Rhizosphere Microbes as Essential Partners for Plant Stress Tolerance

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Ever since plants colonized land, they evolved mechanisms to respond to changing environmental conditions and settle in extreme habitats. Recent studies show that several plant species require microbial associations for stress tolerance and survival. Although many plants lack the adaptive capability to adapt to stress conditions, the ability of a variety of plants to adapt to stress conditions often appears to depend on their association with certain microbes, raising a number of questions: What distinguishes the microbes and plants that can adapt to extreme environmental conditions? Can all plants improve stress tolerance when associated with appropriate microbial partners? Answers to these questions should modify our concepts of plant physiology and could lead to new ways towards a sustainable agriculture.

THE ROLE OF RHIZOSPHERE MICROBES IN PLANT STRESS TOLERANCE

Due to their sessile nature, plants have always been confronted with various abiotic and biotic stresses in their immediate environment. As a consequence, the survival of plants depends on their ability to rapidly adjust their physiology, development, and growth to escape or mitigate the impacts of stress. All plants are known to perceive and respond to stress signals such as drought, heat, salinity, herbivory, and pathogens (Hirt, 2009). Some responses are common to various stresses, including the production of certain proteins and the adjustment of the primary metabolism.

Due to photosynthesis, plants can produce carbohydrates, of which a considerable fraction passes to root-associated microorganisms, commonly denoted as the rhizosphere. Plant growth also requires significant quantities of nitrate, phosphate, and other minerals which are often not available in free form or in limited quantities in the soil. This is where root-associated beneficial microbes are important partners. The best-known beneficial microbes are mycorrhizal fungi and rhizobia. Approximately 80% of all terrestrial plant species interact with mycorrhiza which make phosphate and nitrate available to plants. Free-living or endophytic rhizobia can fix atmospheric nitrogen, but only the family of leguminosae profits from such an interaction

through their ability to house rhizobia in root nodules. The interaction of plants with mycorrhizal fungi and rhizobial bacteria is well documented (Corradi and Bonfante, 2012; Geurts et al., 2012). Non-symbiotic rhizosphere microbes have received much less attention and are therefore treated in this review.

Soil-grown plants are immersed in a sea of microbes and diverse beneficial microorganisms such as plant-growth-promoting bacteria (PGPB) as well as plant-growth-promoting fungi (PGPF) can stimulate plant growth and/or confer enhanced resistance to biotic and abiotic stresses (Lugtenberg and Kamilova, 2009). The establishment of beneficial plant–microbial interactions requires the mutual recognition and a considerable orchestration of the responses at both the plant and the microbial side. Rhizobial and mycorrhizal symbioses share a common plant-signaling pathway that is activated by rhizobial and mycorrhizal factors (Corradi and Bonfante, 2012) and this signaling pathway also seems to be activated by certain beneficial bacteria, suggesting that different beneficial and pathogenic microbes initiate common plant-signaling pathways. Recent evidence indicates that beneficial and pathogenic microbes also suppress the host defense system by various strategies, including the production of effectors, exopolysaccharides, or phytohormones (Zamioudis and Pieterse, 2012).

THE ROLE OF PLANT GROWTH-PROMOTING BACTERIA

PGPBs belong to a number of different bacterial families, including *Rhizobium*, *Bacillus*, *Pseudomonas*, *Burkholderia*, etc. PGPBs can improve the growth of vegetables and crops under abiotic stress conditions (Egamberdieva and Kucharova, 2009) and might therefore open new applications for a

sustainable agriculture. Enhanced salt tolerance of *Zea mays* upon co-inoculation with *Rhizobium* and *Pseudomonas* is correlated with decreased electrolyte leakage and maintenance of leaf water contents (Bano and Fatima, 2009). Some microorganisms produce plant hormones, such as indole acetic acid and gibberellic acid, which induce increased root growth and thereby lead to enhanced uptake of nutrients (Egamberdieva and Kucharova, 2009).

Plants have the ability to acquire a state of induced systemic resistance (ISR) to pathogens after inoculation with PGPBs. In association with plant roots, PGPBs can prime the plant innate immune system and confer resistance to a broad spectrum of pathogens with a minimal impact on yield and growth (Van Hulten et al., 2006). Several PGPBs, including *Pseudomonas fluorescens*, *Pseudomonas putida*, *Bacillus pumilus*, *Serratia marcescens*, *Paenibacillus alvei*, *Acinetobacter lwoffii*, *Chryseobacterium balustinum*, and *Azospirillum brasilense* colonize roots and protect on a large variety of plant species, including vegetables, crops, and even trees, against foliar diseases in greenhouse and field trials (Van Loon, 2007).

PLANT-ASSOCIATED FUNGI CONFER STRESS TOLERANCE TO PLANTS

Mycorrhizal and/or endophytic fungi can interact with many plant species and thereby significantly contribute to the adaptation of these plants to a number of environmental stresses (Rodriguez et al., 2008). These conditions include drought, heat, pathogens, herbivores, or limiting nutrients. Moreover, some plants are unable to withstand stress conditions in the absence of their associated microbes (Redman et al., 2002). It appears that stress tolerance of the host plant can be a habitat-specific feature of the interaction. For example, *Curvularia protuberata* confers heat tolerance to its geothermal host plant *Dichanthelium lanuginosum*. However, neither the fungus nor the plant can survive alone at temperatures above 38°C (Redman et al., 2002). Moreover, only *C. protuberata* isolates from geothermal plants can confer heat tolerance (Rodriguez et al., 2008).

A comparison of different fungal endophytes unravels a further layer of specificity: *C. protuberata* confers heat but neither disease nor salt tolerance. In contrast, *Fusarium culmorum* only confers salt tolerance and *Curvularia magna* only disease tolerance (Rodriguez et al., 2008). It appears that these specific features contribute to the ability of some plants to establish and survive in extreme habitats.

Symbiotically conferred disease tolerance appears to involve different mechanisms depending on the endophyte. For example, a non-pathogenic *Colletotrichum* strain that confers disease resistance does not activate host defense in the absence of pathogen challenge (Redman et al., 1999). Moreover, disease resistance is localized to tissues that the fungus has colonized, but is not systemic.

In contrast, *Piriformospora indica* confers disease resistance systemically. *P. indica* colonizes the roots of many plant species and stimulates growth, biomass, and seed production of the hosts (Fig. 1). *P. indica* promotes nitrate and phosphate uptake and confers resistance against abiotic (Waller et al., 2005) and biotic stress (Stein et al., 2008). Colonization by the fungus stimulates the host to synthesize phosphatidic acid,
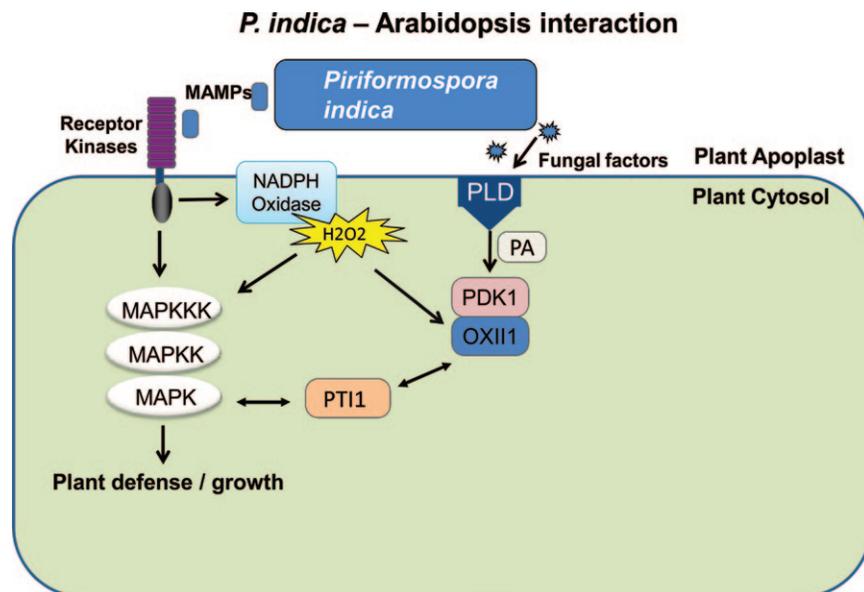

Figure 1. The Beneficial Fungus *Piriformospora indica* Stimulates Phospholipase D to Synthesize Phosphatidic Acid (PA) which Activates the Protein Kinases PDK1 and Subsequently OXI1 and MAPKs.
OXI1 and MAPKs can be activated via recognition of microbe-associated molecular patterns (MAMPs) and also generate H_2O_2 to activate the OXI1–MAPK pathway. On the other hand, fungal auxin production interferes with the activation of plant defense responses, suggesting that the balance between inactivation and activation of the host defense pathways might determine whether plants go into a defense or growth mode, respectively.

which triggers the OXI1 pathway (Camehl et al., 2011). This pathway is usually activated only in response to pathogen attack to activate host defense (Rentel et al., 2004), and a defect in the OXI1 pathway negatively affects plant growth by the fungus, resembling a pathogenic interaction. Overall, the differences between *Colletotrichum* spp.- and *P. indica*-conferred disease resistance indicate that a number of different mechanisms exist that have yet to be elucidated.

Further evidence exists that our present concepts of categorizing microbes as pathogenic or beneficial are inadequate. For example, *F. culmorum* was designated as pathogenic, as it can cause disease on a variety of crop plants. However, the *F. culmorum* isolate FcRed1 functions as a beneficial microbe and confers salt tolerance on its host dunegrass *Leymus mollis*, but isolates from non-coastal dunegrass do not have this property (Rodriguez et al., 2008). *C. protuberata* is a plant pathogen for several monocots, but isolate Cp4666D confers heat and drought tolerance to its host plant *D. lanuginosum* (Rodriguez et al., 2008). While *Curvularia* species are not known to have broad disease-host ranges, *C. protuberata* from the monocot *D. lanuginosum* also confers heat tolerance on tomato (Marquez et al., 2007; Rodriguez et al., 2008).

The ability of pathogenic *Colletotrichum* species to switch to a beneficiary lifestyle reveals that we still understand very little about the molecular basis of the plant–microbe interactions dictating friend–foe relationships. Some microbes can also be present in plants without showing disease symptoms. For example, *Colletotrichum acutatum* can colonize pepper, eggplant, bean, and tomato without causing disease but, with other plants, such as strawberry, disease symptoms become evident (Freeman et al., 2001). So it appears that a number of microbes have a host-dependent lifestyle as pathogenic or beneficiary partner of plants.

CONCLUSIONS

It is important to point out that plant-associated microbial communities also include viruses and algae. Bacteria, fungi, viruses, and algae can all contribute to the outcome of the plant–microbial interaction and hence increase the complexity of studying these interactions. Moreover, fungi may also harbor bacteria and viruses which can affect the outcome of the plant–microbe interaction. For example, the Cp4666D isolate of *C. protuberata*, which was isolated from plants growing in geothermal soils, contains a double-stranded RNA virus that is required for conferring heat tolerance to its host plants (Marquez et al., 2007). In the absence of the virus, *C. protuberata* Cp4666D can still colonize its host, but has lost its ability to confer heat tolerance to the plants. Therefore, in this case, a ménage-a-trois (a virus in a fungus in a plant) is required for heat tolerance.

This discussion shows that the extent and specificity of the existing plant–microbe interactions are poorly known. The complexity of strain-specific interactions within a species suggests the existence of highly specific and complex association

mechanisms and our understanding of what factors govern the choice and selectivity of plant–microbial associations and how microbes confer stress tolerance to plants is still in its infancy. However, enlarging research in this field and applying the gained knowledge to crop plants promises additional avenues to develop agriculture in a sustainable way.

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