

# The Microbially Extended Phenotype of Plants, a Keystone against Abiotic Stress

Mario X. Ruiz-González<sup>1\*</sup>, Oscar Vicente<sup>1</sup>

## Abstract

**Background:** Climate change affects every region across the globe with heterogeneous effects on local temperatures and precipitation patterns. In plants, sessile organisms, climate change imposes more drastic effects leading to loss of yield or even death. However, plants establish mutualistic interactions with microorganisms that boost plant tolerance against abiotic stresses or strengthen the plant immune system against pathogens, thus, enhancing their survival and fitness. Moreover, in the wild, microbial endophytes provide important ecosystem services.

**Purpose and scope:** Little we know about the mechanisms of response against the adverse effects of climate change on natural populations of wild plants and even less about the potential role played by microbial biostimulants. In this article, we review the effects of biostimulants on plant responses against abiotic stresses, with a particular focus on the role of mycorrhizas and leaf endophytes.

**Results:** We have reviewed the effects of the main abiotic stresses in plants, the mechanisms that plants use to face these abiotic challenges, and the interaction plant-biostimulant-abiotic stress, highlighting the primary responses and parameters to evaluate different plant responses.

**Conclusion:** Abiotic stresses can check the phenotypic plasticity of plants and also trigger a complex and heterogeneous array of responses to face different abiotic stresses, and beneficial microorganisms do play an essential role in enhancing such responses. Our laboratory has initiated a project to characterise microbial populations associated with plants from wild areas and analyse their potential role in aiding the plants to cope with abiotic stresses.

**Keywords:** abiotic stress, climate change, phenotypic plasticity, endophyte, mutualism

## Introduction

All organisms are exposed to challenging environmental conditions, whether of abiotic or biotic origins. Those challenges represent the driving force that sorts biologically successful individuals from the populations; that is, those individuals that will reproduce. This process results either in the specialisation of populations to local conditions for particular ecological niches or in generalist strategies, both with important consequences for diversity (1). However, a third evolutionary outcome might occur: when environmental factors change faster than the potential of a species to produce phenotypes able to survive and reproduce in the new conditions, extinction happens. When the extinction rate is hundreds of times quicker than expected, we call the event a mass extinction. The planet and its biodiversity have faced several such episodes of drastically changing environmental conditions (2,3). Moreover, nowadays, there is strong evidence supporting that we are facing a new mass extinction event driven by human activities (4) and strongly accelerated by drastic changes in the abiotic conditions (5). Indeed, some physical parameters have changed drastically since the industrial revolution. For example, the temperature has risen 1.1 °C and is expected to rise above 1.5 °C during the next two decades, as the risk of flooding and heatwaves (5). The effects of climate change cascade from genes to ecosys-

<sup>1</sup> Institute for Conservation and Improvement of Valencian Agrodiversity, Universitat Politècnica de València, Camino de Vera s/n, Valencia 46022, Spain

\* **Corresponding author:**

Mario X. Ruiz-González  
Email: maruigon@upvnet.upv.es  
Orcid ID: 0000-0003-3654-773X

DOI: 10.2478/ebtj-2022-0017

© 2022 Authors. This work was licensed under the Creative Commons Attribution-NonCommercial-NoDerivs 3.0 License.

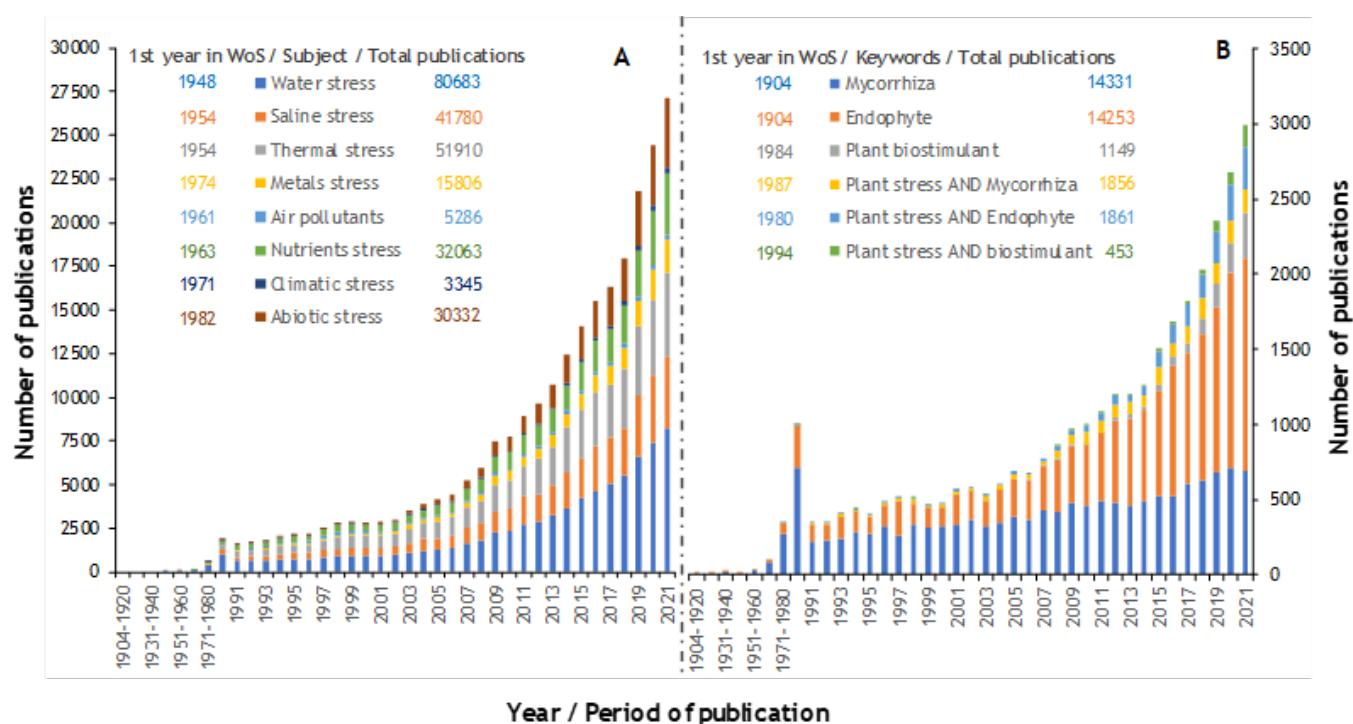
tems with highly heterogeneous but severe impacts on all levels of biological organisation (6).

## 1. Abiotic stresses in plants

Plants are strongly dependent upon the abiotic factors of the environment and prone to suffer the consequences of changing conditions. Thus, abiotic stress is any adverse environmental factor that negatively affects plant fitness, e.g., by restricting growth or productivity. Abiotic factors become stressors depending on their intensity: low and high temperatures, water availability (drought and flood), salinity, metal toxicity, UV,

pH, nutrient availability... Many, if not all, of these stressors can happen due to natural processes or triggered by anthropogenic activities.

The investigation of the effects of abiotic stress on plants has been a research topic for many decades. The oldest and more active fields focused on the effects of moisture and salts on some morphological parameters [since the 1900s; e.g. (7)]. However, as shown in 2022 Clarivate Web of Science®, it was in the 1940s when researchers started using the term stress, first for water stress, followed by the thermal and saline stresses since the 1950s, and a decade later, the nutrient stress, or



**Figure 1.** Number of publications per year related to plant stress and microorganisms in Web of Science. A) Publications on different plant abiotic stresses. B) Publications on mycorrhiza, endophytes, or biostimulants, and their role on plant stress. Legends show the first year a publication on that subject appeared in WoS and the total publications up to 2021. Data included herein are derived from 2022 Clarivate Web of Science®. Keywords to select for publications on plant abiotic stresses: 1) water stress: plant water stress OR plant moisture stress OR plant drought stress; 2) Saline stress: plant saline stress OR plant salt stress OR plant osmotic stress; 3) Thermal stress: plant thermal stress OR plant freezing stress OR plant ice stress OR plant cold stress OR plant heat stress OR plant chilling stress OR plant low temperature stress OR plant high temperature stress; 4) Metals stress: Plant iron stress Fe OR plant cadmium stress Cd OR plant aluminium stress Al OR plant mercury stress Hg OR plant heavy metals stress OR plant nickel stress Ni OR plant copper stress Cu OR plant lead stress Pb OR plant Cobalt stress Co OR plant zinc stress Zn OR plant manganese stress Mn; 5) Air pollutants: plant air pollution stress OR plant carbon dioxide stress CO<sub>2</sub> OR plant oxygen stress O<sub>2</sub> OR plant hydrogen stress H<sub>2</sub> OR plant ozone stress; 6) Nutrients stress: plant nutrient stress OR plant nitrogen stress OR plant nitrate stress OR plant NO<sub>2</sub> stress OR plant phosphorus stress P OR plant potassium stress K; 7) Climatic stress: plant climatic stress. Although some related publications shall be lost in the search, we decided to use 'plant' as a keyword followed by the stress descriptors to avoid overcounting those papers on organisms other than plants. Moreover, because 'plant' and 'stress' are terms used in engineering and mechanical research, we eliminated from the search those publications with the following keywords in all fields: power, equipment, nuclear plant, heating plant, vibrations plant, insulated plant, turbine, welding, pipes, shearing. Finally, some research used plant extracts to analyse animal stress responses; thus, we also restricted the search by eliminating those papers with 'rats', 'echinoderm', 'crustacea' or '*Caenorhabditis*' as a keyword.

other stresses (Fig 1 A). Since the 1981-1990 decade, heterogeneous research on the many different abiotic stresses has risen exponentially. Nowadays, studying plant responses to abiotic stress has become a very active research topic. Initially, it was motivated by mitigating crop loss and boosting productivity (8). However, investigating the mechanisms underlying the response of plants to abiotic stress has become a priority since then (9). Many of the publications researching the effects of water, salt, thermal, and heavy metals stresses have focused on diverse molecular or morphologic parameters in the model plant *Arabidopsis thaliana* and a diverse array of crop species, including grasses (e.g., *Avena* sp., *Oryza sativa*, *Triticum* sp., or *Zea mays*), legumes (e.g., *Cicer arietinum*, *Lens culinaris*, or *Phaseolus* sp.), vegetables (e.g., *Brassica* sp., *Capsicum* sp., or *Solanum* sp.), fruits (e.g., *Citrus* sp., *Fragaria* sp., or *Rubus* sp.), and commodities such as *Coffea* sp., *Gossypium hirsutum*, or *Theobroma cacao*, amongst others. These studies have led to a good knowledge of the basic, general molecular mechanisms of plant responses to abiotic stress (e.g., 8,10).

### 1.1 Temperature

Temperature can stress plants by both extreme heat and cold (chilling and freezing) by altering the fluidity and stability of the phospholipid membranes. As a general response to different stresses, such as mechanical stimulation, pathogen infection, osmotic stress, salt, hydrogen peroxide and too low or too high temperatures, free  $\text{Ca}^{2+}$  concentration increases in the cytosol after modification of the  $\text{Ca}^{2+}$ -permeable channels; thus, acting as a stimulus-specific second messenger (11). Under cold conditions, the  $\text{Ca}^{2+}$ -permeable transporter ANNEXIN1 (AtANN1) mediates plant tolerance to freezing by regulating COR (cold-regulated) gene expression (12). Moreover, the temperature can also modify the stability of proteins by triggering different responses to cold and heat (13). For example, the phytochrome B (phyB) photoreceptor, which participates in temperature perception, can reverse its activity from the active to the inactive state, thus acting as a temperature sensor (14,15). High temperatures also trigger the transcriptional repressor ELF3. Low temperatures can be sensed by membrane  $\text{Ca}^{2+}$  regulator channels and the accumulation of CBF transcription factors to protect the photoreceptor phyB (13,16). Both cold and heat affect the membrane's fluidity, protein folding, and RNA secondary structures, but only high temperatures can damage proteins by denaturation. The latter triggers the overexpression of a different protective mechanism, the synthesis of heat shock proteins (HSPs). There exist five major families of HSPs: HSP100 (Clp proteins), HSP90 (linked to denaturing stress), HSP70 (its expression can be cold and heat stress-induced), small hsp (ATP-independent activity), and chaperonins type I and type II (16). It is noteworthy to mention that the subset of overexpressed heat-induced proteins under high temperatures mirrors the aminoacidic composition of thermophilic organisms, suggesting adaptation to temperature (17). Under temperature stress, plants also produce regulatory factors: heat shock factors (HSFs), dehydration response ele-

ment binding (DREB) proteins, and both transcription (with WRKY domains) and multiprotein binding factors (8,16,18). A process unique to plants consists of the accumulation of complexes made of hsp aggregates, known as heat stress granules (HSGs), which might play a protective and storage role for housekeeping mRNPs (16). Reducing trienoic fatty acids (TA) in chloroplast membranes promotes thermotolerance, possibly because its peroxidation produces cytotoxic compounds, such as ACR and MVK (18). Moreover, high temperatures can, depending on the plant species, suppress seed germination, induce severe necrotic symptoms, accelerate anthesis and maturation but with reduced yield, boost the production of potentially toxic electrophilic compounds, and severely affect fertility and grain quality by downregulating sucrose metabolism and upregulating starch degradation (18). More protective mechanisms against heat are the presence of antioxidants scavenging ROS, the production of protectants such as glycine betaine (GB), or trehaloses; all of the latter can induce thermotolerance (18). In addition, it has been shown the protective role of phytohormones such as abscisic acid (ABA) in cold, salt, and drought stresses, and the brassinosteroid action in increasing thermotolerance in plants (16).

### 1.2 Salinity

Soil salinisation drastically alters soil physicochemical properties, soil respiration activity, and microbial diversity; that is, natural cycles (biological, biochemical, hydrological, and erosional), affecting ecosystem services and food security by decreasing natural resources, goods, and agricultural production (19). Thus, soil salinisation is the most detrimental abiotic stress (20). Salinity can occur in two ways: 1) naturally or primarily salinity, as a result of weathering or as deposition of sea salts in natural soil deposits and coastal areas; and 2) as a by-product of human activity or secondary salinity due to agricultural and irrigation activities (21). Soil salinity can be classified depending on the electrical conductivity of a soil saturation extract: non-saline (0-2  $\text{dS m}^{-1}$ ), with negligible effects on plants; slightly saline (2-4  $\text{dS m}^{-1}$ ), which restricts the more sensitive plants; moderately saline (4-8  $\text{dS m}^{-1}$ ), which affects many plant species and crops; strongly saline (8-16  $\text{dS m}^{-1}$ ), where only tolerant plants prosper; and, very strongly saline (> 16  $\text{dS m}^{-1}$ ), where a few very tolerant species reproduce (22). Salinity affects the ability of plants to acquire water from the soil. The accumulation of  $\text{Na}^+$  induces ionic stress, which inhibits photosynthesis and enzymatic activities, and induces senescence, in addition to hyperosmotic stress, which inhibits water uptake, and decreases plant growth (23). Furthermore, photosystem II (PSII) inhibition negatively affects leaf chlorophyll contents, leaf area and photosynthetic ability. Then, soil salinity further affects the mobilisation of nutrients, induces plant hormonal imbalance and the production of ROS, and reduces the influx of  $\text{K}^+$  (24). Moreover, salinity severely affects soil microbial diversity and health.

Plants have three primary mechanisms for promoting salt tolerance: ion exclusion, osmotic tolerance, and tissue toler-

ance (25). The former occurs in the roots and consists of the transport of Na<sup>+</sup> and Cl<sup>-</sup> to avoid their accumulation at toxic levels; ions can be stored in vacuoles after being removed from the xylem and/or expelled back to the soil. Several gene families have been identified as playing a role in the improvement of salt tolerance: the HKT (high-affinity potassium transporter), Na<sup>+</sup>/H<sup>+</sup> antiporters SOS (salt overly sensitive pathway) and SOD<sub>2</sub> (Superoxide Dismutase 2), and the Na<sup>+</sup> ENA ATPases (25). Tissue tolerance arises through different strategies based on proton pumps or transporters like Na<sup>+</sup>/H<sup>+</sup> antiporters NHX and nhaA or a vacuolar H<sup>+</sup> pyrophosphatase. Another strategy is the accumulation of compatible solutes (trehalose-6-phosphate synthase (TPS), trehalose-6-phosphate phosphatase (TPP), mannitol-1-phosphate dehydrogenase (mt1D), L-myo-Inositol-1-phosphate synthase (MIP), betaine aldehyde dehydrogenase (BADH), choline oxidase/dehydrogenase (codA/betA), Δ1-pyrroline-5-carboxylate synthetase (P5CS) or myo-inositol O-methyltransferase). A third mechanism consists of the degradation of reactive oxygen species (ROS), catalysed by antioxidant enzymes such as ascorbate peroxidase (APX), glutathione S-transferase (GST), superoxide dismutase monodehydroascorbate reductase (MDR), or catalase (25). However, sensing and signalling mechanisms for osmotic tolerance in plants are yet unknown.

### 1.3 Drought

Drought is a major abiotic stress due to its vast potential impact worldwide at the ecological and economic levels. For example, 80 % of agricultural production comes from rainfed croplands (26). Drought induces hyperosmotic stress, as does salinity. Drought produces drastic adverse effects on plant germination, growth, biomass, productivity, and phenology, due to reduced water uptake, deficient enzyme activities, and reduced energy supply (27). Moreover, drought severely affects plant transpiration, stomatal conductance, the photosynthesis rate, the contents of photosynthetic pigments, and the efficiency of PSII (8,28). The OSCA1 (hyperosmolarity-gated calcium channel) family is potentially involved in sensing hyperosmotic stress (13). Some plant responses to promote drought tolerance are similar to those described above for salinity. However, we found an accumulation of osmoprotective solutes such as proline, increased total soluble sugars and amino acids, and higher catalase activity (28). Under drought stress, plants also activate a set of late embryogenesis abundant (LEA) proteins that act as water-binding molecules in membrane and macromolecule stabilisation and ion sequestration; thus, protecting the cells due to their biochemical properties, such as hydrophilicity and glycine content, presence of randomly coiled α-helices, and repeating motifs (29).

## 2. Plants weaponry against abiotic stresses

### 2.1 Biochemical responses for homeostasis

Plants have evolved different strategies to cope with stressful abiotic conditions. The first strategy we can think off is the interconnected complex set of regulatory pathways of molecular

responses that plants activate to enhance their tolerance to hostile abiotic factors in time. Five groups of biochemical responses have been described (10): 1) for protection at the cuticle level, there is overexpression of fatty acid synthases, transporters, and alcohol forming reductases; 2) for the modulation of the membranes, the plant triggers the overexpression of fatty acid desaturases, acyltransferases, enzyme cofactor acyl-carrier proteins (ACPs); 3) to boost the homeostasis of ROS, there is overexpression of peroxidases, glutathione S-transferases (GSTs), and activation of other ROS scavengers; 4) at the protein level, some proteins families such as the heat shock proteins are overexpressed (HSPs); and 5) osmoprotection through the production of compatible solutes such as glycine betaine (GB), proline (Pro), sugars, overexpression of galactinol synthases (GOLSs) or proline dehydrogenases (ProDHs). We have seen in the previous section how different stressors relate to the activation of some of these responses.

A second mechanism for protection against abiotic stressors is that plants can express many important morphological and physiological characters differently, depending on the environmental conditions. That is, the same genotype expresses different phenotypes in different environments, which might be considered an adaptive strategy (30). Finally, plants establish a broad array of fine-tuned biotic interactions with microorganisms than enhance the host performance: growth, tolerance against abiotic stress, and resistance against biotic threats.

### 2.2 Phenotypic plasticity and phenology: plants and ecosystems

Boyer (9) suggested that the adaptation of plants to adverse environments could significantly contribute to agriculture. This requires the study of plants adaptation under different environmental conditions, but very few works have studied the responses to abiotic stresses in wild plants and even fewer in their natural habitats. However, the responses of wild plants living in naturally occurring stressful habitats must be very efficient and might render of particular interest to gain insight on how to boost crops or wild species facing new environmental conditions (31).

Plants, as sessile organisms, cannot escape from the environmental conditions and their changes across time, which is particularly drastic under the current climate change scenarios. However, plants are endowed with the ability of phenotypic plasticity, that is, the alteration of morphological and physiological characters in response to environmental changes (32). The study of the phenotypic plasticity of plants, which started at the end of the 19<sup>th</sup> century (e.g., 33), is pivotal in predicting their fate in different environments. Changes in phenotypic characters might produce two non-exclusive effects: on the one hand, plant responses might boost the individual fitness or its chance for survival in the new conditions (e.g., might help invasive species to prosper in alien environments); on the other hand, however, they might have negative consequences for the biotic interactions, with an impact on the community composition (32,34,35). Many plants' functional traits can

show plasticity: morphological, physiological, reproductive, phenological, etc. One important and reliable bioindicator of the effects of climate on plants is their phenology; it can be measured following the BBCH scales, which have become a powerful tool (36,37). A strongly confirmed result is the tendency for many phenophases or phenological traits to occur earlier each year, which is positively correlated with the annual temperatures, and a tendency for spring events to change more than autumn events (36,38-40). However, the phenological responses might have different ecological impacts, with annual or insect-pollinated plants flowering earlier than perennials or wind-pollinated plants; the species-specific responses must be determined before extrapolating conclusions across geographical areas (39,41).

The investigation of the response of wild plants to abiotic stress is even more important if we think that plants are primary producer species and, often, are keystone species; that is, without them, the ecosystem may not exist (42). Indeed, human well-being depends directly on biodiversity, i.e., the variability of all life forms on Earth, their genes and the ecosystems in which they live. Moreover, the quality of human life is directly related to the health of ecosystems, which in turn depends on the richness of native species (43). Indeed, the sustainability of humans in the biosphere depends on the goods and services derived from ecosystem functions, the valuation of which is fundamental for decision-making and conservation policies and strategies (44). For example, coastal dune and saltmarsh ecosystems provide the most valuable ecosystem services (over 12,000 € per ha): shoreline protection, disturbance regulation (erosion), nutrient cycling, carbon sequestration, water purification, biological control, habitat/refuge, food production, raw materials, and tourism, recreation, culture, education and research (44-46). In the face of increasing risks due to the effects of climate change, some ecosystem services, such as carbon storage by wetlands, take on unique value (46). Although the many ecosystem services these habitats provide are well known, they are globally at risk due to, e.g., agriculture, eutrophication, or land use (47,48).

All natural habitats are threatened by either direct climate change effects or human activities; however, some ecosystems already suffer extreme abiotic conditions: high salinity environments or evaporites soils ecosystems, such as halite (NaCl), gypsum ( $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$ ) or anhydrite ( $\text{CaSO}_4$ ) and arid and semi-arid zones, or permafrost habitats (31,49). These ecosystems host species that display tolerant behaviours to such stressful conditions. Many of them might represent important sources of genetic innovations to promote adaptation to abiotic stresses in cattle or crops (50,51).

### 2.3 Plants and their mutualist biotic interactions

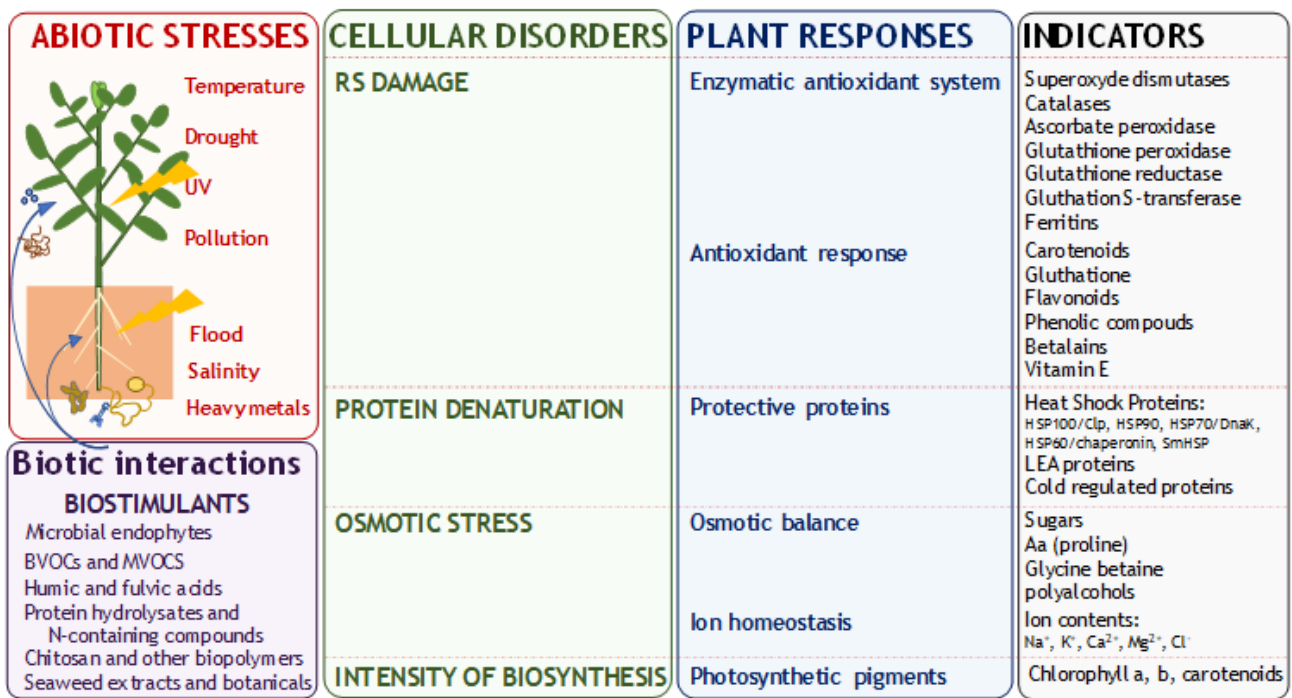
Wild plants represent a complex arena of multipartite interactions in the ecosystem. All possible types of biological interactions occur in plants: predation/herbivory, competition/facilitation (allelopathy), commensalism, parasitism, and mutualism. Albeit plants interact with a broad range of organisms

(other plants, vertebrates, invertebrates, protozoans, fungi, bacteria, and viruses), many of the biotic interactions that the plants establish are with microorganisms. A pivotal interaction for plants and ecosystem ecological and evolutionary dynamics is also established between the plant host and the so-called endophytes, usually non-pathogenic microorganisms (bacteria and fungi). Endophytes invade internal plant tissues such as roots (rhizospheric endophytes), stems or leaves (phyllospheric endophytes); they either remain asymptomatic for long periods or with limited pathogenic effects and often provide important advantages to the plant hosts (52,53). Moreover, well-known examples of endophytes are mycorrhizal fungi, diazotrophic bacteria, and phyllosphere endophytes. Thus, more than 80 % of the plants establish mutualistic interactions with mycorrhizal fungi, an association dating back to 460 Mya that directly affects plant biodiversity and ecosystem properties (54-56). Moreover, these fungi can be found in all major ecosystems (57).

Microbe-plant interactions were firstly described during the 19th century (58). Fungi associated with plants were the first microbial endophyte-plant interaction to be described by Heinrich F. Link in 1809, as species living and fruiting sub-epidermically in either living or dead plants (59). Later, the characterisation of lichens as the resulting association of fungi and alga led to coining of the term "symbiosis" by Albert B. Frank in 1877. Soon afterwards, Franz Kamienski, in 1882, described the association between fungi and plant roots (60), and A. B. Frank in 1885, defined the morphological organ arising after the union of both organisms as a mycorrhiza (61). In 1866, Mikhail S. Woronin (62) described the presence of endophytic bacteria in root nodules. Later in 1888, Martinus W. Beijerinck was able to isolate and culture root nodule bacteria. Recent findings unveiled the existence of beneficial viruses in the newly described multipartite interactions plant-fungus-virus (63,64). Therefore, it is not surprising to find research on microbial endophytes and mycorrhizae as early as 1904 in the literature registered on the Web of Science (<https://www.webofscience.com/wos/woscc/basic-search>), with an exponential increase in the number of publications since the 1960s (Fig. 1B). It was after the 1980s that researchers started investigating the role of microorganisms in mitigating the effects of stress on plants. Moreover, a new concept was born in that decade: the biostimulant.

### 3. Biostimulants and plant response to abiotic stresses

In addition to the classical genetic innovation, plants display an "extended phenotype" to face environmental conditions and maximise their fitness, provided by their microbial associates. This association represents a vital help in boosting the response of wild plants and crops to abiotic stresses (65). However, the definition of plant biostimulants (PBS) encompasses everything that improves plant health, boosts growth and yield, and enhances nutrient uptake and protection against abiotic and biotic stresses. Thus, we find active substances such as humic and fulvic acids, compounds rich in nitrogen, protein hydrolysates,



**Figure 2.** Plant cellular disorders triggered by abiotic stress, plant responses and their indicators, and potential biostimulants to boost plant health. Some indicators easy to measure in the laboratory to provide insight into the primary plant responses to common cellular disorders produced by abiotic stresses. LEA: late embryogenesis abundant proteins; MVOCs: microbial volatile organic compounds.

extracts from algae and other organisms, natural or synthetic polymers, some chemical elements, and both microbial volatile organic compounds (MVOCs), and biogenic volatile organic compounds (BVOCs), as well as mutualist fungi, bacteria, and viruses (64,66-68; Figure 2).

### 3.1 The multilevel cascade effects of microbial mutualists of plants

Mutualist plant-microbe interactions are fine-tuned at the genetic and metabolic levels (57). For example, the plant arbuscular mycorrhizal fungi (AM) form specialised structures, but the interaction starts with the efflux of root-borne signal strigolactones (e.g., 5-deoxy-strigol) that trigger hyphal branching, followed by fungal signals (N-acetylglucosamine tetramers and pentamers and lipochitoooligosaccharides) to trigger a symbiotic response in roots. A set of common genes are expressed to induce the development of both the arbuscular mycorrhiza or the bacteria nitrogen-fixing root nodule symbiosis; after the formation of a prepenetration apparatus (PPA) and the cortical cell entry, the hyphae produce a structure called birdsfoot, that develops into a mature arbuscule and later a collapsed arbuscule (69-70). In these arbuscules takes place the exchange of plant sucrose (that will be transformed into hexose and glycogen), and fungal phosphate (Pi), nitrogen (the fungus transforms arginine into urea, and this into ammonium, NH<sup>4+</sup>), and potassium (58). In the case of bacterial endophytes or diazotrophic bacteria, they transport NH<sup>4+</sup> and nitrate (NO<sup>3-</sup>) to the plant; reduce abiotic stress by metabolising ACC, the pre-

cursor of ethylene, with the 1-aminocyclopropane-1-carboxylate deaminase (ACCd); and boost plant growth, biocontrol and remediation by contributing to iron homeostasis, and siderophore production (58,71). In addition, both fungi and bacteria transfer plant hormones to the host, such as auxins, gibberellins, cytokinins, volatile compounds (VOCs), polyamines (Poly-NH<sub>2</sub>), and secondary metabolites. That is, endophytes promote individual plant development. Moreover, endophytes can further protect the plants against abiotic and biotic stresses, enhancing tolerance to saline and heavy metals conditions, or protecting against root diseases and nematode attacks (72). Therefore, overall, endophytes improve plant fitness.

Biotic mutualist interactions in the wild usually consist of a complex multipartite relationship, with bacteria associated internally with mycorrhizal fungi (endobacteria) or externally on the surface of hyphae with beneficial effects for both partners (54). Amongst mutualist bacteria of mycorrhizal fungi, we find the so-called mycorrhizal helper bacteria (MHB), which promote hyphae development and mycorrhiza establishment (73). At the ecological and evolutionary levels, plants have established endophytic associations with fungi even before the evolution of the roots (74). The diversity of mycorrhiza has evolved from arbuscular mycorrhiza (AM) associated with early land plants and includes now the modern vesicular arbuscular mycorrhizae fungi (VAM, in the zygomycete order Glomales), ectomycorrhizae (ECMs, with more than 6000 species of basidiomycetes, ascomycetes and zygomycetes), ericoid and orchid mycorrhizas (specific mycorrhiza of the Ericales order and the



**Figure 3.** Mechanisms and traits triggered by microbes against abiotic stresses. ACCd (1-aminocyclopropane-1-carboxylate deaminase) is exclusive to bacteria; all other mechanisms can be triggered by bacteria or fungi. Osmotic adjustment, antioxidant enzymes, and phytohormones can be transcribed by virus genes (80).

Orchidaceae family, respectively), or other new mycorrhizae associations(74). Non-mycorrhiza (NM) plants have specialised nutritional strategies or habitats, such as carnivorous, parasite, epiphyte or hydrophyte plants (75). New research has identified very complex coevolutionary plant-fungus interactions involving multipartite species interactions, where the fungus acts as a phyllospheric mycorrhiza (76). The diversity of microbial mutualist species like fungi associated with plants in the ecosystems, alongside to contributing to plant health and diversity, plays a pivotal role in contributing to the valuable ecosystem services (57,77): 1) provisioning (food, drink, materials, water supply, and climate regulation); 2) regulating (water supply, waste treatment – fungal bioremediation, regulation of animal and human diseases, regulation of plant diseases, biological control); 3) supporting services (enhanced photosynthetic primary production, soil formation and stability, nutrient cycling, bioremediation); 4) cultural (recreational); and 5) ecosystem goods (plant biomass for fuel, yeast fermentation, pharmaceuticals, biocides, food additives, enzymes, genetic resources...); with a yearly average value of  $33 \times 10^{12}$  US\$ (78).

### 3.2 The plant-microbe diversity and effects against abiotic stresses

Plant-microbe interactions have an essential effect on plant phenotypic plasticity and fitness when facing environmental

stresses such as nutrient limitation, abiotic stresses, or biotic stresses: 1) nitrogen-fixing bacteria activate NOD factors enhancing plant nitrogen uptake; mycorrhizal fungi activate Myc factors to trigger the transference of phosphorous to the host; plant growth-promoting rhizobacteria (PGPR) produce mVOCs that enhance iron uptake; 2) many fungi, bacteria and virus endophytes change the chemical plasticity of the plant to enhance mechanisms against abiotic stresses (temperature, salt, drought); 3) PGPR and fungal species trigger induced systemic resistance or specific activities against pathogenic species (67; Figure 3).

#### 3.2.1 Plant-associated fungi and abiotic stress

AMF, in addition to mobilising phosphorous and nitrogen from the soil to the host plants, also play a role in resistance to saline stress by improving osmoregulating capacity due to increased soluble sugars and electrolyte concentrations in roots (81). Plants infected with VAM fungi (vesicular-arbuscular mycorrhizal fungi) exhibit higher photosynthetic activity, stomatal conductance, and transpiration. During drought events, these plants show enhanced growth rates, possibly due to more efficient water use, and have more soluble sugars in leaves; the amino acid concentration as the levels of proline vary in VAM plants depending on their ability to avoid drought (82). Some abiotic stresses have a negative effect on the microbial mutu-

alist. Salinity decreases the colonisation ability and activity of AM and inhibits it at extreme concentrations (83). Endophytic fungi and bacteria, however, can induce host plant salt tolerance by enhancing plant growth, preventing Na<sup>+</sup> uptake, and reducing NaCl toxicity (84, 85).

### 3.2.2 Plant-associated bacteria (PAB)

PAB can be found in the rhizosphere, whether intimately associated with hosts roots (rhizobacteria and root endophytes) or in close enough proximity to the roots, and in the phyllosphere, within tissues or on leaves, buds or flowers. The plant - PAB interactions have multiple eco-evolutionary implications. Moreover, PAB can alter critical plant phenological characters such as flowering, fruiting, and senescence due to PAB-induced alterations in the nutritional and hormonal status of the host (86,87). Plants can sort out and recruit particular communities of microorganisms, the rhizobiome, whose association with the plant host provide various benefits (amelioration of abiotic stresses, stimulation of growth and physiological capacities, and protection against biotic challenges) through highly heterogeneous mechanisms of action (88); thus, suggesting the importance of gaining a deeper insight of these intimate interactions.

Bacterial 1-aminocyclopropane-1-carboxylate deaminase (ACCd) regulates de production of ethylene, a stress phytohormone triggered by abiotic and biotic stresses with negative effects on root and plant growth (71). Some plant bacteria (for example, *Bacillus* sp, *Enterobacter* sp, *Proteus* sp, *Pseudomonas* sp) can promote tolerance against heavy metals (phytoextraction of Cd, Cr, Cu, Ni, Pb, Zn) by combining different mechanisms such as the production of phytohormones (IAA), siderophores, the expression of ACC, and phosphate solubilisation (89). Indeed, the presence of PAB species improves soil conditions, promotes the establishment and development of plants, and represents important support against abiotic stresses over short periods (90). However, the same synergistic natural microbial communities might negatively affect plants under more severe abiotic stresses or more extended periods of stress occurrence (91). Nevertheless, some plant multipartite interactions with plant growth promoting rhizobacterium (PGPR) in roots and endophytes in leaves play an unpredicted synergistic role by boosting plant development and triggering plant production of indoles (92); this highlights the importance of setting up more complex experimental approaches to address eco-evolutionary questions in these systems.

In semi-arid regions, plants are exposed to drought, high salinity, and high-temperature stresses, and PGPB (plant growth-promoting bacteria; *Bacillus*, *Bradyrhizobium*, *Enterobacter*, *Pantoea*, *Pseudomonas*, *Rhizobium*) triggers the following mechanisms: ACC deaminase, antioxidant system, IAA production, EPS (Exopolysaccharides) production, nitrogen and phosphate acquisition, drought and salt tolerance (93).

### 3.2.3 Microbial Volatile Organic Compounds - MVOCs

Up to date, a very diverse collection of over 2000 MVOCs with

synergistic effects on plants have been identified and analysed (mVOC 3.0; 94) in more than 1000 species of bacteria (649 species) and yeast and filamentous fungi (385 species); many of them with host species- or pathogen target species-specific effects (67): antifungal and oomycetocidal activities (54% of the cases), plant growth promotion (20%), induction of systemic resistance (7%), increase tolerance to salinity or drought (4%), antibacterial activity, induction of fruit defences, parasitoid attraction, bacteriostatic effect, increase fruit flavour and defence, increased starch synthesis, insect-pest repellent, nematocidal activity, attraction of pest insect predatory wasps, and flowering promotion. On the other hand, some MVOCs produce negative effects, such as toxicity, decreased growth, or inhibition of germination (67).

### 3.2.4 A short note on the virus-fungi-plant interaction under drought or heat stress

Some double-stranded RNA mycovirus (CThTv) infecting fungal endophytes can enhance plant tolerance against heat or drought stresses (63). While the fungus, *Curvularia protuberata*, can only resist up to 38 °C, the plant-fungus-virus system can resist up to 65 °C. The analysis of the viral-induced ESTs under heat stress (VIH-ESTs) unveiled key enzymes involved in osmoprotectants synthesis, such as TauD (taurine catabolism dioxygenase), glycine, betaine, trehalose, and a key enzyme in the melanin pathway, the scytalone dehydratase (SCD) that was overexpressed ten-fold compared to control conditions (95). The VIH-ESTs also had transcripts of several reactive oxygen species detoxification enzymes, such as five glutathione S-transferase (GST) orthologs and a peroxidase/catalase gene.

## Conclusions

Plants face severe abiotic stresses that might become drastic under the new global climate change scenarios. Many different strategies are being implemented to work together against climate change and build up resilience against its harmful effects. One of these strategies is the use of microorganisms as biostimulants in agriculture. However, biotic interactions are fine-tuned amongst the interacting partners, with cascade effects at different ecological levels. Therefore, we must build up basic knowledge about microbial diversity, the specificity of the interactions, and their potential to promote effective tolerance against abiotic stresses.

We have initiated a project to characterise microbial populations associated with plants from wild areas, especially saline environments, and analyse their potential role in aiding the plants to cope with abiotic stresses. We also aim to characterise the phenotypic plasticity of the plant-microbe system to understand the eco-evolutionary dynamics that can enhance resilience against the climate change effects in conservation biology and agriculture.

## Acknowledgements

This study was supported by a Maria Zambrano distinguished researcher contract to MXR-G, and funded by both the Min-



isterio de Universidades (Gobierno de España) and the Next generation EU. The authors have no competing financial interests.

## References

1. Kassen R. The experimental evolution of specialists, generalists, and the maintenance of diversity. *J Evol Biol* 2002; 15:173-190.
2. Elewa AMT & Joseph R. The History, Origins, and Causes of Mass Extinctions. *J Cosmol* 2009; 2: 201-220.
3. Raup D, Sepkowski JJ. Mass Extinctions in the Marine Fossil Record. *Science* 1982; 215:1501-1503. doi:10.1126/science.215.4539.1501
4. Butchart SHM, Walpole M, Collen B, van Strien A, Scharlemann JPW, et al. Global Biodiversity: Indicators of Recent Declines. *Science* 2010; 328:1164-1168. doi:10.1126/science.1187512
5. IPCC. Climate Change 2021. Intergovernmental Panel on Climate Change, Switzerland 2021
6. Scheffers BR, De Meester L, Bridge TCL, et al. The broad footprint of climate change from genes to biomes to people. *Science* 2016; 354(6313):aaf7671.
7. Breazeale JF. A study of the toxicity of salines that occur in black alkali soils. *Ariz Agr Exp Sta Tech Bul* 2021; 14:337-357.
8. Imran QM, Falak N, Hussain A, Mun B-G, Yun B-W. Abiotic Stress in Plants; Stress Perception to Molecular Response and Role of Biotechnological Tools in Stress Resistance. *Agronomy* 2021; 11:1579. doi:10.3390/agronomy11081579.
9. Boyer JS. Plant productivity and environment. *Science* 1982; 218:443-448. doi:10.1126/science.218.4571.443.
10. He M, He C-Q, Ding N-Z. Abiotic stresses: general defenses of land plants and chances for engineering multistress tolerance. *Front Plant Sci* 2018; 9:1771. doi:10.3389/fpls.2018.01771.
11. Martí MC, Stancombe MA, Webb AAR. Cell- and Stimulus Type-Specific Intracellular Free Ca<sup>2+</sup> Signals in *Arabidopsis*. *Plant Physiol* 2013; 163:625-634. doi:10.1104/pp.113.222901
12. Liu Q, Ding Y, Shi Y, Ma L, Wang Y, et al. The calcium transporter ANNEXIN1 mediates cold-induced calcium signaling and freezing tolerance in plants. *The EMBO J* 2021; 40:e104559.
13. Zhang H, Zhu J, Gong Z, Zhu J-K. Abiotic stress in plants. *Nat Rev Genet* 2022; 23:104-119. doi:10.1038/s41576-021-00413-0
14. Jung J-H, Domijan M, Klose C, Biswas S, Ezer D, et al. Phytochromes function as thermosensors in *Arabidopsis*. *Science* 2016; 354:886-889.
15. Legris M, Klose C, Burgie AE, et al. Phytochrome B integrates light and temperature signals in *Arabidopsis*. *Science* 2016; 354:897-900.
16. Krishna P. Plant response to heat stress. In: Hirt H, Shinozaki K, eds. *Plant Responses to Abiotic Stress*, Topics in Current Genetics; 2004:73-101.
17. Alvarez-Ponce D, Ruiz-González MX, Vera-Sirera F, Feyertag F, Perez-Amador MA, Fares MA. *Arabidopsis* Heat Stress-Induced Proteins Are Enriched in Electrostatically Charged Amino Acids and Intrinsically Disordered Regions. *Int J Mol Sci* 2018; 19:2276.
18. Kai H & Iba K. Temperature stress in Plants. In eLS; John Wiley & Sons, Chichester; 2014.
19. Hu Y & Schmidhalter U. Limitation of salt stress to plant growth. In Hock B, Elsner EF, eds. *Plant Toxicology* 4<sup>th</sup> ed. Boca Raton, FL, CRC Press; 2004:191-224
20. Daliakopoulos IN, Tsanis IK, Koutroulis A, et al. The threat of soil salinity: A European scale review. *Sci Total Environ* 2016; 573: 727-739.
21. Parihar P, Singh S, Singh R, Singh, VP, Prasad SM. Effect of salinity stress on plants and its tolerance strategies: a review. *Environ Sci Pollut Res* 2015; 22:4056-4075.
22. Harper RJ, Dell B, Ruprecht JK, Sochacki SJ & Smetten KRJ. Salinity and the reclamation of salinized lands. In: Stanturf JA, Callahan Jr MA eds. *Soils and Landscape Restoration*. Academic Press, Elsevier, 2021:193-208.
23. Al-shareef NO, Tester M. *Plant Salinity Tolerance*. In: eLS. John Wiley & Sons, Ltd: Chichester 2019. doi:10.1002/9780470015902.a0001300.pub3
24. Kumar A, Singh S, Gaurav AK, Srivastava S & Verma JP. *Plant Growth-Promoting Bacteria: Biological Tools for the Mitigation of Salinity Stress in Plants*. *Front Microbiol* 2020; 11:1216.
25. Roy SJ, Negrao S, Tester M. Salt resistant crop plants. *Curr Op Biotechnol* 2014; 26:115-124. doi:10.1016/j.copbio.2013.12.004
26. Molden D, Vithanage M, de Fraiture C, et al. Availability and Its Use in Agriculture. In: Wilderer P, ed. *Treatise on Water Science*, Elsevier; 2011:707-732.
27. Farooq M, Hussain M, Wahid A & Siddique KHM. Drought stress in plants: an overview. In: Aroca R ed. *Plant responses to drought stress*, Springer. 2012.
28. Zargar SM, Gupta N, Nazir M, et al. Impact of drought on photosynthesis: Molecular perspective. *Plant Gene* 2017; 11:154-159. doi:10.1016/j.plgene.2017.04.00
29. Wang W, Vinocur B, Altman A. Plant responses to drought, salinity and extreme temperatures: towards genetic engineering for stress tolerance. *Planta* 2003; 218:1-14. doi:10.1007/s00425-003-1105-5
30. Sultan SE. Phenotypic plasticity for plant development, function and life history. *Trends Plant Sci* 2000; 5(12):537-542.
31. Boscaiu M, Lull C, Lidón A, et al. Plant responses to abiotic stress in their natural habitats. *Bull UASVM, Horticulture* 2008; 65:53-58.
32. Bradshaw AD. Evolutionary Significance of Phenotypic Plasticity in Plants. *Adv Genet* 1965; 13:115-155. doi:10.1016/s0065-2660(08)6004
33. Bonnier G. Les plantes de la région alpine et leurs rapports avec le climat. *Ann Géograph* 1895; 4(17):393-413. doi:

- <https://doi.org/10.3406/geo.1895.5724>.
34. Matesanz S, Gianoli E, Valladares F. Global change and the evolution of phenotypic plasticity in plants. *Ann NY Acad Sci* 2010; 1206:35-55. doi:10.1111/j.1749-6632.2010.05704.x
  35. Schlichting, C. D. 1986. The Evolution of Phenotypic Plasticity in Plants. *Ann. Rev. Ecol. Syst.* 17, 667–693. doi:10.1146/annurev.es.17.1101
  36. Gordo, O. & Sanz J. J. 2010. Impact of climate change on plant phenology in Mediterranean ecosystems. *Global change Biol.* 16, 1082-1106. doi: 10.1111/j.1365-2486.2009.02084.x
  37. Meier U, Bleiholder H, Buhr L, et al. The BBCH system to coding the phenological growth stages of plants – history and publications – *J KULTURPFL* 2009; 61(2):S41–52.
  38. Bradley NL, Leopold AC, Ross J & Huffaker W. Phenological changes reflect climate change in Wisconsin. *PNAS* 1999; 96:9701-9704. doi:10.1073/pnas.96.17.9701
  39. Fitter AH & Fitter RSR. Rapid Changes in Flowering Time in British Plants. *Science* 2002; 296: 1689-1691. doi:10.1126/science.107161
  40. Lee HK, Lee SJ, Kim MK & Lee SD. Prediction of Plant Phenological Shift under Climate Change in South Korea. *Sustainability* 2020; 12:9276.
  41. Primack RB, Ibáñez I, Higuchi H, et al. Spatial and inter-specific variability in phenological responses to warming temperatures. *Biol Conserv* 2009; 142:2569-2577. doi:10.1016/j.biocon.2009.06.003
  42. Bond WJ. Keystone Species. In: Schulze E-D et al. eds. *Biodiversity and Ecosystem Function*, Springer-Verlag Berlin Heidelberg, 1994:237–253. doi:10.1007/978-3-642-58001-7\_11.
  43. Rawat US; Agarwal NK. Biodiversity: Concept, threats and conservation. *Environ Conserv J* 2015; 16:9-28. doi:10.36953/ECJ.2015.16303
  44. Costanza R, et al. The value of the world's ecosystem services and natural capital. *Nature* 1997; 387:253-260.
  45. Barbier EB, Hacker SD, Kennedy C, Koch EW, Stier AC, Silliman BR. The value of estuarine and coastal ecosystem services. *Ecol Mon* 2011; 81(2):169-193.
  46. Gedan KB, Silliman BR & Bertness MD. Centuries of human-driven change in salt marsh ecosystems. *Annu Rev Mar Sci* 2009; 1:117–41.
  47. Adams JB, Raw JL, Riddin T, Wasserman J, Van Niekerk L.. Salt marsh restoration for the provision of multiple ecosystem services. *Diversity* 2021; 13:680. doi:10.3390/d13120680
  48. Stevanović ZD, Ačić S, Stešević D, Luković M, Šilc U. Halophytic vegetation in South-east Europe: Classification, conservation and ecogeographical patterns. In: Hasanuzzaman M, Shabala S, Fujita M, eds. *Halophytes and climate change*. CAB International, UK; 2019:55-68.
  49. Rothschild LJ, Mancinelli RL. Life in extreme environments. *Nature* 2001; 409:1092-1101.
  50. Galuzzi G, Seyoum A, Halewood M, López Noriega I & Welch EW. The Role of Genetic Resources in Breeding for Climate Change: The Case of Public Breeding Programmes in Eighteen Developing Countries. *Plants* 2020; 9:1129. doi:10.3390/plants9091129
  51. Passamonti MM, Somenzi E, Barbato M, et al. The Quest for Genes Involved in Adaptation to Climate Change in Ruminant Livestock. *Animals* 2021; 11:2833. doi:10.3390/ani11102833
  52. Carroll G. Fungal Endophytes in Stems and Leaves: From Latent Pathogen to Mutualistic Symbiont. *Ecology* 1988; 69:2–9. doi:10.2307/1943154
  53. Rodriguez RJ, White Jr JF, Arnold AE, Redman RS. Fungal endophytes: diversity and functional roles. *New Phytol* 2009; 182:314-330. doi:10.1111/j.1469-8137.2009.02773.x
  54. Bonfante P & Anca I-A. Plants, Mycorrhizal Fungi, and Bacteria: A Network of Interactions. *Annu Rev Microbiol* 2009; 63:363-383. doi:10.1146/annurev.micro.091208.073504
  55. Redecker D, Kodner R, Graham LE. Glomalean Fungi from the Ordovician. *Science* 2000; 289:1920-1921. doi:10.1126/science.289.5486.19
  56. van der Heijden MGA, Klironomos JN, Ursic M, et al. Mycorrhizal fungal diversity determines plant biodiversity, ecosystem variability and productivity. *Nature* 1998; 396:69-72.
  57. Chen M, Arato M, Borghi L, Nouri E & Reinhardt D. Beneficial services or arbuscular mycorrhizal fungi – From ecology to application. *Front Plant Sci* 2018; 9:1270.
  58. Haridoim PR, et al. The Hidden World within Plants: Ecological and Evolutionary Considerations for Defining Functioning of Microbial Endophytes. *Microbiol Mol Biol Rev* 2015; 79:293-320. doi:10.1128/MMBR.00050-14.
  59. Link HF. *Observationes in ordinibus plantarum naturalium, dissertatio prima, complectens anandarum ordinibus Epiphytas, Mucedines, Gastromycos et Fungos*. Der Gesellschaft Naturforschender Freunde zu Berlin, Berlin, Germany. 1809:3-42. <https://hdl.handle.net/2027/hvd.32044106318025>
  60. Berch SM, Massicotte HB & Tackaberry LE. Re-publication of a translation of ‘The vegetative organs of *Monotropa hypopitys* L.’ published by F. Kamienski in 1882, with an update on *Monotropa mycorrhizas*. *Mycorrhiza* 2005; 15:323-332. doi: 10.1007/s00572-004-0334-1.
  61. Frank AB. Über die auf Wurzelsymbiose beruhende Ernährung gewisser Bäume durch unterirdische Pilze. *Ber Dtsch Bot Ges* 1885; 3: 128–145.
  62. Woronin MS. Über die bei der schwarzerle (*Alnus glutinosa*) und der gewöhnlichen garten-lupine (*Lupinus mutabilis*) auftretenden Wurzelanschwellungen. *Mém de l'Acad Imp des Sciences de St-Petersbourg* 1866; X6.
  63. Marquez LM, Redamn RS, Rodriguez RJ, Roossinck MJ. A Virus in a Fungus in a Plant: Three-Way Symbiosis Required for Thermal Tolerance. *Science* 2007; 315:513-515. doi:10.1126/science.1136237
  64. Roossinck MJ. The good viruses: viral mutualistic symbi-

- oses. *Nat Rev Microbiol* 2011; 9: 99-108. doi:10.1038/nr-micro2491
65. Lata R, Chowdhury S, Gond S K, White Jr JF. Induction of abiotic stress tolerance in plants by endophytic microbes. *Lett. Appl. Microbiol.* 2018; 66:268-276.
  66. Drobeck M, Frac M & Cybulska J. Plant Biostimulants: Importance of the Quality and Yield of Horticultural Crops and the Improvement of Plant Tolerance to Abiotic Stress—A Review. *Agronomy* 2019; 9:335. doi:10.3390/agronomy9060335
  67. Poveda J. Beneficial effects of microbial volatile compounds (MVOCs) in plants. *Appl Soil Ecol* 2021; 168:104118. doi: 10.1016/j.apsoil.2021.104118
  68. Povero G, Mejoa JF, Di Tommaso D, Piaggese A, Warrior P. A systematic approach to discover and characterize natural plant Biostimulants. *Front Plant Sci* 2016; 7:435.
  69. Akiyama K, Matsuzaki K-I, Hayashi H. Plant sesquiterpenes induce hyphal branching in arbuscular mycorrhizal fungi. *Nature* 2005; 435:824-827.
  70. Gutjar C & Parniske M. Cell and Developmental Biology of Arbuscular Mycorrhiza Symbiosis. *Annu Rev Cell Dev Biol* 2013; 29:593–617.
  71. Souza R, Ambrosini A, Passaglia LMP. Plant growth-promoting bacteria as inoculants in agriculture soils. *Genet Mol Biol* 2015; 38 (4):401-419.
  72. Lee E-H, Eo J-K, Ka K-H & Eom A-H. Diversity of arbuscular mycorrhizal fungi and their roles in ecosystems. *Mycobiology* 2013; 41(3):121-125.
  73. Garbaye J. Helper bacteria: a new dimension to the mycorrhizal symbiosis. *New Phytol* 1994; 128:197–210.
  74. Brundrett MC. Coevolution of roots and mycorrhizas of land plants. *New Phytol* 2002; 154:275-304.
  75. Brundrett MC, Tardersoo L. Evolutionary history of mycorrhizal symbioses and global host plant diversity. *New Phytol* 2018; 220:1108–1115.
  76. Leroy C, Séjalon-Delmas N, Jauneau A, Ruiz-González MX, et al. Trophic mediation by a fungus in an ant–plant mutualism. *J Ecol* 2011; 99:583-590. doi:10.1111/j.1365-2745.2010.01763.x
  77. Moore D, Robson GD, Trinci APJ. 21<sup>st</sup> Century Guidebook to Fungi, 2019.
  78. Gianinazzi S, Gollotte A, Binet M-N, van Tuinen D, Redecker D & Wipf D. Agroecology: the key role of arbuscular mycorrhizas in ecosystems services. *Mycorrhiza* 2010; 20:519-530.
  79. Goh C-H, Veliz Vallejos DF, Nicotra AB & Mathesius U. The impact of beneficial plant-associated microbes on plant phenotypic plasticity. *J Chem Ecol* 2013; 39:826-839.
  80. Poudel M, Mendes R, Costa LAS, et al. The Role of Plant-Associated Bacteria, Fungi, and Viruses in Drought Stress Mitigation. *Front Microbiol* 2021; 12:743512. doi:10.3389/fmicb.2021.743512
  81. Feng G, Zhang FS, Li XL, Tian CY, Tang C, Rengel Z. Improved tolerance of maize plants to salt stress by arbuscular mycorrhiza is related to higher accumulation of soluble sugars in roots. *Mycorrhiza* 2002; 12:185-90.
  82. Augé RM. Water relations, drought and vesicular-arbuscular mycorrhizal symbiosis. *Mycorrhiza* 2001; 11:3-42.
  83. Carvalho LM, Correia PM, Caçador I & Martins-Louçao AM. Effects of salinity and flooding on the infectivity of salt marsh arbuscular mycorrhizal fungi in *Aster tripolium* L. *Biol Fertil Soils* 2003; 38:137-143.
  84. Lubna, Muhammad AK, Sajjad A, Rahmatullah J, Muhammad W, Kyung-Min K & In-Jung L. Endophytic fungus *Bipolaris* sp. CSL-1 induces salt tolerance in *Glycine max*.L via modulating its endogenous hormones, antioxidative system and gene expression. *J Plant Interact* 2022; 17: 319-332. doi: 10.1080/17429145.2022.2036836.
  85. Qing L, Zhehong H, Caisheng D, Kuan-Hung L, Shumei H & ShiPeng C. Endophytic *Klebsiella* sp. San01 promotes growth performance and induces salinity and drought tolerance in sweet potato (*Ipomoea batatas* L.), *J Plant Interact* 2022; 17: 608-619. doi: 10.1080/17429145.2022.2077464
  86. O'Brien AM, Ginnan NA, Rebolleda-Gómez M, Wagner MR. Microbial effects on plant phenology and fitness. *Am J Bot* 2021; 108:1824–1837. doi:10.1002/ajb2.1743
  87. Wagner MR, Lundberg DS, Coleman-Derr D, Tringe SG, Dangl JL, Mitchell-Olds T. Natural soil microbes alter flowering phenology and the intensity of selection on flowering time in a wild *Arabidopsis* relative. *Ecol Lett* 2014; 17:717-726. doi:10.1111/ele.12276
  88. Orozco-Mosqueda MC, Fadji AE, Babalola OO, Glick BR & Santoyo G. Rhizobiome engineering: Unveiling complex rhizosphere interactions to enhance plant growth and health. *Microbiol Res* 2022; 263: 127137. doi: 10.1016/j.micres.2022.127137
  89. Glick BR. Using soil bacteria to facilitate phytoremediation. *Biotechnol Adv* 2010; 28:367-374.
  90. Qian S, Xiaoshuang S, Xun D, Jiayu L, Junkai W, Kai M & Ruiqing S. Effects of plant growth promoting Rhizobacteria microbial on the growth, rhizosphere soil properties, and bacterial community of *Pinus sylvestris* var. *mongolica* seedlings. *Scand J For Res* 2021; 36: 249-262. doi: 10.1080/02827581.2021.1917649
  91. Ulrich DEM, Sevanto S, Ryan M, Albright MBN, Johansen RB & Dunbar JM. Plant-microbe interactions before drought influence plant physiological responses to subsequent severe drought. *Sci Rep* 2019; 9: 249. doi: 10.1038/s41598-018-36971-3
  92. de Almeida JR, Bonatelli ML, Durante Batista B, Teixeira-Silva NS, Mondin M, dos Santos RC, Simoes Bento, JM, Azevedo Jm, Quecine MC. *Bacillus thuringiensis* RZ2MS9, a tropical plant growth-promoting rhizobacterium, colonizes maize endophytically and alters the plant's production of volatile organic compounds during co-inoculation with *Azospirillum brasilense* Ab-V5. *Environ Microbiol Rep* 2021; 13: 812–821 doi: 10.1111/1758-2229.13004
  93. Bonatelli ML, Lacerda-Júnior GV, dos Reis Junior FB, Fernandes-Júnior PV, Soares Melo I & Quecine MC. Beneficial plant-associated microorganisms from semi-arid

- regions and seasonally dry environments: a review. *Front Microbiol* 2021; 11:553223.
94. Lemfack MC, Gohlke B-O, Toguem SMT, et al. mVOC 2.0: a database of microbial volatiles. *Nucleic Acids Res* 2018; 46(D1):D1261-D1265.
  95. Morsy MR, Oswald J, He J, Tang Y & Roossinck MJ. Teasing apart a three-way symbiosis: transcriptome analyses of *Curvularia protuberata* in response to viral infection and heat stress. *Biochem Biophys Res Commun* 2010; 401:225–230. doi:10.1016/j.bbrc.2010.09.034