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Current Opinion in Microbiology

Dissecting endophytic lifestyle along the parasitism/mutualism continuum in *Arabidopsis*

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Mutualistic interactions between plants and fungi often occur in the rhizosphere, although examples exist where shoot-endophytes support host growth and increase resistance to pathogens and herbivores. Fungal endophytes which colonize their hosts without any visible disease symptoms have been recognized to be fundamental components of various ecosystems. Initial efforts have been taken to decipher the genetic basis of beneficial plant–fungus interactions and of lifestyle transitions. This review gives a short overview on well established experimental systems amenable to genetic manipulation and of known genome sequence for dissecting plant–fungal endophyte interactions with a special focus on *Arabidopsis thaliana* associations.

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Current Opinion in Microbiology 2016, **32**:103–112

This review comes from a themed issue on **Host-microbe interactions: fungi**

Edited by **Elaine Bignell** and **Bart PHJ Thomma**

For a complete overview see the Issue and the Editorial

Available online 6th June 2016

http://dx.doi.org/10.1016/j.mib.2016.05.008

Introduction

Fungal endophytes are a ubiquitous and phylogenetically diverse group of organisms that engage in remarkably stable long-term interactions with their hosts. Their effects can vary depending on the plant host (including plant genotype), the physiological status of the host, nutrient availability, environmental conditions and interaction with the microbiome [1,2•,3–6]. Despite their abundance in many ecosystems, their mode of action, ecology and evolution is poorly understood. The broad definition of endophytes which does not specify a functional relationship implies that in addition to commensalistic symbionts, endophytes can span from latent pathogens or latent saprotrophs to mutualistic associations.

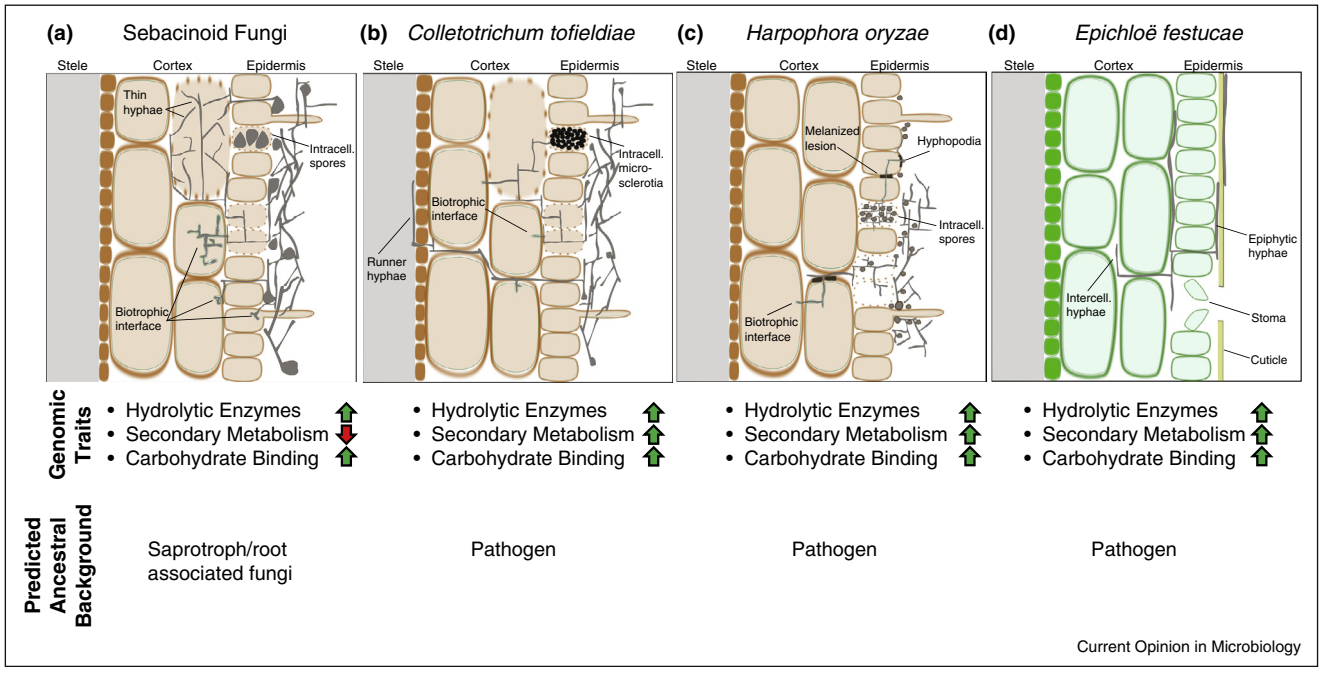
In the past years different experimental systems for endophytes have been established for dissecting mutualistic interactions with plants. These include, among those amenable to genetic manipulation and of known genome sequence, the generalist root endophytic and orchid associated fungi of the order Sebacinales (Basidiomycota) with *Serendipita indica* (syn: *Piriformospora indica*) and *Serendipita vermifera* strains (syn: *Sebacina vermifera*), the recently described root/systemic endophyte *Colletotrichum tofieldiae* (Ascomycota, Sordariomycetes, Glomerellaceae) in natural populations of *Arabidopsis thaliana*, the dark septate rice root endophyte *Harpophora oryzae* (Ascomycota, Sordariomycetes, Magnaporthaceae) and the shoot grass endophytes of the genus *Epichloë* (Ascomycota, Sordariomycetes, Clavicipitaceae) (Figure 1). A different group of beneficial fungi with biocontrol activities [7] is represented by the root associated *Trichoderma* species (Ascomycota, Sordariomycetes, Hypocreaceae), among which *Trichoderma harzianum* has been recently described to colonize *A. thaliana* roots [8,9]. To establish compatibility with their host, endophytic fungi have evolved diverse colonization strategies with distinct morphological, functional and genomic specializations as well as different degrees of interdependence and host specificity.

Sebacinales represent an ancient order of the Agaricomycotina (reconciliation analyses suggest that the divergence of Sebacinales occurred approximately 270 Mya) with a vast array of beneficial lifestyles ranging from ectomycorrhiza to endophytes, but also saprotrophic fungi such as *Paulisebacina*, *Craterocolla*, *Efibulobasidium*, *Chaetospermum* and *Globulisebacina* species have been described [10–12]. The root associated Sebacinales derived most probably from a saprotrophic ancestor [10,12] and no descriptions of pathogenic strains were reported from natural systems (Figure 1a).

C. tofieldiae, originally isolated from asymptomatic *A. thaliana* plants, initiates endophytic growth via the roots and occasionally spreads systemically into shoots without causing discernible disease symptoms (Figure 1b) [13]. This isolate was shown to transfer phosphorus from the fungus via roots to *A. thaliana* shoots and to promote plant growth under phosphorus-deficient conditions [14••]. *C. tofieldiae* is closely related to pathogenic *Colletotrichum* species such as *C. incanum* [15••].

The root endophyte *H. oryzae* was first described among endophytes residing in domestic Chinese wild rice (*Oryza granulata*), where it promotes growth and biomass accumulation [16•]. *H. oryzae* (Figure 1c) can protect rice roots from invasion by the rice blast fungus *Magnaporthe oryzae*

Figure 1



Schematic representation of the colonization strategies of fungal endophytes. **(a)** Sebacinoid root endophytes (*S. indica* and *S. vermifera*) are able to colonize the intercellular space of plant roots and epidermis and cortex cells intracellularly. At early colonization stages the penetrating fungal hyphae are multilobed and bulbous, are enveloped by a plant-derived plasma membrane reflecting the vitality of the colonized cells and form the biotrophic interface. At later stages thinner hyphae are visible that are predominantly found in dead or dying root cells where they massively proliferate and form chlamydospores. The colonization is restricted to the epidermis and the root cortex and fungal hyphae are never found in the endodermis and the central cylinder. Genomic analysis of the sebacinoid fungi revealed the expansion ofgene families encoding proteins with hydrolytic activity and with carbohydrate binding properties (green bottom-up arrow) whereas gene families involved in secondary metabolism are contracted (red top-down arrow). **(b)** *C. tofieldiae* is able to intercellularly and intracellularly colonize the root tissue. Colonized epidermal cells undergo at later stages cell death whereas root cortex cells remain viable for a longer period of time. During colonization of vital cells the fungal hyphae stay enveloped by host derived fungal membrane. Black, melanized microsclerotia are massively deposited within dead cells at later stages. Runner hyphae occasionally reach the central cylinder resulting in a systemic colonization of the aerial plant parts. Genomic traits of *C. tofieldiae* are the expansion of genes encoding hydrolytic enzymes, proteins involved in secondary metabolism and carbohydrate binding proteins (green bottom-up arrow). **(c)** *Harpophora oryzae* forms hyphopodia to penetrate root epidermal cells, where the fungus mostly resides during the whole interaction. *H. oryzae* is furthermore also able to enter the root tissue intercellularly. Penetrated cells remain alive at early stages of the interaction forming the biotrophic interface but display melanized lesions and eventually undergo cell death at later stages of the interaction. When bridging the cell wall during cell to cell growth, *H. oryzae* hyphae form constricted neck-like structures. The hyphae are also able to reach the root cortex layer but never reach the central cylinder, thus the interaction is restricted to the root tissue. At later stages thick-walled, melanized chlamydospores are found within dead root epidermal cells. The *H. oryzae* genome displays an expansion of genes encoding hydrolytic enzymes, proteins involved in secondary metabolism and carbohydrate binding proteins (green bottom-up arrow). **(d)** *Epichloë festucae* is able to grow intercellularly within the leaf tissue or epiphytically on the leaf surface above or below the cuticle. *E. festucae* hyphae are found solely in the extracellular space. Spores are deposited in so called stromas and on the leaf surface or the fungus directly infects seeds for propagation. The *E. festucae* genome displays an expansion of genes encoding hydrolytic enzymes, proteins involved in secondary metabolism and carbohydrate binding proteins (green bottom-up arrow). The putative ancestral background differs for these endophytes.

and can induce systemic resistance. Phylogenetic analyses have shown that *H. oryzae* is closely related to members of the Magnaporthaceae, such as *Gaeumannomyces* and *Magnaporthe*, most of which are plant pathogens [17••].

Clavicipitaceous endophytes of grasses which include *Epichloë* species and their asexual descendants spend most of their life cycle in the intercellular space of stems, leaves, inflorescences and seeds (but not in roots, although *E. coenophialum* has also been detected in root tissues) of their coolseason hosts [18,19•] (Figure 1d). They span the symbiotic continuum from antagonism to mutualism. Sexual *Epichloë* species transmitted horizontally via sexual spores can suppress the development of their host plant's inflorescence whereas the asexual species transmit vertically in healthy grass seeds and can confer benefits to their host, including the production of different herbivore-deterring alkaloids, growth promotion and increased stress resistance to abiotic stress such as drought [20,21]. *Epichloë* species are phylogenetically

placed within a group of endophytic and plant pathogenic fungi, whose common ancestor probably derived from an animal pathogen [22].

Intensive work with these plant-endophyte model systems has suggested that fungal endophytes could be latent or quiescent pathogens where for example a change in the host environment may lead to activation of pathogenic behavior [6,14••,23,24,25••]. *De facto* many of the endophytes described are closely related to pathogens and they still retain a number of pathogenic features. On the other hand in fungi where root endophytism and saprotrophism seem ancestral, like in the Sebacinales where no closely related pathogenic members are known, it has been hypothesized that the close relationship with roots allowed the evolution of tighter beneficial associations such as ectomycorrhizal interactions [11,12]. These endophytes with strong saprotrophic abilities which are able to enter living hosts are speculated to become more active on senescent plant organs, gaining a competitive advantage over other saprotrophs [26]. Both hypotheses are supported by diverse studies [nicely reviewed in [2•,6,27]] but little is known about the mechanisms that trigger the shift from endophytic to pathogenic or to saprotrophic colonization in an immune compromised or senescent host or on a different host species.

Genomic signature of root endophytes

The genome of a fungus contains the signatures of its phylogenetic position and also of the kind of lifestyle to which it is adapted. Although it is probably too early to draw compelling conclusions on which genomic traits are important for the endophytic lifestyle of fungi as only few genomes are available from taxonomically distinct taxa with very diverse ancestor lifestyles, an overall picture is forming. Ectomycorrhizal lifestyle arose independently multiple times from saprotrophic ancestors during evolution. This lifestyle transition was tightly associated with the loss of genes encoding plant cell wall degrading enzymes (PCWDEs) and with the evolution of lineage-specific subsets of symbiosis-induced genes [12]. In contrast with ectomycorrhizal fungi belonging to the class Agaricomycetes, the transition to root endophytism in *C. tofieldiae*, *S. indica* and *H. oryzae* is not accompanied by contraction of their PCWDE repertoires [15••,25••]. Endophytic *Epichlöe* spp., *C. tofieldiae* and *H. oryzae* are beneficial fungi derived from a pathogenic ancestor and embedded within pathogenic species [15••,17••,22]. These fungal endophytes have possibly evolved either by direct evolution from a single pathogenic species, probably due to loss of sexual state as suggested for the asexual *Epichlöe* species or through interspecific hybridization events between either sexual species or distinct sexual and asexual lineages [28], or via loss of genes encoding effectors as suggested for *C. tofieldiae* [15••]. In contrast to this the root endophytes and orchid mycorrhizal fungi from the order Sebacinales are embedded within root associated or saprotrophic but not pathogenic fungi [11]. It can be assumed that the ancestral background plays an important role in the lifestyle-associated genomic adaptations retained by this distinct class of endophytes. Whereas in *C. tofieldiae*, *Epichlöe* spp., and *H. oryzae* genes encoding secondary metabolites are well represented, in the sequenced Sebacinoid taxa these genes are indeed strongly reduced (Figure 1).

Dissecting Arabidopsis–fungal endophyte interactions

The model plant *Arabidopsis thaliana* is by far the best studied model among flowering plants and like most of the Brassicaceae, it has lost the ability to form mycorrhizal symbiosis during evolution [29,30]. The Sebacinoid endophytes *S. indica*, *S. vermifera* and *S. herbamans* gained remarkable interest in the past few years due to their ability to engage in beneficial interaction with *A. thaliana* under controlled laboratory conditions [25••,31,32]. Lately members of Sebacinales were detected in environmental samples of *A. thaliana* roots, suggesting that Sebacinoid fungi can associate with Brassicaceae in natural ecosystems [33] although they are not the most common colonizers in this plant family. Sebacinoid fungi are often described as generalists and ubiquitously distributed. Their sequences can range from 0.44% to 11.3% of all fungal sequences in different natural and managed ecosystems, and thus have been proposed to be important hidden players in terrestrial ecosystems [32–34,35•]. Despite their wide distribution a recent study suggested that they are threatened by conventional agriculture where plant diversity is reduced and pesticides or mineral fertilizers are used [35•]. It is unclear which mechanisms are responsible for the reduction of Sebacinales occurrence in intensively used agricultural sites and which functional consequences this reduction may have, but in accordancewith their broad host range a correlation between plant diversity and Sebacinales occurrence was observed [35•]. The growth promoting effects of Sebacinoid fungi include increased root and shoot biomass and fertility in different hosts including *A. thaliana* [31,36]. These effects are especially (but not solely) evident under low nutrients availability and occur independently of plant common symbiosis genes [37]. Additionally it was shown that root colonization by *S. indica* leads to an enhanced nitrogen and phosphate uptake of the plant [38,39] and to increased drought stress tolerance [40]. Whether these have any functional relevance for plant fitness in natural and managed ecosystems remains to be clarified.

The recent discovery and characterization of the *A. thaliana* endophyte *C. tofieldiae* [13], promoting growth of its natural host under low phosphate [14••], represents an important step towards the understanding of the ecological relevance of endophytic interactions. This endophyte

was isolated from the leaves of *A. thaliana* natural populations in central Spain but was not found in populations in Germany and France [13,14••]. *C. tofieldiae* was also reported as an endophyte in trees and orchids across the Eurasian continent [41,42] suggesting a broad host range. *C. tofieldiae* is closely related to pathogenic *Colletotrichum* spp. and diverged only recently (estimated 8.8 Mya) but its ability to promote the growth of *A. thaliana* under low phosphate conditions suggests a beneficial potential for this endophyte [14••,15••]. Under these conditions colonization of *C. tofieldiae* leads to higher phosphate levels in the shoot of *A. thaliana* and to an induction of plant phosphate transporters [14••]. Surprising for a mutualistic fungus which supports phosphate transfer to the host, *C. tofieldiae* occasionally produces runner hyphae that are able to enter the central cylinder and systemically colonize also the aerial part of the plant [14••] reminiscent of a pathogenic association. *C. tofieldiae* partially retained its pathogenic ability on different hosts (e.g. it strongly inhibits the growth of *Capsella rubella*) [14••], but the narrowed repertoire of secreted effector proteins and the reduction of the *in planta* activated pathogenicity-related genes compared to its pathogenic relative *C. incanum* [43,44] indicate that some of the pathogenic abilities are possibly lost [15••]. Indeed the modification of a phytopathogenic *Colletotrichum* sp. into a nonpathogenic endophyte by a mutation at a single genetic locus was demonstrated [45••]. These authors showed that the WT *C. magna* and the mutant path-1 were both capable of systemic growth in susceptible plants, however path-1 did not produce visible disease symptoms and protected the host from disease caused by virulent pathogens [45••]. These data strongly suggest that mutation of one gene or closely linked genes critical to disease development after infection can change the fundamental biological description of an isolate from a pathogen to an endophytic mutualist [6,45••].

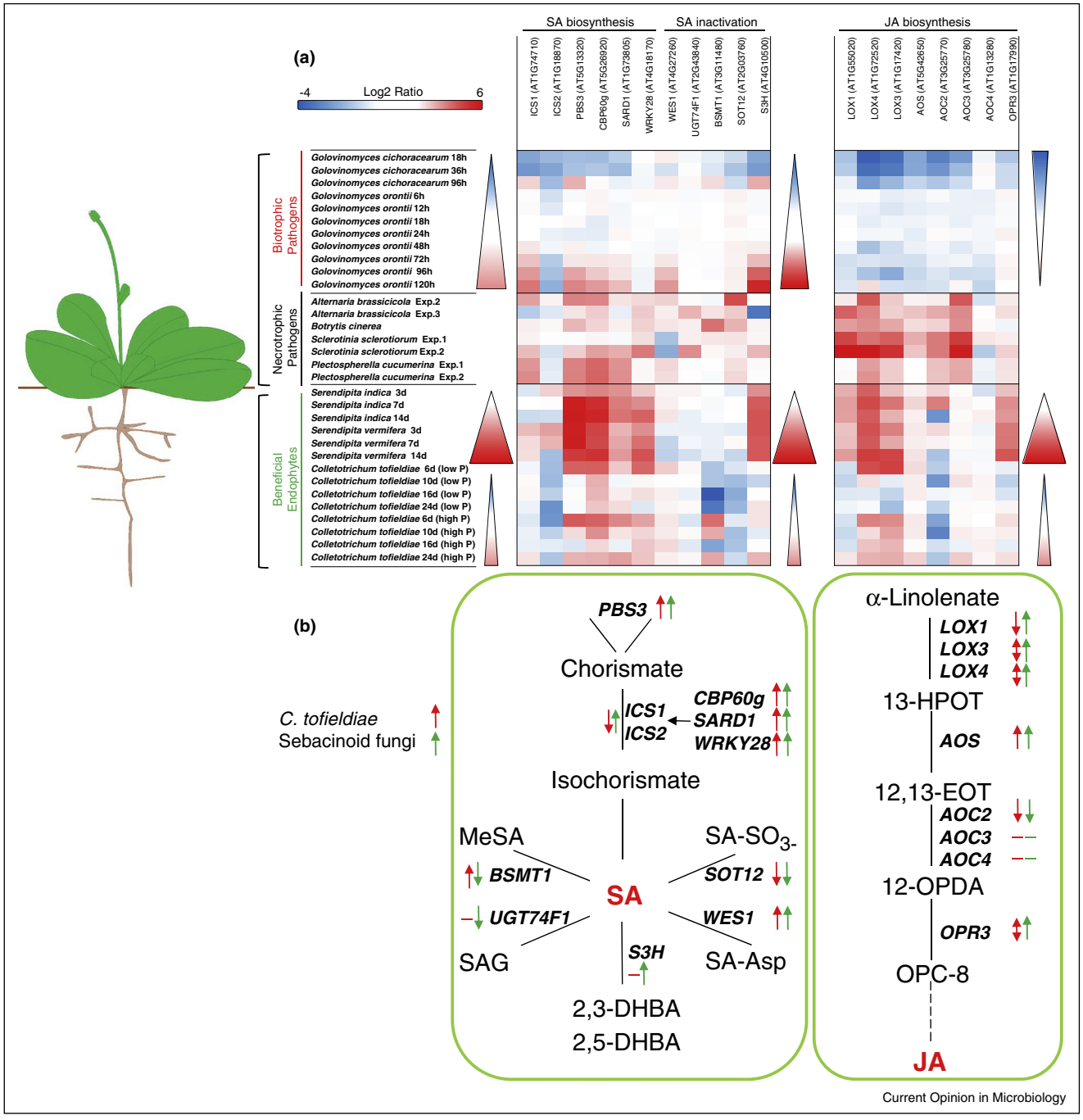
Sebacinoid fungi and *C. tofieldiae* represent attractive models for examining the mechanisms that lead to the evolution of a beneficial endophyte from either root associated fungi with more or less pronounced saprotrophic capabilities or pathogens. Exploiting the genetic repertoire of the model plant *A. thaliana* will pave the way to a deeper understanding of the colonization strategies of fungal root endophytes. The existing knowledge about different pathogenic fungi interacting with *A. thaliana* (Table 1) created a quite complex picture of fungal infection strategies of biotrophic and necrotrophic pathogens and the corresponding response of the plant innate immune system [46,47]. Establishment of fungal endophyte systems which undergo a beneficial association with Arabidopsis roots bear the potential to (i) investigate the pathway that accommodate symbiotic fungi in a non-mycorrhizal plant; (ii) study mechanisms of compatibility in roots in response to pathogens and beneficial endophytes and compare them to the situation in leaves; and (iii) fill the knowledge gap between endophytes, pathogens and mycorrhizal fungi. *S. indica* and *C. tofieldiae* have similar beneficial output *in planta* but their distinct ancestral background suggests a convergent evolution. *A. thaliana* serves as the platform to integrate the gathered knowledge into a larger context. The fact that *A. thaliana* is a non-mycorrhizal plant may be seen as an advantage which helps to uncover alternative fungal and plant strategies that are independent of the common symbiosis genes.

Table 1

Overview of fungi used in interaction-studies with *A. thaliana* clustered by lifestyle

Fungal species	Phyla	Nature of the interaction	Plant organ	Reference
Colletotrichum tofieldiae	Ascomycete	Mutualistic	Systemic	[14••]
Serendipita indica	Basidiomycete	Mutualistic	Root	[31]
Serendipita herbamans	Basidiomycete	Mutualistic	Root	[32]
Serendipita vermifera	Basidiomycete	Mutualistic	Root	[25••]
Trichoderma harzianum	Ascomycete	Mutualistic	Root	[8]
Erysiphe cruciferarum	Ascomycete	Biotrophic	Leaf	[67]
Golovinomyces cichoracearum	Ascomycete	Biotrophic	Leaf	[68]
Golovinomyces orontii	Ascomycete	Biotrophic	Leaf	[69]
Oidium neolycopersici	Ascomycete	Biotrophic	Leaf	[70]
Verticillium longisporum	Ascomycete	Biotrophic	Systemic	[71]
Verticilium dahliae	Ascomycete	Biotrophic	Systemic	[72]
Colletotrichum higginsianum	Ascomycete	Hemibiotrophic	Leaf	[73]
Colletotrichum incanum	Ascomycete	Hemibiotrophic	Systemic	[15••]
Fusarium oxysporum f.sp. lycopersici Fo5176	Ascomycete	Hemibiotrophic	Systemic	[74]
Leptosphaeria maculans	Ascomycete	Hemibiotrophic	Systemic	[75]
Alternaria brassicicola	Ascomycete	Necrotrophic	Leaf	[76]
Botrytis cinerea	Ascomycete	Necrotrophic	Systemic	[77]
Plectosphaerella cucumerina	Ascomycete	Necrotrophic	Systemic	[78]
Rhizoctonia solani	Basidiomycete	Necrotrophic	Systemic	[79]
Sclerotinia sclerotiorum	Ascomycete	Necrotrophic	Leaf	[80]

Figure 2



Expression pattern of a subset of *A. thaliana* genes involved in SA synthesis, SA degradation and JA biosynthesis in response to different plant associated fungi. The lower panel highlights the position of the respective enzymes within the SA and JA biosynthesis pathways. **(a)** Expression data are displayed as log2 fold changes normalized to mock treated *A. thaliana* Col-0 plants and were retrieved from the Genevestigator database (Genevestigator) [59]. *S. indica* (GEO: GSE60736) [25••], *S. vermifera* (GEO: GSE60736) [25••] and *C. tofieldiae* (GEO: GSE70094) [14••,15••] datasets were normalized separately. Except for *C. tofieldiae*, where expression data results are from RNAseq experiments, all other data sets were obtained from Affimetrix Arabidopsis ATH1 Genome Array experiments or from Agilent [25••,48]. The fungi are clustered by their lifestyle. Time course experiments of biotrophic pathogens *Golovinomyces cichoracearum* (18 hours, 36 hours, 96 hours; GEO: GSE26679) [81] and *Golovinomyces orontii* (6 hours, 12 hours, 18 hours, 24 hours, 48 hours, 72 hours, 98 hours, 120 hours; GEO: GSE5686) infecting aerial parts of *A. thaliana*, and necrotrophic pathogens *Alternaria brassicicola* (two experiments; GEO: GSE50526) [82,83], *Botrytis cinerea* (GEO: GSE5684), *Sclerotinia sclerotiorum* (two experiments; Array Express: E-MEXP-3122) [84] and *Plectosphaerella cucumerina* (two experiments; Array Express: E-MTAB-641) [85] are shown. S. *indica* (3 days, 7 days, 14 days), *S. vermifera* (3 days, 7 days, 14 days) and *C. tofieldiae* (6 days, 10 days, 16 days, 24 days; either under high phosphate P and low phosphate conditions) were chosen as root inhabiting beneficial endophytes. The triangles next to the heat

Mutualistic root endophytism in Arabidopsis requires a noncompromised plant innate immunity

Despite differences in taxonomy, distribution, plant host origin and ancestor lifestyle *A. thaliana* endophytic root colonization by Sebacinoid fungi and *C. tofieldiae* share common features. Both types of fungi initially penetrate the epidermis by means of undifferentiated hyphae which then ramify through the root cortex both intercellularly and intracellularly [14••,23,48]. During early penetrations the fungal hyphae are enveloped by the host plasma membrane indicating viability of the host cells (Figure 1a,b). At later stages of the interaction the colonized root cells die, especially in the epidermis. This suggests that epidermal cell death upon fungal penetration in the root is a common mechanism in *A. thaliana*. Importantly, this phenomenon was observed also in other plants such as in barley upon *S. indica* root colonization and also during colonization of rice roots by *H. oryzae* (Figure 1c), suggesting that the root epidermal cells can respond to fungal colonization with a fast cell death phenotype. This cell death which seems to be restricted to the epidermal layer does not lead to browning of the roots and does not hinder the establishment of a long-term beneficial interaction [14••,49]. This root response may not affect concurrent colonization by arbuscular mycorrhizal (AM) fungi as these organisms produce arbuscules in the deeper layers of the root close to the endodermis. Heavy colonization of the dying epidermal layers as observed in root endophytes thus may represent an adaptation to a different root niche, resulting in a competitive advantage compared to AM fungi which depend on living root cells for survival. The question arises how plants balance the colonization by multiple mutualistic fungi that display different/competing colonization strategies. Colonization of *A. thaliana* by Sebacinoid fungi and by *C. tofieldiae* requires a noncompromised plant innate immunity. Using different Arabidopsis indole glucosinolate mutants and measurement of secondary metabolites, the importance of the indole glucosinolate pathway in the growth restriction of *S. indica*, *S. vermifera* and *C. tofieldiae* was demonstrated. These results clearly show that tryptophan-derived secondary metabolites are important key players in the maintenance of a mutualistic interaction with root endophytes and potential determinants of fungal host-range [14••,15••,25••,50]. In conclusion an intact plant immune system is required to restrict the colonization and to keep endophytism balanced.

Salicylic acid (SA) and jasmonic acid (JA) are important plant phytohormones that among others play a crucial role in plant defense mechanisms. Upon *A. thaliana* colonization by the Sebacinoid fungi, genes involved in SA and JA biosynthesis are highly induced. Additionally, the SA degrading enzyme SA 3-hydroxylase (S3H), which converts SA to 2,3-dihydroxybenzoic acid (2,3-DHBA) and 2,5-DHBA [51,52], is consistently induced during all symbiotic stages leading to accumulation of 2,3-DHBA and 2,5-DHBA [25••]. In accordance, JA levels were found to be elevated and SA levels reduced by metabolomic analyses [25••]. The JA and ethylene pathways have been discussed for their role in the responses against wounding and necrotrophic pathogen attack [53–55] and in modulating accommodation of beneficial fungi [56,57]. Traditionally necrotrophic fungi have been shown to be the primary activators of JA-dependent defenses [58]. Conversely in plants colonized by biotrophic fungi, activation of SA-mediated defense and suppression of JA-mediated responses are observed.

Publically available *A. thaliana* transcriptomic data (Genevestigator) of infected plant tissues [59] were compared to those of *S. indica*, *S. vermifera* and *C. tofieldiae* colonized roots (Figure 2). The fungi were grouped according to their lifestyles into biotrophic pathogens (*Golovinomyces cichoracearum* and *G. oronti*), necrotropic pathogens (*Alternaria brassicicola*, *Botrytis cinerea*, *Sclerotinia sclerotiorum* and *Plectospherella cucumerina*) and beneficial root endophytes (*S. indica*, *S. vermifera* and *C. tofieldiae*). Genes involved in the biosynthesis of SA such as the isochorismate synthases 1 and 2 (ICS1 and 2) [60,61] and the positive regulators of SA biosynthesis GH3.12, CBP60g, SARD1 and WRKY28 [62–65] are equally well induced during interaction with necrotrophic as well as biotrophic and endophytic fungi. Interestingly, and in agreement with the conclusion drawn by Hacquard and colleagues, phosphate availability seems to have an impact on the activation of genes encoding SA biosynthetic enzymes upon colonization by *C. tofieldiae* [15••]. This highlights

(Figure 2 Legend Continued) maps summarize the overall trend of gene induction and gene repression during the time course experiments (from top to bottom): for the biotrophic pathogens, for the sebacinoid fungi *S. indica* and *S. vermifera* and *C. tofieldiae*. From left to right the triangles indicate the overall expression trends for genes involved in SA biosynthesis, SA inactivation and JA biosynthesis respectively. **(b)** The lower panel illustrates selected steps of the SA (left box) and JA biosynthesis (right box) pathways of *A. thaliana* with the respective intermediates and the enzymes involved. The arrows and bars next to the enzymes display the way the genes encoding these enzymes are transcriptionally regulated in response to colonization by *C. tofieldiae* (red) and by the sebacinoid fungi *S. indica* and *S. vermifera* (green). Bottom-up arrows display induction of gene expression, top-down arrows display repression of gene expression and horizontal bars display overall unchanged gene expression in response to colonization by the respective fungal endopyhtes. SA derivates are abbreviated as follows: methyl salicylic acid (MeSA), salicylic acid sulfate (SA-SO_3^-), salicylic acid glycoside (SAG), salicylic acid aspartate (SA-Asp) and dihydroxybenzoic acid (2,3-DHBA, 2,5-DHBA). Intermediate products of the JA synthesis pathway are abbreviated as follows: 13-hydroxyperoxyoctadeca-9,11,15-trienoate (HPOT), 12,13-epoxylinolenate (EOT), 12-oxo-10,15-phytodienoate (OPDA) and oxo-2-cyclopentane-1-octanoate (OPC-8). The dotted line between OPC-8 and JA in the box on the right illustrates that intermediate products and enzymatic conversions are left out for simplification reasons.

the importance of SA signaling and the connection between nutrition and immunity in plant–microbe interactions. Induction of genes involved in SA inactivation seems to be a common response to compatible fungal colonization and possibly represents a main strategy of fungi to cope with SA-mediated defense responses. Differences between the SA inactivation pathways used by necrotrophs and biotrophs during compatible interaction are maybe present. In the interaction with necrotrophs, induction of genes involved in glycosylation and methylation of SA are evident, whereas biotrophic pathogens seem to favor degradation to 2,3-DHBA and 2,5-DHBA. *C. tofieldiae* apparently induces both SA methylation and SA conversion to 2,3-DHBA and 2,5-DHBA under sufficient phosphate supply, whereas under low phosphate conditions induction of SA-related genes is only moderate.

The comparative transcriptional analyses substantiate the general model that expression of genes involved in JA synthesis is suppressed during colonization by biotrophic pathogens and induced by necrotrophic fungi. Under sufficient phosphate supply the deregulation pattern for JA biosynthesis induced by *C. tofieldiae* colonization displays some similarity to that of the Sebacinoid fungi. Under low phosphate conditions, where the interaction is beneficial, expression of JA-related genes is low or repressed. For the interaction with *C. tofieldiae* it is still unknown how the fungus affects SA and JA levels and the concentration of their derivatives. It is reasonable to assume that the SA and JA defense activation is required to maintain a balanced interaction as shown for *S. indica* [23].

Conclusions

Under the term endophyte a taxonomically broad collection of fungi with distinct ancestral backgrounds are listed which encompasses commensalistic, latent pathogenic, latent saprotrophic and mutualistic relationships. It is reasonable to assume that beside the taxonomic position, the ancestral background plays a critical role in the genome signatures retained by this heterogeneous group of fungi. It is thus not surprising that evidence for a common toolkit in the analyzed models was not found. The increasing number of genomes will help now to refine sub-categories for fungal endophytes.

The recent data obtained when working with beneficial and pathogenic fungi evince the necessity to address one of the great challenges in the field of plant–microbe interaction which is to develop an integrated molecular concept that explains how plants concomitantly manage pathogenic and beneficial interactions to ensure plant survival and maximize plant fitness. Various conceptual models have been presented to describe the innate immune system of plants that can explain the molecular recognition of pathogenic microorganisms and activation of plant immune responses to limit or terminate pathogen growth [66]. However, these models lead to an apparent paradox as they fall short explaining how plants can discriminate between pathogenic and beneficial microbes to both eliminate foes and accommodate friends. It is accepted that at least part of the innate immune system is necessary for the accommodation of beneficial microbes [14••,25••]. This calls for a conceptual realignment of evolutionary paths and functions of the innate immune system which is especially evident in roots where beneficial and pathogenic interactions often occur alongside one another.

Acknowledgements

We acknowledge the support by CEPLAS (EXC 1028). Dr. Gregor Langen, Prof. Paul Schulze-Lefert and Prof. Jane Parker are gratefully acknowledged for the fruitful discussion. We apologize to all colleagues whose original work could not be cited owing to space constraints.

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- • of special interest
- •• of outstanding interest

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