol. 1), *Chilling, freezing, and high temperature stresses*. Academic press, p. 497.
- Luan, S. (2002) Signalling drought in guard cells. *Plant, Cell & Environment*, 25, 229–237.
- Lukatkin, A.S. & Anjum, N.A. (2014) Control of cucumber (*Cucumis sativus* L.) tolerance to chilling stress—evaluating the role of ascorbic acid and glutathione. *Front. Environmental Sciences*, 2, 62.
- Mahmood, T., Khalid, S., Abdullah, M., Ahmed, Z., Shah, M.K.N., Ghafoor, A. et al. (2020) Insights into drought stress signaling in plants and the molecular genetic basis of cotton drought tolerance. *Cell*, 9, 105.
- Majid, S.A., Asghar, R. & Murtaza, G. (2007) Potassium-calcium interrelationship linked to drought tolerance in wheat (*Triticum aestivum* L.). *Pakistan Journal of Botany*, 39, 1609–1621.
- Manivannan, P., Jaleel, C.A., Sankar, B., Kishorekumar, A., Somasundaram, R., Lakshmanan, G.M.A. et al. (2007) Growth, biochemical modifications and proline metabolism in *Helianthus annuus* L. as induced by drought stress. *Colloids and Surfaces. B, Biointerfaces*, 59, 141–149.
- Martin-Tanguy, J. (2001) Metabolism and function of polyamines in plants: recent development (new approaches). *Plant Growth Regulation*, 34, 135–148.
- McDowell, N., Pockman, W.T., Allen, C.D., Breshears, D.D., Cobb, N., Kolb, T. et al. (2008) Mechanisms of plant survival and mortality during drought: why do some plants survive while others succumb to drought? *The New Phytologist*, 178, 719–739.
- McGregor, I.R., Helcoski, R., Kunert, N., Tepley, A.J., Gonzalez-Akre, E.B., Herrmann, V. et al. (2020) Tree height and leaf drought tolerance traits shape growth responses across droughts in a temperate broadleaf forest. *The New Phytologist*.
- Mukarram, M., Khan, M.M.A. & Corpas, F.J. (2021a) Silicon nanoparticles elicit an increase in lemongrass (*Cymbopogon flexuosus* [Steud.] Wats) agronomic parameters with a higher essential oil yield. *Journal of Hazardous Materials*, 412, 125254. <https://doi.org/10.1016/j.jhazmat.2021.125254>.
- Mukarram, M., Khan, M.M.A., Uddin, M. & Corpas, F.J. (2021b) Irradiated chitosan (ICH): an alternative tool to increase essential oil content in lemongrass (*Cymbopogon flexuosus*). *Acta Physiologiae Plantarum* (In press).
- Nemhauser, J.L., Hong, F. & Chory, J. (2006) Different plant hormones regulate similar processes through largely nonoverlapping transcriptional responses. *Cell*, 126, 467–475.
- Osakabe, Y., Osakabe, K., Shinozaki, K. & Tran, L.S.P. (2014) Response of plants to water stress. *Frontiers in Plant Science*, 5, 86.
- Quan, L.J., Zhang, B., Shi, W.W. & Li, H.Y. (2008) Hydrogen peroxide in plants: a versatile molecule of the reactive oxygen species network. *Journal of Integrative Plant Biology*, 50, 2–18.
- Reddy, A.R., Chaitanya, K.V. & Vivekanandan, M. (2004) Drought-induced responses of photosynthesis and antioxidant metabolism in higher plants. *Journal of Plant Physiology*, 161, 1189–1202.
- Rowland, L., da Costa, A.C.L., Galbraith, D.R., Oliveira, R.S., Binks, O.J., Oliveira, A.A.R. et al. (2015) Death from drought in tropical forests is triggered by hydraulics not carbon starvation. *Nature*, 528, 119–122.
- Sanders, G.J. & Arndt, S.K. (2012) Osmotic adjustment under drought conditions. In: Aroca, R. (Ed.) *Plant responses to drought stress*. Berlin, Heidelberg: Springer, pp. 199–229.
- Schachtman, D.P. & Goodger, J.Q. (2008) Chemical root to shoot signaling under drought. *Trends in Plant Science*, 13, 281–287.
- Schroeder, J.I., Kwak, J.M. & Allen, G.J. (2001) Guard cell abscisic acid signalling and engineering drought hardiness in plants. *Nature*, 410, 327–330.
- Serraj, R. & Sinclair, T.R. (2002) Osmolyte accumulation: can it really help increase crop yield under drought conditions? *Plant, Cell & Environment*, 25, 333–341.
- Singh, M., Kumar, J., Singh, S., Singh, V.P. & Prasad, S.M. (2015) Roles of osmoprotectants in improving salinity and drought tolerance in plants: a review. *Reviews in Environmental Science and Biotechnology*, 14, 407–426.
- Song, W.Y., Zhang, Z.B., Shao, H.B., Guo, X.L., Cao, H.X., Zhao, H.B. et al. (2008) Relationship between calcium decoding elements and plant abiotic-stress resistance. *International Journal of Biological Sciences*, 4, 116.
- Strauss, G. & Hauser, H. (1986) Stabilization of lipid bilayer vesicles by sucrose during freezing. *Proceedings of the National Academy of Sciences of the United States of America*, 83, 2422–2426.
- Taiz, L. & Zeiger, E. (2010) *Plant physiology*, 5th edition. Sunderland, MA: Sinauer Associates.
- Wu, Q., Wang, M., Shen, J., Chen, D., Zheng, Y. & Zhang, W. (2019) ZmOST1 mediates abscisic acid regulation of guard cell ion channels and drought stress responses. *Journal of Integrative Plant Biology*, 61, 478–491.
- Yamaguchi, K., Takahashi, Y., Berberich, T., Imai, A., Takahashi, T., Michael, A.J. et al. (2007) A protective role for the polyamine spermine against drought stress in *Arabidopsis*. *Biochemical and Biophysical Research Communications*, 352, 486–490.
- Yu, D., Wildhagen, H., Tylewicz, S., Miskolczi, P.C., Bhalerao, R.P. & Polle, A. (2019) Abscisic acid signalling mediates biomass trade-off and allocation in poplar. *The New Phytologist*, 223, 1192–1203.
- Zandalinas, S.I., Balfagón, D., Arbona, V. & Gómez-Cadenas, A. (2017) Modulation of antioxidant defense system is associated with

- combined drought and heat stress tolerance in citrus. *Frontiers in Plant Science*, 8, 953.
- Zhang, J., Jia, W., Yang, J. & Ismail, A.M. (2006) Role of ABA in integrating plant responses to drought and salt stresses. *Field Crops Research*, 97, 111–119.
- Zhao, C., Haigh, A.M., Holford, P. & Chen, Z.H. (2018) Roles of chloroplast retrograde signals and ion transport in plant drought tolerance. *International Journal of Molecular Sciences*, 19, 963.

How to cite this article: Mukarram M, Choudhary S, Kurjak D, Petek A, Khan MMA. Drought: Sensing, signalling, effects and tolerance in higher plants. *Physiologia Plantarum*. 2021;1–10.
<https://doi.org/10.1111/ppl.13423>