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Symbiotic Lifestyle Expression by Fungal Endophytes and the Adaptation of Plants to Stress: Unraveling the Complexities of Intimacy

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34.1 INTRODUCTION

The fossil record indicates that fungal symbionts have been associated with plants since the Ordovician period (approximately 400 million years ago), when plants first became established on land (Pirozynski and Malloch, 1975; Redecker et al., 2000; Remy et al., 1994; Simon et al., 1993). Transitioning from aquatic to terrestrial habitats likely presented plants with new stresses, including periods of desiccation. Since symbiotic fungi are known to confer drought tolerance to plants (Bacon, 1993; Read and Camp, 1986), it has been suggested that fungal symbiosis was involved with or responsible for the establishment of land plants (Pirozynski and Malloch, 1975). Symbiosis was first defined by De Bary in 1879, and since that time, all plants in natural ecosystems have been found to be colonized with fungal and bacterial symbionts. It is clear that individual plants represent symbiotic communities with microorganisms associated in or on tissues below- and aboveground.

There are two major classes of fungal symbionts associated with internal plant tissues: fungal endophytes that reside entirely within plants and may be associated with

roots, stems leaves, or flowers; and mycorrhizal fungi that reside only in roots but extend out into the rhizosphere. In addition, fungal endophytes may be divided into two classes: (1) a relatively small number of fastidious species that are limited to a few monocot hosts (Clay and Schardl, 2002), and (2) a large number of tractable species with broad host ranges, including both monocots and eudicots (Stone et al., 2000). While significant resources and research have been invested in mycorrhizae and class 1 endophytes, comparatively little is known about class 2 endophytes, which may represent the largest group of fungal symbionts. This is partially because the symbiotic functionalities of class 2 endophytes have only recently been elucidated and shown to be responsible for the adaptation of some plants to high-stress environments (Redman et al., 1999, 2001, 2002a; Arnold et al., 2003; Dingle and McGee, 2003; Ernst et al., 2003).

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In this chapter, we focus on symbiotic interactions between class 2 endophytes and a variety of monocot and eudicot host species. Specifically, we will discuss the ability of endophytes to express more than one symbiotic lifestyle, fungal taxonomy vs. lifestyle expression, the adaptive nature of symbioses, mechanisms of symbiotically conferred stress tolerance, and the evolutionary implications of adaptive symbiosis. We will refer to class 2 endophytes as fungal endophytes throughout the text.

34.2 SYMBIOSIS: IT IS ALL ABOUT LIFESTYLES

Historically, fungal symbionts were thought to be restricted to specific symbiotic lifestyles (e.g., mutualism, commensalism, or parasitism; Lewis, 1985). However, recent studies indicate that fungi may express different symbiotic lifestyles in response to host genotypes or environmental factors. For example, depending on the physiological status of plants, some mycorrhizal fungi may be mutualistic or parasitic (Francis and Read, 1995; Johnson et al., 1997, Graham and Eissenstat, 1998). Moreover, both pathogenic and nonpathogenic fungi are routinely isolated from asymptomatic plant tissues, suggesting that pathogens either express nonpathogenic lifestyles or infect and remain dormant until plant senescence (Schulz et al., 1999). One of the more interesting aspects of lifestyle expression is that the initial phases of infection and colonization by pathogens, mutualists, and commensals are identical for many fungi. Therefore, lifestyle expression is a postcolonization phenomenon and must involve biochemical and or genetic communication between the symbiont and host.

We began investigating the genetic basis of symbiotic lifestyle expression in the cucurbit pathogen *Colletotrichum magna* (Jenkins, 1963; Winstead et al., 1966). Specifically, we were interested in determining if fungal plant pathogens could express nonpathogenic lifestyles. Mutation studies involving UV light or plasmid integration into the nuclear genome revealed that *C. magna* could be converted from a virulent pathogen to a commensal or mutualist by disrupting single genetic loci (Freeman and Rodriguez, 1993; Redman et al., 1999). The nonpathogenic *C. magna* mutants asymptotically colonized the roots and stems of cucurbit hosts, and mutualism was defined by the ability to confer resistance against a virulent *C. magna* isolate. This was the first demonstration that the symbiotic lifestyle expressed by one fungal isolate could be changed by mutation and that a pathogen could express nonpathogenic lifestyles. It is not known how many pathogenic fungi can be converted to mutualists or commensals by mutation, but this phenomenon appears to be common among *Colletotrichum* species. We have isolated the nuclear DNA responsible for the conversion of *C. magna* (isolate L2.5) to a mutualist (isolate M68) and constructed a gene disruption plasmid pGM68 (Redman et al., in preparation). When transformed into the wild-type *C. magna* isolate L2.5, the plasmid

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integrated by homology (approximately 50%) and generated the same phenotype as expressed by isolate M68. Moreover, when pGM68 was transformed into four other *Colletotrichum* species, the plasmid integrated by homology and generated nonpathogenic mutualistic phenotypes indistinguishable from *C. magna* isolate M68. Although there appear to be few genetic differences between pathogenic and mutualistic lifestyles, lifestyle-altering mutations may be pleiotrophic. Moreover, the ability to express pathogenic lifestyles may require fungi to possess the ability to express nonpathogenic lifestyles. It is not yet known if there is a hierarchy to symbiotic lifestyles or how many pathogenic fungi have the ability to express nonpathogenic lifestyles. Regardless, more extensive mutation and host range/lifestyle expression studies are required to address these issues.

Another observation made with the nonpathogenic *C. magna* mutants was that the host range of a fungus is influenced by symbiotic lifestyle expression (Redman et al., 2001). The nonpathogenic mutants of *C. magna* were able to colonize *Colletotrichum*-resistant cucurbit cultivars that the wild-type isolate L2.5 was incapable of colonizing. This prompted a more extensive host range study of the *C. magna* wild type and nonpathogenic mutants involving several plant families (Redman et al., 2001). We found that the wild-type L2.5 and nonpathogenic mutants were able to asymptotically colonize a variety of plants not previously known to be hosts. Moreover, although the wild-type L2.5 could not colonize as many hosts as the mutant, it could express either parasitic, commensal, or mutualistic lifestyles depending on the host genotype. The nonpathogenic lifestyle expressed by L2.5 was defined as mutualistic based on the ability to confer resistance against virulent pathogens. This was the first demonstration that a fungal pathogen could express a mutualistic lifestyle and confer disease resistance to host plants.

More extensive host range studies with several pathogenic *Colletotrichum* species revealed that fungal pathogens have more flexibility than previously thought in regard to plant host range and symbiotic lifestyle expression (Redman et al., 2001). Our studies indicated that *Colletotrichum* species could be classified into one of three categories:

1. Limited host range capable of only expressing a pathogenic lifestyle
2. Broad host range capable of only expressing a pathogenic lifestyle
3. Broad host range capable of expressing pathogenic, mutualistic, or commensal lifestyles.

It is possible that categories 1 and 2 may be a reflection of the limited number of hosts analyzed, and that all of the *Colletotrichum* species have asymptomatic hosts and are able to express nonpathogenic lifestyles.

Historically, the host range of fungal pathogens encompassed plants that exhibited disease symptoms in response to colonization by a particular fungal species. However, it is clear that pathogenic fungi are able to “switch” lifestyles based on the host genotype (Table 34.1). Symbiotic lifestyle switching of fungal pathogens can occur in genetically divergent species (e.g., cucurbit vs. solanaceous species) or in cultivars of the same species (e.g., tomato), suggesting that relatively subtle host differences can alter the communication responsible for the expression of symbiotic lifestyles (Redman et al., 2001; Table 34.2). This is not surprising considering that single gene modifications in fungal pathogens can result in the expression of mutualistic lifestyles. Regardless, the concept that pathogens express a single lifestyle is no longer valid, and the ability to express nonpathogenic lifestyles may explain the presence of pathogenic fungi in the absence of disease so commonly observed in endophyte studies (Schulz, 1999).

Table 34.1 Lifestyle Switching of Pathogenic *Colletotrichum* Species in Asymptomatic Hosts

Endophyte	Disease Host ^a	Asymptomatic Host ^b	Symbiotic Lifestyle Switch ^c
<i>C. magna</i>	Cucurbits	Tomato	Mutualist
<i>C. coccodes</i>	Tomato	nf	No switch
<i>C. musae</i>	Banana	Pepper	Mutualist
<i>C. orbiculare</i>	Cucurbits	Tomato	Mutualist
<i>C. lindemuthianum</i>	Dry bean	nf	No switch
<i>C. graminicola</i>	Corn	nf	No switch
<i>C. acutatum</i>	Strawberry	Watermelon	Commensal
<i>C. gloeosporioides</i>	Strawberry	Watermelon	Commensal

^a Indicates diseased plants from which the isolates were isolated; some species have several disease hosts.

^b There were several asymptomatic hosts for some species, but only one is listed to indicate lifestyle switching. nf = no asymptomatic hosts found.

^c Mutualists conferred disease resistance to asymptomatic hosts, commensals did not confer disease resistance, and no switch indicates that the isolate was pathogenic on all hosts tested.

Data and methods described in Redman et al., *New Phytologist*, 151, 705–716, 2001.

34.3 LIFESTYLE SWITCHING AND FUNGAL TAXONOMY

Symbiotic lifestyle switching adds a new dimension to fungal taxonomy and fungal ecology. It appears that defining fungi based on lifestyle expression may be a tenuous proposition because of host genotype influence. For example, wild-type *Colletotrichum* species may be classified as pathogens, mutualists, or commensals depending on whether they cause disease symptoms or asymptotically colonize plants and confer disease resistance (Table 34.1). However, when studies are expanded to measure fitness benefits (other than disease resistance) known to be conferred to plants by fungal mutualists, a more complicated pattern of lifestyle expression emerges (Table 34.2). If lifestyle characterization is based on the ability of fungi to confer disease resistance, drought tolerance, or growth enhancement, then *Colletotrichum* species that can switch lifestyles appear to bestow one or more mutualistic benefits. It is too early to tell if all endophytic fungi are capable of switching symbiotic lifestyles and have mutualistic potential. If they do, then it is possible that fungi may have generally evolved as mutualists and that plant disease has become prevalent as a result of transportation of plants around the world. An analysis of the evolution of agriculture on Earth supports this supposition. Between 10,000 and 50,000 years ago, humans began converting from hunting and gathering to crop cultivation. Prior to the era of global conquest and exploration that began more than 2000 years ago, ancient cultures focused on gods thought to be responsible for climate and fertility. During the time of Roman expansion when the spoils of war were returned to what would become Western Europe, a new god, Robigus, the god of crop blight, emerged (Carefoot and Sprott, 1967). Transporting crop plants and seed to new habitats may have introduced the plants to native fungi that were pathogenic on introduced crops, but not on the native plant species. Alternatively, transportation of the new crops and their endophytes into new habitats may have triggered a lifestyle switch. Most likely, both of these scenarios have played themselves out throughout history. Symbiotic lifestyle switching may have started

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Table 34.2 Defining the Symbiotic Lifestyle Expressed by Endophytes Based on Host Fitness Benefits

Endophyte	Tomato, cv Big Beef			Tomato, cv Seattle's Best			Pepper, cv Calif. Wonder		
	Disease Resistance	Drought Tolerance	Growth Enhancement	Disease Resistance	Drought Tolerance	Growth Enhancement	Disease Resistance	Drought Tolerance	Growth Enhancement
<i>C. magna</i>	M	M	M	M	M	M	M	M	C
<i>C. orbiculare</i>	M	M	M	P	nd	nd	NH	nd	nd
<i>C. musae</i>	M	C	C	C	nd	nd	M	M	C
<i>C. gloeosporioides</i>	C	M	M	C	C	C	C	M	M

Note: Abbreviations are as follows: M = mutualist, asymptomatic colonization and confers either disease resistance, drought tolerance, or growth enhancement; C = commensal, asymptomatic colonization with no measurable host fitness benefits; P = pathogenic; nd = not determined; NH = nonhost plant that the fungus was unable to colonize.

Data and methods from Redman et al., *New Phytologist*, 151, 705–716, 2001.

a series of famines that ensued in the following centuries. Although this diatribe is based on conjecture, there are many examples in history indicating that a change in plant–fungal interactions occurred that brought about famines considered to be responsible for restricting population growth in Europe for almost 2000 years (Carefoot and Spratt, 1967). Regardless, until the genetic, biochemical, and ecological bases of lifestyle switching and mutualistic benefits are understood, it will be very difficult to characterize fungi based on lifestyle expression.

34.4 MUTUALISTIC FITNESS BENEFITS VS. ENVIRONMENTAL STRESS

It is well documented that mutualistic fungi collectively may confer several host fitness benefits, such as growth enhancement or tolerance to drought, disease, herbivory, and temperature to plants (Ernst et al., 2003; Redman et al., 2001, 2002a; Clay and Schardl, 2002; Malinowski and Belesky, 2000). But it has been unclear if symbioses are adaptive to habitat-specific stresses and, if so, what the temporal requirements for symbiotic adaptation involve. We compared symbioses in two habitats imposing different stresses to determine if symbioses are adaptive. Fungal endophytes were isolated from crop plants in subtropical and tropical habitats and from native plants in geothermal soils of Yellowstone National Park. Based on perceived habitat stresses, the endophytes were screened for the ability to confer temperature and drought tolerance or disease resistance as described below.

34.4.1 Temperature Tolerance

All plants respond to temperature stress by expressing heat-shock proteins and antioxidant systems, and adjusting osmotic potential and membrane lipids (Iba, 2002). However, few plants thrive in geothermal soils that impose high-temperature stress in the root zone. *Dichanthelium lanuginosum* (panic grass) grows in the geothermal soils that reach temperatures up to 57°C in Yellowstone and Lassen National Parks (Stout et al., 1997; Al-Niemi and Stout, 2002; Stout and Al-Niemi, 2002). These geothermal soils have significant annual temperature fluctuations that are influenced by moisture. Winter snows melt on contact with geothermal soils to decrease temperatures to around 20°C, and a lack of rainfall in summer results in dry, hot soils. Therefore, *D. lanuginosum* is exposed to high root zone temperatures and drought-like conditions on an annual basis.

Two hundred *D. lanuginosum* plants were analyzed and found to be symbiotic with the fungus *Curvularia protuberata* (Redman et al., 2002a). None of the plants analyzed were free of the endophyte, which was isolated from roots, leaves, and seed coats but not seeds. *D. lanuginosum* is the only plant species that occurs in the hot (57°C) geothermal soils and grows as small clusters of individual plants. We surmised that if symbioses were adaptive, then *C. protuberata* may contribute to the thermotolerance and survival of *D. lanuginosum*.

Using a geothermal soil simulator, we observed that the symbiosis between *C. protuberata* and *D. lanuginosum* results in thermotolerance of both symbiont and host plant (Redman et al., 2002a). Nonsymbiotic *D. lanuginosum* has a maximum growth temperature of 40°C, while pure cultures of *C. protuberata* have maximum growth temperatures of 38°C. When these organisms are grown symbiotically, they are able to tolerate daily root temperature regimes of 65°C for 10 h, followed by 37°C for 14 h or sustained 50°C root temperatures (Redman et al., 2002a). After 2 days, symbiotic plants showed no impacts of this temperature regime, while nonsymbiotic plants wilted and died. Five days

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after the nonsymbiotic plants died, symbiotic plants still showed no impacts of the temperature regime and the fungal symbiont was isolated from roots. This indicated that both the host plant and fungal symbiont were protected against heat and that the association was mutualistic. The laboratory results prompted a more complicated test under field conditions. Soil was removed from six locations that ranged in temperature from 35 to 45°C, pastuerized to eliminate resident *C. protuberata* spores or mycelia, and placed back into the geothermal sites (Redman et al., 2002a). Clusters of *C. protuberata* colonized (symbiotic) and uncolonized (nonsymbiotic) *D. lanuginosum* plants were transplanted into the pastuerized soils. Twelve months after transplanting, symbiotic plants had greater biomass than nonsymbiotic plants at all temperatures and the biomass differences increased with temperature. At 45°C soil temperatures, the symbiotic plants survived and the nonsymbiotic plants did not (Redman et al., 2002a). This is the first evidence that fungal endophytes can confer thermotolerance and that symbioses with endophytes may adapt to habitat-specific pressures.

34.4.2 Disease Resistance

Unlike geothermal soil habitats, subtropical and tropical habitats are high in biodiversity. Fungal endophyte surveys indicate that asymptomatic plant tissues are commonly colonized with fungi that are pathogenic in other plant species (Schulz et al., 1999). It is not known if these pathogens are latent or have switched symbiotic lifestyles, but their presence suggests that there is high disease pressure on plant communities in these habitats. One group of pathogenic fungi commonly isolated from asymptomatic plant tissues are *Colletotrichum* species (Cullen et al., 2002; Manaut et al., 2002; Freeman et al., 2001; Pinto et al., 2000). We have demonstrated that several *Colletotrichum* species have the ability to express nonpathogenic lifestyles, including mutualism, depending on the host they colonize (described above). One of the attributes used to classify *Colletotrichum* species as mutualists was the ability to confer disease resistance to asymptomatic hosts (Redman et al., 2001; Table 34.1). In general, disease resistance involved 100% protection against a variety of virulent pathogens. For example, *C. magna* was able to protect plants against pathogenic isolates of *Colletotrichum*, *Fusarium*, and *Phytophthora* (Freeman and Rodriguez, 1993; Redman et al., 2001).

The mechanism of symbiotically conferred disease resistance appears to involve the rapid activation of host defense systems (Redman et al., 1999; ~~Rodriguez et al., 2003~~). The production of lignin and expression of the enzymes peroxidase and phenylalanine ammonia lyase (PAL) correlate with disease resistance in cucurbit species (Ryals et al., 1996). Therefore, these biochemical activities were monitored before and after symbiotic (colonized with the mutualistic *C. magna* mutant Path-1) and nonsymbiotic watermelon plants were exposed to virulent fungal pathogens. After pathogen challenge, lignin biosynthesis, peroxidase, and PAL activities slowly increased over 4 days in nonsymbiotic plants, and by the fifth day the plants were dead. In symbiotic plants, these biochemical activities increased dramatically within 24 h of pathogen challenge, thereby thwarting ingress of the pathogen. The response in symbiotic plants was so strong that the pathogen could not be reisolated from inoculated tissues (Redman et al., 1999). This type of disease protection has been defined as endophyte-associated resistance (EAR) and is localized to tissues colonized by endophytes (Redman et al., 1999).

34.4.3 Drought Tolerance

All plants respond to water deficit through a complex series of biochemical and genetic modifications, including osmotic adjustments, production of antioxidants, altered transcriptional and translational regulation, and altered stomatal activity (Griffiths and Parry,

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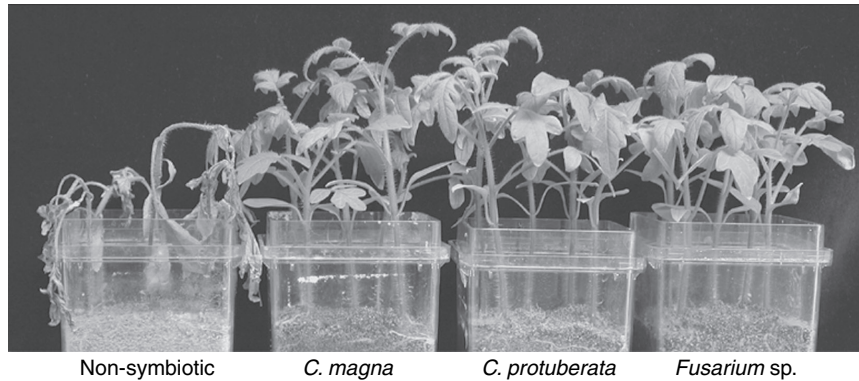


Figure 34.1 Nonsymbiotic and symbiotic plants colonized with the indicated endophytes were grown in sand for 2 weeks with adequate watering. Watering was then stopped and plants left to dry. Nonsymbiotic plants wilted after 4 days of desiccation, while the symbiotic plants stayed hydrated for 9 days before wilting.

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2002; Shinozaki and Yamaguchi-Shinozaki, 1998). Yet, few plant species are drought tolerant and survive in habitats with low moisture (Bray, 1993). It is well documented that fungal endophytes confer some level of drought tolerance to plants (Clay and Schardl, 2002). Although the mechanism of endophyte-conferred drought tolerance is unknown, it is thought to involve adjustments in host osmolyte concentrations or stomatal activity (Malinowski and Belesky, 2000). However, endophyte-conferred drought tolerance has been studied in very few plant species. Interestingly, all of the endophytes we have studied, including pathogenic *Colletotrichum* species that can switch lifestyles, confer some level of drought tolerance to plant hosts (Redman et al., 2001; Table 34.2). For example, *Colletotrichum magna* (expressing a nonpathogenic lifestyle) and *C. protuberata* 4666D confer significant drought tolerance to wheat, tomato (Figure 34.1), and watermelon plants. Therefore, drought tolerance appears to be common among fungal endophytes and the communication involved in this mutualistic benefit is conserved between monocots and dicots, which diverged approximately 200 million years ago. This may reflect the fact that when plants moved onto land ca. 400 million years ago, water relations was one of the most difficult stresses to overcome, and fungal symbioses may have developed as a result of symbiotically conferred drought tolerance (Pirozynski and Malloch, 1975).

34.5 HOW ADAPTIVE ARE SYMBIOSES?

We compared the abilities of fungal endophytes from plants thriving in geothermal soils, coastal beaches, or subtropical/tropical habitats to confer fitness benefits to host plants. Endophytes were screened for the ability to confer temperature and drought tolerance, and disease resistance to the plants they were isolated from or on tomato, a host they all asymptotically colonize (Table 34.3). The endophytes conferred different fitness benefits based on the habitat of isolation. For example, the endophytes from tropical/subtropical habitats, where disease pressure is high, conferred disease resistance, while the endophytes from geothermal soils and coastal beaches did not. Moreover, endophytes from geothermal soils conferred temperature tolerance, while endophytes from the other habitats did not. Endophytes from all three habitats conferred drought tolerance. These

Table 34.3 Mutualistic Benefits vs. Habitat Stress

Endophyte	Habitat	Habitat Stress	Stress Tolerance Conferred by Endophytes ^a		
			Temperature	Disease	Drought
<i>Colletotrichum</i> spp.	Tropical/subtropical agriculture	Disease	0/3	3/3	3/3
<i>Curvularia</i> spp.	Geothermal soils	Heat, desiccation	2/2	0/2	2/2
<i>Fusarium</i> and <i>Alternaria</i> spp.	Coastal beaches	Desiccation	0/3	0/3	3/3

^a The number on the right of the diagonal indicates the number of endophyte species tested from each habitat, and the number on the left is the number of endophytes conferring the respective stress tolerances.

Methods for these analyses are from Redman et al., *New Phytologist*, 151, 705–716, 2001; Redman et al., *Science*, 298, 1581, 2002a. Data for *Fusarium* and *Alternaria* are unpublished.

results suggest that plant–fungal symbioses adapt to stresses in a habitat-specific manner, a phenomenon we describe as adaptive symbiosis. It is clear that adaptive symbiosis involves communication between endophyte and host that not only regulates symbiotic lifestyle expression, but also regulates symbiotically conferred fitness benefits. This is not surprising considering the complex communication involved in plant symbioses with fungal pathogens, agrobacterium, rhizobium, and mycorrhizae. The temporal requirements for adaptive symbiosis and the communication required for this phenomenon are not known. However, the ramifications of adaptive symbiosis are that specific stress tolerances may be conferred to either monocot or eudicot hosts regardless of the endophyte's origin.

34.6 MECHANISMS OF STRESS TOLERANCE

Symbiotically conferred stress tolerance involves at least two mechanisms: (1) activation of host stress response systems soon after exposure to stress, allowing the plants to avoid or mitigate the impacts of the stress (Arnold et al., 2003; Pirttila et al., 2002; Redman et al., 1999; Schulz et al., 1999); and (2) biosynthesis of antistress biochemicals by endophytes (Bacon and Hill, 1996; Siegel and Bush, 1997; Strobel et al., 2001; Miller et al., 2002; Schulz et al., 2002). We have proposed that in addition to antistress chemicals, plant–fungal mutualisms have been maintained over evolutionary time because endophytes control the activation of host stress response systems by acting as biological triggers (Rodriguez et al., 2003).

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34.7 ECOLOGICAL AND EVOLUTIONARY RAMIFICATIONS

There are four major points presented in the discussion above: (1) individual endophytes can switch symbiotic lifestyles and the outcome of symbiosis is influenced by host genotypes, (2) mutualistic benefits conferred by endophytes are also influenced by host genotypes, (3) the host range of endophytes is poorly defined and may include both monocot and eudicot species, and (4) symbioses adapt to habitat-specific stresses, a phenomenon we describe as adaptive symbiosis. It appears that at least some endophytes have evolved with a high degree of flexibility to move between genetically distant plant species and communicate in a manner that allows both organisms to survive environmental conditions they cannot survive on their own. This provides endophytes an opportunity to expand habitat range by dispersion of endophyte-colonized plant tissues (rhizomes, seed coats, seed). For example, *C. protuberata* colonizes the seed coats of *D. lanuginosum* found in geothermal soils of Yellowstone National Park. *D. lanuginosum* seeds are borne on panicles that detach from plants and can be carried by the wind over large distances, allowing both the host and symbiont to spread.

By establishing mutualistic symbioses with endophytes, plants may gain new functionalities allowing them to mitigate the impacts of environmental stresses. This could provide a mechanism for plants to make quantum evolutionary changes allowing for habitat expansion and survival in high-stress habitats. However, the dynamics of this process are probably much more complicated than simply forming new mutualisms and must involve the adaptation, physiology, and ecology of fungal endophytes and plant hosts. Ultimately, it is the symbiotic communication between endophyte and host that dictates the outcome of each association and the ability to survive in high-stress habitats.

34.8 SUMMARY

Although endophytic fungi are responsible for the survival of at least some plants in high-stress habitats, more information is required before the full impacts of lifestyle switching and adaptive symbiosis are understood. We do not know yet the limits of adaptive symbiosis with regard to mutualistic benefits that develop in response to habitat-specific stresses. In addition, the temporal requirements for adaptive symbiosis are not known, nor is the fungal ecology associated with endophyte distribution patterns within and between hosts. Although the interspecies communication involved in mutualistic benefits appears to be conserved between monocots and dicots, the biochemical and genetic bases of this communication are not known. Regardless of these knowledge deficiencies, endophytic symbioses offer novel strategies for mitigating the impacts of global change on native plant communities and agricultural crop production.

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