Mycoscience (2010) 51:44–52
DOI 10.1007/s10267-009-0009-z

FULL PAPER

Mycorrhizal and dark septate fungal associations in shola species of Western Ghats, southern India

Govindaraj Bagyalakshmi · Thangavelu Muthukumar · Kullaiyan Sathiyadash · Vellaisamy Muniappan

Received: 9 December 2008 / Accepted: 22 July 2009 / Published online: 25 December 2009

Abstract We analyzed mycorrhizal types and dark septate endophyte (DSE) fungal associations in a shola vegetation of Western Ghats region, southern India. Plants belonging to 29 species of 19 families were assessed for mycorrhizal type and DSE fungal association. Five mycorrhizal classes were classified based on morphological traits: arbuscular, ecto-, ectendo-, ericoid-, and orchid mycorrhizas. Arbuscular mycorrhizal (AM) association was the most predominant mycorrhizal type, occurring in 16 plant species, followed by orchid (3 species), ericoid- (2 species), and ecto- and ectendomycorrhizas (1 species each). Mycorrhizal association is reported for the first time in 19 plant species. DSE fungal association was found in six plant species. *Arum*- and *Paris*-type AM morphology was found, respectively, in 10 and 5 plant species, with intermediate type recorded in one species. In this study, some new records on the morphological types of AM in some plant families were obtained. Further occurrence of ectendomycorrhizas in *Pinus oocarpa* and dark septate fungal association in *Eleaocarpus munronii*, *Symplocos cochinchiensis*, *Daphniphyllum neilgherrense*, *Euodia roxburghiana*, *Syzygium arnottianum*, and *Syzygium montanum* are reported for the first time. Roots of *Berberis tinctoria*, *Mahonia leschenaultii* (Berberidaceae), *Elaeagnus latifolia* (Elaeagnaceae), and *Elaeocarpus oblongus* (Elaeocarpaceae) lacked any fungal structures.

Keywords AM fungi · DSE fungal endophyte · Ectendomycorrhizas · Ericales · Shola forest

Introduction

Different types of nonpathogenic root fungal associations are encountered in most ecosystems. These root fungal associations play a critical ecological role in plant distribution and abundance in natural ecosystems. Of these, mycorrhizas form a critical link between the plant and the soil by influencing nutrient cycling and soil structure (Korb et al. 2003). Furthermore, the benefits of mycorrhizal associations to plants include enhanced nutrient and water uptake, protection against pathogens, improved resistance to drought, higher tolerance to heavy metals, and increased root surface area (Smith and Read 1997). In species-rich natural communities, colonization of individual plants by mycorrhizal fungi occurs within a few days of emergence of radicle (Read and Birch 1988), and can be important for establishment and survival of compatible plant species (Francis and Read 1994). Because of their effects on individual plant performance, mycorrhizas influence the productivity of plant communities and can affect plant community composition, succession, and species diversity (Janos 1996).

Like mycorrhizal fungi, another group of root fungal symbionts, dark septate endophytic (DSE) fungi, has been characterized as producing a range of effects on their host (Jumpponen 2001). Plants in any ecosystem possess different types of root fungal associations (Thormann et al. 1999; Fontenla et al. 2001). Väre et al. (1992) reported ecto- and ericoid mycorrhizal associations in plant species of Arctic Spitsbergen. Similarly arbuscular mycorrhizal (AM) and DSE fungal associations have been reported in plant communities of alpine, Arctic, Antarctic (Treu et al.

Electronic supplementary material The online version of this article (doi:10.1007/s10267-009-0009-z) contains supplementary material, which is available to authorized users.

G. Bagyalakshmi · T. Muthukumar (✉) · K. Sathiyadash · V. Muniappan
Root and Soil Biology Laboratory, Department of Botany, Bharathiar University, Coimbatore 641 046, Tamil Nadu, India
e-mail: tmkum@yahoo.com

1996; Laursen et al. 1997; Routsalainen et al. 2002), wet lands (Weishampel and Bedford 2006; Kai and Zhiwei 2006), grasslands (Muthukumar and Udaiyan 2002), and epiphytic and terrestrial plant species of Costa Rican cloud forest (Rains et al. 2003). Six mycorrhizal types have been reported from plant communities in coastal nature reserves of the Mediterranean basin (Maremmani et al. 2003). Likewise, five mycorrhizal types were reported from a primary successional volcanic ecosystem on Mount Koma, northern Japan (Tsuyuzaki et al. 2005).

Recently Wang and Qiu (2006) updated information on the occurrence of different types of mycorrhizas in families of land plants (3617 species of 263 families). These authors emphasized that the prime area in mycorrhizal research that deserves more attention is the investigation of more plant species for their mycorrhizal status. Furthermore, Smith and Smith (1997) and Dickson et al. (2007) also suggested the importance of surveying the distribution of different types of mycorrhizal plants in natural ecosystems worldwide and recording their mycorrhizal classes.

Shola is a high-altitude stunted patch of evergreen forests separated by undulating grasslands found above 1800 m altitude in southern India. Shola forests are restricted to the southern part of the Western Ghats and are habitat to some of the most threatened and endemic plant species. Some of the species found here have close relatives only in the distant evergreen forests of northeast India or those in Southeast Asia. The trees found in sholas are evergreen, usually short-boled, less than 12 m in height, with dense round crowns, coriaceous leaves, and branches covered with epiphytes, mosses, and lichens. Woody climbers are also frequent. In the margins of these forests light-demanding trees and shrubs are common but do not penetrate the shola proper (Meher-Homji 1967). Despite the high diversity and unique flora of shola ecosystems, no systematic study has been carried out to analyze root fungal associations in plants of shola forests. In addition there has been a loss of 50% of shola forest cover since 1850 (Somasundaram and Vijayan 2005). In this work, we identified and quantified the presence of mycorrhizal and DSE fungal associations in common plant species in shola forest of Nilgiris.

Materials and methods

Study site

The study site is a shola (~472 ha) located in T. R. Bazar, The Nilgiris (11°28′N and 76°63′E, 2000 m a.s.l), Tamil Nadu, India. The average temperature ranges between 10°C and 25°C in summer and 5°C and 21°C in winter. The vegetation is wet temperate forest type (Nair and Henry 1983) with annual rainfall ranging between 1520 and 1780 mm. The soil is clayey loam with pH (H_2O) of 6.2 (water, 1:1, soil/water), 2.3 mg kg^{-1} of total nitrogen (N), 0.9 mg kg^{-1} of total phosphorus (P), and 2.8 mg kg^{-1} of exchangeable potassium (K), as assessed according to Jackson (1971).

Sample collection

Sampling occurred between August and December 2005. Plant roots and soil samples were collected from five individuals of each plant species. Care was taken during collection of individual plants that roots could be positively identified as belonging to a particular plant. For this, samples of trees or shrubs were usually taken from saplings if available, or the roots were traced back to the stem. For individual trees, roots were collected from five or six regions around the tree and bulked. Roots were gently washed and fixed in formalin–acetic acid–alcohol (FAA) and transported to the laboratory for processing. Plant nomenclature and authorities for angiosperms follow Nair and Henry (1983) and Henry et al. (1989).

Assessment of root fungal associations

Macroscopic characterization of mycorrhizal status was carried out on the basis of morphological features of the fine roots. The roots were washed thoroughly in running tap water and observed under a Leica binocular dissection microscope. Roots showing extraradical colonization were microscopically characterized after free-hand sectioning. The presence of fungal mantle and Hartig net was used to characterize ectomycorrhizal status, whereas the presence of fungal mantle and Hartig net along with intracellular hyphae was used to characterize ectendomycorrhizas. All root samples were cleared and screened for presence or absence of the different fungal associations. The observations on mycorrhizal and DSE fungal associations were checked against previous publications reporting mycorrhizal and DSE fungal status for each plant genus examined and are presented in the Appendix in the Electronic Supplementary Material.

Preparation of roots and mycorrhizal assessment

Fixed roots were washed free of FAA, cut into 1-cm fragments, cleared in 2.5% KOH (Koske and Gemma 1989), acidified with 5 N HCl, and stained with trypan blue (0.5% in lactoglycerol) overnight. Roots that remained dark after clearing were bleached in alkaline H_2O_2 prior to acidification. Fifty 1-cm-long root pieces were examined for each plant with a compound microscope (×200–400) for AM fungal structures, and the percentage of root length

colonization was estimated according to a magnified intersection method (McGonigle et al. 1990).

Sectioning for ecto-, ectendo-, and ericoid mycorrhizal assessment, and data presentation

Free-hand transverse sections of root samples were made and stained with trypan blue. Stained sections were mounted and observed for presence of mycorrhizal structures (mantle, Hartig net, hyphal coils). For assessing root length with ecto-, ectendo-, ericoid, and orchid mycorrhizas the magnified intersection method used for estimating AM fungal colonization was adopted. Data for mycorrhizal variables are presented as mean ± standard error (SE).

Results

Mycorrhizal and dark septate fungal associations

In the shola, 86% of plant species surveyed had root fungal associations (Table 1). On the basis of macro- and microscopic characteristics, six morphotypes of root fungal associations were classified. Of the 29 plant species (belonging to 19 families) examined, AM were present in 16 (55%), orchid mycorrhizas in 3 (10%), ericoid mycorrhizas in 2 (7%), and DSE fungi in 6 (21%). Ecto- and ectendomycorrhizas occurred in only one (3%) species each. All the individuals of a species had the same or combinations of the same root fungal association. Three species [*Daphniphyllum neilgherrense* (Daphniphyllaceae), *Syzygium montanum* (Myrtaceae), and *Euodia roxburghiana* (Rutaceae)] showed both AM and DSE fungal association. *Eucalyptus grandis* (Myrtaceae) had both ectomycorrhizas and AM in different portions of the root system. Fungal structures were absent in roots of *Berberis tinctoria*, *Mahonia leschenaultii* (Berberidaceae), *Elaeagnus latifolia* (Elaeagnaceae), and *Elaeocarpus oblongus* (Elaeocarpaceae).

Ectendomycorrhizas

Ectendomycorrhizal association was found in *Pinus oocarpa* (Fig. 1a–c). The mycorrhizal tips were 3–5 mm in length, possessing sparse to extensive amounts of narrow (1.6–2 μm) darkly pigmented smooth to finely verrucose and regularly septate extramatrical hyphae. The root sections showed dense, continuous mantles (5–50 μm in thickness) and narrow well-developed uniseriate Hartig net. The Hartig net extended to the first layer of epidermal cells and one to three cell layers further into the cortex. Intracellular broad thick-walled hyphal coils characteristic of ectendomycorrhizas were present in the cortical cells. Root length colonization in *P. oocarpa* was 49% (Table 1).

Ectomycorrhizas

Ectomycorrhizal association in *E. grandis* was characterized by the presence of a fungus mantle covering the root and a Hartig net. The Hartig net was limited mainly to the outer cortical region. The fungal hyphae never penetrated the endodermis. Root length colonization in *E. grandis* was 73% (Table 1).

Ericoid mycorrhizas

The mycorrhizal morphology of *Gaultheria fragrantissima* and *Rhododendron arboreum* (Ericaceae) was typical of ericoid species. Some roots exhibited a compact fungal "mantle," whereas others showed a loose arrangement of branching hyphae on the root surface (Fig. 1d, e). Some roots examined, however, showed no obvious external colonization. In all segments examined internal colonization was restricted to epidermal cells. The thickness of the epidermal wall and the shape of epidermal cells varied among hosts and with location along the root axis. Intracellular hyphal complexes were variable in shape and complexity, sometimes revealing fungal entry points. Darkly pigmented, thick-walled hyphae and microsclerotia having a typical appearance of structures formed by dark septate endophytes were frequently observed in cleared hair roots. Two main patterns of epidermal cell colonization were observed: loosely arranged intracellular hyphal coils as in *G. fragrantissima*, and compact intracellular hyphal coils occupying most of the host cell volume as in *R. arboreum*. Hyphal connections between contiguous epidermal cells were frequently observed in roots with loosely arranged intracellular coils. The extent of colonization was 88% in *G. fragrantissima* and 84% in *R. arboreum* (Table 1).

Arbuscular mycorrhizas

Ten species in the present study had typical *Arum*-type and 5 had *Paris*-type AM morphology (Table 1, Fig. 1f–h). Intercellular hyphae, vesicles, and intracellular arbuscules characterized *Arum*-type mycorrhizas (Fig. 1f, h). Hyphal coils, if present, were restricted to few outer cells of root at the point of fungal entry. Root colonization ranged between 25% (*Syzygium montanum*, Myrtaceae) and 89% (*Cinnamomum camphora*, Lauraceae). Intracellular hyphal coil, vesicles, and arbusculate coils characterized *Paris*-type AM morphology (Fig. 1g). *Paris*-type root colonization ranged between 40% (*Eurya japonica*, Ternstroemiaceae) and 98% (*D. neilgherrense*). Intermediate type of AM morphology in *Turpinia nepalensis* (Turneraceae) was characterized by inter- and intracellular hyphae and intracellular arbuscules.

Table 1 Extent of mycorrhizal and nonmycorrhizal fungal colonization in roots of shola species (mean ± SE)

Family	Habit	Mycorrhizal type[a]	Mycorrhizal/DSF colonization[b]				
Plant species			%RLH	%RLA	%RLV	%RLHC/%RLMS	%RLT
Adoxaceae							
Viburnum erubescens Wall.[c]	Tree	AM-*Paris*	36.38 ± 3.19			52.26 ± 2.73	88.64 ± 1.26
Asteraceae							
Eupatorium glandulosum Kunth.[c]	Herb	AM-*Paris*	29.81 ± 5.13		5.71 ± 1.05	35.84 ± 6.79	71.35 ± 1.74
Balsaminaceae							
Impatiens leschenaultii (DC.) Wall. ex. Wight & Arn.[cd]	Shrub	AM-*Arum*	57.22 ± 6.83	12.90 ± 5.13	12.43 ± 1.35		82.56 ± 11.55
Berberidaceae							
Berberis tinctoria Lesch.	Shrub	NF					
Mahonia leschenaultii (Wall. ex. Wight & Arn.) Takeda.[d]	Shrub	NF					
Daphniphyllaceae							
Daphniphyllum neilgherrense (Wight) K. Rosenth.[c]	Tree	AM-*Paris*	11.24 ± 0.91	15.98 ± 1.64	14.93 ± 0.95	56.10 ± 0.94	98.23 ± 1.75
		DSF	6.12 ± 0.31			3.58 ± 0.83	9.70 ± 0.38
Elaeagnaceae							
Elaeagnus latifolia L.	Tree	NF					
Elaeocarpaceae							
Elaeocarpus munronii (Wight) Mast.[d]	Tree	DSF	48.03 ± 6.75			30.09 ± 3.16	78.12 ± 3.66
Elaeocarpus oblongus Gaertn.	Tree	NF					
Ericaceae							
Gaultheria fragrantissima Wall.[c]	Shrub	Ericoid	7.11 ± 1.52			80.98 ± 3.18	88.07 ± 5.18
Rhododendron arboreum J.E.Smith. ssp. *nilagiricum* (Zenker) Tagg.[cd]	Tree	Ericoid	3.33 ± 0.40			80.39 ± 4.40	83.72 ± 5.40
Fabaceae							
Acacia melanoxylon R.Br.[c]	Tree	AM- *Arum*	50.10 ± 1.34	3.20 ± 0.15	12.97 ± 4.77		66.28 ± 4.87
Lauraceae							
Cinnamomum camphora (L.) Presl.[c]	Tree	AM-*Arum*	59.67 ± 4.16	8.57 ± 2.66	21.22 ± 2.38		89.46 ± 3.93
Neolitsea zeylanica Merr.[c]	Tree	AM-*Arum*	12.14 ± 2.31	9.26 ± 1.24	14.28 ± 3.56		35.67 ± 4.69
Myrtaceae							
Eucalyptus grandis Hill ex Maiden.	Tree	Ecto					73.26 ± 3.52
		AM-*Arum*	40.6 ± 7.62	2.10 ± 0.98	13.82 ± 1.07		57.55 ± 8.06

Table 1 continued

Family / Plant species	Habit	Mycorrhizal type[a]	Mycorrhizal/DSF colonization[b]				
			%RLH	%RLA	%RLV	%RLHC/%RLMS	%RLT
Syzygium arnottianum Walp.[c]	Tree	DSF	51.17 ± 2.89				51.17 ± 2.89
Syzygium montanum Gamb.[c]	Tree	AM-*Arum*	12.86 ± 1.34	7.92 ± 1.06	4.31 ± 1.01		25.09 ± 2.03
		DSF	12.38 ± 5.26			1.32 ± 0.15	13.71 ± 1.03
Orchidaceae							
Oberonia platycaulon Wight[c]	Herb	Orchid				83.70 ± 3.87	83.70 ± 3.87
Oberonia ensiformis (J. E. Smith) Lindl.[c]	Herb	Orchid				65.25 ± 6.01	65.25 ± 6.01
Malaxis versicolor L.[c]	Herb	Orchid				90.01 ± 8.97	90.01 ± 8.97
Pinaceae							
Pinus oocarpa Schiede ex Schltdl.	Tree	Ectendo	48.67 ± 10.48				48.67 ± 10.48
Rhamnaceae							
Rhamnus wightii Wight & Arn.[c]	Shrub	AM-*Paris*	32.13 ± 5.14	6.08 ± 2.03		38.07 ± 2.29	76.28 ± 6.96
Rosaceae							
Rubus racemosus Roxb.[cd]	Shrub	AM-*Arum*	29.07 ± 3.42	12.36 ± 3.12	22.17 ± 2.36		63.60 ± 5.77
Rubus moluccanus Hk.f.[c]	Shrub	AM-*Arum*	13.25 ± 1.36	15.92 ± 2.05	7.36 ± 1.21		36.54 ± 3.28
Rubus ellipticus Smith.[c]	Shrub	AM-*Arum*	33.63 ± 6.64	29.81 ± 4.16	15.51 ± 4.07		78.96 ± 10.66
Rutaceae							
Euodia roxburghiana Benth.[c]	Tree	AM-*Arum*	47.26 ± 4.04	21.38 ± 3.96	15.33 ± 2.45		83.98 ± 3.63
		DSF	15.38 ± 1.26			4.31 ± 0.65	19.69 ± 1.56
Symplocaceae							
Symplocos cochinchinensis (Lour.) Moore	Tree	DSF	94.67 ± 1.76				94.67 ± 1.76
Ternstroemiaceae							
Eurya japonica Thumb.	Shrub	AM-*Paris*	7.29 ± 3.54	3.81 ± 1.21		28.65 ± 3.79	39.75 ± 4.23
Turneraceae							
Turpinia nepalensis Wall.[c]	Tree	AM-Intermediate	43.86 ± 3.21	12.52 ± 2.68	19.09 ± 0.93		75.47 ± 2.84

[a] *AM* arbuscular mycorrhizas, *DSF* dark septate fungi, *Ecto* ectomycorrhizas, *Ectendo* ectendomycorrhizas, *Orchid* orchid mycorrhizas, *NF* no fungal association

[b] *%RLH*, *%RLA*, *%RLV*, *%RLHC/%RLMS*, and *%RLT*, root length with hyphae, arbuscules, vesicles, hyphal coils/microsclerotia, pelotons, and total colonization, respectively

[c] First report on mycorrhizal occurrence

[d] Endemic taxa

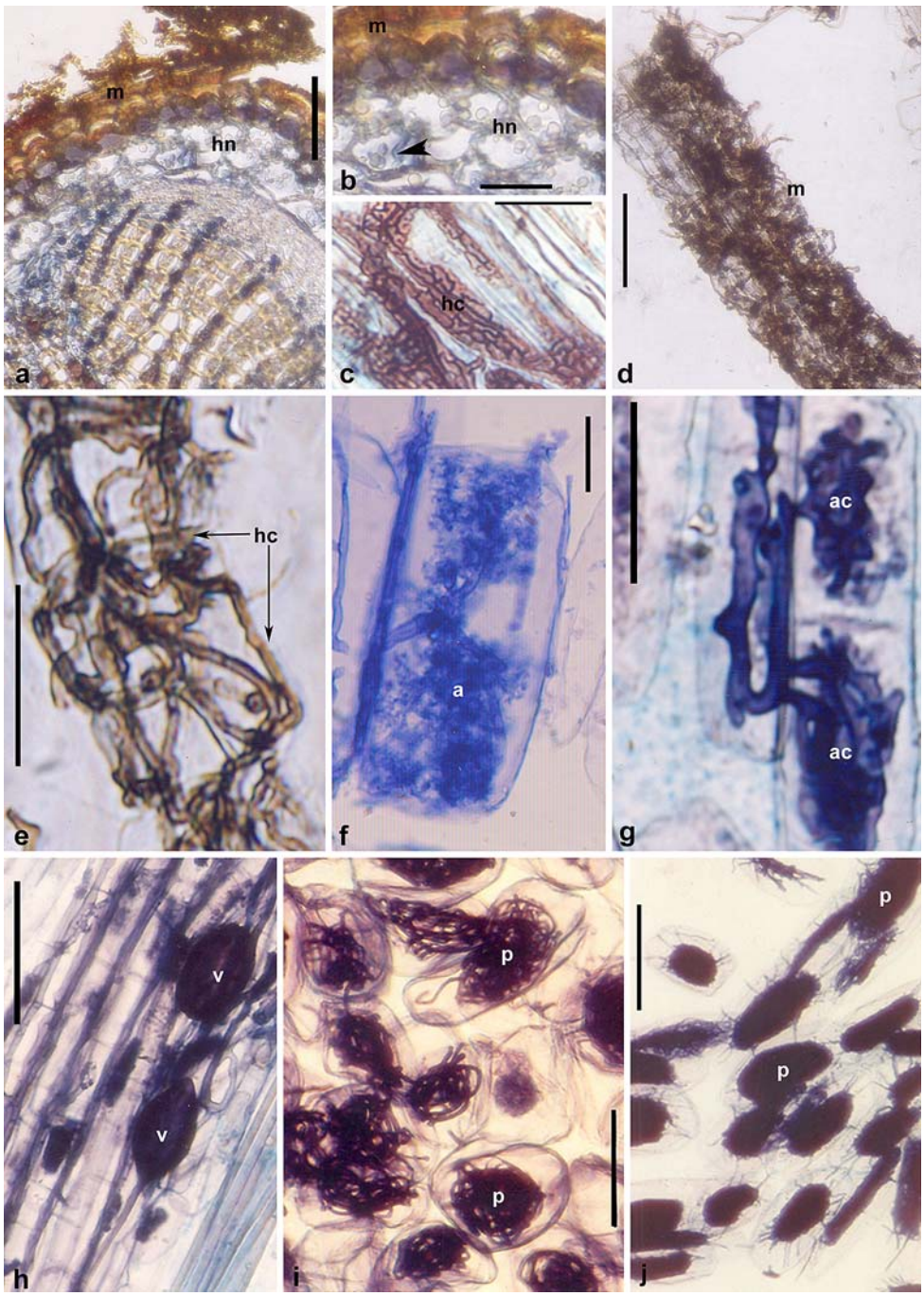

Fig. 1 **a–i** Photomicrographs of mycorrhizal types in roots of shola plant species. **a** Transverse section of *Pinus oocarpa* root showing mantle (*m*) and Hartig net (*hn*); **b** intracellular hyphae (*arrow head*), mantle (*m*), and Hartig net (*hn*) in *P. oocarpa* root; **c** hyphal coils (*hc*) within cortical cells of *P. oocarpa*; **d** root of *Rhododendron arboreum* with surface mantle (*m*); **e** hyphal coils (*hc*) within root cortical cells of *R. arboreum*; **f** *Arum*-type arbuscule (*a*) in *Rubus racemosus*; **g** arbusculate coils (*ac*) in *Viburnum erubescens*; **h** intercellular vesicles (*v*) in *R. racemosus*; pelotons (*p*) in *Oberonia ensiformis* (**i**) and *Malaxis versicolor* (**j**). Scale bars **a**, **c–e**, **g–j** 50 μm; **b**, **f** 25 μm

Orchid mycorrhizas

The three species of orchids examined in the present study were mycorrhizal, although the extent of colonization varied among species (Table 1). Average colonization in epiphytic *Oberonia platycaulon* and *Oberonia ensiformis* was 74%, and that of terrestrial *Malaxis versicolor* was 90%. In epiphytic species, colonization was restricted to those roots that were in contact with the bark. The mycorrhizal morphology was characterized by intact pelotons in cortical cells of young roots and degenerating pelotons in cells of older roots (Fig. 1i, j).

DSE fungal colonization

DSE fungal colonization was observed in *Eleaocarpus munroii* (Elaeocarpaceae), *Symplocos cochinchinensis* (Symplocaceae), and *Syzygium arnottianum* (Myrtaceae), and along with AM in *D. neilgherrense*, *S. montanum* (Myrtaceae), and *E. roxburghiana*. The percentage of root

colonization ranged between 10% in *D. neilgherrense* and 95% in *S. cochincinensis* (Table 1).

DSE fungal association was characterized by narrow runner hyphae, 2–4 μm wide, commonly occurring on the root surface and typically running parallel to the root's long axis. Sometimes a loose hyphal network occurred on the root surface. The runner hyphae were infrequently branched at a 90° angle to the main hyphae, with occasional swollen tips. Hyphae often followed the sulcule between the epidermal cells with terminal and intercalary hyphal swelling oppressed against the epidermal surface. At times individual hyphae grew along the depressions between adjacent epidermal cells and colonized the space between cortical cells along the main axis of the root. The hyphae penetrated the epidermis and formed coils in the outer or inner cortical layers. At these penetration points, root hairs were absent.

Once within the epidermis the hyphae grew parallel to the main axis of the host root and from cell to cell within the epidermis, without any distortion of host root. The hyphae passed through the adjoining epidermal cell walls by narrow penetration tips, which occasionally arose from an appressorium-like structure. Intraradical colonization was characterized by frequently septate intercellular hyphae and intracellular microsclerotia formation. The stele was free of fungal structures and there was no damage to host root tissues. In addition, there was a lack of morphological distinction between intraradical and extramatrical hyphae.

Discussion

We investigated root fungal associations in 16 trees, 9 shrubs, and 4 herbaceous shola plant species. Five of these (*Elaeocarpus munronii*, *Impatiens leschenaultii*, *Mahonia leschenaultii*, *Rhododendron arboreum* ssp. *nilagiricum*, and *Rubus racemosus*) are endemic to Western Ghats (Ahmedullah and Nayar 1986). Five different mycorrhizal types and DSE fungal associations were classified, confirming previous reports on the co-occurrence of diverse root fungal associations within the same plant community (Maremmani et al. 2003; Tsuyuzaki et al. 2005).

The mycorrhizal type of most plant species surveyed confirmed data reported by other workers. Arbuscular mycorrhiza was found in greater than 50% of the plant species examined, showing the predominance of this mycorrhizal type in Western Ghats region (Muthukumar and Udaiyan 2000; Muthukumar et al. 2006). The occurrence of mycorrhizas in 19 species and the presence of ectendomycorrhizas in *P. oocarpa* have been reported for the first time. The data in the Appendix provide the type of root fungal association reported in related plant species for which the root fungal association has been reported for the first time in this study. The occurrence of ectendomycorrhizas in several *Pinus* species has been previously reported (Rudawska et al. 2001; Yu et al. 2001), and structures observed in *P. oocarpa* roots are similar to those observed in other *Pinus* species (Yu et al. 2001). Yu et al. (2001) indicated that occurrence of ectendomycorrhizas in field-collected roots should be interpreted with caution as they may represent senescent ectomycorrhizas. However, in the present study, care was taken to avoid sampling old or senescing roots, and the observations presented are for young intact short roots. This report on the existence of ectendomycorrhizas in *P. oocarpa* extends the global distribution of ectendomycorrhizal association in diverse habitats, as found in other studies, and clearly suggests that they play an important and more significant role in host growth that has yet to be ascertained. The occurrence of both ectomycorrhizal and AM in *E. grandis* is in line with the general phenomenon of different mycorrhizal association in this *Eucalyptus* (Bellei et al. 1992; Olivera et al. 1997; dos Santos et al. 2001), and a succession over time of two overlapping systems has been described (Bellei et al. 1992). When AM and ectomycorrhizas co-occur, AM fungi most likely colonize the root first, followed by the formation of mantle and Hartig net (Chilvers et al. 1987; Wagg et al. 2008).

A group of plant species belonging to mycorrhizal families such as Berberidaceae, Elaeagnaceae, Elaeocarpaceae, Myrtaceae, and Symplocaceae (Wang and Qiu 2006) were devoid of any mycorrhizal association according to our findings. Of the 16 AM plant species, *Arum*-type morphology was present in 10, and 5 species had *Paris*-type morphology. It has been generally believed that the *Arum*-type of AM morphology is more common than the *Paris*-type, since most cultivated herbaceous plants that have been used in studies form the *Arum*-type. However, Smith and Smith (1997) indicated that *Paris*-type AM morphology is found in a rather wide range of plant taxa. *Arum*-type AM morphology was reported to be widespread in plant species growing in different forest types in Xishuangbanna, southwest China (Muthukumar et al. 2003). New records on the AM morphological type in some plant families were obtained, such as *Arum*-type in Lauraceae and *Paris*-type in Ternstroemiaceae and Rhamnaceae. Similarly, deviations from normal morphological types reported were obtained. Members of Asteraceae and Turneraceae (Staphyleaceae), which are known to form typical *Arum*-type AM morphology, had *Paris*- and intermediate-type AM morphology, respectively, in this study. Similarly, members of Caprifoliaceae (Adoxaceae), which are known to form intermediate type of AM morphology, formed typical *Paris*-type in the present study.

We compared the distribution of different AM morphological types in relation to plant habit. Our results showed

that 10 of the shrubs and trees examined had *Arum*-type morphology and 4 had *Paris*-type morphology. Yamato (2005) also reported predominance of *Arum*-type AM morphology in tree species at an oil palm farm in Indonesia. Although physiological or functional differences between the two morphological types have not yet been fully elucidated, it has been shown that the development of *Paris*-type is slower than that of *Arum*-type (Cavagnaro et al. 2001). Brundrett and Kendrick (1990) discussed that slower colonization in *Paris*-type AM morphology might be beneficial for the host plants to keep the energy supply to the fungi reduced and might be desirable for plants growing slowly in a woodland environment.

Members of Orchidaceae examined had similar morphology, with intact pelotons in the young roots and degenerating pelotons in the older roots, as described by Zelmer et al. (1996). Restriction of colonization to roots in contact with the substratum in epiphytic species is in accordance with the observation of Hadley and Williamson (1972) in Malaysian epiphytic orchids, where aerial roots that were not in contact with the substratum were free of fungal colonization. The high levels of colonization observed in epiphytic orchids contradict the widely held view that epiphytic orchids have low levels of colonization (Rasmussen 2002). In addition, the higher colonization levels observed in the terrestrial *M. versicolor* than the epiphytic *O. platycaulon* and *O. ensiformis* corroborate the findings that terrestrial orchid species are reported to be more highly mycotrophic than are epiphytic species. Kaliamoorthy (2007) recorded higher levels of colonization in terrestrial *Calanthe triplicata* than in epiphytic *Aerides maculosum*. Though it is generally believed that adult phase of photosynthetic orchids are least or not dependent on mycorrhizal fungi for their existence (Smith and Read 1997), the observations of the present study clearly indicates that orchids sustain mycorrhizal association throughout their lifecycle under natural conditions.

DSE fungal endophytes were found in six plant species studied. This is expected, since dark septate endophytes are widespread among angiosperms (Jumpponen and Trappe 1998). Despite the assumption of the widespread occurrence of DSE in angiosperms, their occurrence has been reported in only 59 tropical plant species (Jumpponen and Trappe 1998), and this is the first report of DSE fungal association in all the six plants reported in this study. The functional relationship between DSE fungi and plants may be comparable to that between ectendomycorrhizal fungi or AM fungi and their host. Infection with DSE fungi seems to be mutualistic rather than parasitic. Studies have shown that DSE are more efficient than AM fungi at colonizing roots in cold soils (Kohn and Stasovski 1990; Väre et al. 1992), but their efficiency in semiarid and arid soils is unknown. Further, increased N and P uptake in response to DSE fungal colonization has been reported in different plant species (Muller et al. 1986; Newsham 1999).

This study clearly shows that root fungal associations are an important component of shola plant community, and it is likely that their occurrence is important to plant success and survival in shola habitat.

Acknowledgments We thank University Grants Commission, New Delhi for financial assistance.

References

Ahmedullah M, Nayar MP (1986) Endemic plants of the Indian region. Botanical Survey of India, Calcutta

Bellei MM, Garbaye J, Gil M (1992) Mycorrhizal succession in young *Eucalyptus viminalis* plantations in Santa Catarina (southern Brazil). For Ecol Manag 54:205–213

Brundrett MC, Kendrick B (1990) The roots and mycorrhizas of herbaceous woodland plants. II. Structural aspects of morphology. New Phytol 114:469–479

Cavagnaro TR, Gao L-L, Smith FA, Smith SE (2001) Morphology of arbuscular mycorrhizas is influenced by fungal identity. New Phytol 151:469–475

Chilvers GA, Lapeyrie FF, Horan DP (1987) Ectomycorrhizal vs endomycorrhizal fungi within the same root system. New Phytol 107:441–448

Dickson S, Smith FA, Smith SE (2007) Structural differences in arbuscular mycorrhizal symbioses: more than 100 years after Gallaud, where next? Mycorrhiza 17:375–393

dos Santos VL, Muchovej RM, Borges AC, Neves JCL, Kasuya MCM (2001) Vesicular arbuscular-/ecto-mycorrhiza succession in seedlings of *Eucalyptus* spp. Braz J Microbiol 32:81–86

Fontenla S, Puntieri J, Ocampo JA (2001) Mycorrhizal associations in the Patagonian Steppe, Argentina. Plant Soil 233:13–29

Francis R, Read DJ (1994) The contributions of mycorrhizal fungi to the determination of plant community structure. Plant Soil 159:11–25

Hadley G, Williamson B (1972) Features of mycorrhizal infection in some Malaysian orchids. New Phytol 71:1111–1118

Henry AN, Kumari GR, Chitra V (eds) (1989) Flora of Tamil Nadu, India, 2nd edn. Botanical Survey of India, Coimbatore

Jackson ML (ed) (1971) Soil chemical analysis. Prentice–Hall, New Delhi

Janos DP (1996) Mycorrhizas, succession and the rehabilitation of deforested lands in the humid tropics. In: Frankland JC, Magan N, Gadd GM (eds) Fungi and environmental change. Cambridge University Press, Cambridge, pp 129–162

Jumpponen A (2001) Dark septate endophytes—are they mycorrhizal? Mycorrhiza 11:207–211

Jumpponen A, Trappe JM (1998) Dark septate endophytes: a review of facultative biotrophic root-colonizing fungi. New Phytol 140:295–310

Kai W, Zhiwei Z (2006) Occurrence of arbuscular mycorrhizas and dark septate endophytes in hydrophytes from lakes and streams in southwest China. Int Rev Hydrobiol 91:29–37

Kaliamoorthy S (2007) Pattern of mycorrhizal infections in the roots of *Aerides maculosum* Lindl. and *Calanthe triplicata* (Willem.)Ames. Mycorrhiza News 19:14–18

Kohn LM, Stasovski E (1990) The mycorrhizal status of plants at Alexandra Fiord, Ellesmere Island, Canada, a high arctic site. Mycorrhiza 82:23–25

Korb JE, Johnson NC, Covington WW (2003) Arbuscular mycorrhizal propagule densities respond rapidly to ponderosa pine restoration treatments. J Appl Ecol 40:101–110

Koske RE, Gemma JN (1989) A modified procedure for staining roots to detect VA mycorrhizas. Mycol Res 92:486–488

Laursen G, Treu R, Seppelt RD, Stephenson SL (1997) Mycorrhizal assessment of vascular plants from subantartic Macquarie Island. Arct Alp Res 29:483–491

Maremmani A, Bedini S, Matoševic I, Tomei PE, Giovannetti M (2003) Type of mycorrhizal associations in two coastal nature reserves of the Mediterranean basin. Mycorrhiza 13:33–40

McGonigle TP, Miller MH, Evens DG, Fairchild GL, Swan JA (1990) A method which gives an objective measure of colonization of roots by vesicular-arbuscular mycorrhizal fungi. New Phytol 115:495–501

Meher-Homji VM (1967) Phytogeography of the South Indian hill stations. Bull Torrey Bot Club 48:43–52

Muller WC, Tessier BJ, Englander L (1986) Immunocytochemical detection of fungi in the roots of *Rhododendron*. Can J Bot 64:718–723

Muthukumar T, Udaiyan K (2000) Arbuscular mycorrhizas of plants growing in Western Ghats region, southern India. Mycorrhiza 9:297–313

Muthukumar T, Udaiyan K (2002) Seasonality of vesicular-arbuscular mycorrhizae in sedges in a semi-arid tropical grassland. Acta Oecol 23:337–347

Muthukumar T, Sha L, Yang X, Cao M, Tang J, Zheng Z (2003) Mycorrhiza of plants in different vegetation types in tropical ecosystems of Xishuangbanna, southwest China. Mycorrhiza 13:289–297

Muthukumar T, Senthilkumar M, Rajangam M, Udaiyan K (2006) Arbuscular mycorrhizal morphology and dark septate fungal associations in medicinal and aromatic plants of Western Ghats, southern India. Mycorrhiza 17:11–24

Nair NC, Henry AN (eds) (1983) Flora of Tamil Nadu, India, vol 1. Botanical Survey of India, Coimbatore

Newsham KK (1999) *Phialophora graminicola*, a dark septate fungus, is a beneficial associate of the grass *Vulpia ciliata* ssp. *ambigua*. New Phytol 144:517–524

Olivera VL, Schmidt VDB, Bellei MM (1997) Patterns of arbuscular- and ecto-mycorrhizal colonization of *Eucalyptus dunnii* in southern Brazil. Ann For Sci 54:473–481

Rains KC, Nadkarni NM, Bledose CS (2003) Epiphytic and terrestrial mycorrhizas in a lower montane Costa Rica cloud forest. Mycorrhiza 13:257–264

Rasmussen HN (2002) Recent developments in the study of orchid mycorrhiza. Plant Soil 244:149–163

Read DJ, Birch CPD (1988) The effects and implications of distribution of mycorrhizal mycelial systems. Proc R Soc Edinburgh 94:13–24

Routsalainen AL, Väre H, Vestberg M (2002) Seasonality of root fungal colonization in low-alpine herbs. Mycorrhiza 12:29–36

Rudawska M, Leski T, Gornowicz R (2001) Mycorrhizal status of *Pinus sylvestris* L. nursery stock in Poland as influenced by nitrogen fertilization. Dendrobiology 46:49–58

Smith SE, Read DJ (eds) (1997) Mycorrhizal symbiosis. Academic, London

Smith SE, Smith FA (1997) Structural diversity in (vesicular)-arbuscular mycorrhizal symbiosis. New Phytol 114:1–38

Somasundaram S, Vijayan L (2005) Conservation of endemic birds in the shola forests of the Palni Hills, south India. In: Vijayan L, Vasudeva R, Priyadarsanan DR, Borges R, Krishnaswamy J, Pramod P, Rao J, Ranjit Daniels RJ (eds) Integrating science and management of biodiversity in Western Ghats. Salim Ali Centre for Ornithology, Coimbatore, pp 11–12

Thormann MN, Currah RS, Bayley SE (1999) The mycorrhizal status of the dominant vegetation along a peatland gradient in southern boreal Alberta, Canada. Wetlands 19:438–450

Treu R, Laursen GA, Stephenson SL (1996) Mycorrhizae from Denali National Park and Preserve, Alaska. Mycorrhiza 6:21–29

Tsuyuzaki S, Hase A, Niinuma H (2005) Distribution of different mycorrhizal classes on Mount Koma, northern Japan. Mycorrhiza 15:93–100

Väre H, Vestberg M, Eurola S (1992) Mycorrhiza and root associated fungi in Spitsbergen. Mycorrhiza 1:93–104

Wagg C, Pautler M, Massicotte HB, Peterson RL (2008) The co-occurrence of ectomycorrhizal, arbuscular mycorrhizal, and dark septate fungi in seedlings of four members of Pinaceae. Mycorrhiza 18:103–110

Wang B, Qiu YL (2006) Phylogenetic distribution and evolution of mycorrhizas in land plants. Mycorrhiza 16:299–363

Weishampel PA, Bedford BL (2006) Wetland dicots and monocots differ in colonization by arbuscular mycorrhizal fungi and dark septate endophytes. Mycorrhiza 11:495–502

Yamato M (2005) Morphological types of arbuscular mycorrhizas in pioneer woody plants growing in an oil palm in Sumatra, Indonesia. Mycoscience 46:66–68

Yu TE, Egger KN, Peterson LR (2001) Ectendomycorrhizal associations—characteristics and functions. Mycorrhiza 11:167–177

Zelmer CD, Cuthbertson L, Currah RS (1996) Fungi associated with terrestrial orchid mycorrhizas, seeds and protocorms. Mycoscience 37:439–448