Chapter 10
Microbial Alleviation of Abiotic and Biotic Stresses in Rice

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Abstract More than 90% of the cultivated area is affected globally by environmental constraints. For instance, abiotic and biotic stresses are major processes that decline agricultural production. Drought, salinity, heat, cold, acidity, and sodicity are major abiotic factors, while insects and pathogens are biotic factors. Rice, a staple food for more than half of the world's population, is highly susceptible to abiotic and biotic stresses. Here, we review stresses in rice and mitigation strategies, with focus on microbes to alleviate stresses. Abiotic stresses in rice are alleviated by microbes belonging to genus *Bacillus, Pseudomonas, Enterobacter, Ochrobactrum, Alcaligens, Paecilomyces, Burkholderia, Achromobacter, Azospirillum,* and *Glomus.* This alleviation proceeds through an accumulation of ascorbate, proline, ethylene, auxin, and stomata conductance of leaf, and by producing antioxidant enzymes, 1-aminocyclopropane-1-carboxylate deaminase, β-aminobutyric acid, salicylic acid and siderophores. Biotic stresses in rice include brown spot, leaf blast, blunt, leaf blight, sheath blight, sheath rot, root rot and seedling disease. They are suppressed by *Pseudomonas*, *Streptomyces, Bacillus, Trichoderma*, *Aspergillus* by inhibiting mycelia growth, iron competition, producing antibiotics, phytohormones, metabolites, and enzymes.

Keywords Rice · Microbial interventions · PGPMs · Abiotic stress · ACC deaminase · Rice diseases · Biocontrol · Induced systemic tolerance · Siderophore · Stress enzymes

Abbreviations

ACC	1-aminocyclopropane-1-carboxylate
ACS	1-aminocyclopropane-1-carboxylate synthase
IAA	Indole acetic acid

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PGPMs	Plant growth-promoting microorganisms
PGPR	Plant growth-promoting rhizobacteria

10.1 Introduction

Presently, the world population is about 7.6 billion which is expected to increase by 20.8% to 9.6 billion in 2050 (UN Report 2013). Most of this increase (93%) will occur in developing countries, whose share of population is projected to increase from 78% in 1990s to 83% in 2020. Rice is a staple crop for over half of the world's population and is prone to a variety of abiotic and biotic stresses (Lafitte et al. 2004; Kumar et al. 2016, 2018a). High salinity, submergence, cold and drought stresses are the major abiotic factors, whereas insects and pathogens are the major biotic factors causing threat to rice crop thereby reducing food security for growing human population (Sanghera et al. 2011; Shanker and Venkateswarlu 2011; Wani et al. 2013; Kumar et al. 2018b). According to various estimates, we have to produce 40% more rice by 2030 and 70% more by 2050 to satisfy the growing demand without affecting the resource base adversely (FAO 2009; Tilman et al. 2011). We have to achieve this demand from less land, labour, water and fewer chemicals.

To meet the challenge of producing more rice from affected lands, a wide range of adaptations and mitigation strategies are required. Efficient resource management and rice crop improvement for evolving transgenic may be one of the alternatives to overcome abiotic and biotic stresses to some extent. However, such strategies being long drawn and cost intensive, there is a need to develop simple and low-cost biological methods for the management of abiotic stress and it can be used on short term basis (Kumar et al. 2017a, 2019). Plant growth-promoting microorganisms (PGPMs) are one of the best options to alleviate abiotic and biotic stresses in agricultural crops including rice with higher yield potential and greater yield stability, if we can exploit their unique properties of tolerance to extremities, ubiquity, genetic diversity, and their interaction with agricultural crops (Kumar et al. 2016). Researchers from all over the world have made great efforts in understanding the mechanisms of PGPM responses to abiotic and biotic stresses in rice (Sarkar et al. 2018; Pandey et al. 2013; Khan et al. 2016; Kakar et al. 2016; Reddy et al. 2007; Law et al. 2017; Saravanakumar et al. 2007). In this chapter, we emphasized a different abiotic and biotic stress mitigation strategy through microbial intervention particularly for rice crop and its mechanistic understanding is represented in Fig. 10.1.

10.2 Plant Stress

Stress can be defined as any unfavorable condition or substance affecting or blocking the metabolism, growth or development of a plant (Lichtenthaler 1996). Accordingly, climate and environmental factors regulate the geographical

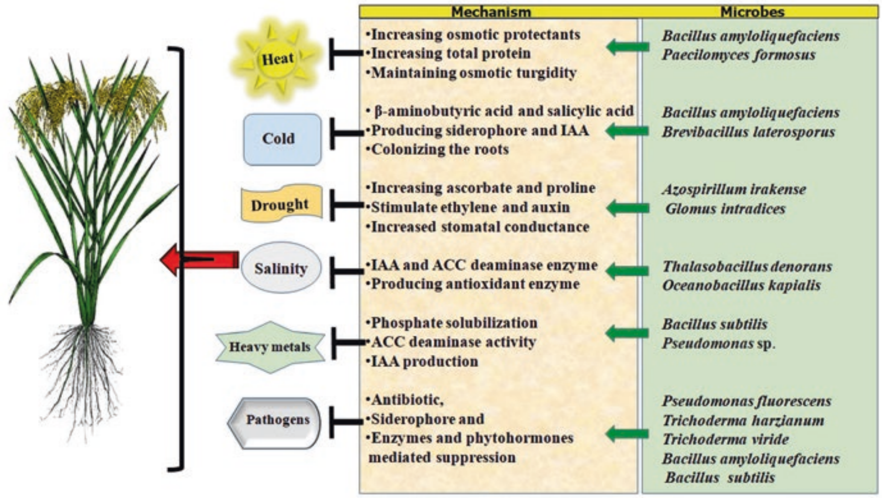

Fig. 10.1 Mode of action of plant growth-promoting microorganisms in rice under biotic and abiotic stresses. Lines with bar indicates inhibition of those environmental stresses by means of plant growth-promoting microorganisms and arrows represent secreted compounds and elicitors by plant growth-promoting microorganisms. IAA: Indole acetic acid; ACC: 1-aminocyclopropane-1-carboxylate

distribution of plants (Walther et al. 2002). Thus, unfavorable environmental changes can affect plant growth and crop yield (Duque et al. 2013). Reactive oxygen species molecules are generally formed in response of oxidative stress (Kumar et al. 2019). Drought, heat shock and salinity are the major oxidative stresses responsible to release reactive oxygen species in the system. Some of the well-known reactive oxygen species molecules that result in membrane and macromolecular damage include hydrogen peroxide (H_2O_2), hydroxyl ion (OH^-) and superoxide anion (O_2^-) (Kumar et al. 2019; Blokhina et al. 2003; Karim 2007; Farnese et al. 2016). In order to increase rice stress tolerance and decrease the detrimental effect of toxic reactive oxygen species compounds, they utilize several antioxidant defense mechanisms in order to scavenge reactive oxygen species.

Several antioxidants that plant mainly uses are namely, ascorbate peroxidase, superoxide dismutase, glutathione reductase and catalase (Kumar et al. 2019) and non-enzymatic antioxidants such as carotenoids, glutathione, ascorbate and anthocyanin (Karim 2007; Mittler 2002; Blokhina et al. 2003; Gould et al. 2002). Whereas biotic stress includes parasitic organisms that are pathogenic and causes plant diseases; this involves a wide spectrum of microbes (fungi, bacteria, viruses, nematodes, protozoa and insects) (Adhya et al. 2018). Every year pathogenic diseases cause significant crop losses all over the world (Agrios 2005; Karim 2007). As we know, the nature of the parasitic organisms is to utilize the host plant for feeding, sheltering, multiplying and growing that causes significant host damage and ultimately leads to death. In these conditions, Plant growth-promoting microorganisms may act as biocontrol agents and mitigates the biotic stress in the plant (Kumar et al. 2013).

10.3 Plant Growth-Promoting Microorganisms

Plant growth-promoting microorganisms (PGPMs) are beneficial microbes that have the distinctive ability to support plant development directly and indirectly. They live in the rhizosphere zone which is rich with plant exudates such as sugars and amino acids or some microbes establish themselves as endophytes within the plants in order to survive in the root rhizosphere by means of penetrating/burrowing tissues of plants, that contributes to plant's nutrition, environment adaptability and survivability. These microbes extend their biological activities in order to survive in the rhizosphere, influencing plant survival and development (Kumari et al. 2015; Khan et al. 2016; Babalola 2010; Kumar et al. 2013). The process in which PGPMs play a role in stimulating variety of abiotic stress tolerance in plants is referred to as induced systemic tolerance (Kumar et al. 2012; Yang et al. 2009).

These PGPMs include multiple bacterial determinants such as *Bacillus amyloliquefaciens* and *Brevibacillus laterosporus*, *Azospirillum brasilense* that are involved in induced systemic tolerance by means of production of indole-3-acetic acid (IAA), 1-aminocyclopropane-1-carboxylic acid (ACC)-deaminase, phosphate solubilization, and volatile, exo-polysaccharides, siderophores production (Farag et al. 2013; Kumari et al. 2015; Nadeem et al. 2016).

These traits help the plants to overcome stress. Certain PGPMs function is to synthesize ACC deaminase that catalyzes the transformation of ACC (ethylene biosynthesis precursor) to ammonia and α-ketobutyrate. Thus, plants with decreased concentrations of ethylene would finally overcome the inhibition of abiotic stress by associating with ACC deaminase-producing bacteria such as *Pleosporalean ascomycete, Alcaligenes*, *Rhodococcus* and *Variovorax* (Barnawal et al. 2014; Nadeem et al. 2010a; Senthilkumar et al. 2009; Glick et al. 2007; Mayak et al. 2004). Considerable attention has been made to the isolation of ACC deaminase-producing microbes for their utilization in direct plant growth promotion under unfavourable environments (Ali et al. 2016; Hardoim et al. 2008; Nadeem et al. 2010b).

In addition to ACC deaminase enzyme, they also produce a variety of substances such as plant hormone–indole acetic acid (Enebe and Babalola 2018), siderophore (Stajkovic-Srbinovic et al. 2014), PO_4^{2-} solubilizing enzyme, salicylic acid (Ekinci et al. 2014) and microbiocidal/biostatic enzyme (Moustaine et al. 2017). By trapping and integrating nitrogen into the plant *via* nitrogen fixation, some of these microbes contribute to plant nutrition (Kumar et al. 2017b; Richardson et al. 2009). The subsequent impacts of this specific form of plant-bacterial association would provide plants with a source of nitrogen (ammonia) (Hardoim et al. 2008). PGPMs also help to sustain the plant's inherent resistance to pathogenic and environmental problems. Some of these organisms are excellent in secretion of polysaccharide substances and formation of biofilm that helps to maintain stability during stress conditions (Kumar et al. 2013; Kasim et al. 2016).

The presence of microbes as bio-inoculant decreases metal stress in plants as they can produce metal rich solution through the biological oxidation of sulfur containing ore and plays a crucial role in metal immobilization and make them unavailable using polymeric substances and other chemicals such as siderophore production (Fashola et al. 2015) and have a significant contribution to bio-hydrometallurgy.

10.4 Role of Microbes in Alleviating Abiotic Stresses in Rice

10.4.1 Drought Stress

World's 64% of the total land area has been affected by water deficit/drought stress (Mittler 2006; Cramer et al. 2011). It has a major impact on soil nutrients availability and transportation through water to the roots. Thus, drought stress reduces the movement of nutrients and water-soluble supplements, such as, NO^{3-}, SO_4^{2-}, Ca, Si, and Mg which are considered essential for growth (Nasim et al. 2017). It also forms free radicals and reactive oxygen species that can further damage the rice plant by membrane lipid peroxidation or degradation of important structural and functional proteins (Kumar et al. 2019; Nair et al. 2008).

Drought stress have direct effects on plant physiology in rice as it ceases the cell growth because of altering the cellular turgidity and regular growth processes (Hsiao and Xu 2000; Rahdari and Hoseini 2012; Jabran et al. 2017). Among the various crops, rice is likely to be more vulnerable to drought stress (Showler 2016). Drought stress restricts the plants growth and development by interrupting biochemical processes such as low nitrate uptake from dry soils, which further reduces the rate of photosynthetic pigmentation, is an indication of photo-oxidation. It also influences some enzymatic activities such as nitrate reductase activity, due of low uptake of NO_3^- from dry soils which restricts plant growth and development (Ali et al. 2016; Awais et al. 2017a, b).

Furthermore, the grain filling stage of rice is adversely affected due to water shortage that favors the remobilization of stored carbohydrates into grains (Nasim et al. 2016a; Yang et al. 2012). Four components are assumed to be mainly involved in this procedure: (1) starch formation; (2) ADP-glucose-pyrophosphorylase; (3) sucrose formation; (4) starch branched compound (Taiz and Zeiger 2002). Under drought stress, decreased sucrose synthase activity lower the rate of grain filling and it also leads to inactivation of ADP-glucose-pyrophosphorylase which in turn causes developmental losses (Ahmadi and Baker 2001; Nasim et al. 2016b). Thus, drought conditions result in diminished photosynthesis, stomata closure and disturb cellular ionic balance because of low water content of soil (Flexas et al. 2004), consequently, reducing plant growth and development, obstructing grain filling and ultimately reducing grain yield.

One of the major weapons to mitigate this abiotic stress is beneficial microbes and some examples of these are presented in Table 10.1. Bacterial inoculation in rice enhanced the production of plant hormones such as IAA that improved lateral

Table 10.1 Beneficial microbes for alleviation of abiotic stress and plant growth-promotion in rice

Beneficial microbes	Abiotic stresses	Impact on plant-growth promotion in rice	Reference
Azospirillum brasilense, *Glomus intraradices*	Drought	Increased stomata conductance, photosynthesis, shoots fresh weight and plant vigor	Ruíz-Sánchez et al. (2011)
Bacillus amyloliquefaciens Bk7, *Brevibacillus laterosporus* B4	Drought, cold	Improved the seedling height and shoot number; alleviate chlorosis, wilting, necrosis and rolling of leaves	Kakar et al. (2016)
Bacillus pumilus	Salt	Improved growth and nutrient uptake	Khan et al. (2016)
Pseudomonas sp., *Burkholderia caryophylli, Achromobacter piechaudii*	Salt, drought	Reduce endogenous ethylene levels in plants and promotes root growth	Wu et al. (2009)
Ochrobactrum sp., *Bacillus* sp. (CdSP9, PbSP6, and AsSP9)	Heavy metals (cadmium, lead, arsenic)	Increase in germination percentage, relative root elongation, amylase and protease activities	Pandey et al. (2013)
Pseudomonas strain (TDK1)	Salt	Increases plant height, root length and leaf area	Sen and Chandrasekhar (2014)
Bacillus amyloliquefaciens	Salt, drought, desiccation, heat, cold	Increased accumulation of osmolytes (proline, soluble sugars, glycine betaine, trehalose, etc.); helped plant to overcome abiotic stresses by maintaining osmotic turgor	Tiwari et al. (2017)
Pseudomonas fluorescens	Drought	Encouraged the expression of abscisic acid synthetic genes particularly at the stage of reproduction by the plant	Saakre et al. (2017)
Thalassobacillus devorans (NCCP-58), *Oceanobacillus kapialis* (NCCP-76)	Salt	Increased germination ability, root and shoot growth, protein, and chlorophyll contents as well as nutrient contents with reduced sodium ion accumulation in the plant	Shah et al. (2017)
Bacillus sp.	Salt	Aided the alleviation of salt stress by increasing the biomass and growth of rice seedling via production of indole acetic acid and ACC deaminase enzyme	Misra et al. (2017)
Bacillus thuringiensis	Salt	Promotes plant-growth	Raheem and Ali (2015)

(continued)

Table 10.1 (continued)

Beneficial microbes	Abiotic stresses	Impact on plant-growth promotion in rice	Reference
Alcaligenes faecalis, Bacillus pumilus, Ochrobactrum sp.	Salt	Results in shoot and root elongation	Bal et al. (2013)
Pseudomonas pseudoalcaligenes, Bacillus pumilus	Salt	Increase in root length and promotes growth and yield	Jha et al. (2013)
Burkholderia pyrrocinia, Pseudomonas fluorescens	Water	Induces increased production of carotenoids and chlorophyll b and promotes plant growth by maintaining the integrity of enzymes and proteins of cell wall	Rêgo et al. (2018)
Azospirillum brasilense	Osmotic	Increases the root elongation, root surface area, root dry matter, and development of lateral roots and root hairs	Cassan et al. (2009)
Bacillus subtilis, Bacillus megaterium, Bacillus sp.	Heavy metals	Promotes plant growth and development along with increased dry matter, grain yield and phosphorus uptake	Asch and Padham (2005) and Becker and Asch (2005)
Enterobacter sp.	Salt	Promoted the growth of rice seedling and reduced ethylene production and antioxidant enzyme activities in the plant	Sarkar et al. (2018)
Pseudomonas fluorescence, P. jessenii, P. synxantha, Bacillus cereus, Arthrobacter nitroguajacolicus	Drought	Enhances plant growth by induction of stress related enzymes and activation of antioxidant defense systems and improves stability of membranes of plant cells	Gusain et al. (2015)
Bacillus amyloliquefaciens NBRISN13	Salt	Enhanced proline accumulation and upregulation or repression of set of stress responsive genes in leaf blade.	Nautiyal et al. (2013)

roots formation and root growth which ultimately increased leaf water content and decreased leaf water potential by increasing water uptake (Dossa et al. 2017). IAA produced by *Azospirillum* enhances tolerance of rice under drought stress, resulting in higher mineral quality and better grain yield (Dimkpa et al. 2009). Inoculation with arbuscular mycorrhizal fungus, *Azospirillum brasilense*, considerably enhances rice growth by increase stomatal conductance that improved growth parameter by 80% under water deficit condition (Ruíz-Sánchez et al. 2011).

During water stress conditions, lipid peroxidation increases with decrease in glutathione contents in plants; while inoculation with arbuscular mycorrhizal fungus, ascorbate and proline contents (as protective components) increase to bypass the deleterious effect of water limitation (Ruíz-Sánchez et al. 2011). Inoculation of rice plants with *Bacillus amyloliquefaciens* Bk7 and *Brevibacillus laterosporus* B4 in water deficit conditions, improve shoot number, seedling height and showed least symptoms of chlorosis, necrosis, wilting and rolling of leaves (Kakar et al. 2016). The endophytic Plant growth-promoting rhizobacteria (PGPR), *Azospirillum irakense* under drought stress trigger the expression of polygalacturonase encoding genes in rice inoculated roots (Sekar et al. 2000). Rice roots inoculated with endophytic PGPR, *Herbaspirillum seropedicae* stimulate the gene expression receptive to ethylene and auxin and results in suppression of defense-related thionins and proteins PBZ1 (Brusamarello-Santos et al. 2012). Therefore, above reports suggest that drought stress in rice might be mitigated through different microorganisms by modulating plant defense responses.

10.4.2 Cold Stress

One of the most significant environmental factors that hamper agricultural production by affecting plant growth is cold stress which affects 57% of the total land area of the world (Mittler 2006; Cramer et al. 2011; Hashimoto and Komatsu 2007). Low temperature impacts the agronomic development of crops including rice. The survivability of plants at extreme low temperature relies upon it's adaptability to cold stress (McKhann et al. 2008). Plants exposed to low temperatures showed increased penetrability that is correlated with the injury of the plasma layer, a major problem for maintaining ionic equilibrium and reversing the damage caused due to cold stress. The unsaturated or saturated fatty acids tend to rearrange themselves that causes a change in the plasma layer *viz.* thickness alterations that result in declining turgidity of the cell (Hughes and Dunn 1996). This plasma layer modification tends to be cold sensitive in several rice varieties that initiates a response by specific gene expression during cold stress (Chinnusamy et al. 2006). Thus, cold stress directly or indirectly hampers geographical distribution of rice that overall reduces the rate of harvest (Pearce and Fuller 2001). Microbes mediated stress responses are one of the best ways to cope up with this cold stress (Table 10.1).

It was reported that, PGPR consortium of two different bacterial strains *Brevibacillus laterosporus* B4 and *Bacillus amyloliquefaciens* Bk7 attributed to the production of high amount of siderophore and IAA and effectively colonized the roots of the plant under cold stress (Kakar et al. 2016). They also induced systemic tolerance in rice under chilling stress and enhanced growth and development. This strain is also well known for the biofilm formation and the production of biochemical elicitors (β-aminobutyric acid and Salicylic acid) in rice for cold stress tolerance. Catalase and superoxide dismutase activities in plants increased by 3.6- and 3.0-fold respectively, after inoculation of Bk7. *Bacillus amyloliquefaciens* NBRI-SN13 (SN13) improved growth of rice seedling under cold stress by increasing proline content (Tiwari et al. 2017). It has been reported that, some phytohormones like abscisic acid, jasmonates, salicylic acid, and ethylene play a key role in cold, salt, heat and drought stresses response in several plants including rice to sustain a balanced and healthy growth of plant (Lata et al. 2011; Kohli et al. 2013).

10.4.3 Heat Stress

Most of the cereal crops especially rice, are at major risk due to annual increase in temperature and its deleterious effect on overall growth, development and productivity (Fahad et al. 2015a, b, 2016a, b, c, d, 2018; Watanabe and Kume 2009; Mahmood et al. 2007). It is expected that the rate of rice yield will decrease by 41% before the end of twenty-first century due to drastic increase in temperature (Ceccarelli et al. 2010). Even it is predicted that all the cropping zone of rice could completely wipe out if the temperature continues to be this extreme (Aghamolki et al. 2014).

The ideal temperature for appropriate rice growth and development ranges between 27 °C to 32 °C (Yin et al. 1996). Further higher temperatures than the given range could have severe impact on all the stages of rice; from growth stage to maturation and then till harvesting. Heat tolerance ability of rice plant is very sensitive at different growth stages. It is highly temperature sensitive particularly during generation and blossoming which could lead to permanent damage and reduced yield (Porter 2005). Heat stress also widely influences both vegetative as well as reproductive stages of rice; like at vegetative stage, a prolonged exposure to high day temperature can damage leaf properties, while a short time period of warmth could cause premature birth of botanical buds and open blooms in the middle of conceptive stage (Guilioni et al. 1997). Blooming and booting stages of rice are found to be more sensitive to high temperatures *i.e.,* conceptive stage is more susceptible to temperatures than the vegetative stage (Ali et al. 2016; Shah et al. 2011; Peng et al. 2004).

Microbes mediated mitigation strategy is one of the alternate ways to alleviating the heat stress. Tiwari et al. (2017) reported that inoculation of *Bacillus amyloliquefaciens* in rice increased accumulation of osmotic protectants such as proline, soluble sugars, glycine betaine, trehalose under heat stress conditions which helps rice plant to overcome inert stresses by maintaining osmotic turgidity. Inoculation with endophytic fungus, *Paecilomyces formosus* LWL1 in rice grown under no stress and high heat stress conditions, improved growth attributes *viz.* plant fresh weight, height, chlorophyll content and dry weight. Additionally, it also effectively mitigated heat stress by minimizing the endogenous level of stress-indicating components such as jasmonic acid, abscisic acid and increasing total proteins content by 18.76%–33.22% (Waqas et al. 2015). Such beneficial microbes might be very useful at high environmental temperature stresses to maintain an effective and sustainable production of rice.

10.4.4 Salinity Stress

Globally 6% of the total land area has been affected by salinity (Mittler 2006; Cramer et al. 2011). Salinity affected area has been increased by almost 34 million ha of irrigated land (FAO 2009). Increased annual loss of crop production in irrigated lands is due to land degradation by salinity (Qadir et al. 2014). Saline soils have a number of soluble salts such as Ca^{2+}, Na^+, $Mg^{2+}$and anions SO_4^{2-}, HCO_3^-, Cl^- with large amounts of K^+, NO_3^-, CO_3^-. A soil can be referred as saline if it has an osmotic pressure of approximately 0.2 M Pa (~40 mM NaCl) or the EC 4 dS m^{-1} or more (USDA-ARS 2008). The pH of saline soils ranges between 7–8.5 (Mengel et al. 2001; Ghosh et al. 2016). Increased salt accumulation is very common in arid and semi-arid zones, where high evaporation and low precipitation occurs. Moreover, this process of salt deposition has been also favored by weathering of the parental rocks (Rengasamy 2002).

Rice is considered to be one of the most sensitive crops to salinity (Rahnama et al. 2010). Salt stress causes change in plants physiological processes by suppressing seed germination (Shannon and Grieve 1998). The damage caused by Cl^- initialization in rice can be figured by broad leaf cutting, indicates burning whereas Na^+ accumulation can be characterized by rolling and molting of leaves (Acosta-motos et al. 2017). Salt stress reduces the rice leaf development, which leads to stomatal closure and in turn decreases the rate of photosynthesis (Rahnama et al. 2010). The major components that regulate salt accumulation are reduced salt uptake, improved Na^+/K^+ proportion, antioxidant regulation system, tissue resistance, proficiency of water utilization to minimize the concentration of NaCl in plants (Ismail et al. 2007; Hashmi et al. 2017). During the whole life cycle of plants, several phytohormones play a crucial role as they regulate the key processes of response in plants under abiotic stresses, including plant responses to salinity stress.

Salinity stress responses involve the synthesis of ethylene, as stress hormone, which also regulates the plant growth and development (Hardoim et al. 2008). Biosynthetic pathway of ethylene involves the conversion of S-adenosyl-methionine by the enzyme ACS (1-aminocyclopropane-1-carboxylate synthase) into ACC (1-aminocyclopropane-1-carboxylate), which is the immediate precursor of ethylene to α-ketobutyrate and ammonia. However, in rice plants, under salt stress conditions, ethylene involves in endogenous regulation of plants stable equilibrium which results in reduced growth of root and shoot, which finally impacts on yield productivity.

In plants, ACC is degraded and sequestered by bacteria producing ACC deaminase in order to supply energy and nitrogen under salt stress (Glick 2005). Further, by eliminating ACC, the harmful effect of ethylene is reduced by the bacteria that improves plants stress tolerance and promotes growth by inhibiting salt-induced growth. Soil microbes belonging to genera *Bacillus*, *Alcaligenes*, *Rhodococcus* and *Variovorax* have ACC deaminase producing activity which is effective to confer salt stress in rice (Belimov et al. 2005). *Ochrobactrum* sp. was also previously reported to have ACC deaminase producing ability (Jia et al. 2013). It was reported that under salt stress, the rice seedlings showed improved plant biomass and salt tolerance capability by inoculation with class 2 endophyte Ascomycota (*Fusarium culmorum* FcRed1) (Redman et al. 2011). Rice root inoculated with *Pleosporalean ascomycete*, isolated from the roots of halophyte *Suaeda salsa* belongs to family Amaranthaceae, significantly increased the proline accumulation followed by increased photosynthetic pigment (chlorophyll and carotenoids) levels under salt stress condition (Jogawat et al. 2013; Kumar et al. 2012). The fungal isolate from roots of halophyte *Suaeda salsa* could endophytically colonize rice roots and improved plant health under salt stress (Qin et al. 2016). A report also showed that, inoculation of strain *Pseudomonas fluorescens* MSP-393 in rice under salt stress, favored root colonization, the potential strain also synthesizes complex osmolytes such as glycine, alanine, serine, glutamine, asparagine and threonine in their cytosol along with increased production of salt stress protein for effective nullification of the negative impact of high osmolarity (Paul and Nair 2008).

Three promising isolates with multiple plant growth promoting traits *viz.* *Bacillus*, *Alcaligenes* and *Ochrobactrum* sp. promoted rice growth at 150 mM NaCl under axenic conditions and showed increased root elongation assay (Bal et al. 2013). Inoculation of *Bacillus pumilus* in rice seedlings under salt stress showed a progressive potential for the limitation of Na^+ concentration in rice leaves that favored several antioxidant enzyme activities *viz.* superoxide dismutase, catalase, peroxidase that reversed the effect of salinity stress and enhanced plant growth (Khan et al. 2016). Furthermore, it was reported that inoculation of the strain *Enterobacter* sp. P23 in rice seedling showed potential traits of IAA production, siderophore production, phosphate solubilization, ACC and NH_3 production, which decreases stress-induced ethylene and promoted growth and development (Sarkar et al. 2018). Inoculation of two more promising strains of *Bacillus i.e., Oceanobacillus kapialis* (NCCP-76) and *Thalassobacillus devorans* (NCCP-58) in rice, improved root elongation and shoot length under NaCl stress (Shah et al. 2017).

10.4.5 Heavy Metal Stress

Metal industries, agrochemical industries mainly pesticides, sewage sludge and other various sources discharge metalloids and heavy metals, which causes a critical threat to the environment as well as human health (Kumar et al. 2017a). The concentration of the toxic metals in soil results in absorption by the roots which is then transported to different parts of the plant leading to diminished plant metabolism, impaired growth and reduced yield production in rice (John et al. 2012). In rice plants, some of the heavy metals play a major role in supplement of micronutrients (Prasad 2013); although presence of some heavy metals (Cd, Pb, Ni, Cu, Al, Zn) in small quantities have harmful impact on rice crop (Kovács et al. 2009; Lakho et al. 2017). Plants exposed to heavy metal stress have shown penetrability expansion in plasma layer, as metal ions bind to OH^-group of phospholipids and SH-group of proteins and further replaces Ca^{+2} at the initial cell growth level. Altogether these conditions lead to imbalance in ionic homeostasis of cell and disturb the integrity layer of the cell (Lakho et al. 2017).

Among all the heavy metals present in soils, cadmium (Cd) is considered as the toxic one, as it reduces root and shoot growth of the plant and directly hamper productivity by reducing essential nutrient uptake and disrupting homeostasis as well (Hashmi et al. 2017). Increased accumulation of cadmium in the soil causes impaired growth and development of root, nutrients disruption as well as low metabolism of carbohydrate which result in reduced yield and biomass (Akram et al. 2019). Among the metals, lead (Pb) is considered as one of the abundant metals on earth and its ingestion also results in severe health issues in humans. Even its minimal concentration in rice soil leads to yield loss by disturbing seed germination, rate of photosynthesis, nutrition uptake, plant-water balance, activity of enzyme as well as cells proliferation (Patra et al. 2004). Several reports focused on the activity of enzymes under heavy metal stress and it is observed that metal stresses (Cd, Pb, Ni, Cu, Al) altered enzymatic activities. During seed germination the presence of heavy metals such as Cd, Pb, Zn and Cu severely impacts on the ratio of root/shoot length as well as height of young seedlings (Mahmood et al. 2007). Moreover, increased concentration of heavy metals has a major impact on vegetative growth, seed germination and rice yield.

In such circumstances, PGPR plays an important role in removal of metal toxicity and improve plant nutrition and development (Table 10.1). Many previous reports on bacteria in soil play a major role in mobilization and immobilization of metals for metals tolerant (Glick et al. 1998). PGPR helps in reduction of metal toxicity by two ways: (i) decrease in plants ethylene stress level in metal toxic soil by ACC deaminase activity resulting in longer roots development that allows better plant establishment during initial growth stages (Glick 2005), (ii) release of siderophores, an iron chelating compound that causes the increased accumulation of iron in roots of the plant in the metal polluted conditions (Fig. 10.1). The rice variety ‘Satabdi’ inoculated with cadmium resistant *Ochrobactrum* sp. CdSP9, arsenic resistant *Bacillus* sp. AsSP9 and lead resistant *Bacillus* sp. PbSP9, increased percent germination, overall biomass, relative root elongation, protease and amylase activity. It was also observed that all the three bacterial strains were positive to catalase and ACC deaminase activity (Pandey et al. 2013).

Several plant growth-promoting rhizobacteria, *Pseudomonas* spp., *Bacillus* spp., *Azotobacter* spp., *Phosphobacteria* spp., *Azospirillum* spp., *Aspergillus niger, Penicillium* spp. and *Gluconacetobacter* spp., isolated from rice roots rhizosphere were investigated for their role in heavy metal stress mitigation by production of IAA and catalase as well as growth enhancement in rice under heavy metal stress (Samuel and Muthukkaruppan 2011). Potent plant growth-promoting rhizobacterial strain *Enterobacter aerogenes,* isolated from heavy metal contaminated rice rhizosphere found to be resistant to high degree of Pb^{2+}, Cd^{2+}, As^{3+} up to 3800 μg mL^{-1}, 4000 μg $mL^{-1}$and 1500 μg mL^{-1}, respectively. Upon screening of the strains, it was found that they had different plant growth-promoting rhizobacterial attributes like ACC deaminase activity, phosphate solubilization, IAA production and nitrogen fixation which helped in enhancement of rice growth and development (Pramanik et al. 2018). Han et al. (2015) reported that inoculation of rice plants exposed to heavy metal stress (*viz*. 0.3 mM Cu^{2+}, Zn^{2+}, Co^{2+} or Ni^{2+}) with strain of wild type *Pseudomonas stutzeri* A1501 resulted in increased plant biomass, root length, fresh and dry weight of root and plant height of rice plant. Seed inoculation of two basmati rice cultivars (B-385 and KSK-282) grown in different concentration of nickel contaminated soil (0, 100, 250, 500, and 1000 ppm), with *Bacillus licheniformis* NCCP-59 showed enhanced seed germination and biochemical traits which reverses the effect of nickel toxicity; such strains can be used for the phytoremediation of Ni contaminated soil (Jamil et al. 2014).

10.5 Biotic Stresses in Rice

The term biotic stress described as "interactions between living organisms and plants that leads to partial plant damage which can cost upon plants survivability". Plants are utilized as host by the parasitic organisms for their feeding, sheltering, multiplying and growing purpose; which ultimately leads to senescence of the plants. Plant pathogens obtain nutrients by feeding on host plant organs and causes physical damage to the plant (Kumar et al. 2016). Biotic stresses can hence be referred to as external biological factors affecting plants by damaging the cells, tissues, organs, organelles or even whole plant. Biotic stresses generally include pathogenic-organisms *viz*., bacteria, fungi, viruses or even nematodes as well as insects (Kranner et al. 2010). Every year the reason behind major crop losses is due to attack of these disease-causing pathogens (Karim 2007). This is an interaction between pathogen-host at molecular and biochemical levels that causes certain physiological and metabolic changes which further leads to morphological disorders and even death of the plant host (Karim 2007). The stress responsive mechanisms for pathogen suppression can be categorized as (i) antibiotic mediated suppression, (ii) siderophore mediated suppression, (iii) enzymes and phytohormones mediated suppression (Dreher and Callis 2007).

10.5.1 Antibiotic-Mediated Suppression

Pseudomonas fluorescens can produce several antibiotic compounds *viz*. phenazine, 2, 4-diacetylphloroglucinol, pyoluteorin, pyrrolnitrin etc. (Kumar et al. 2018a; Mageshwaran et al. 2012; Meera and Balabaskar 2012). Balasubramanian (1994) reported that leaf and neck blast of rice can be controlled by *P. fluorescens* through production of Phenazine-1-Carboxylic acid. By producing these compounds, *P. fluorescens* not only enhances its own growth but also play a major role in protection of crops from pathogens. It inhibited the growth of *Xanthomonas oryzae pv. oryzae,* the causative agent of bacterial leaf blight disease of rice thereby maintains soil health (Kumar and Mishra 2014; Vasudevan et al. 2002; Velusamy et al. 2006). It was also reported that strain *P. aeruginosa* PUPa3 successfully suppressed the disease caused by *Sarocladium oryzae* and *Rhizoctonia solani* by producing Phenazine-1- Carboxamide antibiotics in rice (Megha et al. 2007). The causative agent of rice sheath blight, *S. oryzae* was found highly susceptible to the antibiotics produced by *P. fluorescence* (Nathan et al. 2011). *P. fluorescens* isolated from the rice rhizosphere showed effective antifungal activity and suppressing mycelial growth by 62–85% against *Rhizoctonia solani*, *Sarocladium oryzae*, *Magnaporthe grisea* and *Drechslera oryzae* (Reddy et al. 2007).

Streptomyces vinaceusdrappus is reported to inhibit the growth of rice blast disease causing agent, *Magnaporthe oryzae* (anamorph *Pyricularia oryzae*), by inhibiting mycelial growth up to 88.73% (Law et al. 2017). Besides these, *Streptomyces* are well known producers of prolific and bioactive antibiotic compounds. Blasticidin-S and Kasugamycin isolated from *Streptomyces griseochromogenes* and *Streptomyces kasugaensis*, respectively; are often used as active fungicides for controlling rice blast (Fukunaga et al. 1955; Tapadar and Jha 2013; Copping and Duke 2007). *Streptomyces* sp. PM5 isolated from rice rhizosphere having the ability to produce two aliphatic compounds SPM5C-1 and SPM5C-2 with a ketone and lactone carbonyl unit, which was effective against rice disease causing pathogen *R. solani* and *P. oryzae* as they showed active antifungal activity and suppressed the growth of these pathogens at concentrations of 25, 50, 75 and 100 μg mL^{-1} (Prabavathy et al. 2006). Omura et al. (1984) found that, 20 membered macrolides produced by *Streptomyces flavus* subsp. *irumaensis* showed potent activity against *P. oryzae*, however, an antifungal metabolite dapiramycin, obtained from *Micromonospora* sp. found to be effective against *R. solani* (Nishizawa et al. 1984). Three isolates namely *Enterobacter agglomerans*, *Xanthomonas luminescens* and *Serratia liquefaciens* were isolated from rice rhizosphere grown in Bali, effectively inhibited the growth of *P. oryzae* cv. that causes rice blast (Suprapta 2012) (Table 10.2).

10.5.2 Siderophore-Mediated Suppression

Siderophores are extracellular iron binding compounds having low molecular weights and higher ferric iron affinity, produced by microbes for the uptake of iron from the environment (Saha et al. 2016). This iron sequestration ability of microorganisms offers them a competitive advantage over pathogens. Siderophores serve as vehicle for transportation of ferric ions by chelating the ions into the microbial cell with a high specific activity (Neilands 1981). The ferric siderophore complex formed is particularly recognized by a membrane receptor that mediates the transportation of iron into the cell (Mercado-Blanco and Bakker 2007). In various oxido-reductive enzymatic reactions, iron acts as a co-factor and a crucial element in binding with siderophore. Thus, binding of iron with siderophores creates an

Table 10.2 Microbial biocontrol agents for disease suppression and growth promotion of rice

Microbial biocontrol agents	Pathogens	Diseases	References
Pseudomonas fluorescens	*Cnaphalocrocis medinalis*	Brown spot	Saravanakumar et al. (2007)
P. fluorescens, Trichoderma spp.	*Pyricularia oryzae*	Blast	Singh (2014)
Trichoderma harzianum, T. viride, T. virens, T. deliquescens	*Neovossia indica*	Blunt	Singh (2014)
Pseudomonas fluorescens, P. putida, T. harzianum, T. viride, T. virens, Aspergillus niger	*Rhizoctonia solani*	Sheath blight	Kumar and Mishra (2014) and Singh (2014)
T. viride	*Drechslera oryzae*	Brown spot	Singh (2014)
Bacillus spp.	*Xanthomonas oryzae*	Bacterial leaf blight	Singh (2014)
T. viride (Tv2), *T. harzianum* (Th5), *T. reesei* (Tr3)	*Cochliobolus miyabeanus*	Brown spot	Harish et al. (2008)
Streptomyces sp. PM5	*P. oryzae, Rhizoctonia solani*	Blast & sheath blight	Prabavathy et al. (2006)
P. fluorescens	*Magnaporthe grisea*	Blast	Reddy et al. (2007)
	Drechslera oryzae	Brown leaf spot	
	Rhizoctonia solani	Sheath blight	
	Sarocladium oryzae	Sheath rot	
Streptomyces vinaceusdrappus	*Magnaporthe oryzae*	Blast	Law et al. (2017)
P. fluorescens	*Magnaporthe oryzae*	Leaf blast	De Vleesschauwer et al. (2008)
Bacillus amyloliquefaciens RWL-1	*Fusarium spp.*	Root rot	Shahzad et al. (2016)
P. fluorescens (S3), *P. tolaasii* (S20), *P. veronii* (S21), *Sphingomonas trueperi*	*Achlya klebsiana, Pythium spinosum*	Seedling disease	Adhikari et al. (2001)

artificial deficiency of iron in the soil, which results in disease suppression through iron competition with the pathogen of rice (Bakker et al. 2007; Duiff et al. 1997). Siderophore production by *P. fluorescens* was initially reported by Kloepper and Schroth (1981) and its plant pathogenic suppression was reported by Becker and Cook (1988). *Fusarium oxysporum*, causative agent of wilt diseases in rice was effectively controlled by *P. fluorescens* through iron competition (Shahzad et al. 2016). Root application of *P. fluorescens* WCS374r in rice successfully controlled *M. oryzae,* a causative agent of leaf blast in rice, through triggering the ISR, siderophore and pseudobactin production, which accelerated the complex defense system. Thus, by generating multiple blast-effective pathways *P. fluorescens* successfully induced resistance (De Vleesschauwer et al. 2008).

10.5.3 Enzymes and Phytohormones-Mediated Suppression

Several defense enzymes *viz.*, cellulase, chitinase, β-1,3 glucanase also play a key role in pathogenic (fungal pathogens) disease suppression in rice by means of cell wall degradation through breakdown of glycosidic bonds, chitin and β −1,3 glucan (Chet et al. 1990; Lorito et al. 1996; Schroth and Hancock 1981). Microbes involved in excretion of chitinase are categorized as effective biocontrol agents (Inbar and Chet 1991; Ordentlich et al. 1988). Chitinase produced by *P. fluorescens* suppressed the phytopathogenic fungi by breaking and fragmenting cell wall of fungus (Narayanan et al. 2009). *P. oryzae* causing blast disease was inhibited by *P. fluorescens* (AUPF25) through production of proteases and phytohormones such as IAA and siderophore, which inhibited mycelial growth (Shyamala and Sivakumaar 2012; Antoun and Prévost 2005). The endophytic bacterial strain, *Bacillus amyloliquefaciens* RWL-1 isolated from rice seed suppressed the pathogenic effect of *Fusarium* spp. by producing phytohormones such as gibberellic acids GA4, GA12, and GA20. Two pathogenic rice seedling diseases caused by *Pythium spinosum* and *Achlya klebsiana* was inhibited by *P. tolaasii* (S20), *P. fluorescens* (S3), *Sphingomonas trueperi* (S12) and *P. veronii* (S21). However, other biocontrol agents such as *Trichoderma virens*, *B. subtilis* and *P. fluorescens*, respectively showed 80%, 63% and 93% reduction of the pathogenic fungi *Aspergillus flavus* (Reddy et al. 2009). Thus, several beneficial microbes along with active plant growth promoting traits in rice also give an immense contribution in the field of biocontrol through modulation of enzymes and endogenous hormones.

10.6 Conclusion

Seven decades ago, there was a drastic increase in global agricultural production which was possible because of the green revolution era that saved billions of people from undernourishment and starvation. This triggered the introduction of chemical fertilizers and pesticides by human that marked the dawn of environmental damage. This injury further extended to the dome of abiotic and biotic stresses that added to environmental disturbances. These stresses are of a major threat and concern to rice productivity. The present chapter concludes a positive trend that could be set by the use of plant growth-promotion microorganisms in terms of conferring abiotic stresses to alleviate different stress effect on rice. Additionally, several researchers strongly advocated the use of bio-control agents to manage insect and diseases in rice without affecting the soil health. Moreover, their use in sustainable production for rice exists but more efforts are required to explore and spread awareness of these eco-friendly, non-harmful and omnipotent use of microbes. Thus, the use of these beneficial stress mitigating microbes will become safeguard for the stability and productivity of agro-ecosystem, which could uplift the global agricultural sustainability and lead us towards to become one of the ideal agricultural producing nations.

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