

Plant Growth Promoting Rhizobacteria (PGPR) mediated temperature, drought and pesticide stress tolerance of crop plants through multidisciplinary approach

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Abstract

Root colonizing, beneficial, soil bacteria are usually referred to as PGPR, influences plant growth by various direct or indirect mechanisms. Global warming, drought and pesticide stress causes a major challenge to crop productivity. The present study is aimed to review the PGPR mediated temperature, drought and pesticide stress tolerance of crop plants, through application of potential PGPR. Recent studies revealed that rhizospheric and endophytic bacteria such as *Azospirillum brasilense*, *Achromobacter piechaudi*, *P. polymyxa*, *Herbaspirillum* sp. and *Gluconacetobacter diazotrophicus* significantly reduce drought stress of crop plants. Pre-inoculation of heat resistant endospore of *Bacillus* sp. and *Burkholderia phytofirmans* significantly enhance plant growth at high temperature. The water retention capacity of some exo-polysaccharides can exceed several-fold of their mass through the PGPR mediated biofilm formation. PGPR *Pseudomonas* sp. *Flavobacterium* sp. *Rhizobium* sp. *Mesorhizobium* strain MRC4, *Pseudomonas diminuta* MG, *Pseudomonas maltophilia* and *Azotobacter* sp. revealed resistance to various pesticides. Taken together, the beneficial plant growth promoting and abiotic stress tolerance potentials of PGPR, simultaneously could be of agricultural significance and warrants further investigations.

Key words: PGPR, Pesticide, Drought, Heat tolerance

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Introduction

Over the last decade, a significant research interest has been generated around PGPR, because of their capacity to increase plant growth, through various direct/indirect mechanisms. The benefits of plant growth, by the addition of PGPR include increases in germination rates, root growth, yield including grain size, leaf area, chlorophyll content, hydraulic activity and tolerance to drought (Lucy *et al.*, 2004). Plant growth in agricultural soils is influenced by many abiotic and biotic factors. Abiotic stresses such as drought,

hazardous pesticide and environmental temperature are the major limiting factors for crop growth and productivity. Abiotic stress may cause physiological, biochemical, morphological and molecular changes in the crop plants. The rhizospheric soil is more nutrient rich as compared to the bulk soil due to the accumulation of a variety of plant exudates, such as amino acids and sugars (Vurukonda *et al.*, 2016). The number of rhizobacteria in rhizospheric soil is generally 10 to 100 times higher than that of the bulk soil (Weller and Thomashow, 1994). This is due to various chemicals secreted from microorganisms and plant's roots, such as amino acids, organic acids, flavonols, glucosinolates, indole compounds, fatty acids, polysaccharides and proteins in the rhizosphere that acts as signals (Vurukonda *et al.*, 2016). It is well established that only 1 to 2% of bacteria promote plant growth in the rhizosphere (Antoun and Kloepper, 2001). PGPR of genera *Bacillus* sp. and *Pseudomonas* sp. have been identified as the most predominant in rhizospheric soil (Podile and Kishore, 2006).

Crops are highly sensitive to environmental and climatic variations. Heat stress may cause serious injuries to the cell membrane, cell membrane permeability, cell differentiation, cell elongation, cell expansion and microtubular organization. Moreover, heat stress may also cause alteration in the carbon flux of the chloroplast stroma, thylakoid membrane system, Rubisco enzymes that may lead to reduced plant productivity (Bita and Gerats 2013). Furthermore, exposure to low temperatures and high temperature induces inactivation of enzymes, proteins, disturbed leaf and active shoot and reactive oxygen species accumulation in wheat crop plants, causing an oxidative stress.

Many workers have acknowledged the role of PGPR in drought, temperature and pesticide stress tolerance of crop plants and significant advancements have been made in this area but there are still gaps left. Bacterially mediated tolerance to these abiotic stresses and the modes of action are still remaining elusive. Studies documenting role of PGPR in abiotic stress tolerance are limited. Temperature, draught and pesticide stress tolerance using PGPR could be an emerging field of stress management. However, molecular mechanisms leading to PGPR mediated stress tolerance are poorly understood. Thus, we strongly recommend that PGPR could be further developed as a possible potential antistress agent, which is not only cost effective but also easily available and ecologically safe. Thus, application of PGPR may provide a mechanism through which rhizobacteria ameliorate phytostimulatory and stress tolerance potential, simultaneously. Therefore, the present study aimed to review the PGPR mediated alleviation of drought, temperature and pesticide stress of crop plants.

Drought tolerance:

Drought stress affects the water relations of a plant at cellular and whole plant level. PGPR are adapted to adverse conditions and protect plants from the deleterious effects of some environmental stresses (Glick and Bashan 1997). Timmusk and Wagner (1999) were the firstly showed that inoculation with *Paenibacillus polymyxa* in *Arabidopsis thaliana* confers drought tolerance, through the induction of drought responsive *ERD15* gene. Moreover, inoculation of *Azospirillum brasilense* Sp245 in wheat under drought stress resulted in a better water status and an elastic adjustment leading to better grain yield and mineral quality of magnesium, potassium and calcium (Creus *et al.*, 2004). Furthermore, plants treated with EPS producing bacteria display increased resistance to water stress (Bensalim *et al.*, 1998). The EPS producing strain *Pseudomonas putida* forms biofilm on the

root surface of sunflower seedlings and impart tolerance to plants against drought stress. The inoculated seedlings showed improved soil aggregation and root-adhering soil and higher relative water content in the leaves (Sandhya *et al.*, 2009). Moreover, Jackson *et al.*, (1994) reported that ACC deaminase containing soil bacteria decrease a significant portion of physiological damage to plants, high temperature and drought exposure. Mayak *et al.*, (2004) reported that the ACC deaminase activity of *Achromobacter piechaudi* confer draught tolerance in tomato and pepper plants. Moreover, pre-inoculation of maize seedling with *Azospirillum brasilense* under water deficient condition up to 75%, showed a significant root growth, total aerial biomass, foliar area, accumulation of proline in leaves and root, improved relative and absolute water content as compared to control plant. *Azospirillum brasilense* significantly prevent drop in water potential (Casanovas *et al.*, (2002). In another study pre inoculation of wheat seedling with *Azospirillum* sp. significantly increase relative water content, water potential and apoplastic water fraction, lower volumetric cell wall moduli of elasticity values (Creus *et al.*, 2004). PGPR mediated changes in the elasticity of the root cell membranes could be one of the first steps towards an enhanced tolerance to water deficiency. It is believed that the potential drought tolerant *Azospirillum* inoculation enhance host plant's root morphology, due to release of hormone like substance which stimulate endogenous hormone levels of the crop plants (Dobbelaere *et al.*, 1999; Cassan *et al.*, 2001). Studies revealed enhanced shoot, leaf biomass and photosynthetic activity of drought challenged grapevine by *Acinetobacter* and *Pseudomonas* bacteria (Rolli *et al.*, 2014). Razi and Sen (1996) reported that foliar application of *Klebsiella* sp. increased the grain yield, increased nutrient uptake and L-Proline content in rice crop plants.

On the other hand, under aerobic conditions, *A. brasilense* produces significant amounts of the small diffusible gas, nitric oxide, which has been shown to act as a signaling molecule in an IAA induced pathways involved in adventitious, lateral root and root hair development in *Azospirillum*-inoculated tomato plants (Creus *et al.*, 2004; Molina-Favero *et al.*, 2008). Sueldo *et al.*, (1996) reported that under water deficit condition in wheat seedlings, there are certain changes in phospholipid composition in the root, increase in phosphatidylcholine content and a reduction of phosphatidylethanolamine takes place. However, inoculation with *Azospirillum* prevented these changes (Pereyra *et al.*, 2006). Pre-inoculation of PGPR reduce the damage of cell membrane and lipid composition of plants caused by water deficiency (Moran *et al.*, 1994). Changes in proton efflux activities in wheat and cowpea, reduced membrane potentials in wheat seedlings, as well as changes in phospholipid content in the cell membranes of cowpea, have been observed upon inoculation with *Azospirillum* (Bashan *et al.*, 1992, 2014).

Rocha *et al.*, (2007) reported that the treatment of the sugarcane plant with beneficial endophytic bacteria *Herbaspirillum* spp. and *Gluconacetobacter diazotrophicus* resulted in the induction of drought resistance. Studies of Callaghan (2016) showed that *Rhizobium* enhance the drought tolerance by intracellular accumulation of osmoprotectant compounds such as trehalose, glycine betaine. Seed treatment with drought tolerant isolates of *Trichoderma harzianum* reduce the severity of drought stress in wheat plants and improved seed germination. Moreover, studies revealed that when drought-tolerant *Bacillus* spp. as *Bacillus amyloliquefaciens*, *Bacillus licheniformis*, *Bacillus thuringiensis*, *Paenibacillus favisporus* and *Bacillus subtilis* inoculated with maize seedlings, showed increased plant

biomass, increased relative water content, increased leaf water potential, root adhering soil/root tissue ratio, aggregate stability, decreasing leaf water loss. Such inoculation reduced the activity of antioxidant enzymes such as ascorbate peroxidase, catalase, glutathione peroxidase. *Bacillus* sp. effect on osmoregulation through increased proline, sugars, free amino acids and decreased electrolyte leakage at minimal water potential (Vardharajulaa *et al.*, 2011).

High temperature tolerance:

Heat stress adversely affects the growth and development of crop seedlings. Studies well documented that a temperature increase of 3-4⁰C could cause crop yields to fall by 15–35% in Africa and Asia and by 25–35% in the Middle East (Ortiz *et al.*, 2008). There are few physiological and molecular factors involved in signaling cascades of transcriptional control for tolerance of temperature stress such as stress proteins, osmoprotectants, free-radical scavengers, ion transporters (Wang *et al.*, 2004). Kennedy *et al.*, (1991) reported that *Pseudomonas putida*, able to survive at low soil moisture potential, colonized the rhizoplane and soil adhering to wheat and sunflower roots and increased the percentage of stable soil aggregates. Moreover, Bensalim *et al.*, (1998) reported the pre-inoculation with *Burkholderia phytofirmans* of potato showed the increase in stem length, shoot and root biomass at high temperature. Yanez-Mendizabal *et al.*, (2012) reported that *Bacillus* spp. have proven to be ideal candidates for development as stable and efficient biological products because of their ability to produce heat-resistant endospores. Certain PGPR produce multifunctional polysaccharides such as exo polysaccharides, an active signaling molecule plays pivotal role in biofilm formation; root colonization, interaction of microbes with roots appendages and constitute shielding from desiccation and abiotic stress conditions (Qurashi and Sabri 2012).

Although the exact mechanisms of plant heat stress tolerance enhancement by PGPR remain largely speculative, possible explanations include formation of bacterial biofilm i.e. extracellular matrix (Vurukonda *et al.*, 2016). In particular, an extracellular matrix formed by bacterial biofilm can provide almost infinite range of macromolecules beneficial for plant development and growth. Biofilms contain sugars and oligo- and polysaccharides that can play various roles in bacteria-plant interactions such as in improving water availability in root medium. The water retention capacity of some polysaccharides can exceed several-fold their mass. It has been demonstrated that even small polysaccharide alginate content in the biofilm facilitates maintenance of hydrated microenvironment (Chang *et al.*, 2007). *Pseudomonas aeruginosa* strain 2CpS1, has been reported to grow at a temperature as high as 42°C. Studies revealed that wheat seed treatment with *P.aeruginosa* shown significant increase in plant height, root length, leaf area, total dry matter, total chlorophyll content, relative water content and decline in the cell membrane injury under high temperature condition (Meena *et al.*, 2015).

Pesticide tolerance:

Exposure of pesticides poses serious threats to human health. The presence of the toxic metals, above critical concentration in soil adversely affects the environment. Ahmed and

Khan (2011) reported that *Mesorhizobium* sp. revealed resistance to herbicides (metribuzin and glyphosate), insecticides (imidacloprid and thiamethoxam) and fungicides (hexaconazole, metalaxyl and kitazin) up to certain level. Tolerance of organophosphorus pesticides (guthion, methyl parathion and dimethoate) on *Pseudomonas* and *Flavobacterium* sp. has also been well documented (Nazarian and Mousawi, 2005). Likewise, both *Rhizobium* sp. specific to chickpea and *Rhizobium* sp. specific to green gram tolerated aldrin (Juneja and Dogra, 1978). Moreover, Boldt and Jacobsen (1998) also reported a tolerance *Pseudomonas* strains to sulfonylurea herbicides (metsulfuron methyl, chlorsulfuron and thifensulfuron methyl). The variation in tolerance to pesticide by rhizobacteria could probably be due to the fact that rhizobacteria adopt different strategies to overcome the toxic effects of pesticides and such mechanisms included biodegradation (Yang and Lee, 2008) and enzymatic hydrolysis (Dumas *et al.*, 1989; Herman *et al.*, 2005) of pesticide. For instance, organophosphorus hydrolase, an enzyme isolated from *Pseudomonas diminuta* and *Flavobacterium* sp. possesses the ability to hydrolyze different organophosphorus insecticides (Dumas *et al.*, 1989). Similarly, dicamba monooxygenase, an enzyme extracted from *Pseudomonas maltophilia* completely inactivated the herbicidal activity of dicamba (Herman *et al.*, 2005). Studies revealed that phytotoxicity caused by clodinafop-propargyl, herbicide can be overcome by phytohormone such as 2,4-Dichlorophenoxyacetic acid. On the other hand, glyphosate induced phytotoxicity on fava bean can be reversed by GA3 alone or in a mixture with cytokinin. Brassinosteroids reduce the damaging effect of herbicide namely simazine, butachlor, or pretilachlor in rice (Varshney *et al.*, 2012). Studies revealed that some of the everlasting pesticides may cause changes in the soil microbial community including inhibition of beneficial potential of PGPR. Some of the species are known to either tolerate or degrade various pesticides. That includes several PGPR species such as *Pseudomonas*, *Flavobacteria*, *Azotobacter*, *Acetobacter*, *Arthrobacter*, *Alcaligenes*, *Bacillus*, *Enterobacter* and *Klebsiella*. Many species of *Pseudomonas*, *Bacillus* and *Azotobacter* can grow and survive at extreme environmental conditions. by producing cyst. *Azotobacter* species can tolerate and survive in extreme environmental condition by producing cysts (Chennappa *et al.*, 2014). Chennappa *et al.*, (2014) reported that several species of PGPR *Azotobacter* sp. were resistant to 4 different pesticides which are generally used for the paddy cultivation are 1 to 5 % of Pendimethalin, Chloropyrifos, Glyphosat and Phorate. Presence of these PGPR in crop fields may reduce the use of chemical fertilizers and pesticides, which are otherwise hazardous as well as costlier. In order to remove toxic and hazardous effect of costlier fertilizer and pesticide or to reduce N and P fertilizer rates to crop plant, potential PGPR can be used for sustainable agriculture. Application of PGPR can significantly overcome the problems due to drought and temperature stress induced crop yield loss.

Conclusion

In this review we are presenting the beneficial potentials of PGPR in context with stress tolerant response. PGPR could play a significant role in alleviation of drought, temperature and pesticide stress in plants. After thorough review of literature, it appears that the PGPR are of great relevance and multi-potential in terms drought, temperature and

pesticide tolerance, as well as their phytostimulatory potential for agricultural productivity. Application of these PGPR may be useful as bio inoculum for high crop yield as well as drought, temperature and pesticide tolerance for sustainable agricultural production are of great relevance.

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