

## Stress combination---when two negatives may become antagonistic, synergistic, or additive for plants: A review

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### ABSTRACT

Plants in their natural environment are constantly subjected to various abiotic and biotic stressors and therefore have developed several defence mechanisms to maintain fitness. Stress responses are intricate and require various physiological, biochemical, and cellular changes in plants. The reaction mechanisms in plants subjected to drought, salinity, and heat stresses individually are explained in numerous studies. However, the field condition is far different from the controlled conditions of the labs. In the fields, crops or plants are simultaneously exposed to two or more abiotic and/or biotic stress conditions, such as a combination of salinity and heat, drought and cold, etc. or any of the significant abiotic stresses combined with pathogens' infection. Studies have shown that plants' reactions to combinations of more than two stress factors are distinct and cannot be explicitly deduced from their responses over different stresses when applied separately. Therefore, additional efforts are needed to understand the complete mechanism of plants' responses to stress by analysing single stress with data on multiple stress responses. This review article aims to include an overview of current researches concentrating on plants' responses to a combination of various stress conditions and their influence on the metabolic, transcriptional and physiological characteristics of plants.

*Key Words:* ABA, abiotic stress, biotic stress, multifactorial stress, positive and negative interaction, ROS

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### INTRODUCTION

It is a great challenge for farmers to continuously supply food to the world with an increasing population while the natural resources remain the same. According to FAO (Food and Agriculture Organization), about 1/3rd of the total land area is acknowledged as potentially suitable for arable agriculture, of which merely 3.5% of the total land area remained unaltered by any harsh environmental factors (FAO, 2017). Multifactorial stresses affecting plants are prominent in most agricultural fields worldwide and are characterised as one of the most persistent threats in the field. According to IPCC (Intergovernmental Panel on Climate Change), abiotic stress situations, mainly drought, salinity, and high temperature are the immediate attention of maximum agricultural research as the losses they inflict each year are projected to be \$14-19 million worldwide (IPCC, 2014). A wide variety of biotic components (fungi, bacteria, nematodes, phytoplasmas, viruses, viroids, etc.) and abiotic stress components (drought, heat, salinity, heavy metals, metalloids etc.) regularly and simultaneously challenge plants in their natural habitats (Mantri *et al.*, 2014; Stork, 2018). Consistent exposure to biotic and abiotic stresses alters the

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morpho-physiological, biochemical, and molecular changes in plant cells, which work orderly to control reactive oxygen species (ROS) levels. These affect their growth and productivity (Hasanuzzaman *et al.*, 2020).

Based on the number of interacting factors, stresses are broadly classified into three groups: single, multiple individuals, and combined. Only one stress factor called single stress affects plants' growth and development. In the case of multiple individuals, the consequences of two or more stresses exist at separate time duration without any overlap. However, when there are at least some degrees of overlap between the stresses, it is called combined stress. The concurrence of drought and high temperature in hot seasons is a suitable example of combined abiotic stress. The invasion of plants by bacterial and fungal pathogens at the same period is an example of combined biotic stress. However, plant biologists have conventionally researched abiotic or biotic stress by introducing a single stressor and studied plants' responses to it in order to develop cultivars that are more tolerant of that particular stressor (Matsui *et al.*, 2008; Shaik and Ramakrishna, 2013). Such studies have greatly aided our knowledge about how plants respond to environmental stressors. Though, this kind of investigation, on the other hand, may not mimic the actual field circumstances where plants have to face multiple stresses. In this regard, recent researches have revealed that plants' reaction to the combination of stresses is unique, and thus cannot be inferred from the reactions attained from these stresses when applied separately (Atkinson and Urwin, 2012; Prasad and Sonnewald, 2013; Pandey *et al.*, 2015; Zhang and Sonnewald, 2017).

It is ambiguous whether coexisting stresses are additive, antagonistic, or synergistic, causing a particular form of stress to be more or less sensitive (Maher *et al.*, 2019). Sinha *et al.* (2017) studied the interaction of drought and pathogens on chickpea. They noticed an increased occurrence of fungal infections such as dry root rot (causal agent-*Rhizoctonia bataticola*) and black root rot (causal agent-*Fusarium solani*) under extreme drought, compared to irrigated field conditions. Studies on the simultaneous pathogen and high temperature suggest that elevated temperature enhances the vulnerability of plants to diseases. Sharma *et al.* (2007) observed the amplification in the intensity of Spot blotch, a fungal leaf disease of wheat caused by *Cochliobolus sativus* for six consecutive years, and it was concluded that the rise in average temperatures during night hours was responsible for the disease amplification. However, one beneficial consequence of a plant's exposure to multiple stresses is that it can defend itself against one stress and become immune to several others. This phenomenon is called cross-tolerance, demonstrating that plants develop a complex regulatory mechanism to adapt effectively to the capricious surroundings (Rejeb, 2018). For example, high temperature and stripe rust disease (causal agent- *Puccinia striiformis* f. sp. *tritici*) strengthen disease resistance in *Triticum aestivum* because of high-temperature adult-plant resistance (HTAP), which is only expressed during the adult-plant stage. When the temperatures are warm, it provides durable protection against stripe rust (Carter *et al.*, 2009).

Furthermore, in a concentration-dependent manner, salinity is reported to improve resistance towards the powdery mildew disease (causal agent- *Blumeria graminis*) in *Hordeum vulgare* (Weise *et al.*, 2004). The ion and osmotic toxicity influenced by the salinity stress can feasibly curtail the intensification of the pathogens. The combined influence of two stresses on plants may have a detrimental or beneficial effect; the subsequent stress is typically the one that triggers considerable suffering (Atkinson and Urwin, 2012). Studies on multiple stresses have attempted to simulate the natural environment; however, surrounding conditions could not be managed in the field. The primary stress protection response of plants can be strongly regulated by one stressor (Fujita *et al.*, 2006). It is usually challenging to design convincing combined stress experiments, and the results of combinatorial stress are highly reliant on the experimental setting. The outcomes of the experiments will be determined by plants' growth stage, duration and frequency of stress applications (consecutive or parallel stresses), magnitude and nature of individual

stresses. Regardless of all these differences, some common reactions have been recorded among different treatments. Subsequent exposure with different stresses causes the activation of specific ion channels and kinase cascades (Fraire-Velázquez *et al.*, 2011). This leads to the accumulation of ROS, plant hormones and reorganisation of genetic mechanism assist in many defensive responses along with the increase in plant resistance in order to reduce the crop yield losses (Rejeb *et al.*, 2014).

This article will discuss an overview of the recently published studies focusing on plants' responses towards the combinatorial exposure of abiotic and/or biotic stresses. Aptly, this review aims to enhance and clarify the existing perspective of stress combinations by outlining certain basic principles relevant to them, pointing out their worldwide prevalence, and evaluating their impact on metabolic, transcriptional and physiological characteristics. We will also include a general update of the various stress combinations and address their impact on crucial crops.

## STRESS COMBINATION RESEARCH: A REALITY CHECK

In previous years, a significant number of studies have focused on the responses of plants towards environmental factors. The maximum of which is executed under the restricted environment, especially in laboratory conditions, is the best way to perform reproducible research. Most studies focus solely on the responsiveness of plants towards exposure to a single stress. However, aside from prominent research on the impact of synchronised drought and heat stress (Rizhsky *et al.*, 2004; Mittler, 2006), the effect of stress combinations has been little studied. Until now, the characterisation and investigation of physiological, biochemical, and molecular pathways associated with plants' reactions are primarily concentrated on the stress studied individually, while researches associated with multiple stresses are inadequately represented (Suzuki *et al.*, 2014; Pandey *et al.*, 2017; Zhang and Sonnewald, 2017). In recent decades, several review articles have discussed the promising effects of climatic variation on plants' pathogens and diseases (Juroszek *et al.*, 2020). Unpredictably, Juroszek and co-researchers discovered a reduction in the number of reviews in research concerned with climatic change's impact on plants' pathogens and agricultural diseases since 2014, indicating a loss of attention in this area. In a few decades, there might be a substantial gap endured amid the data accomplished by these studies and the necessary information requisite to evolve plants and crops with improved resistance to field circumstances. These gaps could illustrate how several transgenic plants grew well under lab conditions with improved resistance for specific biotic or abiotic stress situations but failed to justify this in field conditions. Hence, to bridge those gaps and assist the growth of crops and plants by enhancing tolerance at field stress conditions, an emphasis on molecular, physiological, and metabolic prospects of stress combination is required.

Thankfully, some researchers have begun to overcome the technical difficulties of developing the experimental set-up of combinatorial stress factors in the lab under controlled conditions. Hence, in addition to two revolutionary investigations on the combined influence of heat and drought stresses on genomic profiles of tobacco and *Arabidopsis* (Rizhsky *et al.*, 2002, 2004), many current genomic studies have turned to analyse plants' responses to synchronized stresses (Shaar-Moshe *et al.*, 2017; Sinha *et al.*, 2017; Osthoff *et al.*, 2019; Sewelam *et al.*, 2020; Lopez-Delacalle *et al.*, 2021).

## THE NECESSITY OF STUDYING STRESS COMBINATION

According to the IPCC, temperature will be increased between 1.8 and 4.0 °C by the year 2100. Therefore, the conditions will be drier, hotter, wetter, frequent droughts and/or extreme temperature, wind, and precipitation events that will alter the global agricultural system (IPCC, 2014). The dwelling territory

of pests and pathogens would also be affected by climate change as elevated temperature promotes pathogen transmission (El-Sayed and Kamel, 2020). For instance, elevated temperatures are acknowledged to promote pathogen spread (Luck *et al.*, 2011; Madgwick *et al.*, 2011). Furthermore, numerous abiotic stress situations have been proven to decrease plants' defence mechanisms, making them more susceptible to pathogens' infection (Mittler and Blumwald, 2010; Atkinson and Urwin, 2012).

Consequently, primary crops in future fields are supposed to be exposed to a broader range of abiotic and biotic factors, as well as their combinations. Due to climate change, longer growing seasons will lengthen its time for pathogens to reproduce and spread. For example, studying the consequence of temperature on life-history traits of powdery mildew fungus (*Podosphaera plantaginis*) naturally infecting *Plantago lanceolata* displayed hastening of spore germination and stimulation of spore production at higher temperatures (Vaumourin and Laine, 2018). This study shows that contrary to sexual traits, all asexual traits perform better in such situations.

Lopez-Delacalle *et al.* (2021) demonstrate that an exclusive redesigning of metabolic pathways occurs when salinity and heat stress are combined, including alterations in the expression of 1388 genes and the accumulation of 568 molecular characteristics. Furthermore, transcriptomics and metabolic results showed that the proline and ascorbate pathways perform concurrently to uphold cellular redox homeostasis. They recognized important transcription factors from the basic Leucine Zipper Domain (bZIP), Zinc Finger Cysteine-2/Histidine-2 (C2H2) and Trihelix families, which are possible controllers of the reported up-regulated genes in the presence of salinity and heat combination. Such research explains how plants adapt to multifactorial environmental challenges and highlights the synergy between critical cell metabolic pathways for efficient ROS detoxification.

In a large-scale microarray experiment, Rasmussen *et al.* (2013) studied both abiotic and biotic stresses alone and in combinations. They correlate transcriptomic changes in 10 *Arabidopsis thaliana* ecotypes using cold, heat, high-light, salt, and flagellin treatments as single stress factors and double combinations. Approximately 61% of the alternation in combination stresses cannot be figured from the single stress treatment. They also demonstrated that plants favoured only 5--10% of the responding transcripts between potentially antagonistic responses. On analysing the transcriptomic profile of plants exposed to different abiotic stresses, it was revealed that different stresses elicit a substantially unique reaction, and only minor similarities can be created in transcript expression between plants' responses to abiotic stress such as heat, water, cold, salt or mechanical stress (Fracasso *et al.*, 2016; Baillo *et al.*, 2019; Demirel *et al.*, 2020; Kang *et al.*, 2020; Liu *et al.*, 2020). Caixia and his colleagues in 2015 studied the consequence of combined salt and drought stresses on maize (*Zea mays* L.) plants by treating them either singly or simultaneously with soil water deficiency (60% soil water content) and high salinity (5 mL, 250 mM NaCl) for seven days. They observed that the effect of combined stresses differs at both metabolic and physiological levels compared to plants treated individually with drought or salt stress. Maize plants exhibit a completely different paradigm of metabolic reaction to the combined stresses. A few metabolites have responded significantly to it and differ from those caused by individual stress.

Sewelam *et al.* (2020) studied the responses of *Arabidopsis* to salt, osmotic, and heat stresses individually and in combinations. The metabolite profile investigated specified the generation of explicit compatible solutes determined by the type of stress employed. Furthermore, they reported that the treatments that include heat stress lowered transcription of genes that code for abundant photosynthetic proteins and proteins regulating the cell life cycle while up-regulating the genes involved in protein degradation. On exposure to combined stresses, the plants switched their metabolism to endurance, characterised by minimum yield. Studies on multifactorial stresses provide molecular data for the threats to plant yield and the anticipated world food security modelled by heatwaves arising from global warming.

These findings, along with those from other researches on this topic, contemplate abiotic stress combinations for engineering or breeding plants' tolerance to the actual field scenarios.

The information about stress combinations from the fields, laboratories and molecular studies is increasing day by day. These results suggest that plants counter the stress-combination in a non-additive way, generating outcomes that cannot be understood by studying the effect of individual stress (Kissoudis *et al.*, 2014). Multifactorial stress combination could have unforeseeable consequences for agricultural areas or ecosystems. For instance, because of a small level of individual stressor, it might not be possible to detect a distinct reduction in crop productivity. However, despite a small level, the introduction of new stressors might negatively interact with one another and prompt striking reduction in crop productivity and shift the ecosystem to a speedy decline.

## CROSS-TOLERANCE BETWEEN ABIOTIC AND BIOTIC STRESSES

Plants acclimatised to biotic and abiotic stresses by inducing a cascade or network of series that start with stress recognition and finish with various target genes' transcription. These include stress stimuli, signal transduction, transcription regulators, target genes, and stress responses, including changes in morphology, biochemistry, and physiology. Plant responses to different stresses are synchronized by complicated and generally interrelated signalling pathways that regulate several metabolic systems (Rejeb *et al.*, 2014). This process, acknowledged as cross-tolerance, helps plants adjust or acclimatise to various stresses following experience to particular stress. Such interactions have perhaps emerged as a mechanism to allow plants with minimum and suitable biological approaches to counter stress. Signalling cascades stimulate ion channels, kinase cascades, ROS, hormones accumulation, apprehend biotic and abiotic stresses. Recent studies show that defence genes that are usually associated in response to the combination of abiotic or biotic stress show a substantial overlap either shared or as overall stress-reaction genes (Massa *et al.*, 2013; Narsai *et al.*, 2013; Shaik and Ramakrishna, 2013; Sham *et al.*, 2014, 2015; Zhang *et al.*, 2016). Plants, however, direct considerable transcriptional reprogramming in response to a combination of stresses and demonstrate a unique transcript response that is in a way different from either of the stresses separately, despite the significant overlap in responsiveness towards the combination of ecological stresses (Atkinson, 2011; Prash and Sonnewald, 2013; Rasmussen *et al.*, 2013; Sham *et al.*, 2015).

In general, environmental stress can induce resistance in plants through abiotic and biotic stresses. In addition, specific atmospheric stresses can predispose plants by permitting them to react swiftly and in a resistant way to further threats. Cross-tolerance between environmental and biotic stress will thus, cause a beneficial impact by improving tolerance in plants allowing convincing agriculture ideas. Fascinatingly, abiotic stress regulates the defence mechanism at the pathogen infectivity site and other components, thus amplifying plants' resistant mechanism (Venegas-Molina *et al.*, 2020). Prior studies on the interaction of drought with various pathogens in trees found that drought and pathogens' infection had a synergistic effect (Desprez-Loustau *et al.*, 2006). However, few plants that encountered single stress were described as more vulnerable than the simultaneous treatment of different stresses (Suzuki *et al.*, 2014).

Mota *et al.* (2021) studied the transcriptional dynamics elicited by root-knot nematode infection combined with drought stress through the meta-analysis of wild *Arachis* RNA-Sequencing data. *A. stenosperma* overexpressed the cross-stress tolerant endochitinase-encoding gene (AsECHI), which led to the reduction of *Meloidogyne incognita* infection by approximately 30% and stimulated post-drought improvement in *Arabidopsis* plants exposed to both stresses. Biotic and abiotic factors individually arouse hormone-responsive genes majorly in the jasmonic acid (JA) and abscisic acid (ABA) pathways, whereas

multiple stressors appear to stimulate ethylene hormone pathways. The revelation of a network of cross-stress regulated genes in *Arachis* assists in a more excellent knowledge for the complicated regulation of biotic and abiotic stresses in plants, allowing for more effective crop breeding for combined stress tolerance. Davis *et al.* (2015) reported that cereal virus (Barley yellow dwarf virus) infection increases the level of phytohormone and salicylic acid (SA) in *Triticum aestivum* in a time-dependent manner, signifying a possible biochemical basis for virus-induced hormonal responses that modifies plant reaction to environmental stress. In another study, the introduction of bacteria (*Bacillus pumilus*) increased the morphological parameters and elevated drought tolerance in *Glycyrrhiza uralensis* by protecting chloroplast submicroscopic structure, thus increased the chlorophyll content, photosynthetic rate, and water status (Zhang *et al.*, 2019). Various plant growth-promoting bacteria species are reported to enhance drought resistance in maize (Vardharajula *et al.*, 2011), wheat (El-Afry, 2012), bean (Sarma and Saikia, 2014), and *Brassica* species (Saeed *et al.*, 2016). Similarly, numerous mycorrhizal fungi and rhizobacteria are found to induce stress tolerance in many crops by various defence mechanisms, including the production of antioxidants, restricting ethylene generation, and improving ABA regulation (Grover *et al.*, 2011). Application of these microorganisms will offer an innovative approach for gene modification and plant breeding to deliver rapid, however successful, enhancement in crop stress resistance (Grover *et al.*, 2011).

## EFFECT OF STRESS COMBINATION ON THE PHYSIOLOGY

### *Examples of beneficial interaction from combinatorial stress*

Certain stress combinations may have positive effects on plants compared to individual stresses when applied separately. Some recent studies that reported the positive interaction of combined stress are listed in Table I.

TABLE I

Recent experimental studies on the positive effects of combinatorial stress on plants.

Stress combination	Plant	Responses	Reference
Salinity+ Heat	Tomato	reduces the ROS accumulation	Rivero <i>et al.</i> 2013
Drought + Ozone	<i>Medicago truncatula</i>	transcriptome study revealed that genes involved in glucan metabolism, temperature responses, and light signalling play a significant role in moderating ozone responses caused by drought-induced stomatal closure in stress-combination	Iyer <i>et al.</i> (2013)
High CO <sub>2</sub> + High light	Lettuce	increased biomass production, increased the antioxidant capacity	Perez-Lopez <i>et al.</i> (2013)
Drought + Barley yellow dwarf virus	<i>Triticum aestivum</i>	plant's performance was not reduced from virus infection, and infected plants recovered from severe stress events more readily than non-infected plants	Davis <i>et al.</i> (2015)
Salinity + <i>Oidium neolycopersici</i>	<i>Solanum lycopersicum</i>	decreased pathogens' symptoms under severe stress due to accumulation of Na <sup>+</sup> and Cl <sup>-</sup>	Kissoudis <i>et al.</i> (2016)
Soil moisture stress + <i>Sclerotium rolfsii</i>	Chickpea	the expression of defence response genes was highly up-regulated in combined stress treated plants	Tarafdar <i>et al.</i> (2018)
Drought + Waterlogging	<i>Zea mays</i>	enhanced growth parameters (leaf area, plant height and stem diameter) and improved the tolerant mechanism of maize plants	Rafique <i>et al.</i> (2019)

Iyer *et al.* (2013) studied the reaction of *Medicago truncatula* to ozone, drought, and its combination. The authors noticed that short-term ozone stress ( $70 \text{ nmol mol}^{-1}$ ) for 6 h per day for six consecutive days caused chlorosis and small necrotic lesions, while drought (withholding water for a period of 10 d) caused the entire leaves to wilt and break down together with chlorosis. Surprisingly, when those stresses were combined, the impact caused by ozone or drought alone was dramatically wiped out. However, it is believed that the important factor that might be linked to a reduction in the mutual effects of ozone and drought stress could be minimal stomatal conductance. The individual exposure to drought or ozone caused an upsurge in ROS levels. In contrast, the ROS level reduced to a value equivalent to the controlled state when exposed to combinatorial stress. Similarly, in tomato plants, the combinational exposure of salinity and heat stress (Fig. 1) reduces the ROS accumulation to a considerable level. It protects the plants from the damages caused by salt stress (Rivero *et al.*, 2013).

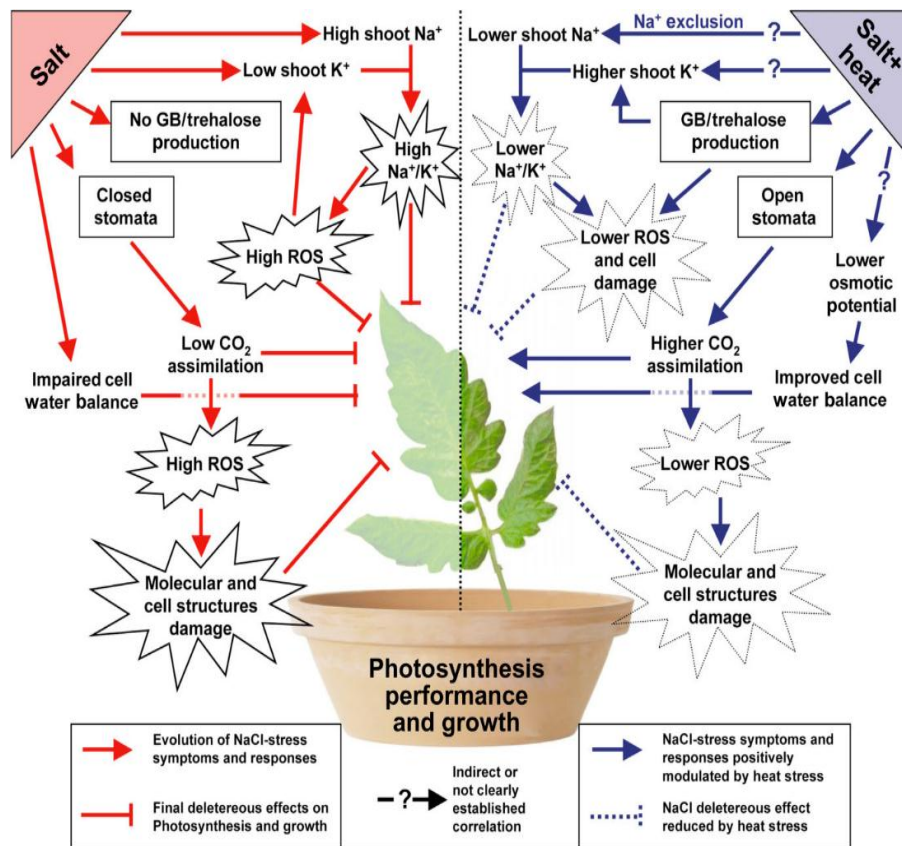


Fig. 1 Evaluation of the physiological reaction of tomato plants to salt stress or a combination of salt and moderate heat stress conferred from the outcomes acquired by Rivero *et al.* (2013) and others. Major reactions promptly adapted by the combination of salt and heat stresses are boxed: stomatal opening, and glycine betaine (GB), and trehalose production. Heat stress prompted stomatal opening to assist cooling of the leaves, permitting elevated transpiration rate and consequently high CO<sub>2</sub> assimilation rate, elevated photosystem II (PSII) function, decreased reactive oxygen species (ROS) generation, and less destruction of the photosynthetic apparatus. Elevated GB elicitation in salt and heat stress combination might improve tolerance by suppressing ROS amassing and also *via* better adjustment of PSII (Chen and Murata, 2011). Adapted from Colmenero-Flores and Rosales, 2014

Abiotic stress, primarily drought and salinity stress, decreases pathogen resistance (Suzuki *et al.*, 2014). However, in some of the experiments conducted by researchers, it has also been observed that specific abiotic stresses often promote resistance in plants to pathogens. For example, to investigate the outcomes of NaCl on the relationship existing amongst tomatoes and powdery mildew, Kissoudis *et al.* (2016) analyzed three different conditions of salt stress: mild, moderate and severe (50, 100, and 150 mM NaCl, respectively) on powdery mildew resistance and general performance of tomato introgression lines with contrasting levels of partial resistance, as well as near-isogenic lines (NILs) having the resistance gene *Ol-1* (associated with a slow hypersensitivity response; HR), *Ol-2* (a *mlo* mutant associated with papilla formation), and *Ol-4* (an *R* gene associated with a fast HR). Increased susceptibility was seen in susceptible and partial resistant lines during mild salt stress (50 mM), followed by increased senescence. On the other hand, severe salt stress (150 mM) lowered the disease symptoms. Under severe stress, Na<sup>+</sup> and Cl<sup>-</sup> concentration in the leaves was linearly associated with reducing pathogens' symptoms. The consequence of simultaneous salinity and powdery mildew on tomato plants is determined by the intensity of the salt stress and the disease-resistance mechanism. Under mild salt stress, negative interactions such as greater powdery mildew susceptibility, leaf senescence, and reduced biomass were seen in most agricultural scenarios. Under extreme salt stress, these effects were partially reversed, but this significantly influenced plant biomass. The expression of ethylene and JA pathway genes and the cell wall invertase gene LIN6 was attributed to greater sensitivity and senescence in NIL-*Ol-1* to the combination of stress. Na<sup>+</sup> and Cl<sup>-</sup> build-up distinguishes salt stress from other abiotic conditions such as drought or heat. This accumulation generally has harmful consequences on the plant, but so is for fungus. NaCl is known to be an antifungal agent (Blomberg and Adler, 1993).

A weak negative connection was found amongst SO<sub>4</sub><sup>2-</sup> and Ca<sup>2+</sup> concentration and enhanced disease resistance, in conjunction with Na<sup>+</sup> and Cl<sup>-</sup>. Because both SO<sub>4</sub><sup>2-</sup> and Ca<sup>2+</sup> promote disease resistance (Kruse *et al.*, 2007; Jiang *et al.*, 2013), disrupting their equilibrium under combined stress may lead to plants' defence failure. These results highlight the significance of stress severity and resistance type on plants' performance under the combination of abiotic and biotic stresses. Drought stress has also been found to augment resistance in tomatoes against *Botrytis cinerea* which stimulates possibly overlapping, pathogen-defence pathways that might not essentially comprise ABA (Achuo *et al.*, 2006). Similar positive interactions have also been reported by other abiotic stresses, such as heavy metal stress. Some non-toxic metal ions induce defence responses to non-hyperaccumulator plants, which are not competent enough to flourish in heavy metal stressed soils. Introduction of healthy *Triticum aestivum* var Sonalika seeds with a mild dose of cadmium (Cd<sup>2+</sup>) given as 50 μM CdCl<sub>2</sub> for 48 h and then washing off Cd<sup>2+</sup> offered resistance to the following infection by *Fusarium oxysporum* inoculum. It is correlated with Cd-binding protein expression; subsequently, an increase in ROS level prompted by metal may also activate defence responses (Mittra *et al.*, 2004).

#### *Examples of negative interaction from combinatorial stress*

In general, the ability of plants to recognise and adapt to different stress combinations is crucial when these particular stresses can have a detrimental influence on the growth and reproduction of plants. Studies on the negative impact of the combinational exposure of drought and heat stresses on plants' development and productivity are given in Table II. One such example is the damages caused by multiple stresses on spring wheat, where the drought stress hampered the grain weight, flower growth, and ovule activity, whereas the pollen fertility and grain quantity were prompted by heat stress (Prasad *et al.*, 2011). The consequences of these multiple stresses are much on reproductive tissues than by drought or heat stress alone, causing a more significant harmful impact on crop yield. Drought and heat stress



combination in *Populus yunnanensis* resulted in much decline in photosynthetic activity with higher ROS generation; however, in *Festuca arundinacea* and *Lolium perenne*, it caused a significant decline in photosystem II (PSII) photochemical efficiency (Jiang and Huang, 2001; Li *et al.*, 2014). It has also been testified that drought situations in England and Wales are associated with increases in the occurrence of common scab (causal agent-*Streptomyces scabiei*) in potatoes (Potato Council News, 2011).

TABLE II

Recent experimental studies on the negative effects of combinatorial stress on plants.

Stress combination	Plant	Responses	References
Salt stress + <i>Pseudomonas syringae</i> pv <i>lachrymans</i>	Cucumber	intensified the negative impact of NaCl on plant growth	Chojak-Kozniewska <i>et al.</i> (2017)
Heat + High Light	<i>Triticum aestivum</i>	quick stomatal closure which leads to fall in photosynthetic rate when CO <sub>2</sub> assimilation and Rubisco activity decrease	Chen <i>et al.</i> (2017)
Drought + Heat	Lentil	decreased the concentrations of sucrose and starch in leaves and seeds by inhibiting the enzymes sucrose synthase and starch phosphorylase	Sehgal <i>et al.</i> (2017)
Salinity + Heat	<i>Carrizo citrange</i>	The enhanced Cl absorption in leaves was due to the high transpiration rate generated by high temperatures, which counteracted physiological responses of plants to salt stress	Balfagón <i>et al.</i> (2018)
Drought + Heat	Canola	oil production was significantly lowered due to a decrease in photosynthetic carbon absorption.	Elferjani and Soolanayakanahally (2018)
Drought + Heat	<i>Solanum lycopersicum</i>	reduced electron transport rate (ETR) and $\phi$ PSII	Zhou <i>et al.</i> (2019)
Drought+ Heat	Barley	severe yield loss (over 95%)	Mahalingam and Bregitzer (2019)
Hight temperature + Drought	<i>Triticum aestivum</i>	Pollen susceptibility and impairment to female reproductive organs resulted in a nearly 55%t reduction in yield.	Fábiá <i>et al.</i> (2019)
Drought + Heat	<i>Triticum aestivum</i>	Response was hypo-additive, water relation characteristics were negatively affected	Sattar <i>et al.</i> (2020)

In contrast to individual stresses, synchronised treatment of drought and *Turnip mosaic virus* (TuMV) to *Arabidopsis* contributed to a greater decline in plants' weight and leaf number (Prasch and Sonnewald, 2013). Silva *et al.* (2013) assessed the contrasting primary physiological reaction of *Jatropha curcas* with salt stress singly and in combination with heat stress. Before and after exposure to 43 °C for 6 hours, the plants were treated with salt stress (100 mM NaCl). The outcomes of salt stress were much more severe than heat stress, and when all these stress factors were combined, salinity augmented. Substantial reduction in CO<sub>2</sub> assimilation rate and stomatal conductance were involved in the adverse effects of the combined treatments. Stress-combination increased Na<sup>+</sup> and Cl<sup>-</sup> build-up in the leaves because of membrane destruction and lipid peroxidation. In another study, Mahalingam and Bregitzer (2019) studied the influence of short-term heat, drought and its combination on barley varieties. They explained that susceptibility to the stress-combination was generally higher than that of heat or drought stress alone. Also, severe yield loss (over 95%) was reported when stress combination was imposed during the heading stage.

In another study performed by Zhou *et al.* (2019), the activity of the photosynthetic apparatus was reduced in tomato plants when treated with a combination of drought and heat stress. Regarding control set-ups, tomato plants lowered their electron transport rate (ETR) and quantum yield of photosystem II ( $\phi$ PSII) when subjected to drought (combined or not with heat stress). Interestingly, plants under drought conditions could repair ETR and  $\phi$ PSII following a recovery period, but plants under stress-combinations could not. Hence, the photosynthetic apparatus was irreversibly damaged due to the combined stress, and tomato plants could not recover PSII performance following the combined stress period. In a similar study, two citrus genotypes, *Carrizo citrange* and *Cleopatra mandarin*, demonstrated different capabilities to handle elevated temperatures individually or with drought stress. The leaf relative water content (RWC) in both genotypes had a similar reduction in water scarcity, showing that the effect was identical in both genotypes for drought. Contrarily, heat stress increases transpiration in both citrus genotypes. However, there was an extreme decline in RWC when both stresses were introduced in combination. This might be because of the additive effect of the individual stresses (drought-induced water loss and high temperatures increased transpiration). Likewise, the compatible osmolyte proline accumulation was also maximum in drought + heat stress treatments (Zandalinas *et al.*, 2016b). The osmotically active molecule, proline (García-Sánchez *et al.*, 2007; Moustakas *et al.*, 2011) is also concentrated in response to other different stresses.

Consequently, in addition to its well-known function as a compatible osmolyte, proline has a number of other defending properties, such as redox balance and radical scavenging, protein structure preservation, performing as a molecular chaperonin, improving the activities of various enzymes, and helping to reduce cell membrane damage (Shao *et al.*, 2008; Szabados and Saviouré, 2010). In the above study, an increase in proline concentration was linked to water loss brought up by drought conditions, whereas the increase in transpiration rates was connected to the high temperatures or both. As a result, large regulatory amounts of proline inhibit further osmolyte production unless more extreme stress conditions are encountered, enabling the linear relationship between the RWC and the biochemical response to be disrupted.

Sattar *et al.* (2020) investigated the combined and individual effect of drought and heat stress on water relations, photosynthetic pigments, osmolytes accumulation and antioxidants defence mechanism in the flag leaf of bread wheat. It was found that the collaborative outcomes of both stresses were hypo-additive. Similarly, water stress and fungal infection (*Erysiphe cruciferarum*) had an additive effect on *Alliaria petiolata* growth. Though water-stress lessened disease expansion (percentage of infected leaf area dropped under water stress circumstances), plants under drought stress were significantly smaller than those appropriately watered, so powdery mildew covered the whole leaf area by the termination of the experiment (Enright and Cipollini, 2007).

## UNIQUE TRANSCRIPTOMIC FEEDBACK OF THE PLANTS TO THE STRESS COMBINATION

Despite having a plethora of studies committed to single-stress conditions, quite a few are focused on understanding the molecular mechanism adapted by plants against the combination of two or more different stresses. Fortunately, a current rise in the number of transcriptomic studies intending to interpret the reaction of plants to stress combinations (biotic and abiotic) has been seen. Some of these studies are cold + drought (Zheng *et al.*, 2016), salinity + heat (Suzuki *et al.*, 2016), drought/flooding + herbivore attack (Nguyen *et al.*, 2016), drought + pathogen (Sinha *et al.*, 2017), drought + heat (Ashoub *et al.*, 2018; Liu *et al.*, 2018; Wang *et al.*, 2018), salinity + ozone (Natali *et al.*, 2018), light + heat (Balfagón *et al.*, 2019). In the response of plants with high temperatures and different other stresses, the heat shock factors (HSF) family of transcriptional regulators play a pivotal function by influencing both immediate

responses and epigenetic control of heat stress memory (Jacob *et al.*, 2017; Ohama *et al.*, 2017). Shaar-Moshe *et al.* (2017) studied the transcriptional patterns and morpho-physiological acclimatization of *Brachypodium dystachion* to salinity, drought, and heat stresses individually and in their double and triple combinations. As plants were exposed to combined stresses, hierarchical clustering analysis of morpho-physiological acclimatization revealed that various attributes displayed continuous deteriorating outcomes. Only 37% of differentially expressed genes conserved their responses under single and combined stresses, demonstrating restricted expression constancy among partly overlapping stresses. When common stress genes were compared to the ones exclusively expressed to the combination of stresses, a substantial switch from augmented intensity to antagonistic responses was discovered. The varied transcriptional fingerprints reflect a change in the mechanism of action when multiple stresses are combined and inadequate potential to anticipate plants' responses as different stresses are combined.

Osthoff *et al.* (2019) reveal that the intricate transcriptomic responses regulate the recognition and signalling of stress-combination in barley. They studied the impact of drought and salt stresses individually and its combination on barley seminal roots' development and transcriptomic plasticity. Drought and combined stress notably decreased the total root length when exposed for more than four days. Furthermore, the transcriptome sequence illustrates that 60--80% of stress type-specific gene expression responses noticed six hours following exposure was available too after 24 h of stress treatment. Subsequently, in 24 h of stress treatments, 100s of extra genes were stress-regulated compared to 6 h of application. The combination of salinity and drought results in a distinctive transcriptomic reaction.

Moreover, transcription factors promoted global reprogramming with the persistent preponderance of basic helix-loop-helix (bHLH) transcription factors, HSF, and ethylene response factors (ERF). Rizhsky *et al.* (2002) employed cDNA arrays, together with physiological parameters, to analyse the consequence of drought and heat shock on tobacco plants. Due to drought + heat shock application, the closing of stomata with a reduction of photosynthesis and increased respiration was reported. During their combinatorial treatment, those transcripts triggered at the time of drought exposure (such as those encoding catalase and dehydrin), and heat shock (thioredoxin peroxidase and ascorbate peroxidase) were eliminated. However, in the combination of drought and heat shock exposure, additional transcript expression such as glutathione-peroxidase, pathogen-related proteins, a WRKY transcription factor, ethylene response transcriptional co-activator was explicitly activated. Photosynthesis genes have been silenced, while transcripts expressing certain enzymes of the glycolysis and pentose phosphate pathway have been triggered, indicating the use of sugars during stress by these pathways.

Balfagón *et al.* (2019) studied the responses of *A. thaliana* plants to high light and heat stress-combination. High light and heat stress-combination caused unalterable destruction to PSII, decreased D1 (encoded by PsbA gene) protein levels, and elevated transcriptional response suggestive of PSII repair activation. High light + heat stress-specific transcripts are engaged in several other processes, such as redox mechanism, transportation of protein, protein catabolic processes, or photosynthesis and associated with cadmium-responses, salinity, or involvement in the citric acid cycle. They noticed that HSFs exhibited an additive expression, of which HSFA2, HSFA7A, HSFB1, HSFB2A, and HSFB2B display the maximum expression values to high light + heat stress conditions. A mutant deficient in JA biosynthesis (allene oxide synthase) showed increased susceptibility to high light and heat stress conditions, indicating that JA is necessary for altering numerous transcriptional responses exclusive to stress combination. Johnson *et al.* (2014) studied drought and heat stress conditions singly or in combination to understand the transcriptional response of *Sorghum bicolor* using microarrays. Following drought and heat stress, microarrays with 28585 gene probes observed gene expression alterations corresponding to 4% and 18% of genes on the chip, respectively. Approximately 20% of probes were

differentially expressed in response to drought and heat stress-combination. In *Sorghum* combined stress response, ontological investigation of these 'unique' transcripts revealed a possible involvement for particular transcription factors like MYB78 and ATAF1, chaperones such as HSPs, and biochemical functions like polyamine synthesis.

## SIGNALLING PATHWAY INDUCED BY MULTIPLE STRESSES' RESPONSE

Earlier researches on biotic and/or abiotic stress combinations concerning two or three different stress factors (Rasmussen *et al.*, 2013; Zhou *et al.*, 2019; Zandalinas *et al.*, 2020) confirmed that plants respond to stress combinations in an exclusive means and their responses do not imitate the sum of the plants' responses to each of the two or three individual stresses that contribute in combination. Though in some cases of stress-combination, the reaction to one of the stressors involved could be more prevalent than the other (Zhou *et al.*, 2019; Sewelam *et al.*, 2020), or the different stresses might have an overall additive effect (Vile *et al.*, 2012; Bansal *et al.*, 2013; Shaar-Moshe *et al.*, 2017). Nearly all researches accomplished until recently about plants' responsiveness to stress combinations include transcripts, proteins, and metabolites that are unique to the combination (Prasch and Sonnewald, 2013; Kissaudis *et al.*, 2014; Sewelam *et al.*, 2020).

### *Abscisic acid (ABA) signalling*

ABA acts as chemical stimuli that stimulate alternations in plants' physiological and development approaches in response to atmospheric factors, hence, contributing to the acclimatisation of plants to different abiotic and biotic stresses in combination and alone (Ramegowdaa and Senthil-Kumar, 2015; Berensa *et al.*, 2019; Gull *et al.*, 2019). Stress-combination studies demonstrated that ABA might play a role in the modified response of plants towards drought and heat stress exposures either in combination or alone. For instance, on exposure to drought or heat stress alone, the accumulation of 9-cis epoxy-carotenoid dioxygenase (NCED) protein essential for ABA synthesis was significantly up-regulated in poplar (Li *et al.*, 2014). In contrast, in drought + heat stress, NCED protein accumulation elevated at first and then decreased. These findings show that ABA has different regulatory activities under single and combinatorial stressors. ABA serves as a pivotal regulator that manages water status and stomatal activities. During water stress, plants elicit and accumulate ABA that brings about stomata closure leading to water conservation. The cellular and molecular pathways concerning stomatal closure prompted by ABA have been comprehensively studied and reviewed (Lim *et al.*, 2015; Zhao *et al.*, 2017; Niu *et al.*, 2018; Bharath *et al.*, 2021). In addition to contributing to water management through drought conditions, stomatal closure likewise acts as a protective mechanism in stopping pathogen incursions. In addition to ABA's involvement in stomatal closure that restricts pathogen entry, ABA also influences pathogens' interactions by intervening with other hormones involved in plants' defence mechanisms (Oide *et al.*, 2013; Lim *et al.*, 2015; Boba *et al.*, 2020).

Plants identify pathogen-associated molecular patterns (PAMP), which activate their innate immunity. PAMPs participate in closing stomata and curb the entry of pathogens (Choi and Klessig, 2016). However, in some studies, it is investigated that the closing of stomata is not caused by PAMPs in the mutant plants using *ost1* mutants that do not respond to ABA (Mustilli *et al.*, 2002) and ABA-deficient *aba3-1* mutant (Leon-Kloosterziel *et al.*, 1996). It is therefore suggested that in guard cells, PAMP-regulated stomatal closure involves intensive ABA signalling pathways. Consequently, by activation of stomatal closure, ABA does indeed have a beneficial impact on disease resistance. Tossi *et al.* (2014) reported that UV-B irradiation in *A. thaliana* enhanced the production of ABA and the

synthesis of nitric oxide (NO) and ROS, thus contributing to the closing of stomata. ABA-induced stomatal closing might be more significant for acclimating plants to the drought + UV-B stress-combination than the modified plant responses to the drought + heat stress conditions.

Several plant hormones, such as SA, JA, and ethylene play a significant role in resisting pathogens. Generally, SA is associated with systemic acquired resistance (SAR) and biotrophic pathogen resistance, while JA and ethylene are associated with induce systemic resistance (ISR) and necrotrophic pathogen resistance (Glazebrook, 2005; Pieterse *et al.*, 2009). Meanwhile, ABA is associated with the SA, JA and ethylene signalling pathways and functions either synergistically or antagonistically with these hormones (Anderson *et al.*, 2004; Mosher *et al.*, 2010). At the gene transcription level, ABA has been shown to have a deleterious influence on JA and ethylene-dependent pathogen resistance (Anderson *et al.*, 2004). ABA deficient mutants triggered the usual JA and ethylene marker genes, including PDF1.2 and CHI, more intensely than wild-type *Arabidopsis*; however, exogenous ABA application down-regulated these genes (Anderson *et al.*, 2004). Transcriptomic study of salinity and heat stresses individually and in combination in *A. thaliana* found that the expression of 699 transcripts was explicitly stimulated in reaction to stress-combination (Suzuki *et al.*, 2016). Strangely, in these 699 transcripts, transcripts that correlated with the ABA-signalling pathway were extensively characterised. In comparison, transcripts associated with SA and gibberellic acid, which might antagonise ABA's signalling mechanism, were the least influential (Ishibashi *et al.*, 2012).

Recent researches have discovered a complicated coordination mode between various hormone signals in *Arabidopsis* and other plants in response to drought, heat, and their combinations. For instance, ABA accumulated swiftly in citrus plants in response to individual drought treatments. (Zandalinas *et al.*, 2016a). Drought + heat stress caused a surge in ABA accumulation, though to a considerably lesser amount than drought alone. Unlike ABA, a higher level of SA may be implicated in the signalling pathway antagonising ABA, accumulated under the drought + heat stress combination compared to its individual application (Moeder *et al.*, 2010). Under these single and combined stresses, citrus plants showed a different ABA and SA accumulation profile than *Arabidopsis*, which showed the maximum or least amount of ABA or SA accumulation, respectively, under identical scenarios (Zandalinas *et al.*, 2016a).

### *Reactive oxygen species (ROS)*

The main variables that limit crop productivity and yield are abiotic stresses such as heavy metals, water stress, salinity, high temperature, etc. These stresses are linked to the generation of a specified harmful chemical entity known as ROS (Fig. 2), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), superoxide radical (O<sub>2</sub><sup>-</sup>), hydroxyl radical, etc. ROS can induce cellular damage through protein degradation, enzyme inactivation, gene alterations, and interference with various metabolically essential pathways. It has been concluded from multiple studies that during stress-combination, the specific sequence of ROS concentration, antioxidants, and expression of different scavenging enzymes was shown concerning the sequence obtained by introducing those stresses individually. These modifications have been displayed in levels of O<sub>2</sub><sup>-</sup>, H<sub>2</sub>O<sub>2</sub>, expression of enzymes such as peroxidases, glutathione-S-transferase, glutathione reductase, and concentration of antioxidants and osmoprotectants such as proline, glycine betaine, trehalose, and sucrose (Rivero *et al.*, 2013; Suzuki *et al.*, 2014; Jin *et al.*, 2015; Pandey *et al.*, 2015; Vuleta *et al.*, 2015; Carvalho *et al.*, 2016; Martinez *et al.*, 2016).

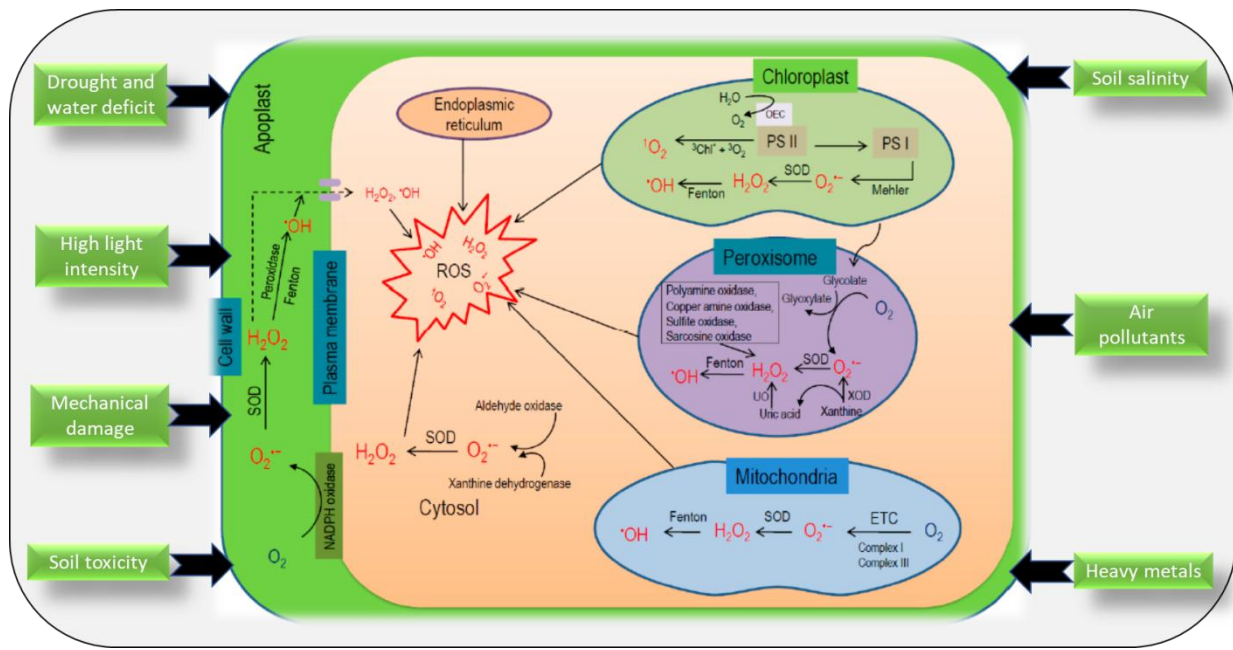


Fig. 2 Production of reactive oxygen species (ROS) in plants by different abiotic stress factors. (Adapted from Hasanuzzaman *et al.*, 2020)

As shown by reports that transgenic plants overexpressing ROS-scavengers or mutants with greater ROS-scavenging capacity exhibit improved resistance to environmental stresses, it is thus concluded that ROS leads to stress damage (Hasegawa *et al.*, 2000; Kocsy *et al.*, 2001). Upon penetration with the vascular pathogen *Verticillium* spp, ROS is implicated in stress-induced tolerance in *A. thaliana* by escalating drought tolerance because of the development of *de-novo* xylem and the consequently increased water flow (Xia *et al.*, 2012). In a study, brassinosteroids treated cucumber plants caused an escalation in  $H_2O_2$  levels and prepared the plant for biotic and abiotic stress tolerance (Xia *et al.*, 2012). The salt resistance activation of  $H_2O_2$  in citrus raised the concentration of oxidised and S-nitrosylated proteins significantly, and the level persisted like that after stress treatment, but non-treated plants were more stress-sensitive (Tanou *et al.*, 2009). Hypoxic or anoxic conditions caused by flooding or waterlogging (WL) produce toxic compounds that impede plant metabolism, resulting in ROS overproduction and oxidative damage (Loreti *et al.*, 2016). Anee *et al.* (2019) studied the WL-sensitive *Sesamum indicum* for different time periods (2, 4, 6, and 8 d) of WL stress and analysed that malondialdehyde (MDA) and  $H_2O_2$  increased in a time-dependent manner. *Solanum lycopersicum* showed an equivalent increase in these oxidative stress indicators (Rasheed *et al.*, 2018). After exposure to WL conditions for 7 d, an Antarctic plant called *Deschampsia antarctica* generated increased MDA and  $H_2O_2$  (Park and Lee, 2019). Furthermore, mutants lacking in the function of the ABA and ROS-regulated protein PP2Cs (*abi-1*) have shown to be particularly susceptible to drought and heat stress combination, along with salinity and heat, indicating the involvement of ROS-ABA interactions for plant tolerance to stress combinations (Suzuki *et al.*, 2016; Zandalinas *et al.*, 2016a).

A vast number of studies have shown increased ROS response transcripts as a significant constituent of the stress-combination-acclimatisation response pathway, highlighting the importance of ROS for plants' acclimation to stress combinations (Suzuki *et al.*, 2014). In this expanding and essential line of plant stress research, novel roles for ROS in plant adaptation to stress combinations will likely be highlighted in upcoming studies. Moreover, ROS and ABA appeared to be two of the major influencers that facilitate plants' acclimatisation to stress combinations for the time being.

## CONCLUSIONS

In conclusion, plant sciences have currently achieved major advances to elucidate multiple abiotic and biotic stress effects. We have described the versatile role of abiotic stress and biotic stress conditions when plants are exposed to stress combinations, such as the simultaneous impact of abiotic stress conditions (high temperature, high salt concentrations and drought, etc.) along with biotic stress conditions. The transcriptomics, metagenomics, and metabolomics data were used to address the bottleneck of plant-stress interactions research. Interestingly, transcriptome data of plants exposed to combination of stress stimuli indicated that numbers of genes are differentially expressed than might be predictable from the combination of the single stress. However, key knowledge gaps remain, especially regarding the biological relevance of the rhizospheric microbiomes changes under different stress conditions. Understanding how plants' responses to stress conditions are initiated will allow us to answer fundamental questions on how plants induce biochemical, physiological and molecular modifications regulated by abiotic, biotic and both stress types. It will also help us to understand what is required for a plant to tolerate a stressful condition. Such an increased understanding of plant-stress connection in natural systems is strongly needed, as our review article pointed out that molecular research efforts in crops have to be strengthened considerably.

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## REFERENCES

- Achuo E A, Prinsen E, Hoftte M. 2006. Influence of drought, salt stress and abscisic acid on the resistance of tomato to *Botrytis cineria* and *Oidium neolycopersici*. *Plant Pathol.* **55**: 178--186.
- Anderson J P, Badruzaufari E, Schenk P M, Manners J, Desmond O J, Ehlert C, Maclean D J, Ebert P R, Kazan K. 2004. Antagonistic interaction between abscisic acid and jasmonate-ethylene signaling pathways modulates defense gene expression and disease resistance in *Arabidopsis*. *Plant Cell.* **16**: 3460--3479.
- Anee T I, Nahar K, Rahman A, Mahmud J A, Bhuiyan T F, Alam M U, Fujita M, Hasanuzzaman M. 2019. Oxidative damage and antioxidant defense in *Sesamum indicum* after different waterlogging durations. *Plants.* **8**: 196.
- Ashoub A, Müller N, Jiménez-Gómez J M, Brüggemann W. 2018. Prominent alterations of wild barley leaf transcriptome in response to individual and combined drought acclimation and heat shock conditions. *Physiol Plant.* **163**:18--29.
- Atkinson N J. 2011. Plant molecular response to combined drought and nematode stress. Ph.D. thesis, University of Leeds, England. <http://etheses.whiterose.ac.uk/2131/>
- Atkinson N J, Urwin P E. 2012. The interaction of plant biotic and abiotic stresses: from genes to the field. *J Exp Bot.* **63**: 3523--3543.
- Baillo E H, Kimotho R N, Zhang Z, Ping Xu P. 2019. Transcription Factors Associated with Abiotic and Biotic Stress Tolerance and Their Potential for Crops Improvement. *Genes.* **10**: 771.
- Balfagón D, Zandalinas S I, Baliño P, Muriach M, Gómez-Cadenas A. 2018. Involvement of ascorbate peroxidase and heat shock proteins on citrus tolerance to combined conditions of drought and high temperatures. *Plant Physiol Bioch.* **127**: 194--199.
- Balfagón D, Sengupta S, Gómez-Cadenas A, Fritschi F B, Azad R K, Mittler R, Zandalinas S I. 2019. Jasmonic acid is required for plant acclimation to a combination of high light and heat stress. *Plant Physiol.* **181**: 1668--1682.

- Bansal S, Hallsby G, Löfvenius M O, Nilsson M C. 2013. Synergistic, additive and antagonistic impacts of drought and herbivory on *Pinus sylvestris*: leaf, tissue and whole-plant responses and recovery. *Tree Physiol.* **33**: 451--463.
- Berensa M L, Wolinskaa K W, Spaepena S, Zieglerb J, Noboria T, Krülera A N A V, Winkelmüllera T M, Wanga Y, Minea A, Beckera D, Garrido-Otera R, Schulze-Leferta P, Tsudaa K. 2019. Balancing trade-offs between biotic and abiotic stress responses through leaf age-dependent variation in stress hormone cross-talk. *PNAS.* **116**: 2364--2373.
- Bharath P, Gahir S, Raghavendra A S. 2021. Abscisic Acid-Induced Stomatal Closure: An Important Component of Plant Defense Against Abiotic and Biotic Stress. *Front Plant Sci.* **12**: 615114.
- Blomberg A, Adler L. 1993. Tolerance of fungi to NaCl. In Jennings D H (ed.) Stress tolerance of fungi. New York: Marcel Dekker pp. 209--232.
- Boba A, Kostyn K, Kozak B, Wojtasik W, Preisner M, Prescha A, Gola E M, Lysh D, Dudek B, Szopa J, Kulma A. 2020. *Fusarium oxysporum* infection activates the plastidial branch of the terpenoid biosynthesis pathway in flax, leading to increased ABA synthesis. *Planta.* **251**: 50.
- Caixia S, Gao X, Fu J, Zhou J, Wu X. 2015. Metabolic response of maize (*Zea mays* L.) plants to combined drought and salt stress. *Plant Soil.* **388**: 99--117.
- Carter A H, Chen X M, Garland-Campbell K, Kidwell K K. 2009. Identifying QTL for high-temperature adult-plant resistance to stripe rust (*Puccinia striiformis* f. sp. *tritici*) in the spring wheat (*Triticum aestivum* L.) cultivar 'Louise'. *Theor Appl Genet.* **119**: 1119--1128.
- Carvalho L C, Coito J L, Goncalves E F, Chaves M M, Amancio S. 2016. Differential physiological response of the grapevine varieties *Touriga nacional* and *Trincadeira* to combined heat, drought and light stresses. *Plant Biol.* **18**: 101--111.
- Chen T H H, Murata N. 2011. Glycine betaine protects plants against abiotic stress: mechanisms and biotechnological applications. *Plant Cell Environ.* **34**: 120.
- Chen Y E, Zhang C M, Su Y Q, Ma J, Zhang Z W, Yuan M, Zhang H Y, Yuan S. 2017. Responses of photosystem II and antioxidative systems to high light and high temperature co-stress in wheat. *Environ Exp Bot.* **135**: 45--55.
- Choi H W, Klessig D F. 2016. DAMPs, MAMPs, and NAMPs in plant innate immunity. *BMC Plant Biol.* **16**: 232.
- Chojak-Kozniewska J, Linkiewicz A, Sowa S, Radzioch M A, Kuzniak E. 2017. Interactive effects of salt stress and *Pseudomonas syringae* pv *lachrymans* infection in cucumber: involvement of antioxidant enzymes, abscisic acid and salicylic acid. *Environ Exp Bot.* **136**: 9--20.
- Colmenero-Flores J M, Rosales M A. 2014. Interaction between salt and heat stress: when two wrongs make a right. *Plant Cell Environ.* **37**: 1042--1045.
- Davis T S, Bosque - Pérez N A, Foote N E, Magney T, Eigenbrode S D. 2015. Environmentally dependent host-pathogen and vector-pathogen interactions in the Barley yellow dwarf virus pathosystem. *J App Ecol.* **52**: 1392--1401.
- Demirel U, Morris W L, Ducreux L J M, Yavuz C, Asim A, Tindas I, Campbell R, Morris J A, Verrall S R, Hedley P E, Gokce Z N O, Caliskan S, Aksoy E, Caliskan M E, Taylor M A, Hancock R D. 2020. Physiological, Biochemical, and Transcriptional Responses to Single and Combined Abiotic Stress in Stress-Tolerant and Stress-Sensitive Potato Genotype. *Front Plant Sci.* **11**: 169.
- Desprez-Loustau M L, Marçais B, Nageleisen L M, Piou D, Vannini A. 2006. Interactive effects of drought and pathogens in forest trees. *Ann For Sci.* **63**: 597--612.
- El-Afry M M. 2012. Anatomical studies on drought-stressed wheat plants (*Triticum aestivum* L.) treated with some bacterial strains. *Acta Biol Szeg.* **56**: 165--174.
- Elferjani R, Soolanayakanahally R. 2018 Canola responses to drought, heat, and combined stress: shared and specific effects on carbon assimilation, seed yield, and oil composition. *Front Plant Sci.* **9**: 1224
- El-Sayed A, Kamel M. 2020. Climatic changes and their role in emergence and re-emergence of diseases. *Environ Sci Poll Res.* **27**: 22336--22352.
- Enright S M, Cipollini D. 2007. Infection by powdery mildew *Erysiphe cruciferarum* (Erysiphaceae) strongly affects growth and fitness of *Alliaria petiolata* (Brassicaceae). *Am J Bot.* **94**: 1813--1820.
- Fábiá A, Sáfrán E, Szabó-Eitel G, Barnabás B, Jäger K. 2019. Stigma functionality and fertility are reduced by heat and drought co-stress in wheat. *Front Plant Sci.* **10**: 244.
- Food and Agriculture Organization (FAO). 2017. Voluntary Guidelines for Sustainable Soil Management Food and Agriculture Organization of the United Nations Rome, Italy. <http://www.fao.org/home/en/>
- Fracasso A, Trindade L M, Amaducci S. 2016. Drought stress tolerance strategies revealed by RNA-Seq in two sorghum genotypes with contrasting WUE. *BMC Plant Biol.* **16**: 115.
- Fraire-Velázquez S, Rodríguez-Guerra R, Sánchez-Calderón L. 2011. Abiotic and biotic stress response crosstalk in plants. In Shanker A (ed) Abiotic and Biotic Stress Response in Plants Physiological, Biochemical and Genetic Perspectives. InTech Open Access Company: Rijeka, Croatia. pp. 1--26.



- Fujita M, Fijita Y, Noutoshi Y, Takahashi F, Narusaka Y, Yamaguchi-Shinozaki K, Shinozaki K. 2006. Crosstalk between abiotic and biotic stress responses: A current view from the points of convergence in the stress signaling networks. *Curr Opin Plant Biol.* **9**: 436--442.
- García-Sánchez F, Syvertsen J P, Gimeno V, Botía P, Perez-Perez J G. 2007. Responses to flooding and drought stress by two citrus rootstock seedlings with different water-use efficiency. *Physiol Plant.* **130**: 532--542.
- Glazebrook J. 2005. Contrasting mechanisms of defense against biotrophic and necrotrophic pathogens. *Ann Rev Phytopathol.* **43**: 205--227.
- Grover M, Ali S Z, Sandhya V, Rasul A, Venkateswarlu B. 2011. Role of microorganisms in adaptation of agriculture crops to abiotic stresses. *World J Microbiol Biotechnol.* **27**: 1231--1240.
- Gull A, Lone A A, Wani N U I. 2019. Biotic and abiotic stresses in plants. In Oliveira A O (ed) *Abiotic and Biotic Stress in Plants*. IntechOpen London, United Kingdom. pp 1--5.
- Hasanuzzaman M, Bhuyan M H M B, Zulfiqar F, Raza A, Mohsin S M, Al-Mahmud J, Fujita M, Fotopoulos V. 2020. Reactive oxygen species and antioxidant defense in plants under abiotic stress: revisiting the crucial role of a universal defense regulator. *Antioxidants.* **9**: 681.
- Hasegawa P M, Bressan R A, Zhu J K, Bohnert H J. 2000. Plant cellular and molecular responses to high salinity. *Annu Rev Plant Mol Plant Physiol.* **51**: 463--499.
- Intergovernmental Panel on Climate Change (IPCC). 2014. Synthesis Report. Contribution of Working Groups I, II and III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. Geneva, Switzerland. IPCC. 151.
- Ishibashi Y, Tawaratsumida T, Kondo K, Kasa S, Sakamoto M, Aoki N, Zheng SH, Yuasa T, Iwaya-Inoue M. 2012. Reactive oxygen species are involved in gibberellin/abscisic acid signaling in barley aleurone cells. *Plant Physiol.* **158**: 1705--1714.
- Iyer N J, Tang Y, Mahalingam R. 2013. Physiological, biochemical and molecular responses to a combination of drought and ozone in *Medicago truncatula*. *Plant Cell Environ.* **36**: 706--720.
- Jacob P, Hirt H, Bendahmane A. 2017. The heat-shock protein/chaperone network and multiple stress resistance. *Plant Biotechnol J.* **15**: 405--414.
- Jiang J F, Li J G, Dong Y H. 2013. Effect of calcium nutrition on resistance of tomato against bacterial wilt induced by *Ralstonia solanacearum*. *Eur J Plant Pathol.* **136**: 547--555.
- Jiang Y, Huang B. 2001. Drought and heat stress injury to two cool-season turfgrasses in relation to antioxidant metabolism and lipid peroxidation. *Crop Sci.* **41**: 436--442.
- Jin R, Wang Y, Liu R, Gou J, Chan Z. 2015. Physiological and metabolic changes of Purslane (*Portulaca oleracea* L.) in response to drought, heat, and combined stresses. *Front Plant Sci.* **6**: 1123.
- Johnson S M, Lim F L, Finkler A, Fromm H, Slabas A R, Knight M R. 2014. Transcriptomic analysis of *Sorghum bicolor* responding to combined heat and drought stress. *BMC Genomics* **15**: 1--19.
- Juroszek P, Racca P, Link S, Farhumand J, Kleinhenz B. 2020. Overview on the review articles published during the past 30 years relating to the potential climate change effects on plant pathogens and crop disease risks. *Plant Pathol.* **69**: 179--193.
- Kang W H1, Sim Y M, Koo N, Nam J Y, Lee J, Kim N, Jang H, Kim Y M, Yeom S I. 2020. Transcriptome profiling of abiotic responses to heat, cold, salt, and osmotic stress of *Capsicum annuum* L. *Sci Data.* **7**: 17.
- Kissoudis C, van deWiel C, Visser R G F, van der Linden G. 2014. Enhancing crop resilience to combined abiotic and biotic stress through the dissection of physiological and molecular crosstalk. *Front Plant Sci.* **5**: 1--20.
- Kissoudis C, Sunarti S, van de Wiel C, Visser R G F, van der Linden C G, Bai Y. 2016. Responses to combined abiotic and biotic stress in tomato are governed by stress intensity and resistance mechanism. *J Exp Bot.* **67**: 5119--5132.
- Kocsy G, Galiba G, Brunold C. 2001. Role of glutathione in adaptation and signaling during chilling and cold acclimation in plants. *Physiol Plant.* **113**: 158--164.
- Kruse C, Jost R, Lipschis M, Kopp B, Hartmann M, Hell R. 2007. Sulfur-enhanced defence: effects of sulfur metabolism, nitrogen supply, and pathogen lifestyle. *Plant Biol. (Stuttgart)* **9**: 608--619.
- Leon-Kloosterziel K M, Gil M A, Ruijs G J, Jacobsen S E, Olszewski N E, Schwartz S H, Zeevaart J A, and Koornneef M. 1996. Isolation and characterization of abscisic acid-deficient Arabidopsis mutants at two new loci. *Plant J Cell Mol Biol.* **10**: 655--661.
- Li X, Yang Y, Sun X, Lin H, Chen J, Ren J, Hu X, Yang Y. 2014. Comparative physiological and proteomic analyses of poplar (*Populus yunnanensis*) plantlets exposed to high temperature and drought. *PLoS One.* **9**: e107605.
- Lim C W, Baek W, Jung J, Kim J H, Lee S C. 2015. Function of ABA in stomatal defense against biotic and drought stresses. *Int J Mol Sci.* **16**: 15251--15270.
- Liu X, Meng P, Yang G, Zhang M, Peng S, Zhai M Z. 2020. Genome-wide identification and transcript profiles of walnut heat stress transcription factor involved in abiotic stress. *BMC Genom.* **21**: 474.

- Liu Z, Qin J, Tian X, Xu S, Wang Y, Li H, Wang X, Peng H, Yao Y, Hu Z, Ni Z. 2018. Global profiling of alternative splicing landscape responsive to drought, heat and their combination in wheat (*Triticum aestivum* L.). *Plant biotechnol J*. **16**: 714--726.
- Lopez-Delacalle M, Silva C J, Mestre T C, Martinez V, Blanco-Ulate B, Rivero R M. 2021. Synchronization of proline, ascorbate and oxidative stress pathways under the combination of salinity and heat in tomato plants. *Environ Exp Bot*. **183**: 104351.
- Loreti E, van Veen H, Perata P. 2016. Plant responses to flooding stress. *Curr Opin Plant Biol*. **33**: 64--71.
- Luck J, Spackman M, Freeman A, Trebicki P, Griffiths W, Finlay K, Chakraborty S. 2011. Climate change and diseases of food crops. *Plant Pathol*. **60**: 113--121.
- Madgwick J, West J, White R, Semenov M, Townsend J, Turner J, Fitt BL. 2011. Impacts of climate change on wheat anthesis and fusarium ear blight in the UK. *Eur J Plant Pathol*. **130**: 117--131.
- Mahalingam R, Bregitzer P. 2019. Impact on physiology and malting quality of barley exposed to heat, drought and their combination during different growth stages under controlled environment. *Physiol Plant*. **165**: 277--289.
- Maher R L, Rice M M, McMinds R, Burkepile D E, Thurber R V. 2019. Multiple stressors interact primarily through antagonism to drive changes in the coral microbiome. *Sci Rep*. **9**: 6834.
- Mantri N, Patade V, Pang E C K. 2014. Recent advances in rapid and sensitive screening for abiotic stress. In Ahmad P, Wani M R, Azooz M M, Tran L S P (eds.) *Improvement of Crops in the Era of Climate Change*. Springer, New York. pp. 37--47.
- Martinez V, Mestre T C, Rubio F, Girones-Vilaplana A, Moreno D A, Mittler R, Rivero R M. 2016. Accumulation of flavonols over hydroxycinnamic acids favors oxidative damage protection under abiotic stress. *Front Plant Sci*. **7**: 838.
- Massa A N, Childs K L, Buell R. 2013. Abiotic and biotic stress responses in *Solanum tuberosum* group Phureja DM1-3 516 R44 as measured through whole transcriptome sequencing. *Plant Genome*. **6**: 1--10.
- Matsui A, Ishida J, Morosawa T, Mochizuki Y, Kaminuma E, Endo TA, Okamoto M, Nambara E, Nakajima M, Kawashima M, Satou M. 2008. *Arabidopsis* transcriptome analysis under drought, cold, high-salinity and ABA treatment conditions using a tiling array. *Plant Cell Physiol*. **49**: 1135--1149.
- Mittler R. 2006. Abiotic stress, the field environment and stress combination. *Trends Plant Sci*. **11**: 15--19.
- Mittler R, Blumwald E. 2010. Genetic engineering for modern agriculture: challenges and perspectives. *Annu Rev Plant Biol*. **61**: 443--462.
- Mittra B, Ghosh P, Henry S L, Mishra J, Das T K, Gosh S, Babu C R, Mohanty P. 2004. Novel mode of resistance to *Fusarium* infection by a mild dose pre-exposure of cadmium in wheat. *Plant Physiol Biochem*. **42**: 781--787.
- Moeder W, Ung H, Mosher S, Yoshioka K. 2010. SA-ABA antagonism in defense responses. *Plant Signal Behav*. **5**: 1231--1233.
- Mosher S, Moeder W, Nishimura N, Jikumaru Y, Joo S H, Urquhart W, Klessig D F, Kim S K, Nambara E, Yoshioka K. 2010. The lesion-mimic mutant cpr22 shows alterations in abscisic acid signaling and abscisic acid insensitivity in a salicylic acid-dependent manner. *Plant Physiol*. **152**: 1901--1913.
- Mota A P, Brasileiro A C, Vidigal B, Oliveira T N, Martins A D, de Passos Saraiva M A, de Araújo A C, Togawa R C, Grossi-de-Sá M F, Guimaraes P M. 2021. Defining the combined stress response in wild *Arachis*. *Sci Rep*. **27**: 1--2.
- Moustakas M, Sperdouli I, Kouna T, Antonopoulou C I, Therios I. 2011. Exogenous proline induces soluble sugar accumulation and alleviates drought stress effects on photosystem II functioning of *Arabidopsis thaliana* leaves. *Plant Growth Regul*. **65**: 315--325.
- Mustilli A C, Merlot S, Vavasseur A, Fenzi F and Giraudat J. 2002. *Arabidopsis* OST1 protein kinase mediates the regulation of stomatal aperture by abscisic acid and acts upstream of reactive oxygen species production. *Plant Cell*. **14**: 3089--3099.
- Narsai R, Wang C, Chen J, Wu J, Shou H, Whelan J. 2013. Antagonistic, overlapping and distinct responses to biotic stress in rice (*Oryza sativa*) and interactions with abiotic stress. *BMC Genomics*. **14**: 93.
- Natali L, Vangelisti A, Guidi L, Remorini D, Cotrozzi L, Lorenzini G, Nali C, Pellegrini E, Trivellini A, Vernieri P, Landi M. 2018. How *Quercus ilex* L. saplings face combined salt and ozone stress: a transcriptome analysis. *BMC Genomics*. **19**: 1--8.
- Nguyen D, D'Agostino N, Tytgat TO, Sun P, Lortzing T, Visser EJ, Cristescu SM, Steppuhn A, Mariani C, van Dam NM, Rieu I. 2016. Drought and flooding have distinct effects on herbivore - induced responses and resistance in *Solanum dulcamara*. *Plant Cell Environ*. **39**: 1485--1499.
- Niu M, Xie J, Chen C, Cao H, Sun J, Kong Q, Shabala S, Huang Y, Bie Z. 2018. An early ABA-induced stomatal closure, Na<sup>+</sup> sequestration in leaf vein and K<sup>+</sup> retention in mesophyll confer salt tissue tolerance in *Cucurbita* species. *J Exp Bot*. **69**: 4945--4960.
- Ohama N, Sato H, Shinozaki K, Yamaguchi-Shinozaki K. 2017. Transcriptional regulatory network of plant heat stress response. *Trends Plant Sci*. **22**: 53--65.

- Oide S, Bejai S, Staal J, Guan N, Kaliff M, Dixelius C. 2013. A novel role of PR2 in abscisic acid (ABA) mediated, pathogen-induced callose deposition in *Arabidopsis thaliana*. *New Phytol.* **200**: 1187--1199.
- Osthoﬀ A, Rose P D, Baldauf J, Piepho H P, Hochholding F. 2019. Transcriptomic reprogramming of barley seminal roots by combined water deficit and salt stress. *BMC Genomics.* **20**: 325.
- Pandey P, Irulappan V, Bagavathiannan M V, Senthil-Kumar M. 2017. Impact of combined abiotic and biotic stresses on plant growth and avenues for crop improvement by exploiting physio-morphological traits. *Front Plant Sci.* **8**: 537.
- Pandey P, Ramegowda V, Senthil-Kumar M. 2015. Shared and unique responses of plants to multiple individual stresses and stress combinations: physiological and molecular mechanisms. *Front Plant Sci.* **6**: 1--14.
- Park J S, Lee E J. 2019. Waterlogging induced oxidative stress and the mortality of the Antarctic plant, *Deschampsia antarctica*. *J Ecol Environ.* **43**: 29.
- Perez-Lopez U, Miranda-Apodaca J, Munoz-Rueda A, Mena-Petite A. 2013. Lettuce production and antioxidant capacity are differentially modified by salt stress and light intensity under ambient and elevated CO<sub>2</sub>. *J Plant Physiol.* **170**: 1517--1525.
- Pieterse C M, Leon-Reyes A, Van der Ent S, Van Wees S C. 2009. Networking by small-molecule hormones in plant immunity. *Nature Chem Biol.* **5**: 308--316.
- Potato Council News. 2011. Available at: <http://www.potatopro.com/news/2011/continuing-drought-increases-common-scab-threat-uk-potato-growers> (verified on September 19, 2021).
- Prasad P V V, Pisipati S R, Momcilovic I, Ristic Z. 2011. Independent and combined effects of high temperature and drought stress during grain filling on plant yield and chloroplast EF-Tu expression in spring wheat. *J Agron Crop Sci.* **197**: 430--441.
- Prasch C M, Sonnewald U. 2013. Simultaneous application of heat, drought, and virus to *Arabidopsis* plants reveals significant shifts in signaling networks. *Plant Physiol.* **162**: 1849--1866.
- Rafique S, Abdin M Z, Alam W. 2019. Response of combined abiotic stresses on maize (*Zea mays* L.) inbred lines and interaction among various stresses. *Maydica.* **64**: 1--8.
- Ramegowda V, Senthil-Kumar M. 2015. The interactive effects of simultaneous biotic and abiotic stresses on plants: Mechanistic understanding from drought and pathogen combination. *J Plant Physiol.* **176**: 47--54.
- Rasheed R, Iqbal M, Ashraf M A, Hussain I, Shafiq F, Yousaf A, Zaheer A. 2018. Glycine betaine counteracts the inhibitory effects of waterlogging on growth, photosynthetic pigments, oxidative defence system, nutrient composition, and fruit quality in tomato. *J Horticult Sci Biotechnol.* **93**: 385--391.
- Rasmussen S, Barah P, Suarez-Rodriguez M C, Bressendorff S, Friis P, Costantino P, Bones A M, Nielsen H B, Mundy J. 2013. Transcriptome responses to combinations of stresses in *Arabidopsis*. *Plant Physiol.* **161**: 1783--1794.
- Rejeb I B, Pastor V, Mauch - Mani B. 2014. Plant responses to simultaneous biotic and abiotic stress: molecular mechanisms. *Plants.* **3**: 458--475.
- Rejeb I B. 2018. BABA in priming tomato for enhanced tolerance to drought, salinity and fungal stress and combinations thereof. D.Sc thesis, University of Neuchâtel, CH pp. 81.
- Rivero R M, Mestre T C, Mittler R, Rubio F, Garcia-Sanchez F, Martinez V. 2013. The combined effect of salinity and heat reveals a specific physiological, biochemical and molecular response in tomato plants. *Plant Cell Environ.* **37**: 1059--1073.
- Rizhsky L, Liang H, Shuman J, Shulaev V, Davletova S, Mittler R. 2004. When defense pathways collide. The response of *Arabidopsis* to a combination of drought and heat stress. *Plant Physiol.* **134**: 1683--1696.
- Rizhsky L, Liang H, Mittler R. 2002. The combined effect of drought stress and heat shock on gene expression in tobacco. *Plant Physiol.* **130**: 1143--1151.
- Saeed M, Noshin I, Abida A, Naveed R, Roomina M, Fatima B, Nazima B. 2016. Effect of drought stress on *Brassica* crops and its mitigation by inoculation of PGPR. *Inter J Biosci.* **9**: 282--291.
- Sarma R K, Saikia R. 2014. Alleviation of drought stress in mung bean by strain *Pseudomonas aeruginosa* GGRJ21. *Plant Soil.* **377**: 111--126.
- Sattar A, Sher A, Ijaz M, Ul-Allah S, Rizwan MS, Hussain M, Jabran K, Cheema M A. (2020) Terminal drought and heat stress alter physiological and biochemical attributes in flag leaf of bread wheat. *PLoS ONE.* **15**: e0232974.
- Sehgal A, Sita K, Kumar J, Kumar S, Singh S, Siddique KHM, Nayyar H. 2017. Effects of drought, heat and their interaction on the growth, yield and photosynthetic function of lentil (*Lens culinaris* medikus) genotypes varying in heat and drought sensitivity. *Front Plant Sci* **8**: 1776.
- Sewelam N, Brilhaus D, Bräutigam A, Alseekh S, Fernie A R, Maurino V G. 2020. Molecular plant responses to combined abiotic stresses put a spotlight on unknown and abundant genes. *J Exp Bot.* **71**: 5098--5112.
- Shaar-Moshe L, Blumwald E, Peleg Z. 2017. Unique physiological and transcriptional shifts under combinations of salinity, drought, and heat. *Plant Physiol.* **174**: 421--434.
- Shaik R, Ramakrishna W. 2013. Genes and co-expression modules common to drought and bacterial stress responses in *Arabidopsis* and rice. *PLoS ONE.* **8**: e77261.

- Sham A, Al-Azzawi A, Al-Ameri S, Al-Mahmoud B, Awwad F, Al-Rawashdeh A, Iratni R, AbuQamar S. 2014. Transcriptome analysis reveals genes commonly induced by *Botrytis cinerea* infection, cold, drought and oxidative stresses in *Arabidopsis*. *PLoS ONE*. **25**: e113718.
- Sham A, Moustafa K, Al-Ameri S, Al-Azzawi A, Iratni R, AbuQamar S. 2015. Identification of *Arabidopsis* candidate genes in response to biotic and abiotic stresses using comparative microarrays. *PLoS ONE*. **10**: e0125666
- Shao H, Chu L, Shao M, Jaleel C A, Mi H. 2008. Higher plant antioxidants and redox signaling under environmental stresses. *C R Biol*. **331**: 433--441.
- Sharma R C, Duveiller E, Ortiz-Ferrara G. 2007. Progress and challenge towards reducing wheat spot blotch threat in the Eastern Gangetic Plains of South Asia: is climate change already taking its toll? *Field Crop Res*. **103**: 109--118.
- Silva E N, Vieira S A, Ribeiro R V, Ponte L F A, Ferreira-Silva S L, Silveira J A G. 2013. Contrasting physiological responses of *Jatropha curcas* plants to single and combined stresses of salinity and heat. *J Plant Growth Regul*. **32**: 159--169.
- Sinha R, Gupta A, Senthil-Kumar M. 2017. Concurrent drought stress and vascular pathogen infection induce common and distinct transcriptomic responses in chickpea. *Fron Plant Sci*. **8**: 333.
- Stork N E. 2018. How many species of insects and other terrestrial arthropods are there on earth? *Annu Rev Entomol*. **63**: 31--45.
- Suzuki N, Bassil E, Hamilton J S, Inupakutika M A, Zandalinas S I., Tripathy D, Luo Y, Dion E, Fukui G, Kumazaki A, Nakano R, Rivero R M, Verbeck G F, Azad R K, Blumwald E, Mittler R. 2016. ABA is required for plant acclimation to a combination of salt and heat stress. *PLoS ONE*. **11**: e0147625.
- Suzuki N, Rivero R M, Shulaev V, Blumwald E, Mittler R. 2014. Abiotic and biotic stress combinations. *New Phytol*. **203**: 32--43.
- Szabados L, Savaure A. 2010. Proline: a multifunctional amino acid. *Trends Plant Sci*. **15**: 89--97.
- Tanou G, Molassiotis A, Diamantidis G. 2009. Hydrogen peroxide and nitric oxide-induced systemic antioxidant prime-like activity under NaCl-stress and stress-free conditions in citrus plants. *J Plant Physiol*. **166**: 1904--1913.
- Tarafdar A, Rani T S, Chandran U S S, Ghosh R, Chobe D R, Sharma M. 2018. Exploring combined effect of abiotic (soil moisture) and biotic (*Sclerotium rolfsii* Sacc.) stress on collar rot development in chickpea. *Front Plant Sci*. **9**: 1154.
- Tossi V, Lamattina L, Jenkins G I, Cassia R O. 2014. Ultraviolet-B-induced stomatal closure in *Arabidopsis* is regulated by the UV RESISTANCE LOCUS8 photoreceptor in a nitric oxide-dependent mechanism. *Plant Physiol*. **164**: 2220--2230.
- Vardharajula S, Zulfikar Ali S, Grover M, Reddy G, Bandi V. 2011. Drought-tolerant plant growth promoting *Bacillus* spp.: effect on growth, osmolytes, and antioxidant status of maize under drought stress. *J Plant Interact*. **6**: 1--14.
- Vaumourin E, Laine A L. 2018. Role of temperature and coinfection in mediating pathogen Life-History traits. *Front Plant Sci*. **9**: 1670.
- Venegas-Molina J, Proietti S, Pollier J, Orozco-Freire W, Ramirez-Villacis D, Leon-Reyes A. 2020. Induced tolerance to abiotic and biotic stresses of broccoli and *Arabidopsis* after treatment with elicitor molecules. *Sci Rep*. **10**: 10319.
- Vile D, Pervent M, Belluau M, Vasseur F, Bresson J, Muller B, Granier C, Simonneau T. 2012. *Arabidopsis* growth under prolonged high temperature and water deficit: independent or interactive effects? *Plant Cell Environ*. **35**: 702--718.
- Vuleta A, Jovanovic S M, Tucic B. 2015. How do plants cope with oxidative stress in nature? A study on the dwarf bearded iris (*Iris pumila*). *Acta Physiol Plant*. **37**: 1711--1719.
- Wang L, Liu L, Ma Y, Li S, Dong S, Zu W. 2018. Transcriptome profiling analysis characterized the gene expression patterns responded to combined drought and heat stresses in soybean. *Comp Biol Chem*. **77**: 413--429.
- Weise J, Kranz T, Schbert S. 2004. Induction of pathogen resistance in barley by abiotic stress. *Plant Biol*. **6**: 529--536.
- Xia X J, Wang Y J, Zhou Y H, Tao Y, Mao W H, Shi K, Asami T, Chen Z, Yu J Q. 2012. Reactive oxygen species are involved in brassinosteroid-induced stress tolerance in cucumber. *Plant Physiol*. **158**: 1034--1045.
- Zandalinas S I, Balfagón D, Arbona V, Gómez-Cadenas A, Inupakutika M A, Mittler R. 2016a. ABA is required for the accumulation of APX1 and MBF1c during a combination of water deficit and heat stress. *J Exp Bot*. **67**: 5381--5390.
- Zandalinas S I, Fritschi F B, Mittler R. 2020. Signal transduction networks during stress combination. *J Exp Bot*. **71**: 1734--1741.
- Zandalinas S I, Rivero R M, Martínez V, Gómez-Cadenas A, Arbona V. 2016b. Tolerance of citrus plants to the combination of high temperatures and drought is associated to the increase in transpiration modulated by a reduction in abscisic acid levels. *BMC Plant Biol*. **16**: 105.
- Zhang F, Zhang F, Huang L, Vera Cruz C, Ali J, Xu J, Zhou Y, Li Z. 2016. Overlap between signalling pathways responsive to *Xanthomonas oryzae* pv. *oryzae* infection and drought stress in rice introgression line revealed by RNA-Seq. *J Plant Growth Regul*. **35**: 345--356.
- Zhang H, Sonnwald, U. 2017. Differences and commonalities of plant responses to single and combined stresses. *Plant J*. **90**: 839--855.

- Zhang W, Xie W Z, Zhang X, Lang D, Zhang X. 2019. Growth-promoting bacteria alleviates drought stress of *G. uralensis* through improving photosynthesis characteristics and water status. *J Plant Interac.* **14**: 580--589.
- Zhao Y, Gao J, Im Kim J, Chen K, Bressan R A, Zhu J K. 2017. Control of plant water use by ABA induction of senescence and dormancy: an overlooked lesson from evolution. *Plant Cell Physiol.* **58**: 1319--1327.
- Zheng C, Wang Y, Ding Z, Zhao L. 2016. Global transcriptional analysis reveals the complex relationship between tea quality, leaf senescence and the responses to cold-drought combined stress in *Camellia sinensis*. *Front Plant Sci.* **7**: 1858.
- Zhou R, Kong L, Wu Z, Rosenqvist E, Wang Y, Zhao L, Zhao T, Ottosen C O. 2019. Physiological response of tomatoes at drought, heat and their combination followed by recovery. *Physiol Plant.* **165**: 144--154.