

## Enhancing Climate Resilience of Crop Plants : An Approach using Endophytes

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### ABSTRACT

The importance of increasing crop production in light of extreme events due to climate change and a human population growth projected to reach nine billion this century is a major challenge. Endophytic fungi and bacteria offer a novel approach to enhance agricultural productivity while reducing environmental costs. Endophytes have been shown to aid several plant growth processes. Endophytes could also help increase crop production and reduce yield losses by improving resistance to both biotic and abiotic stresses. With climate change projected to have drastic impact on agriculture especially in arid and semi-arid regions, endophytes could play a major role in sustaining agricultural production. In concert with other novel agronomic technologies and management, endophytes could help in mitigating the impacts of climate change. This review focuses on the ability of endophytes in promoting growth as well as in imparting stress tolerance in the context of climate change.

WATER and food security are two of the key issues that are threatened by climate change. The impacts of climate change on water resources and agricultural yield, worldwide, are beginning to be documented (Wheeler and von-Braun, 2013). The average global temperatures are expected to increase by 1.4–5.8 °C and this could result in substantial reduction in agricultural yield by the end of the 21<sup>st</sup> century (Misra, 2014). Increase in temperatures could also result in variations in precipitation patterns and river flows besides rise in sea level (Solomon *et al.*, 2007, IPCC report 2008). Quite obviously, countries in the arid and semi-arid regions that are largely dependent on precipitation for their agriculture are likely to be most affected. Erratic and extreme rainfall events could result respectively, to frequent drought and floods. Evidence is mounting to suggest that, in recent years, both the frequency and intensity of drought have increased in many parts of the world (Solomon *et al.*, 2007). A few studies have indicated that agricultural yields will be severely affected due to unprecedented rates of change in the climate system (Thornton *et al.*, 2011). At a landscape level, global warming is predicted to increase desertification and double the loss of arable area in the world (IPCC, 2008). In summary, climate change, coupled with the demands made by a steadily increasing human population, which is expected to rise to 9 billion by 2050, poses a serious challenge to maintaining world food security. Unless appropriate mitigation and adaptation strategies are

developed to ensure a climate-secure agriculture, it is feared that global food production will be deeply compromised by climate change (Wheeler and von-Braun 2013, Davidson, 2016).

### **I. Plant responses to stress and crop improvement program**

Abiotic stresses such as extremes of temperatures, salinity, drought, and flooding limit crop productivity worldwide. Despite plants being sessile in nature and lacking mechanisms to escape these adverse conditions, they have over evolutionary period of time, developed unique and sophisticated responses to environmental stresses. These mechanisms include tolerance, resistance or avoidance. Plants that develop tolerance to a given stress can, over time, overcome the effects of abiotic stress without injury. In case of resistance, plants have developed counter measures to overcome the stressful environment. Similarly, plants have also developed avoidance mechanisms to prevent exposure to stress.

In recent years, understanding the molecular mechanisms of abiotic stress tolerance as well as inducing stress tolerance some crops has been explored (Parvathi *et al.*, 2013; Sajeevan and Nataraja, 2016; Parvathi and Nataraja, 2017). A number of efforts have been made to understand molecular, physiological and metabolic aspects of stress tolerance to facilitate the development of crops with an inherent capacity to withstand abiotic stresses. These include molecular

breeding for new varieties by exploiting the genetic variability existing for these traits, screening and selection of the existing germplasm of potential crops, production of genetically modified (GM) crops, exogenous use of osmoprotectants etc. (Kathuria, *et al.*, 2009; Pruthvi *et al.*, 2014).

## II. Endophytes and their role in climate resilient agriculture

As opposed to an active modulation of plant responses to abiotic stresses, through conventional and molecular genetic approaches, a little or lesser known approach that is recently gaining attention is modulation of plant growth by plant microbiome. Plants are intimately associated with a diversity of microbiome and these have been known for long to influence plant growth and productivity (Redmann, 2002). The association of microbiome with plants span relationships including commensalism, mutualism and in the extreme cases, to symbiosis. One such microbiome is the endophyte.

Endophytes (both bacterial as well as fungal) are ubiquitous plant symbionts that reside in intercellular spaces of stems, petioles, roots, reproductive parts and leaves of plants without causing any overt negative effects (Mohana-Kumara *et al.*, 2013). Many plant processes have been attributed to be shaped by endophytes. Endophytic fungi play a major role in structuring plant communities and in shaping processes such as colonization, competition, coexistence and soil nutrient dynamics (Schulz *et al.* 2002). Several studies have also demonstrated the role of endophytic fungi in imparting tolerance to plants against abiotic and biotic stresses (Mohana-Kumara *et al.*, 2013). Besides their role in aiding several plant growth processes, endophytic fungi are known to produce a large number of secondary metabolites (Tan and Zou, 2001). Endophytes have been recognized as an important repository of novel bioactive products including alkaloids, steroids, terpenoids, isocoumarins, quinones, flavonoids, phenylpropanoids, lignans, peptides, phenolics, aliphatics, and volatile organic compounds with potential application in agriculture, medicine, and food industry (Suryanarayanan *et al.*, 2017). In the context of the association of endophytes with plants, it would be interesting to explore their

possible implications in ensuring a climate-secure agriculture where, crop plants, could be made climate resilient. In this paper, we briefly review the role of endophytes in ameliorating abiotic stress in plants and discuss how this knowledge could offer exciting possibilities of using endophytes as a possible approach to make agriculture climate resilient.

## III. Modulating abiotic stress tolerance using endophytes

a) *Modulation of plant responses to temperature and drought* : Among abiotic stresses, drought is a major stress that adversely affects crop growth and productivity worldwide. Global warming is projected to increase the severity and frequency of drought in the future leading to a possible decrease in global food production. It is estimated that more than 50 per cent of the arable land would be affected by drought by 2050, severely affecting plant growth (Vinocur and Altman, 2005). Drought stress affects water relations at cellular, tissue and organ levels impairing biochemical and physiological mechanisms in plants and reducing their growth and development (Beck *et al.*, 2007). Similarly, heat stress interrupts plant growth by impairing important physiological and morphological processes such as germination, photosynthesis and flowering (Hasanuzzaman *et al.*, 2013; Sgobba *et al.*, 2015).

In plants, drought stresses could be relieved to some extent by the infecting endophytes, which evoke various natural mechanisms to help plants sustain their growth even under stressful conditions (Vardharajula *et al.*, 2011). Under both drought and heat stress, endophytes have been able to mitigate the stress by producing a number of phytohormones (Khan *et al.*, 2012; Waqas *et al.*, 2014). Endophytes also benefit the host plants by improving the uptake of nutrients and water, water-use efficiency and endogenous hormone levels (Khan *et al.*, 2012). The endophytic association with host plants has been reported to positively alter primary and secondary metabolites (Khan and Lee, 2013) under abiotic stress. Free amino acids, particularly proline, glutamine, and leucine accumulated in endophyte-infested soybean plants. Among these, the osmo-protectant proline is important for growth restoration and tolerance under abiotic stress (Khan and Lee, 2013).

Heat stress due to global warming is often associated with drought stress (Sgobba *et al.*, 2015). Global temperatures are predicted to rise by 2-5 °C by the end of this century affecting crop growth and productivity. The occurrence of heat shock waves will further affect agricultural production (Fragkoste fanakis *et al.*, 2014). As in case of drought stress, endophytes have also been reported to impart thermo-tolerance to plants as reported in case of *Dichanthelium lanuginosum* (panic grass), which thrives in the geothermal soils of the Yellowstone National Park, Wyoming (Redman *et al.*, 2002). The endophyte *Curvularia* sp. is reported to confer thermotolerance to plant and this plant/fungal symbiosis is responsible for survival of both, in geothermal soils. When grown separately, the plant and endophytic fungus exhibit maximum growth at 40 °C and 38 °C respectively. However, when the the plant and the fungus are grown together in an symbiotic association, they are able to tolerate temperature regimes upto 70 °C.

One of the mechanisms of endophyte mediated thermo-tolerance involves interaction with the heat shock proteins of the host (McLellan *et al.*, 2007). The endophyte *Paraphaeosphaeria quadriseptata* can affect expression of heat shock proteins and enhance thermotolerance in *Arabidopsis thaliana* (McLellan *et al.*, 2007). Similarly, another endophyte *Morchella* increased biomass and fecundity of its local cheat grass (*Bromus tectorum*) host, as well as survival of seeds exposed to heat in the seed bank during a cheat grass fire (Baynes *et al.*, 2012).

b) *Modulation of plant responses to salinity* : The effects of climate change on the quantity and, to a lesser extent, the variability of water supplies and the consequent impact on soil salinity may pose an additional, albeit lesser-known, challenge. Nearly one-third of the irrigated land worldwide is affected by salinization (Schwabe *et al.*, 2006). Increased irrigation to meet the growing world food demand has further led to more arable land becoming saline (Connor *et al.*, 2012). Global climate change has also resulted in sea water level rise, resulting in inundation and salinity intrusion in many low lying areas. Salinity is thus expected to pose a serious threat to the

sustainable agricultural development and food security (Connor *et al.*, 2012). Saline environments tend to hinder agricultural production by lowering crop yields, often quite substantially. Applying excess water was one of the traditional approaches to leach out the excess salts from the root zone. However, in case of climate change, reduced water supplies in arid and semi-arid regions where salinity is an issue work against such a response. Climate change, furthermore, may compound salinity challenges in basins where declining inflows provide less dilution. Without reductions in salt loads, lower flows result in higher salt concentrations (Connor *et al.*, 2008).

The use of fungal endophytes can be an excellent opportunity to minimize the negative effect of abiotic factors, such as salinity, on crop yield. *Association with endophytes has been shown to ameliorate plant responses to salinity stress* (Singh *et al.*, 2011). *The gibberellic acid producing fungal strains, Pennicillium funiculosum and Aspergillus fumigatus*, significantly improved growth of soybean under moderate-/high-salinity stress (Khan *et al.*, 2011). In the presence of the endophyte, the host plant reprograms its salinity-stress response by regulating phytohormones and antioxidant enzymes (peroxidases, catalases) that scavenge reactive oxygen species to minimize cellular toxicity from the secondary oxidative stress (Khan *et al.*, 2015a; Khan *et al.*, 2015b). Contreras-Cornejo *et al.* (2014) reported the salinity-stress tolerance induction potential of *Trichoderma* species in *Arabidopsis* seedlings. Plant-growth promotion under saline and normal conditions was related to increase in endogenous auxin (IAA) by fungal association that promotes a larger number of lateral roots and hairs. Inoculation of *Trichoderma* spp. also enhanced the antioxidant and osmo-protectant status of *Arabidopsis* seedlings under salinity stress (Contreras-Cornejo *et al.*, 2014).

c) *Modulation of plant responses to flooding*: Apart from drought and salinity, flooding is another major abiotic stress determining agricultural productivity worldwide (Ahmed *et al.*, 2013). Flooding results in waterlogging and is regarded as one of the most hazardous natural occurrences in low-lying countries (Ahmed *et al.*, 2013), and accumulating

evidence suggests that climate change will increase the risk of geographic coverage of floods in the future (Woodruff *et al.*, 2013). Water logging, results in either soil hypoxia (deficiency of O<sub>2</sub>) or anoxia (absence of O<sub>2</sub>) (Zabalza *et al.*, 2009), and alters soil physiochemical properties such as soil pH and redox potential of the soil (Ashraf, 2012).

Plant responses to waterlogging, in which roots and some portion of the shoot are submerged, vary with species as well as with water level, duration and timing of waterlogging (Pucciariello and Perata, 2013). Waterlogging has been shown to induce leaf senescence, reduce chlorophyll content and leaf area, inhibit photosynthesis and plant growth (Gibbs *et al.*, 2011). Song *et al.*, (2015), show that *Hordeum brevisubulatum* plants which are infected with *Epichloe* endophyte are able to have higher resistance to water logging compared to endophyte free plants. The endophyte infected plants showed significantly less damage to waterlogging and produced significantly greater content of chlorophyll, more tillers, higher shoots and higher under-ground biomass compared to endophyte free plants. Water logging induced osmoprotective proline production particularly in endophyte infected plants and had lower malondialdehyde content and electrolyte leakage, suggesting that endophyte infection positively affects osmotic potential and oxidative balance of the host plant.

#### IV. Enhancing biotic tolerance

Climate change is likely to influence disease epidemics in cultivated plants and natural vegetation and threaten global food security and natural ecosystems. Climate also affects agricultural pests. The spatial and temporal distribution and proliferation of insects, weeds, and pathogens is determined, to a large extent, by climate, because temperature, light, and water are major factors controlling their growth and development. Increasing infestation by insect herbivores and pathogenic fungi in response to climate change will inevitably impact agricultural production (Rosenzweig, 2001). Plant disease and pests can cause substantial losses to agricultural production, causing famine in some cases, and also damage to natural plant systems. Temperature influences

infection processes for pathogens as well as determining rates of reproduction of arthropods that vector pathogens (Coakley *et al.*, 1999). As weather patterns change, disease risk also changes, requiring that strategies for management be updated to new conditions.

Association with endophytes aid the plant hosts to tolerate various biotic stressors. Endophyte infected leaves are often not defoliated by leaf-cutting ants (Estrad *et al.*, 2015). Introducing entomopathogenic endophytes *Purpureocillium lilacinum* and *Beauveria bassiana* in cotton inhibited aphid reproduction in greenhouse and field conditions (Lopez, *et al.*, 2014). Similarly, the Bt cotton plants which experience lesser insect visitations support less endophyte load in their tissues suggesting a positive correlation between insect damage and density of endophyte colonization of plants (Suryanarayanan *et al.*, 2011). While the exact mechanism of insect deterrence by endophytes is not known, it is hypothesized that production of secondary metabolites by these endophytes could make the plants toxic/distasteful to the insects or reduce fitness of insects (Bittleston *et al.*, 2011). Endophytes have also been reported to reduce disease severity caused by fungal pathogens by up-regulating many defence-related genes of the plant host (Mejía *et al.*, 2014). *Piriformospora indica*, a root endophyte of many plants confers resistance to some pathogens by stimulating the host's OXII pathway, which activates the host defense reaction (Camehl *et al.*, 2011).

Endophytes with biocontrol effect have also received attention as an alternative to chemical disease control, substantially reducing the use of hazardous chemicals (Porrás-Alfaro and Bayman, 2011). Bacterial endophytes from maize (*Bacillus subtilis* and *Bacillus mojavensis*) have shown great potential in the biocontrol of *Fusarium moniliforme* in maize and reduced seedling blight of wheat caused by *Fusarium graminearum* and related species respectively (Bacon *et al.*, 2001; Bacon and Hinton, 2007). Similarly, *Trichoderma* spp. and other endophytic fungi isolated from *Theobroma cacao* have shown antagonistic effect against a number of important pathogens of cacao (*Phytophthora*

*palmivora*, *Monilophthora roreri*, and *Monilophthora perniciosa*) (Bailey *et al.*, 2006; Mejía *et al.*, 2008). It is demonstrated in several pathosystems that the endophytes are effective biocontrol agents that reduce disease severity of plant diseases (Wicklow *et al.*, 2005).

## V. Enabling growth and increasing yield

Climate change is expected to drastically alter global patterns of food supply and demand. Reduced crop production, as expected due to warming temperatures and greater incidence of extreme weather events, could lead to significant reductions in crop yields and increased vulnerability to malnutrition and hunger in developing countries. Growing population especially in the developing countries and the shortage of land and water could pose serious challenges for enhancing agricultural production. Further, the cost of production is also likely to rise in a changing climate as farmers need to change their crop varieties and species, schedule more operations for land and water management, and invest in new technologies and infrastructure.

In recent years, there has been a growing evidence to suggest that the endophytes not only impart biotic and abiotic stress tolerance to crop plants but also are able to enhance plant growth as well as yield by promoting nutrient availability, biological nitrogen fixation, and the production of phytohormones (Shishido *et al.*, 1999; Ryan *et al.*, 2008; Kim *et al.*, 2011). They have also been reported to enhance growth by indirectly reducing microbial populations that are pathogenic to plants acting as agents of biological control through competition, antibiosis, or systemic resistance induction (Sturz *et al.*, 2000; Ramamoorthy *et al.*, 2001). For example, *Serratia marcescens*, an important rice endophyte (Gyaneshwar *et al.*, 2001), induces plant growth by stimulating phytohormone production and phosphate solubilization (Chen *et al.*, 2006; Selvakumar *et al.*, 2008) along with improvement of nitrogen supply in non-symbiotic associations (Islam *et al.*, 2010). Similarly, this endophyte has also been reported to increase plant height, new leaf production, and leaf biomass in tea seedlings (Chakraborty *et al.*, 2010). Endophytes have been reported to confer benefits to their host plants

via biological nitrogen fixation (Pankievicz *et al.*, 2015, Doty *et al.*, 2016), enhancing the bioavailability of phosphorous (P), iron (Fe) and other mineral nutrients (Bulgarelli *et al.*, 2013), production of phytohormones including indole acetic acid, abscisic acid, gibberellic acid, brassinosteroids, jasmonates, salicylic acid (Sharma and Abrams, 2005; Javid *et al.*, 2011; Straub *et al.*, 2013; Fahad *et al.*, 2015), generation of antioxidants (Mitter *et al.*, 2013). Although the interaction between endophytes and their host plants is not fully understood, several studies have demonstrated the positive effects of inoculation of endophytes to increase plant productivity.

## VI. Adaptation to climate variability

Application of endophytes to crop plants to modulate their growth under climate variability as mentioned above should be factored in as part of the ongoing technological development in agriculture, including molecular breeding, irrigation management, application of information and communication technology etc. To avoid or at least reduce negative effects of climate change and enhance possible positive effects, endophytes should be exploited in large scale especially in crop plants as well as in horticultural plants. Endophytic fungi associated with distinct plants and habitats (such as medicinal plants, land races/wild relatives of crop plants/extreme habitats) could be expected to have evolved novel bioactive molecules and intrinsic mechanisms to tolerate abiotic and biotic stresses and other such habitat specific adaptations. As integral component of plants, the endophytic fungi could passively or actively facilitate their host plants to tolerate abiotic and biotic stress over and above the plants' own defenses. They could also provision their hosts with a number of secondary metabolites that directly and indirectly may help enhance the host fitness. Thus, enrichment of plants with endophytic fungi can actually enhance crop productivity under both biotic and abiotic stress (Fig. 1). Endophytes could also be exploited to enhance early seedling vigor as in forest nurseries and during transplantations to overcome transplanting stress. With increasing stress on recovering saline soils and wastelands, endophytes could play a role in enhancing the establishment of large-scale planting. However, research will also have to deal with some unknown aspects that due to their

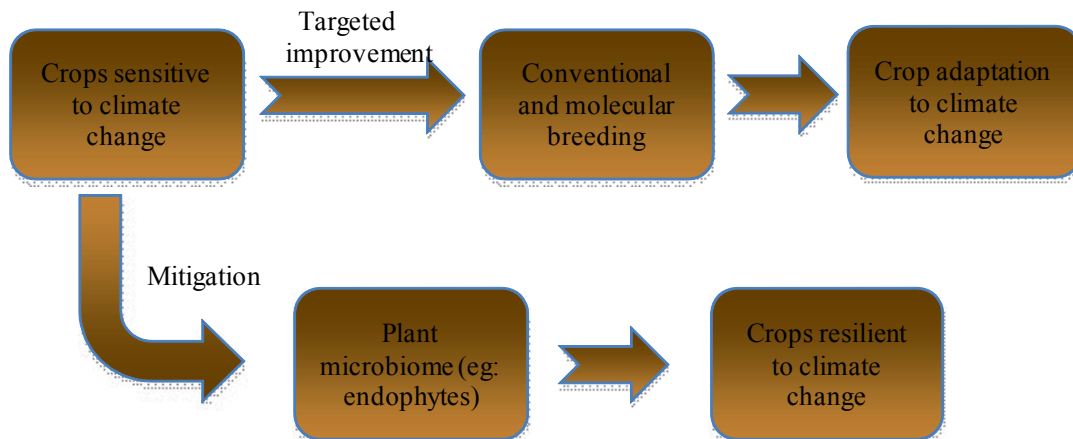


Fig. 1: Enhancing climate resilience of crop plants through conventional genetic approaches and through use of plant microbiome

complexity have not yet been studied in detail. These include the effect of endophytes on secondary factors of agricultural production (e.g. weeds), the effect on the quality of crop, the effect of changes in frequency of isolated and extreme weather events on agricultural production, and the interaction with the surrounding natural ecosystems.

The ability of endophytes to confer stress tolerance to plants may provide a novel strategy for mitigating the impacts of global climate change on agricultural and native plant communities. Endophytes could play a significant role in stress management, once their unique properties of tolerance to extremities, their ubiquity, and genetic diversity are understood and methods for their successful deployment in agriculture production are developed. These endophytes also provide excellent models for understanding stress tolerance mechanisms that can be subsequently engineered in crop plants. In summary, the use of endophytes, besides other conventional approaches, could help accelerate efforts towards developing crop plants resilient to climate change.

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