Harnessing symbiotic plant–fungus interactions to unleash hidden forces from extreme plant ecosystems

Marta-Marina Pérez-Alonso1, Carmen Guerrero-Galán1, Sandra S. Scholz2, Takatoshi Kiba3,4, Hitoshi Sakakibara3,4, Jutta Ludwig-Müller5, Anne Krapp6, Ralf Oelmüller2, Jesús Vicente-Carbajosa1,7, and Stephan Pollmann1,7,*

1 Centro de Biotecnología y Genómica de Plantas, Universidad Politécnica de Madrid (UPM)–Instituto Nacional de Investigación y Tecnología Agraria y Alimentación (INIA), Campus de Montegancedo, 28223 Pozuelo de Alarcón (Madrid), Spain
2 Matthias Schleiden Institute of Genetics, Bioinformatics and Molecular Botany, Department of Plant Physiology, Friedrich-Schiller-University Jena, 07743 Jena, Germany
3 RIKEN Center for Sustainable Resource Science, 1-7-22, Suehiro, Tsurumi, Yokohama, 230-0045, Japan
4 Department of Applied Biosciences, Graduate School of Bioagricultural Sciences, Nagoya University, Nagoya, 464-8601, Japan
5 Institute of Botany, Technische Universität Dresden, 01062 Dresden, Germany
6 Institut Jean-Pierre Bourgin, INRA, AgroParisTech, CNRS, Université Paris-Saclay, 78000 Versailles, France
7 Departamento de Biotecnología-Biología Vegetal, Escuela Técnica Superior de Ingeniería Agronómica, Alimentaria y de Biosistemas, Universidad Politécnica de Madrid (UPM), 28040 Madrid, Spain

* Correspondence: stephan.pollmann@upm.es

Received 5 November 2019; Editorial decision 15 January 2020; Accepted 21 January 2020

Editor: Peter Doerner, University of Edinburgh, UK

Abstract

Global climate change is arguably one of the biggest threats of modern times and has already led to a wide range of impacts on the environment, economy, and society. Owing to past emissions and climate system inertia, global climate change is predicted to continue for decades even if anthropogenic greenhouse gas emissions were to stop immediately. In many regions, such as central Europe and the Mediterranean region, the temperature is likely to rise by 2–5 °C and annual precipitation is predicted to decrease. Expected heat and drought periods followed by floods, and unpredictable growing seasons, are predicted to have detrimental effects on agricultural production systems, causing immense economic losses and food supply problems. To mitigate the risks of climate change, agricultural innovations counteracting these effects need to be embraced and accelerated. To achieve maximum improvement, the required agricultural innovations should not focus only on crops but rather pursue a holistic approach including the entire ecosystem. Over millions of years, plants have evolved in close association with other organisms, particularly soil microbes that have shaped their evolution and contemporary ecology. Many studies have already highlighted beneficial interactions among plants and the communities of microorganisms with which they coexist. Questions arising from these discoveries are whether it will be possible to decipher a common molecular pattern and the underlying biochemical framework of interspecies communication, and whether such knowledge can be used to improve agricultural performance under environmental stress conditions. In this review, we summarize the current knowledge of plant interactions with fungal endosymbionts found in extreme ecosystems. Special attention will be paid to the interaction of plants with the symbiotic root-colonizing endophytic fungus Serendipita indica, which has been developed as a model system for beneficial plant–fungus interactions.

Keywords: Abiotic stress, crosstalk, endosymbiosis, plant performance, Serendipita indica.
Introduction

Agriculture is the principal means of livelihood in many regions of the world. However, current agricultural practice is not sustainable and is known to cause serious ecological damage, such as soil erosion, eutrophication of water bodies, increasing salinity of soils, and desertification, which in the long run translates into substantially reduced agricultural productivity owing to further loss of arable land. Global climate change represents an additional hazard that aggravates the negative effects of current agricultural practices and causes further dramatic losses in agricultural production.

Given the increasing demand for agricultural products resulting from the growing world population, which is predicted to reach 9.5 billion people by 2050 (ISAAA, 2017), the combination of non-sustainable agricultural practices and climate change effects will not only entail economic losses for the agricultural industry but put the general food supply for humankind in considerable jeopardy. With respect to current estimates, 800–925 million people will be undernourished in 2020; ‘hidden hunger’ due to a lack of vitamins and minerals is estimated to be the most common form of malnutrition, affecting approximately 2 billion people (FAO et al., 2015; Fedoroff, 2015). Hence, immediate measures must be taken to limit the damage already caused to the environment and to secure food availability in the future. Advanced agricultural biotechnology methods, including the so-called ‘new breeding techniques’ consisting of directed genome editing (i.e. CRISPR/Cas), are expected to be able to alleviate the effects of climate change and to ensure a more sustainable agriculture (Aglawe et al., 2018). These approaches include, for example, the generation of more drought- and salt-tolerant crops, or plants with improved nitrogen use efficiency or pathogen resistance, by cloning genes from plants found in high-stress environments into stress-sensitive but highly productive species. It is anticipated that these actions will reduce the application of pesticides and mineral fertilizers and limit the need for irrigation (Gartland and Garland, 2018). According to our estimations, such exclusively plant-focused strategies might be insufficient to solve the issue of food security entirely. Thus, in this review we would like to draw attention to the symbiosis of plants with commensal microorganisms, an often underestimated factor that can substantially affect plant performance under unfavorable growth conditions.

Throughout the course of evolution plants have been constantly confronted with changing environmental conditions, forcing them to adapt in order to survive. These changing conditions included temperature fluctuations (Fig. 1), scarce water resources, and high UV radiation. Although it is extremely difficult to precisely determine the temporal dynamics of prehistoric climate changes, it is undeniable that climate change has accelerated in the past century (Murray, 1997; Petit et al., 1999). The fact that anthropogenic greenhouse gas emissions considerably contribute to this acceleration raises the question of whether plants will be able to adapt to the imposed environmental stress conditions. It is possible that they will run out of time to develop appropriate responses to counteract the detrimental effects. However, assuming that this is not the first time that plants have faced such conditions, they may already have suitable molecular mechanisms at their disposal, generated during previous challenges, to withstand the foreseen unfavorable conditions of increased temperatures and water shortages.

The majority of plant studies focus only on plant responses toward abiotic stresses and disregard the fact that plants normally live in close association with a plethora of different microorganisms, such as bacteria, fungi, oomycetes, and protists, and that millions of years of co-evolution have led to the establishment of highly specialized ecosystems in which plants constantly interact with their surrounding communities of commensal, symbiotic, and pathogenic microorganisms. In the climate change context, symbiotic relationships are of particular interest as they are supposed to translate, or already have translated, into mutually advantageous associations that can provide important fitness improvements. The concept of a mutual coexistence between dissimilar organisms, referred to as symbiosis (from the ancient Greek συμβίοσις, ‘living together’), was first described by Heinrich Anton de Bary (1879). Later, the terms symbiosis, symbiont, and symbiote were further defined by Hertig et al. (1937). Since these early observations, our insight into symbiotic associations of plants has substantially advanced to indicate that plant–microbe interactions are important to the structure, function, and health of plant communities and that symbiotic fungi contribute to—and may be even responsible for—the adaptations of plants to environmental stresses (Rodriguez et al., 2004).

![Fig. 1. Record of isotopic temperature changes of the atmosphere extracted from an Antarctic ice core (Pett et al., 1999). BP, Before present.](image)
Given that practically all plants are symbiotic with fungi, which depend either partially (endophytes) or entirely (mycorrhizae) on the interaction with their host plant, we asked ourselves the question of whether it would be potentially interesting to investigate symbiotic interactions in plant–fungal communities known to inhabit ecosystems in extreme environments. Such ‘extreme’ communities may allow us to identify patterns of crosstalk or molecular mechanisms that facilitate the creation of more sustainable agricultural systems with increased performance levels, either by harnessing identified molecular mechanisms or by soil microbiome engineering approaches. There are plenty of examples of mutualistic fungi that can confer stress tolerance upon plants. For example, a morphologically defined group of fungi within the class Ascomycota, the so-called dark septate endophytes, are well known as endosymbionts of numerous plant species, including crop plants (Junpponen and Trappe, 1998; Andrade-Linares et al., 2011). Intriguingly, Marco Molina-Montenegro and colleagues recently reported the symbiosis of two fungal endophytes, Penicillium chrysogenum and Penicillium brevicompactum, with Colobanthus quitensis (Caryophyllaceae) and Deschampsia antarctica (Poaceae), two plants that are found in the Antarctic region. It was demonstrated that the isolated root endophytes are able to increase plant performance under UV-B radiation. Moreover, the inoculation of lettuce with the isolated fungi was shown to significantly improve the ecophysiological performance and yield of the plant under both normal and drought conditions (Molina-Montenegro et al., 2016; Ramos et al., 2018). Another highly interesting example of an ‘extreme’ symbiosis comes from the isolation of a Fusarium culmorum strain that has been identified as an endophytic symbiont of dunegrass (Leymus mollis) collected from coastal beach habitats. Remarkably, the fungal strain isolated from plants collected from this salt-stress environment was more efficient in conferring salt-stress tolerance to dunegrass and tomato plants than a F. culmorum isolate purchased from the American Type Culture Collection; this reflects an ecological phenomenon referred to as habitat-adapted symbiosis. This habitat-specific phenomenon is suggested to provide an intergenomic epigenetic mechanism for plant adaptation and survival in high-stress environments (Rodriguez et al., 2008). We made a similar observation with a Fusarium sp. strain isolated from sea-blime (Suaeda maritima) collected from saline ponds along coastal plains in India (R. Oelmüller, unpublished results).

Possibly the most extensively studied endosymbiotic fungus from an extreme environment is Serendipita indica (formerly named Piriformospora indica). S. indica (Agaricomycetes, Basidiomycota) is an axenically cultivable, root-colonizing endophytic fungus that was first isolated from two xerophytic woody shrubs, Ziziphus nummularia and Prosopis juliflora, in the Thar desert in India (Verma et al., 1998). Closely related endophytic species have been isolated from western European and Namibian Fabaceae, Poaceae, and Araceae (Weiß et al., 2011). The initially identified isolate of S. indica is deposited at the Deutsche Sammlung von Mikroorganismen und Zellkulturen (DSMZ), Braunschweig, Germany (DSM 11827). The fungus possesses exceptional ability to substantially promote plant growth and performance, especially under stress conditions, and an outstanding versatility to colonize a very broad range of plant species (Waller et al., 2005; Jacobs et al., 2011; Gill et al., 2016; Su et al., 2017). The latter feature suggests the existence of conserved molecular mechanism(s) that facilitate promiscuous host selection, which makes S. indica such an interesting microorganism to study and led to a rapid suggestion that it could be applied to increase plant production (Varma et al., 1999). In this review, we will provide a summary on the current knowledge of the molecular and biochemical crosstalk between S. indica and its hosts, and reflect on the possibilities to apply this knowledge to improve agricultural productivity.

### The fungus Serendipita indica

Although the basidiomycete S. indica is classified as an endophyte, the fungus possesses many characteristics that are generally attributed to arbuscular mycorrhizal fungi (AMF), for example, its lifestyle maintaining hyphae outside the host root. Like AMF, S. indica is able to promote the growth of host plants and increase the resistance of colonized plants to fungal pathogens and various abiotic stresses (Harman, 2011; Abdelaziz et al., 2019). Additionally, S. indica is known to affect the secondary metabolism of colonized host plants and to promote seed production by host plants, some of which are of economic importance (Waller et al., 2005; Strehmel et al., 2016). However, unlike AMF, S. indica can be easily cultivated (Oelmüller et al., 2009) and is capable of colonizing the roots of the model dicot plant Arabidopsis thaliana (Peškan-Berghöfer et al., 2004). In fact, S. indica belongs to the newly described family of Sebacinaeae and the new order Sebacinales (Agaricomycetes) (Weiss et al., 2004, 2011; Qiang et al., 2012a; Weiß et al., 2016).

Owing to the great scientific interest in this mutualistic symbiont over the past two decades, great advances have been made with respect to the genomic assessment and manipulation of S. indica. As early as 2009, the group of Alga Zuccaro described a method for the stable genetic transformation of the fungus (Zuccaro et al., 2009), and this was followed by a report on the deep genomic study of its 25 Mb genome (Zuccaro et al., 2011). These experimental advances represent highly valuable tools for further investigation of the molecular mechanisms that allow the fungus to grow readily on diverse media and to colonize an extremely wide range of mono- and dicotyledonous plants.

### Establishment of symbiosis

The genomic study additionally revealed that root colonization by S. indica proceeds in two steps (Zuccaro et al., 2011). The interaction begins with a short biotrophic phase, which is followed by a saprophytic phase in which the fungus feeds on dead host plant cells. During the initial phase of root colonization, the host plant responds with increased programmed cell death triggered by the simultaneous induction of endoplasmic reticulum stress and repression of the unfolded protein response. Moreover, it is hypothesized that the endoplasmic reticulum stress translates into the induction of a vacuolar processing enzyme/caspase 1-like activity-dependent vacuolar
cell death program (Deshmukh et al., 2006; Qiang et al., 2012b). Furthermore, the host plant responds by secreting symbiosis-specific proteins, including proteins related to growth, development, biotic and abiotic stress responses, and mucilage (Thürich et al., 2018). At the same time, plant innate immunity is suppressed by the manipulation of a handful of different plant hormone signaling pathways to overcome plant defenses and establish a compatible interaction between the fungus and the host (Schäfer et al., 2009; Jacobs et al., 2011). In particular, levels of abscisic acid (ABA), salicylic acid, and jasmonic acid, as well as jasmonoyl-l-isoleucine, are reported to substantially increase during very early phases of the interaction. The induction of stress- and defense-related hormones in the host plant can be triggered by chemical compounds released by the fungus before physical contact. It may be concluded that these transient metabolic reactions serve to prepare the plant for the symbiotic interaction. Notably, the plant response is not restricted to the root system, but spreads to aerial tissues as well. Moreover, the response is only short lived, as hormone levels return to normal levels after 6 days of co-cultivation (Vahabi et al., 2015).

The exact role of phytohormones in the establishment of the symbiotic interaction is still a matter of debate (Vadassery et al., 2008; Schäfer et al., 2009; Lee et al., 2011; Hilbert et al., 2012). The broad host range of S. indica suggests that it evolved highly sophisticated colonization strategies, recruiting a similar or identical hormone signaling pathway from host plants in order to successfully establish a symbiotic relationship. However, recent work has cast doubts on this hypothesis of a simple reprogramming of the plant hormone-signaling machinery. A comprehensive study of the role of jasmonic acid and gibberellic acid during the colonization of seven different plant species with S. indica highlighted considerable differences in root colonization and plant hormone action, suggesting a high degree of species specificity in the establishment of symbiosis (Liu et al., 2019).

The controversial discussion regarding the contribution of auxins to fungus-mediated growth promotion and the establishment of the mutual interaction is also noteworthy. In particular, the root growth-promoting effect of S. indica implied the involvement of auxins, possibly the best-characterized class of plant hormones (Fig. 2). Auxins orchestrate virtually every aspect of plant growth and development (Davies, 2010). In plant roots, changes in local auxin levels cause a number of well-described phenotypes, including a dose-dependent increase in the length of epidermal-derived root hairs, a bimodal effect on primary root elongation, and a dose-dependent increase in the number of lateral root primordia (Overvoorde et al., 2010). More importantly in this context, however, is the fact that indole-3-acetic acid (IAA) and several of its precursor molecules, such as indole-pyruvic acid (IPA), indole-3-acetamide (IAM), and tryptamine (TAM), are readily synthesized by various plant-interacting fungi and appear to contribute to fungal-plant interactions (Spaepen et al., 2007; Fu et al., 2015). S. indica was shown to synthesize IAA via IPA and indole-3-acetaldehyde (IAD), while other conceivable indolic intermediates, namely IAM, TAM, and indole-3-acetonitrile, were neither detected as endogenous compounds nor converted into IAA by the fungus (Hilbert et al., 2012).

Further experiments provided strong evidence that mycelium-derived IAA has no significant impact on growth promotion, but rather represents a compatibility factor that is important for the establishment of the biotrophic interaction (Hilbert et al., 2012). More recently, however, another report demonstrated that S. indica hyphae contain considerable amounts of IAM and that both IAM and IAA levels increase during the colonization of Brassica campestris roots (Hua et al., 2017). In any case, it seems that IAA levels in host plants temporarily increase during the early phases of the interaction, only to return to the levels of non-colonized roots shortly after (Vadassery et al., 2008; Hilbert et al., 2012). Genome-wide expression studies and gene ontology (GO) analyses did not provide clear-cut indications of a significant enrichment of genes listed in GO terms related to auxin biosynthesis or signaling during the first 14 days after infection (Lahrmann et al., 2015). Interestingly, the content of conjugated auxin increases moderately over the course of colonization in infected roots (Vadassery et al., 2008). This observation may suggest that the fungus either inactivates free IAA by itself or induces the expression of Gretchen Hagen 3 (GH3) genes. GH3 genes encode IAA-amidinesynthases that catalyze the conjugation of free IAA to amino acids, thereby physiologically inactivating the plant hormone (Staswick et al., 2005; Böttcher et al., 2010, 2012). Remarkably, a recent study employing state-of-the-art live cell imaging techniques and liquid chromatography/mass spectrometry-based plant hormone analyses provided unequivocal evidence that the initial fungus-mediated increase of IAA in plant roots is seemingly adequate to induce lateral root formation within a very short timeframe (Meents et al., 2019). In view of these findings, it appears tempting to speculate that the fungus-derived increase of IAA in the early phase of colonization is sufficient to trigger alterations in the developmental program of the host root.
which result in morphological changes in the root architecture, facilitating the improved nutrition of the host plant through the extended root system in the long term.

A more detailed analysis of transcriptional changes of a group of approximately 140 genes related to auxin metabolism, transport, and signaling over the first 14 days of co-cultivation with S. indica, however, contradicts the notion that auxin-related genes are not considerably affected. The analysis revealed the induction of a set of GH3 genes, namely GH3.2, GH3.3, and GH3.15, in the infected plants, which neatly matches the observed induction of IAA–amino acid conjugates (Vadassery et al., 2008). Interestingly, the UGT84B1 gene, which codes for a UDP-glycosyltransferase that has been described to catalyze the conjugation of free IAA to glucose (Jackson et al., 2001, 2002), is also significantly induced. In line with these findings, it has been observed that infection with S. indica is sufficient to rescue the high-auxin phenotype of the sur1-1 mutant through the reduction of free auxin levels (Vadassery et al., 2008). Together, these experiments support the hypothesis that cellular auxin levels are increased and need to be actively intercepted by conjugation with either sugar or amino acids to prevent the over-accumulation of physiologically active free IAA. The latter assumption is further strengthened by the observed induction of PIN3 expression. PIN5 is an auxin transporter that contributes to the maintenance of subcellular auxin homeostasis by mediating the flow of auxin from the cytoplasm into the lumen of the endoplasmic reticulum (Mravec et al., 2009). Notably, two other PIN genes, PIN3 and PIN4, also appear to be induced upon infection with S. indica (Fig. 3). While PIN3, together with auxin response factor 7 (ARF7), drives early steps in lateral root formation, PIN4 is known to be involved in the generation of auxin gradients and auxin canalization in root tips (Friml et al., 2002; Chen et al., 2015b; Laskowski and ten Tusscher, 2017). Thus, it is suggested that they are involved in triggering the root growth-promoting effect observed in S. indica-infected plants. A detailed study of the role of PIN3 and PIN4 in the infection process is currently under way in our laboratories.

Most notable, however, is the increased flux of metabolites into the production of the defense compounds camalexin and glucobrassicin. As can be seen in Fig. 3, a large proportion of the genes encoding relevant enzymes of the two corresponding biosynthetic pathways (Fig. 4) are significantly induced upon infection with S. indica. The biosynthesis of camalexin and glucobrassicin could be essential to limit colonization of the host plant with S. indica, thus allowing the beneficial interaction while preventing over-colonization (Nongbri et al., 2012). The corresponding metabolic pathways are orchestrated by a small number of transcription factors, namely MYB34, MYB51, MYB122, and WRKY33 (Birkenbihl et al., 2012; Frerigmann and Gigolashvili, 2014; Frerigmann et al., 2015). Consistently, three out of the four transcription factors appear to be substantially induced over the course of infection. However, glucobrassicin and camalexin originate from a common precursor molecule, indole-3-acetaldoxime (IAOx), which is assumed to represent an intermediate in a Brassicaceae-specific metabolic shunt (Pollmann et al., 2006; Lehmann et al., 2017). Although some recent publications have suggested that IAOx also occurs outside the Brassicaceae (Irmisch et al., 2013a, b; Luck et al., 2016; Buezo et al., 2019), a more general role for these compounds in the initial phase of the infection has to be doubted as long as a broader occurrence of IAOx in the plant kingdom is not unequivocally confirmed. Hence, metabolic engineering approaches on the basis of the IAOx shunt appear to offer only little prospect of improving agricultural productivity.

### The induction of cytosolic calcium in plant–fungus interactions

The rapid detection of specific external stimuli, as well as a timely and adequate response to them, is often instrumental to guarantee plant survival. Second messenger molecules, such as

---

**Fig. 3.** Hierarchical clustering analysis of auxin-related genes differentially regulated during the first 2 weeks of co-cultivation with S. indica. The gene expression levels were extracted from publicly available data (GSE60736, Lahrmann et al., 2015) deposited in the Gene Expression Omnibus (GEO) repository for high-throughput microarray and next-generation sequence functional genomic datasets (Barrett et al., 2013). A P-value of 0.05 after adjustment for multiple testing and log2 ratio >0.75 were arbitrarily chosen to select 45 differentially expressed genes in Arabidopsis seedlings co-cultivated with S. indica relative to mock-treated control plants.
cyclic AMP or inositol triphosphate, are intracellular signaling molecules that are released by the cell in response to the perception of extracellular signals. They play important roles in the integration and transduction of signals, triggering physiological responses at the cellular level. Calcium (Ca\(^{2+}\)) is a highly conserved and very versatile intracellular second messenger linking several extracellular cues with appropriate cellular responses, including growth and defense (Dodd et al., 2010). In addition, Ca\(^{2+}\) is involved in providing tolerance to biotic and abiotic stresses (Kudla et al., 2018). Owing to the functional conservation of Ca\(^{2+}\) signaling in plants, it is not at all surprising that the recruitment of Ca\(^{2+}\) signaling by a colonizing fungus represents another well-characterized mechanism that plays a key role in the establishment of plant–fungus interactions.

The interaction of microbes with plant roots often involves a type of chemical communication that commences with the recognition of chemical mediators at the cell surface of the host plant and engages sophisticated downstream plant immunity mechanisms that act as surveillance systems to detect the invasion of microbes into the host cell. In the recognition process, microbe-associated molecular patterns (MAMPs) or invasion patterns are perceived by specific receptors, which, in turn, activate pattern-triggered responses (van’t Padje et al., 2016; Zipfél and Oldroyd, 2017). The response cascades that are triggered can either be suppressed, for example, by biotrophs, or employed, in the case of necrotrophs, to continue symbiosis (Cook et al., 2015).

Cell wall extracts (CWEs) from S. indica have been described to promote the elevation of cytosolic Ca\(^{2+}\) concentration ([Ca\(^{2+}\)]\(_{cyt}\)), thereby mimicking the presence of the fungus and promoting plant growth in the initial phase of the interaction (Vadassery et al., 2009). Intriguingly, this observation suggests that growth promotion can be uncoupled from the colonization of the root system with S. indica and that growth promotion depends on Ca\(^{2+}\). This is further supported by the fact that the CWE-mediated growth–promotion response can be blocked by the additional application of Ca\(^{2+}\)-specific chelators such as BAPTA or LaCl\(_3\). However, it should be noted that even repeated treatment of roots with CWEs has not been sufficient to entirely replace the interaction with the endosymbiont (Vadassery et al., 2009). Hence, it may be concluded that further factors or processes, apart from the perception of the
active elicitor in the CWEs and the downstream Ca\(^{2+}\) signal, also play important roles in this context. The hydrolysis of extracellular ATP (eATP) by specialized fungus-derived ecto-5'-nucleotidases and the resulting interference with the perception of eATP by the lectin receptor kinase DORN1 (Does not Respond to Nucleotides 1) could be such a missing process that apparently forms an integral part of the establishment of the symbiosis (Choi et al., 2014; Nizam et al., 2019).

Mutant analyses revealed that CWE-mediated growth promotion in the S. indica—Arabidopsis interaction involves the activation of MITOGEN-ACTIVATED PROTEIN KINASE 6 (MAPK6), since the mapk6 mutant shows no growth promotion upon treatment with CWEs. The Ca\(^{2+}\) influx-dependent activation of MAPK6 is induced by numerous microbial elicitors (Nühse et al., 2000; Lecourieux et al., 2002) and represents a common theme in plant resistance to biotic stresses (Menke et al., 2004; Takahashi et al., 2007). Furthermore, camalexin biosynthesis is reported to be controlled through the MAPK3/MAPK6 cascade, activating WRKY33 through phosphorylation (Ren et al., 2008; Li et al., 2012), which functionally links the increase of [Ca\(^{2+}\)]\(_{\text{cyt}}\) to the observed plant defense response upon S. indica colonization (Figs 3, 4).

Recent work identified the molecular nature of the active elicitor in S. indica CWEs. During root colonization, cellooligomer (CT) is released by the fungus to initiate the symbiotic interaction of the fungus with the root (Johnson et al., 2018). Plant roots perceive and respond to very low concentrations of short-chain β(1→4)-linked d-glucose units (cellooligomers). Cellooligomers are generally released after structural changes to the cell wall caused by either environmental or plant integral signals. They are suggested to give an account of the state and integrity of the cell wall, which facilitates the activation of appropriate local and distal responses (Souza et al., 2017; Oelmüller, 2018).

The perception of CT by an as yet unidentified receptor results in a dose-dependent rapid and transient increase in [Ca\(^{2+}\)]\(_{\text{cyt}}\), which induces mild defense responses including the induction of reactive oxygen species, changes in membrane potential, and the expression of genes associated with growth regulation and root development (Johnson et al., 2018). CT perception is independent of BRI1-ASSOCIATED RECEPTOR KINASE 1 (BAK1), which is a well-known co-receptor contributing to the perception and integration of multiple MAMP-triggered responses, for instance, those toward bacterial flg22 or elf18 or fungal chitin, inducing [Ca\(^{2+}\)]\(_{\text{cyt}}\) elevation and downstream immune responses (Lu et al., 2010; Zhang et al., 2010; Shi et al., 2013; Li et al., 2014; Kadota et al., 2015). Moreover, mutants of the proposed Ca\(^{2+}\) channels GLU-LIKE RECEPTOR (GLR) 2.4, GLR2.5, GLR3.3, and the vacuolar TWO PORE CHANNEL 1 (TPC1) were not impaired in CT signaling. This strongly suggests that they are also not involved in the process, although they have previously been reported to contribute to wound signaling and wounding-induced systemic Ca\(^{2+}\) elevations (Peiter et al., 2005; Li et al., 2013; Manzoor et al., 2013; Choi et al., 2014b; Kiep et al., 2015). However, the mechanisms that are involved in the rapid Ca\(^{2+}\) influx have yet to be identified.

In addition to the question of how the Ca\(^{2+}\) influx is brought about on the molecular level, the complexity of Ca\(^{2+}\) signaling involves a large number of different Ca\(^{2+}\)-binding proteins to specifically decode incoming Ca\(^{2+}\) signals, makes it tremendously difficult to address the question of which molecular components contribute to the integration of the Ca\(^{2+}\) signals induced during S. indica infection. Complicating the situation even further, different Ca\(^{2+}\) signatures appear to trigger a variety of functions via the signature-specific activation of corresponding Ca\(^{2+}\)-sensor proteins (McAinsh and Pittman, 2009; Johnson et al., 2011). All Ca\(^{2+}\)-sensor proteins, however, contain at least one so-called EF-hand motif (helix-loop-helix domain) in their primary sequence, which mediates the binding of Ca\(^{2+}\) to the sensor. The Arabidopsis proteome contains approximately 250 proteins with EF-hand motifs, and at least 100 have been classified as Ca\(^{2+}\)-sensor proteins (Day et al., 2002; Hashimoto and Kudla, 2011). With respect to the nature of their response domain, which can be either a kinase or a transcription regulation domain, sensor proteins are further divided into sensor relay proteins or sensor responders. The latter class of sensors are particularly versatile, as they combine a sensor and response domain in a single protein and can thus directly transduce the Ca\(^{2+}\) signal to downstream target proteins by phosphorylation.

CALCIUM-DEPENDENT PROTEIN KINASEs (CDPKs) represent an important family of these sensor responder proteins, and 34 members of this family have been identified in Arabidopsis (Cheng et al., 2002). CDPKs have been demonstrated to be activated by Ca\(^{2+}\) signals during the course of interactions of plant roots with biotrophic microbes. They are speculated to control host entry and accommodation in the plant through the efficient suppression of corresponding plant defense responses (Freymark et al., 2007; Chen et al., 2015a). Recent work has suggested the involvement of CDPKs in the S. indica—plant interaction as well, as S. indica CWE-mediated [Ca\(^{2+}\)]\(_{\text{cyt}}\) elevation has been shown to promote tuberization in potato via a pathway that involves the potato CDPK1 sensor responder (Upadhyaya et al., 2013). The work by Johnson et al. (2018) supports this hypothesis, because their microarray data provide strong evidence for the induction of a number of CDPK proteins upon CT treatment. A more detailed analysis of CDPKs involved in the establishment and maintenance of the fungus—plant symbiosis has yet to be carried out, although it might be very difficult to achieve this task owing to the large number of CDPK family members in plants and their presumably partially overlapping functions. Available microarray data also revealed the differential expression of a small number of sensor relay proteins—CALMODULIN-LIKE 37 (CML37) and CML38 as well as CBL-INTERACTING PROTEIN KINASE 13 (CIPK13) and CIPK20—but their role in fungus—plant interaction is even less well investigated and remains a matter of debate (Vahabi et al., 2015; Johnson et al., 2018).

**Serendipita indica** and the growth—defense tradeoff enigma

The initial phase of root colonization by S. indica involves the mounting of defense responses by the host plant (Zuccaro
et al., 2011; Vahabi et al., 2015; Johnson et al., 2018). Such defense responses frequently come at the cost of a substantial reduction in growth and reproduction, which carries important implications for agriculture. However, despite the obvious importance of this tradeoff between growth and defense in shaping plant productivity in agricultural ecosystems, the molecular mechanisms that connect plant growth with plant defense responses are only poorly understood. From the classical point of view, plants employ physiological tradeoffs to allocate their limited metabolic resources between the generation of defense-related protective compounds on the one hand and associated morphological structures on the other hand. In other words, plants face a dilemma: to decide whether to grow or to defend themselves (Herms and Mattson, 1992; Züst et al., 2015). Although most ecological studies of plant resistance to herbivores or pathogens still use the concept of growth–defense tradeoffs as their major paradigm (Denancé et al., 2013; Huot et al., 2014), more recent studies have questioned this simplistic view (Kliebenstein, 2016; Schuman and Baldwin, 2016). They point out that growth inhibition in response to, for example, herbivory is likely not the result of the simple channeling of photoassimilates from growth to defense but rather due to a conserved transcriptional network that serves the purpose of attenuating growth upon wounding (Campos et al., 2016).

However, whichever point of view is chosen, it seems as if plant defense is always realized at the expense of growth, whether through the reallocation of metabolic resources or the activation of specific gene regulatory networks hardwired to confront biotic stress situations. When inspecting the beneficial effect of the co-cultivation of plants with S. indica it becomes obvious that there is a discrepancy between the observed effects and the classical growth–defense tradeoff concept. Although S. indica infection triggers defense responses, plant biomass production and productivity in terms of seed yield are increased (Oelmüller et al., 2009; Achatz et al., 2010). It therefore has to be concluded that, along with the basic initial plant defense response, further mechanisms are triggered by the fungus, allowing the host plant to grow despite all of the metabolic restrictions. In order to sustain growth without paying the price of increased stress susceptibility, the underlying mechanisms likely involve the improvement of plant nutrition, which is essential to mitigate the provoked metabolic deficit. The pronounced extension of the root system of infected plants (Fig. 2) suggests that the penetration and exploration of new areas of soil plays an important role in this context.

In their natural soil habitats, plants interact with a broad variety of microorganisms. In particular, microbes in the rhizosphere play important roles in the acquisition of soil nutrients, including the most important macronutrients, nitrogen (N) and phosphorus (P) (Hayat et al., 2010; Hacquard et al., 2015, 2017). Soil microbes can increase nutrient uptake by converting insoluble complexes, which represent unavailable forms of nutrients in the soil, to their ionic forms, which are more suitable for assimilation via the roots. In addition, the beneficial relationships of AMFs with their host plants have been extensively studied. The mycorrhizal symbiosis is known to facilitate improved access to soil nutrients, particularly phosphate (Chiu and Paszkowski, 2019). As previously pointed out, S. indica and other Sebacinales resemble AMFs in many aspects. Thus, it does not appear improbable that those mutualistic fungi may also impact nutrient uptake in their host plants, and indeed similar effects have been reported (Malla et al., 2004; Shahollari et al., 2005; Saddique et al., 2018). In mycorrhizal communities, nutrient exchange involves a number of specific transporters for both the uptake of nutrients by the fungus and their subsequent exchange with the host (Wang et al., 2017). For most other beneficial endophytic fungi, including S. indica, the molecular mechanisms involved in nutrient exchange are only poorly understood and need to be further investigated. Although it is generally assumed that an effective mechanism exists for nutrient absorption and translocation to the host plant by S. indica, in exchange for plant-derived carbon sources (Khalid et al., 2019), the translocation of inorganic phosphate by S. indica is controversial. S. indica has been reported to contain a high-affinity phosphate transporter (PiPT) that is presumably involved in the transfer of inorganic phosphate to the host plant (Yadav et al., 2010; Kumar et al., 2011). However a more recent study claimed that S. indica interferes primarily with P_i distribution and metabolism, rather than directly promoting phosphate uptake from the soil (Bakshi et al., 2017).

With respect to N, it has been suggested that the inoculation of Arabidopsis and Nicotiana tabacum with S. indica induces N uptake and translocation from the culture medium to the aerial parts of the plant. The reported induction of N uptake is seemingly linked to the stimulation of nitrate reductase activity, a key step in nitrate assimilation, and the transcriptional activation of NITRATE REDUCTASE 2 (NIA2) expression in Arabidopsis (Sherameti et al., 2005). In summary, the available data support the idea that S. indica encroaches on the primary metabolism in host plant roots by delivering nutrients necessary for increased growth and development. The exact mechanisms by which S. indica achieves this, however, remain largely unknown.

### Induced drought and salt resistance

In the global climate change scenario, plant growth and productivity are greatly affected by drought stress, and plants have to adapt to prevailing unfavorable conditions in order to survive (Shinozaki and Yamaguchi–Shinozaki, 2007). Drought provokes a number of interconnected physiological and biochemical responses, including stomatal closure, repression of plant growth and photosynthetic activity, and activation of respiration (Schroeder et al., 2001; Flexas et al., 2004; Roelfsema and Hedrich, 2005; Rennenberg et al., 2006). Work conducted over recent years has identified a large number of drought-inducible genes, which can be divided into two major groups: (i) genes that encode proteins directly involved in conveying abiotic stress tolerance, and (ii) genes encoding regulatory proteins, which interfere with signal transduction or the expression of stress-responsive genes. ABA is known to play a central role in the conversion of abiotic stress signals into appropriate cellular responses, although some ABA-independent signaling
pathways have also been shown to contribute to the transcriptional control of abiotic stress-response genes.

S. indica was initially isolated from the roots of shrubs growing in the Thar desert in India. Hence, it is not surprising that this endosymbiont is also capable of attenuating the negative effects of drought stress effects in its host plant. Experiments with Arabidopsis, rice, and maize revealed that plants inoculated with S. indica performed considerably better under drought conditions (Sherameti et al., 2008; Hosseini et al., 2018; Saddique et al., 2018; Zhang et al., 2018). Microarray analyses performed by Zhang et al. (2018) indicated that a quite diverse set of stress-related genes (up to 2037 genes after 12 h of drought stress treatment) responded differentially in plants co-cultivated with S. indica. The detailed breakdown of the transcriptomic data revealed that S. indica promotes maize root growth under drought stress conditions largely through the stimulation of microtubular processes and the strengthening of the plant’s redox capacity by adjusting its carbon–sulfur balance.

Apart from drought stress, soil salinity represents an increasingly serious environmental threat that affects plant growth and yield. Soil salinity disturbs nutrient absorption by the roots, leading to osmotic and ionic imbalances and oxidative damage (Flowers, 2004; Ruiz-Lozano et al., 2012). Similar to drought stress, salt stress leads to stomatal closure because it reduces the water potential of leaves, which results in decreased photosynthetic activity and increased photodamage (Baker and Rosenqvist, 2004). Several studies have indicated that S. indica is able to increase plant tolerance to salinity, partly through the increased conservation of photosynthetic pigments to reduce photodamage (Waller et al., 2005; Jogawat et al., 2013; Sharma et al., 2014; Ghorbani et al., 2018). However, the precise mechanism by which S. indica improves plant growth under conditions of salt stress has yet to be elucidated. The most recent studies underpin the idea that S. indica colonization of roots improves potassium (K+)/sodium (Na+) homeostasis through the transcriptional regulation of the cyclic nucleotide-gated channel CNGC15 and plant cation/proton antiporter (NHX) genes, including SOS1. In addition, the water uptake potential of host plant roots is increased by the transcriptional regulation of aquaporins, in particular PIPs (plasma membrane intrinsic proteins) and TIPs (tonoplast intrinsic proteins), which have been suggested to be involved in water use efficiency and the maintenance of water equilibrium in plants subjected to diverse environmental stresses (Sade et al., 2010; Ghorbani et al., 2019). Intriguingly, the ion homeostasis-and water status-related effects triggered in the host plant are accompanied by an active Na+ detoxification of plant cells by S. indica. The reduction of Na+ contents in plants colonized with S. indica under saline conditions has been attributed to the induction of two ENA ATPases, SiENA1 and SiENA5, which are involved in K+/Na+ and Na+ efflux, respectively (Lanza et al., 2019). In consequence, this active Na+ detoxification suggests the existence of a barrier effect evoked by S. indica that prevents the accumulation of cations in the plant root. Taken together, recent work sheds some light on the molecular bases and mechanisms of S. indica-mediated improved salt tolerance.

Nonetheless, there are still significant gaps in our current understanding of the processes involved, most importantly concerning the underlying gene regulatory networks.

Prospects and conclusions

Fungi of the order Sebacinales have been identified all around the world, including in extreme ecosystems such as deserts. The root endophyte S. indica is capable of associating with all plants tested so far, transferring growth benefits and increased stress tolerance to its host plants under a broad range of different climate, temperature, and growth conditions. As is evident from the data discussed in this review, the molecular mechanisms that confer the beneficial effects upon host plants are highly complex and multi-layered, which makes it unlikely that a single master regulator will be identified that could be used in biotechnological approaches to unleash the full repertoire of fungal effects exerted during symbiosis in transgenic plants. The increasing insight into the molecular bases of the plant–fungal symbiosis will most probably provide evidence for suitable target genes that drive isolated facets of the interaction, such as growth promotion or increased biotic and abiotic stress tolerance.

A particularly interesting starting point for the improvement of agricultural productivity using information gained from studies of plant–microbe interactions is provided by the identification of the cellooligomer CT, which is the active elicitor of [Ca2+]cyt elevations in S. indica CWEs. Apart from the induction of diverse defense responses, cellular Ca2+ signatures have recently been reported to directly contribute to primary root growth and development (Leitão et al., 2019). However, the application of CT does not provide the full effect of symbiosis, and additionally CT is far too expensive for application in the field to be cost-effective. Nevertheless, some more investigation in this direction, for example, by taking a chemical genetics approach to identify reagents that trigger similar physiological responses and can be applied to soils as plant biostimulants to improve plant productivity, would be valuable. Subsequent studies should include a comprehensive examination of the general applicability of identified compounds and the best way to formulate them. A formulation employing slowly degrading coated beads that persist over a long period of time, as is used for enhanced-efficiency fertilizers, could be an effective way to apply putative CT-like reagents (Timilsena et al., 2015). By using such a formulation, the micro-dosage present in the symbiosis could be emulated. The biochemical nature of the compound(s) also has to be considered. CT itself is an energy-rich carbon source that can most likely be used by other saprophytic fungi present in the soil that are able to degrade the triose. In this respect, a slow-release formulation could also help to contain the growth of unwanted fungal pathogens through limiting its availability as a source of carbon. At best, the chemical genetics approach may provide evidence for chemical compounds with CT-like effects on crops that are not digestible by microbes and, therefore, do not represent an easily accessible carbon source. However, with respect to organic farming practices in particular, the application
of synthetic compounds to the field does not come without disadvantages.

In conclusion, the direct application of the fungus \textit{S. indica} as a biostimulant is currently possibly the most suitable method to make use of the beneficial traits transferred by endophytic fungi to their host plants. \textit{S. indica} offers the huge advantage that it will grow axenically and without a host plant. Moreover, it can be propagated at large scale and used as a biocontrol agent \citep{Sun2014}, which underlines its high potential for biotechnological and agricultural applications.

Acknowledgements

The authors are grateful for financial assistance received from the collaborative IPSC research project realized in the framework of the EIG CONCERT-Japan call on Food Crops and Biomass Production Technologies and the related national funding agencies: grant PCIN-2016–037 from the Ministry of Economy and Competitiveness (MINECO), Spain, to SP and JVC; grants 01DR17007A and 01DR17007B from the Federal Ministry of Education and Research (BMBF), Germany, to J–L–M and RO, respectively; grant JPMJSC16C3 from the Japan Science and Technology Agency (JST) to HS; and grant EIG-JC1JAPAN-045 from the Centre National de la Recherche Scientifique (CNRS), France, to AK. CGG was supported by the Severo Ochoa Program for Centers of Excellence in R&D from the Agencia Estatal de Investigación, Spain, grant SEV-2016–0672 (2017–2021) to the Centro de Biotecnología y Genómica de Plantas.

References

Abdelaziz ME, Abdelhattab M, Abdeldayem EA, Atia MAM, Mahmoud AW, Saaed MM, Hirt H. 2019. \textit{Piriformospora indica} alters \textit{Na}+/\textit{K}+ homeostasis, antioxidant enzymes and \textit{LeNHX1} expression of greenhouse tomato grown under salt stress.\textit{ Scientia Horticulturae} 256, 108532.

Achatz B, Kogel KH, Franken P, Waller F. 2010. \textit{Piriformospora indica} mycorrhization increases grain yield by accelerating early development of barley plants.\textit{ Plant Signaling & Behavior}, 1685–1687.

Aglawe SB, Barbadikar KM, Mangrauthia SK, Madhav MS. 2018. New breeding technique “genome editing” for crop improvement: applications, potentials and challenges. 3 Biotech 8, 336.

Andrade-Linares DR, Grosch R, Franken P, Rexer KH, Kost G, Restrepo S, de Garcia MC, Maximova E. 2011. Colorization of roots of cultivated \textit{Solanum lycopersicum} by dark sepatate and other ascomycetous endophytes. Mycologia 103, 710–721.

Baker NR, Rosenqvist E. 2004. Applications of chlorophyll fluorescence can improve crop production strategies: an examination of future possibilities.\textit{ Journal of Experimental Botany} 55, 1607–1621.

Bakshi M, Sherameti I, Meichsner D, Thürich J, Varma A, Johri AK. 2017. \textit{IАОx} induces \textit{Ca}2+ waves are associated with rapid, long-distance root-to-shoot signaling in plants.\textit{ Proceedings of the National Academy of Sciences, USA} 114, 6497–6502.

Böttcher C, Dennis EG, Booker GW, Polyak SW, Boss PK, Davies C. 2012. A novel tool for studying auxin-metabolism: the inhibition of grapevine indole-3-acetic acid-amido synthetase by a reaction intermediate analogue. PLoS One 7, e37632.

Böttcher C, Keyzers RA, Boss PK, Davies C. 2010. Sequestration of auxin by the indole-3-acetic acid-amido synthetase \textit{GH3-1} in grape berry \textit{(Vitis vinifera L.)} and the proposed role of auxin conjugation during ripening.\textit{ Journal of Experimental Botany} 61, 3615–3625.

Buezo J, Estebar R, Cornejo A, López-Gómez P, Marino D, Chamizo-Ampudia A, Gil MJ, Martínez-Merino V, Moran JF. 2019. IАОx induces the SUE phytohormone and differential signaling from IAA under different types of nitrogen nutrition in \textit{Medicago truncatula} roots.\textit{ Plant Science} 287, 110176.

Campos ML, Yoshida Y, Major IT, et al. 2016. Rewiring of jasmonate and phytochrome B signalling uncouples plant growth-defense tradeoffs.\textit{ Nature Communications} 7, 12570.

Chen J, Gutjahr C, Bleckmann A, Dresselhaus T. 2015a. Calcium signaling during reproduction and biotrophic fungal interactions in plants.\textit{ Molecular Plant} 8, 595–611.

Chen Q, Liu Y, Maere S, et al. 2015b. A coherent transcriptional feed-forward motif model for mediating auxin-sensitive PIN2 expression during lateral root development.\textit{ Nature Communications} 6, 8821.

Cheng SH, Willimm MR, Chen HC, Sheen J. 2002. Calcium signaling through protein kinases. The Arabidopsis calcium-dependent protein kinase gene family.\textit{ Plant Physiology} 129, 469–485.

Chiu CH, Paszkowski U. 2019. Mechanisms and impact of symbiotic phosphate acquisition.\textit{ Cold Spring Harbor Perspectives in Biology} 11, a034603.

Choi J, Tanaka K, Cao Y, Qi Y, Qiu J, Liang Y, Lee SY, Stacey G. 2014a. Identification of a plant receptor for extracellular ATP.\textit{ Science} 343, 290–294.

Choi WG, Toyota M, Kim SH, Hilleary R, Girloy S. 2014b. Salt stress-induced \textit{Ca}2+ waves are associated with rapid, long-distance root-to-shoot signaling in plants.\textit{ Proceedings of the National Academy of Sciences, USA} 111, 6497–6502.

Cook DE, Mesarich CH, Thomma BP. 2015. Understanding plant immunity as a surveillance system to detect invasion.\textit{ Annual Review of Phytopathology} 53, 541–563.

Davies PJ. 2010. \textit{Plant hormones. biosynthesis, signal transduction, action!} Dordrecht: Springer Netherlands.

Day IS, Reddy VS, Shad Ali G, Reddy AS. 2002. Analysis of EF-hand-containing proteins in \textit{Arabidopsis}.\textit{ Genome Biology} 3, RESEARCH0056.

de Bary A. 1879. \textit{Die Erscheinung der Symbiose}. Vortrag auf der Versammlung der Naturforscher und Ärzte zu Cassel. Strassburg: Verlag von K. J. Trübner, 1–30.

D'Onofrio N, Sánchez-Vallet A, Goffner D, Molina A. 2013. Disease resistance or growth: the role of plant hormones in balancing immune responses and fitness costs.\textit{ Frontiers in Plant Science} 4, 155.

Deshmukh S, Häckelhoven R, Schönfer P, Imani J, Sharma M, Weiss M, Waller F, Kogel KH. 2006. The root endophytic fungus \textit{Piriformospora indica} requires host cell death for proliferation during mutualistic symbiosis with barley.\textit{ Proceedings of the National Academy of Sciences, USA} 103, 18450–18457.

Dodd AN, Kudla J, Sanders D. 2010. The language of calcium signaling.\textit{ Annual Review of Plant Biology} 61, 593–620.

FAO, IFAD, WFP. 2015. The state of food insecurity in the world 2015. Rome: Food and Agriculture Organization of the United Nations.

Fedorov RV. 2015. Food in a future of 10 billion. Agriculture & Food Security 4, 11.

Flexas J, Bota J, Loreto F, Cornic G, Sharkey TD. 2004. Diffusive and metabolic limitations to photosynthesis under drought and salinity in \textit{C}₄ plants.\textit{ Plant Biology} 6, 269–279.

Flowers TJ. 2004. Improving crop salt tolerance.\textit{ Journal of Experimental Botany} 55, 307–319.

Freirigmann H, Gigaloshvili T. 2014. \textit{MYB34}, \textit{MYB51} and \textit{MYB122} in the regulation of camalexin biosynthesis in \textit{Arabidopsis thaliana}.\textit{ Molecular Plant} 7, 814–828.

Freirigmann H, Glawischig N, Gigaloshvili T. 2015. The role of \textit{MYB34}, \textit{MYB51} and \textit{MYB122} in the regulation of camalexin biosynthesis in \textit{Arabidopsis thaliana}.\textit{ Frontiers in Plant Science} 6, 654.

Freymark G, Diehl T, Miklis M, Remeis T, Panstruga R. 2007. Antagonistic control of powdery mildew host cell entry by barley calcium-dependent protein kinases (CDPKs).\textit{ Molecular Plant-Microbe Interactions} 20, 1213–1221.

Friml J, Benková E, Billou I, et al. 2002. \textit{AIPIN4} mediates sink-driven auxin gradients and root patterning in \textit{Arabidopsis}.\textit{ Cell} 108, 661–673.

Fu SF, Wei JY, Chen HW, Liu YY, Lu HY, Chou JY. 2015. Indole-3-acetic acid: a widespread physiological code in interactions of fungi with other organisms.\textit{ Plant Signaling & Behavior} 10, e1048082.
Gartland KMA, Gartland JS. 2018. Opportunities in biotechnology. Journal of Biotechnology 282, 38–45.

Ghorbani A, Omran VOG, Razavi SM, Pirdashti H, Ranbar M. 2019. Piriformospora indica confers salinity tolerance on tomato (Lycopersicon esculentum Mill.) through amelioration of nutrient accumulation, K+/Na+, homeostasis and water status. Plant Cell Reports 38, 1151–1163.

Ghorbani A, Razavi SM, Ghasemi Omran VO, Pirdashti H. 2018. Piriformospora indica inoculation alleviates the adverse effect of NaCl stress on growth, gas exchange and chlorophyll fluorescence in tomato (Solanum lycopersicum L.). Plant Biology 20, 729–736.

Gill SS, Gill R, Trivedi DK, et al. 2016. Piriformospora indica: potential and significance in plant stress tolerance. Frontiers in Microbiology 7, 332.

Hacquard S, Garrido-Oter R, González A, et al. 2015. Microbiota and host nutrition across plant and animal kingdoms. Cell Host & Microbe 17, 603–616.

Hacquard S, Spaepen S, Garrido-Oter R, Schulze-Lefert P. 2017. Interplay between innate immunity and the plant microbiota. Annual Review of Phytopathology 55, 685–689.

Harman GE. 2011. Multifunctional fungal plant symbionts: new tools to enhance plant growth and productivity. New Phytologist 189, 647–649.

Hashimoto K, Kudia J. 2011. Calcium decoding mechanisms in plants. Biochimie 93, 2054–2069.

Hayat R, Ali S, Amara U, Khalid R, Ahmed I. 2010. Soil beneficial bacteria and their role in plant growth promotion: a review. Annals of Microbiology 60, 579–598.

Herms DA, Mattson WJ. 1992. The dilemma of plants: to grow or defend. Quarterly Review of Biology 67, 283–335.

Hertig M, Taliaferro WH, Schwartz B. 1992. The dilemma of plants: to grow or defend. Quarterly Review of Biology 67, 326–329.

Hilbert M, Voll LM, Ding Y, Hofmann J, Sharma M, Zuccaro A. 2012. Indole derivative production by the root endophyte Piriformospora indica is not required for growth promotion but for biotrophic colonization of barley roots. New Phytologist 196, 520–534.

Hosseini F, Mosaddegh MR, Dexter AR, Sepehr M. 2018. Maize water status and physiological traits as affected by root endophytic fungus Piriformospora indica under combined drought and mechanical stresses. Planta 247, 1220–1246.

Hua MD, Senthil Kumar R, Shyur LF, Cheng YB, Tian Z, Oelmüller R, Yeh KW. 2017. Metabolomic compounds identified in Piriformospora indica-colonized Chinese cabbage roots delineate symbiotic functions of the interaction. Scientific Reports 7, 9291.

Hucu B, Yao J, Montgomery BL, He SY. 2014. Growth-defense tradeoffs in plants: a balancing act to optimize fitness. Molecular Plant 7, 1267–1287.

Irmisch S, McCormick AC, Boeckler GA, et al. 2013a. Two herbivore-induced cytochrome P450 enzymes CYP79D6 and CYP79D7 catalyze the formation of volatile aldoximes involved in poplar defense. The Plant Cell 25, 4737–4754.

Irmisch S, Unsicker SB, Gershenzon J, Köllner TG. 2013b. Identification and biochemical characterization of CYP79D6v4, a cytochrome P450 enzyme producing aldoximes involved in poplar defense. The Plant Cell 25, 4737–4754.

Leeyc Johnson JM, Chien CT, Sun C, Cai D, Lou B, Oelmüller R, Yeh KW. 2011. Growth promotion of Chinese cabbage and Arabidopsis by Piriformospora indica is not stimulated by mycelium-synthesized auxin. Molecular Plant-Microbe Interactions 24, 421–431.

Lehmann T, Janowitz T, Sánchez-Parra B, Alonso MP, Trompetter I, Piotrowski M, Pollmann S. 2017. Arabidopsis NITRILASE 1 contributes to the regulation of root growth and development through modulation of auxin biosynthesis in seedlings. Frontiers in Plant Science 8, 36.

Leitão N, Dansveille P, Carter R, Charpentier M. 2019. Nuclear calcium signals are associated with root development. Nature Communications 10, 4865.

Li F, Wang J, Ma C, Zhao Y, Wang Y, Hasi A, Qi Z. 2013. Glutamate receptor-like channel3.3 is involved in mediating glutathione-triggered cytosolic calcium transients, transcriptional changes, and innate immunity responses in Arabidopsis. Plant Physiology 162, 1497–1509.

Li G, Meng X, Wang R, Mao G, Han L, Liu Y, Zhang S. 2012. Dual-level regulation of ACC synthase activity by MPK3/MPK6 cascade and its downregulation by constitutively active receptor-like channel 3.3 is involved in mediating glutathione-triggered cytosolic calcium transient. Plant Physiology 162, 3364–3378.

Leitão N, Dansveille P, Carter R, Charpentier M. 2019. Nuclear calcium signals are associated with root development. Nature Communications 10, 4865.

Leeyc Johnson JM, Chien CT, Sun C, Cai D, Lou B, Oelmüller R, Yeh KW. 2011. Growth promotion of Chinese cabbage and Arabidopsis by Piriformospora indica is not stimulated by mycelium-synthesized auxin. Molecular Plant-Microbe Interactions 24, 421–431.

Lehmann T, Janowitz T, Sánchez-Parra B, Alonso MP, Trompetter I, Piotrowski M, Pollmann S. 2017. Arabidopsis NITRILASE 1 contributes to the regulation of root growth and development through modulation of auxin biosynthesis in seedlings. Frontiers in Plant Science 8, 36.

Leitão N, Dansveille P, Carter R, Charpentier M. 2019. Nuclear calcium signals are associated with root development. Nature Communications 10, 4865.

Li F, Wang J, Ma C, Zhao Y, Wang Y, Hasi A, Qi Z. 2013. Glutamate receptor-like channel3.3 is involved in mediating glutathione-triggered cytosolic calcium transients, transcriptional changes, and innate immunity responses in Arabidopsis. Plant Physiology 162, 1497–1509.

Li G, Meng X, Wang R, Mao G, Han L, Liu Y, Zhang S. 2012. Dual-level regulation of ACC synthase activity by MPK3/MPK6 cascade and its downstream WRKY transcription factor during ethylene induction in Arabidopsis. PLOS Genetics 8, e1002767.

Li L, Li M, Yu L, et al. 2014. The FLS2-associated kinase BIK1 directly phosphorylates the NADPH oxidase RboH1 to control plant immunity. Cell Host & Microbe 15, 329–339.

Liu H, Smythikumar R, Ma G, Zou Q, Zhu K, Shen X, Tian D, Hua MS, Oelmüller R, Yeh KW. 2019. Piriformospora indica-induced phytocrome changes and root colonization strategies are highly host-specific. Plant Signaling & Behavior 14, 1632888.

Lu D, Wu S, Gao X, Zhang Y, Shan L, He P. 2010. A receptor-like cytoplasmic kinase, BIK1, associates with a flagellin receptor complex to initiate plant innate immunity. Proceedings of the National Academy of Sciences, USA 107, 496–501.
Hormonal and physiological changes driven by fungal endophytes

Ramos P, Rivas N, Pollmann S, Casati P, Molina-Montenegro MA. 2004. Phosphorus solubilizing symbiotic fungus: Piriformospora indica. Endocytobiosis Cell Research 15, 579–600.

Manzoor H, Kelloniemi J, Chiltz A, Wendehenne D, Pugin A, Poinssot S, Garcia-Brugger A. 2013. Involvement of the glutamate receptor ATGLR5.3 in plant defense signaling and resistance to HyaIoperonospora arabidopsidis. The Plant Journal 76, 466–480.

McAinsh MR, Pittman JK. 2009. Shaping the calcium signature. New Phytologist 181, 275–294.

Meents AK, Furch ACU, Almeida-Trapp M, et al. 2019. Beneficial and pathogenic Arabidopsis root-interacting fungi differentially affect auxin levels and responsive genes during early infection. Frontiers in Microbiology 10, 380.

Menke FL, van Pelt JA, Pieterse CM, Kissig DF. 2004. Silence of the mitogen-activated protein kinase MPK6 compromises disease resistance in Arabidopsis. The Plant Cell 16, 897–907.

Molina-Montenegro MA, Oses R, Torres-Diaz C, Atala C, Zurita-Silva A, Ruíz-Lara S. 2016. Root-endophytes improve the ecophysiological performance and production of an agricultural species under drought conditions. AoB Plants 8, pw062.

Mavec J, Škupá P, Ballay A, et al. 2009. Subcellular homeostasis of phytohormone auxin is mediated by the ER-localized PIN5 transporter. Nature 459, 1136–1140.

Murray DR. 1997. Carbon dioxide and plant responses. Taunton, Research Studies Press.

Nizam S, Qiang X, Wawra S, Nostadt R, Getzke F, Schwanke F, Dreyer I, Langen G, Zuccaro A. 2019. Serendipita indica E5NT modulates extracellular nucleotide levels in the plant apoplast and affects fungal colonization. EMBO Reports 20, e47430.

Nongbri PL, Johnson JM, Sherameti I, Glawischng E, Halkier BA, Oelmüller R. 2012. Indole-3-acetamide-derived compounds restrict root colonization in the beneficial interaction between Arabidopsis roots and the endophyte Piriformospora indica. Molecular Plant-Microbe Interactions 25, 1186–1197.

Nühse TS, Peck SC, Hirt H, Boller T. 2000. Microbial elicitors induce activation and dual phosphorylation of the Arabidopsis thaliana MAPK 6. Journal of Biological Chemistry 275, 7521–7526.

Oelmüller R. 2013. Sensing environmental and developmental signals via callosegomerases. Journal of Plant Physiology 229, 1–6.

Oelmüller R, Sherameti I, Tripathi S, Varma A. 2009. Piriformospora indica, a cultivable root endophyte with multiple biotechnological applications. Symposium 49, 1–17.

Overvoorde P, Fukaki H, Beeckman T. 2010. Auxin control of root development. Cold Spring Harbour Perspectives in Biology 2, a010537.

Peiter E, Maathuis FJ, Mills LN, Knight H, Pellaux J, Hetherington AM, Sanders D. 2005. The vacuolar Ca2+–activated channel TPC1 regulates germination and stomatal movement. Nature 434, 404–408.

Peškan-Berghöfer T, Shahollari B, Giong PH, Hehl S, Markert C, Blanke V, Kost G, Varma A, Oelmüller R. 2004. Association of Piriformospora indica with Arabidopsis thaliana roots represents a novel system novel to study beneficial plant–microbe interactions and involves early plant root modifications in the endoplasmic reticulum and at the plasma membrane. Physiologia Plantarum 122, 465–477.

Petit JR, Jouzel J, Raynaud D, et al. 1999. Climate and atmospheric history of the past 240,000 years from the Vostok ice core, Antarctica. Nature 399, 429–436.

Pollmann S, Müller A, Weiler EW. 2006. Many roads lead to “auxin”: of nitrilases, synthases, and amidases. Plant Biology 8, 326–333.

Qiang X, Weiss M, Kogel KH, Schäfer P. 2012a. Piriformospora indica—a mutualistic basidiomycete with an exceptionally large plant host range. Molecular Plant Pathology 13, 508–518.

Qiang X, Zechmann B, Reitz MU, Kogel KH, Schäfer P. 2012b. The mutualistic fungus Piriformospora indica colonizes Arabidopsis roots by inducing an endoplasmic reticulum stress-triggered caspase-dependent cell death. The Plant Cell 24, 794–809.

Ramos P, Rivas N, Pollmann S, Casati P, Molina-Montenegro MA. 2018. Hormonal and physiological changes driven by fungal endophytes increase Antarctic plant performance under UV-B radiation. Fungal Ecology 34, 76–82.

Ren D, Liu Y, Yang K, Han L, Mao G, Glazebrook J, Zhang S. 2008. A fungal-responsive MAPK cascade regulates phytoalexin biosynthesis in Arabidopsis. Proceedings of the National Academy of Sciences, USA 105, 5638–5643.

Rennenberg H, Loreto F, Polle A, Brilli F, Fares S, Beniwal RS, Gessler A. 2006. Physiological responses of forest trees to heat and drought. Plant Biology 8, 556–571.

Rodriguez RJ, Henson J, Van Volkenburgh E, Hoy M, Wright L, Beckwith F, Kim YO, Redman RS. 2008. Stress tolerance in plants via habitat-adapted symbiosis. The ISME Journal 2, 404–416.

Rodriguez RJ, Redman RS, Henson JM. 2004. The role of fungal symbioses in the adaptation of plants to high stress environments. Mitigation and Adaptation Strategies for Global Change 9, 261–272.

Roelfsema MR, Hedrich R. 2005. In the light of stomatal opening: new insights into the ‘Watergate’. New Phytologist 167, 665–691.

Ruiz-Lozano JM, Porcel R, Azcón A, Arcosa R. 2012. Regulation by arbuscular mycorrhiza of the integrated physiological response to salinity in plants: new challenges in physiological and molecular studies. Journal of Experimental Botany 63, 4033–4044.

Saddique MAB, Ali Z, Khan AS, Rana IA, Shamsi IH. 2018. Inoculation with the endophyte Piriformospora indica significantly affects mechanisms involved in osmotic stress in rice. Rice 11, 34.

Sade N, Gebretsadik M, Seigmann R, Schwach A, Wallach R, Moshelion M. 2010. The role of tobacco Aquaporin1 in improving water use efficiency, hydraulic conductivity, and yield production under salt stress. Plant Physiology 152, 245–254.

Schäfer P, Piffii S, Voll LM, et al. 2009. Manipulation of plant innate immunity and gibberellin as factor of compatibility in the mutualistic association of barley roots with Piriformospora indica. The Plant Journal 59, 461–474.

Schroeder JI, Kwak JM, Allen GJ. 2001. Guard cell abscisic acid signalling and engineering drought hardness in plants. Nature 410, 327–330.

Schuman MC, Baldwin IT. 2016. The layers of plant responses to insect herbivores. Annual Review of Entomology 61, 373–394.

Shahollari B, Varma A, Oelmüller R. 2005. Expression of a receptor kinase in Arabidopsis roots is stimulated by the basidiomycete Piriformospora indica and the protein accumulates in Triton X-100 insoluble plasma membrane microdomains. Journal of Plant Physiology 162, 945–958.

Sharma P, Kharkwal AC, Abdin MZ, Varma A. 2014. Piriformospora indica improves micropropagation, growth and phytochemical content of Aloe vera L. plants. Symbiosis 64, 11–23.

Sherameti I, Shahollari B, Venus Y, Altschmied L, Varma A, Oelmüller R. 2005. The endophytic fungus Piriformospora indica stimulates the expression of nitrate reductase and the starch-degrading enzyme glucon-water dikinase in tobacco and Arabidopsis roots through a homeomdomain transcription factor that binds to a conserved motif in their promoters. Journal of Biological Chemistry 280, 26241–26247.

Sherameti I, Tripathi S, Varma A, Oelmüller R. 2008. The root-colonizing endophyte Piriformospora indica confers drought tolerance in Arabidopsis by stimulating the expression of drought stress–related genes in leaves. Molecular Plant-Microbe Interactions 21, 799–807.

Shi H, Shen Q, Qi Y, Yan H, Nie H, Chen Y, Zhao T, Katagiri F, Tang D. 2013. BR-SIGNALING KINASE1 physically associates with FLAGELLIN SENSING2 and regulates plant innate immunity in Arabidopsis. The Plant Cell 25, 1143–1157.

Shinozaki K, Yamaguchi-Shinozaki K. 2007. Gene networks involved in drought stress response and tolerance. Journal of Experimental Botany 58, 221–227.

Souza CA, Li S, Lin AZ, Boutot F, Grossmann G, Zipfel C, Somerville SC. 2017. Cellulose-derived oligomers act as damage-associated molecular patterns and trigger defense-like responses. Plant Physiology 173, 2383–2398.

Speapen S, Vanderleyden J, Remans R. 2007. Indole-3-acetic acid in microbial and microorganism-plant signaling. FEMS Microbiology Reviews 31, 425–448.

Staswick PE, Serban B, Rowe M, Tiryaki I, Maldonado MT, Maldonado MC, Suzá W. 2005. Characterization of an Arabidopsis enzyme family that conjugates amino acids to indole-3-acetic acid. The Plant Cell 17, 616–627.
Plant–fungus interactions in extreme ecosystems | 3877

Strehmel N, Monchgesang S, Herklotz S, Kruger S, Ziegler J, Scheel D. 2016. Piriformospora indica stimulates root metabolism of Arabidopsis thaliana. International Journal of Molecular Sciences 17, 1091.

Su ZZ, Wang T, Shrivastava N, Chen YY, Liu X, Sun C, Yin Y, Gao QK, Lou BG. 2017. Piriformospora indica promotes growth, seed yield and quality of Brassica napus L. Microbiological Research 199, 29–39.

Sun C, Shao Y, Vahabi K, et al. 2014. The beneficial fungus Piriformospora indica protects Arabidopsis from Verticillium dahliae infection by downregulation plant defense responses. BMC Plant Biology 14, 268.

Takahashi F, Yoshida R, Ichinomiya K, Mizoguchi T, Seo S, Yonezawa M, Maruyama K, Yamaguchi-Shinozaki K, Shinozaki K. 2007. The mitogen-activated protein kinase cascade MKK3–MPK6 is an important part of the jasmonate signal transduction pathway in Arabidopsis. The Plant Cell 19, 805–818.

Thürich J, Meichsner D, Furch ACU, Pfalz J, Krüger T, Kniemeyer O, et al. 2005. The Plant part of the jasmonate signal transduction pathway in mitogen-activated protein kinase cascade MKK3–MPK6 is an important part of the jasmonate signal transduction pathway in Arabidopsis. The Plant Cell 19, 805–818.

Timmerse Y, Adhikari R, Casey P, Muster T, Gill H, Adhikari B. 2015. Enhanced efficiency fertilisers: a review of formulation and nutrient release patterns. Journal of the Science of Food and Agriculture 95, 1131–1142.

Upadhyaya CP, Gururani MA, Prasad R, Verma A. 2013. A cell wall extract from Piriformospora indica promotes tuberization in potato (Solanum tuberosum L.) via enhanced expression of Ca2+ signaling pathway and lipoxynagenase gene. Applied Biochemistry and Biotechnology 170, 743–755.

Vadassery J, Ranf S, Drzewiecki C, Mithöfer A, Mazars C, Scheel D, Lee J, Oelmüller R. 2009. A cell wall extract from the endophytic fungus Piriformospora indica promotes growth of Arabidopsis seedlings and induces intracellular calcium elevation in roots. The Plant Journal 59, 193–206.

Vadassery J, Ritter C, Venus Y, Camehl I, Varma A, Shahollari B, Novák O, Strnad M, Ludwig-Müller J, Oelmüller R. 2008. The role of auxins and cytokinins in the mutualistic interaction between Arabidopsis and Piriformospora indica. Molecular Plant-Microbe Interactions 21, 1371–1383.

Vahabi K, Sherameti I, Bakshi M, Mrozinska A, Ludwig A, Reichelt M, Oelmüller R. 2015. The interaction of Arabidopsis with Piriformospora indica shifts from initial transient stress induced by fungus-released chemical mediators to a mutualistic interaction after physical contact of the two symbionts. BMC Plant Biology 15, 58.

van’t Padje A, Whiteside MD, Kiers ET. 2016. Signals and cues in the evolution of plant-microbe communication. Current Opinion in Plant Biology 32, 47–52.

Varma A, Savita Verma, Sudha, Sahay N, Butehorn B, Franken P. 1999. Piriformospora indica, a cultivable plant-growth-promoting root endophyte. Applied and Environmental Microbiology 65, 2741–2744.

Verma S, Varma A, Lexer K-H, Hassel A, Kost G, Sarbhoy A, Bisen P, Bütehorn B, Franken P. 2016. Piriformospora indica, gen. et sp. nov., a new root-colonizing fungus. Mycologia 90, 896–903.

Waller F, Achatz B, Baltruschat H, et al. 2005. The endophytic fungus Piriformospora indica reprograms barley to salt-stress tolerance, disease resistance, and higher yield. Proceedings of the National Academy of Sciences, USA 102, 13386–13391.

Wang W, Shi J, Xie Q, Jiang Y, Yu N, Wang E. 2017. Nutrient exchange and regulation in arbuscular mycorrhizal symbiosis. Molecular Plant 10, 1147–1158.

Weiss M, Selosse MA, Lexer KH, Urban A, Oberwinkler F. 2004. Sebacinales: a hitherto overlooked cosmr of heterobasidiomycetes with a broad mycorrhizal potential. Mycological Research 108, 1003–1010.

Weiß M, Sykorová Z, Garnica S, Riess K, Martos F, Krause C, Oberwinkler F, Bauer R, Redecker D. 2011. Sebacinales everywhere: previously overlooked ubiquitous fungal endophytes. PLoS One 6, e16793.

Weiß M, Waller F, Zuccaro A, Selosse MA. 2016. Sebacinales - one thousand and one interactions with land plants. New Phytologist 211, 20–40.

Yadav V, Kumar M, Deep DK, Kumar H, Sharma R, Tripathi T, Tuteja N, Saxena AK, Johri AK. 2010. A phosphate transporter from the root endophytic fungus Piriformospora indica plays a role in phosphate transport to the host plant. Journal of Biological Chemistry 285, 26532–26544.

Zhang J, Li W, Xiang T, et al. 2010. Receptor-like cytoplasmic kinases integrate signaling from multiple plant immune receptors and are targeted by a Pseudomonas syringae effector cell. Host & Microbe 7, 290–301.

Zhang W, Wang J, Xu L, Wang A, Huang L, Du H, Qiu L, Oelmüller R. 2018. Drought stress responses in maize are diminished by Piriformospora indica. Plant Signaling & Behavior 13, e1414121.

Zipfel C, Oldroyd GE. 2017. Plant signalling in symbiosis and immunity. Nature 543, 328–336.

Zuccaro A, Baslewiecz M, Zurawska M, Biedenkopf D, Kogel KH. 2009. Karyotype analysis, genome organization, and stable genetic transformation of the root colonizing fungus Piriformospora indica. Fungal Genetics and Biology 46, 543–550.

Zuccaro A, Lahmann U, Güldener U, et al. 2011. Endophytic life strategies decoded by genome and transcriptome analyses of the mutualistic root symbiont Piriformospora indica. PLOS Pathogens 7, e1002290.

Züst T, Rasmann S, Agrawal AA. 2015. Growth–defense tradeoffs for two major anti-herbivore traits of the common milkweed Asclepias syriaca. Oikos 124, 1404–1415.