Tropical and Subtropical Agroecosystems

ASSEMBLAGE AND DIVERSITY OF FUNGI ASSOCIATED WITH MANGROVE WILD LEGUME Canavalia cathartica

[DIVERSIDAD DE HONGOS ASOCIADOS A LA LEGUMINOSA SILVESTRE DE MANGLAR *Canavalia cathartica*]

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SUMMARY

Assemblage and diversity of fungi in five tissues (root, stem, leaf, pod and seed) of mangrove wild legume Canavalia cathartica on surface sterilization and without surface sterilization has been studied. The surface sterilized tissues yielded 36 endophytic fungi with a highest 15 species in stem followed by 14 species in root and pod tissues. Aspergillus niger was the most dominant core-group endophytic fungus (29.3%). The diversity and evenness were highest in roots. The highest similarity of fungi was seen between pods and seeds (41%) followed by roots vs. pods and stem vs. pods (40%), while it was least between roots and leaves (22%). Tissues without surface sterilization yielded a total of 40 fungi with a highest of 25 species in leaf followed by 23 species in pod tissue. Aspergillus niger was the most dominant fungus in all tissues (48.5%). Diversity was highest in leaf, while the similarity ranged between 16% (stem vs. seed) and 50% (stem vs. leaf). Sixteen fungi were common to sterilized and unsterilized tissues. Aspergillus flavus, A. niger, Fusarium oxysporum and Penicillium chrysogenum were the top ranked coregroup fungi common to sterilized and unsterilized tissues. The percent frequency of occurrence of fungi did not differ significantly between surface sterilized and unsterilized tissues.

Key words: *Canavalia cathartica*; wild legume; mangroves; wetlands; fungal diversity; endophytes

INTRODUCTION

Mangroves are known for their ecological services in tropical and subtropical regions of the world by providing niches for a variety of flora, fauna and microbes. Mangroves of the southwest coast of India support several underexploited wild legumes (e.g. *Canavalia* spp., *Sesbania* spp.). Among them, *Canavalia* cathartica Thouars [(common name:

RESUMEN

Se estudió la diversidad de hongos en cinco tejidos (raíz, tallo, hoja, vaina y semilla) de la leguminosa silvestre de maglar Canavalia cathartica con o sin esterilización de la superficie. En la superficie esterilizada se obtuvo 36 hongo endofitos con un total de 15 especies en el tallo, seguido 14 en la ráiz, y vaina. Aspergillus niger fue la especie dominante (29.3%). La diversidad y regularidad fue mayor en las raíces. La mayor similaridad de hongos se encontró entre las vainas y semillas (41%) seguido de raíces vs. Vainas (40%), mientras que la menor fue entre raíces y hojas (22%). En el tejido sin esterilización de superficie se encontraron 40 hongos con una total de 25 especies en la hoja, seguido de 23 en las vaina. Aspergillus niger fue el hongo dominante en todos los tejidos (48.5%). La diversidad fue mayor en la hoja, mientras que la similaridad fue de 16% (tallo vs. Semilla) y 50% (tallo vs. Hoja). La frecuencia de ocurrencias de los hongos no fue diferente entre tejidos esterilizados y no. Diez y seis hongos fueron comunes a los tejidos esterilizados y no esterilizados. Aspergillus flavus, A. niger. Fusarium oxysporum y Penicillium chrysogenum fueron los más frecuentes en ambos tipos de muestra.

Palabras clave: *Canavalia cathartica*; leguminosas silvestres; manglar; humedales; diversidad fungal; endofitas.

Maunaloa; synonyms: *C. microcarpa* (DC.) Piper; *C. turgida* Graham *ex* A. Gray; *C. virosa* (Roxb.) Wight *et* Arn.; *Dolichos virosus* Roxb.; *Lablab microcarpus* DC.)] are of special importance. It is a wild ancestor of *C. gladiata* (Jacq.) DC. (synonym: *Dolichos gladiatus* Jacq.), distributed throughout the tropical Asia and Africa (Purseglove, 1974). It is native to mangroves and disseminates seeds or propagules through drift to coastal sand dunes. Although *C. cahartica* is a wild

legume, coastal farmers utilize this germplasm in their plantations and agricultural fields in order to improve the soil fertility as mulch, green manure and its ability to fix atmosphoric nitrogen through rhizobia (Arun and Sridhar, 2004, 2005). Sastrapradja et al. (1981) have reported a natural hybrid of C. cathartica and C. gladiata, and artificial hybridization of C. cathartica with C. ensiformis decreased the pollen fertility in F1 and F2 progenies. Kathiravan and Ignacimuthu (1999) transplanted tissue cultured plants of C. cathartica to the field successfully after hardening. Gamma irradiation (4 and 6 krad) improved the germination ability of C. cathartica seeds (Rodrigues, 1993). Recently dry seeds, ripened beans and tender pods have been evaluated for their nutritional and anitnutritional properties (Seena et al., 2006; Bhagya and Sridhar, 2007; Bhagya et al., 2007). However, cultivability, nutritional versatility and symbiotic potentiality with microbes of the C. cathartica landrace still remain as challenge (Sridhar and Seena, 2006). Along the mangrove wetlands of the River Nethravathi, Southwest coast of India, C. cathartica grows well and useful mainly as mulch and green manure. Aim of the present study is to document fungal assemblage and diversity of C. cathartica growing at the Nethravathi wetlands. This study assesses the fungal diversity in five tissue classes of mature plants with and without surface sterilization to compare the endophytic fungi with epiphytic/saprophytic fungi.

MATERIALS AND METHODS

Study site

Study site selected at the Nethravathi mangrove area of Southwest coast of India (12°50'27"N, 74°51'45"E) consists of perennial wild legume Canavalia cathartica in large expanses of wetlands (Fig. 1a-d). This legume grows luxuriously in cultivated and uncultivated areas of the mangrove habitat. Being nitrogen-fixing, farmers allow this legume to grow and utilize as cover crop, mulch and green manure for plantations (coconut and areca) and other commercial crops (paddy, sugarcane and vegetable). This legume also partially serves as fodder for livestock. These plants flower during monsoon and post-monsoon season (June-October) followed by formation of tender pods and the pods dry up during the summer months (February-May) and spread seeds. The study has been carried out during the post-monsoon season (October, 2006 - January, 2007).

Canavalia cathartica

Five mature plants each about 50 m apart were selected, uprooted, transferred to the laboratory and processed the tissues within four hours duration. Four

tissue types (root, stem, leaf, tender pod) from each plant were separated and cut into five segments each of one cm length and washed in distilled water to remove the extraneous matter. From each plant five dry seeds were selected to evaluate the fungal component. The tissue segments and seeds were surface sterilized using 95% ethanol (1 min), 6% sodium hypochlorite (5 min) and 95% ethanol (0.5 min) followed by rinse in sterile distilled water. The tissue segments and seeds were plated on antibiotic amended (tetracycline, 250 mg/l) 1.5% potato dextrose agar (PDA) medium. The tissues and seeds without surface sterilization were also plated on the antibiotic amended PDA medium. The plates were incubated at 23±2°C up to four weeks at 12 hr light and dark regime. Periodically they were screened for the growth of mycelia or discrete colonies on the medium or on the segments or seeds. The growing mycelial portions were transferred to fresh antibiotic-free PDA medium. Fungi were identified based on the spore morphology and colony characteristics using standard monographs and taxonomic keys.

Data analysis

The percent frequency of colonization and mean percent frequency of all fungi and core-group fungi (frequency of occurrence $\geq 10\%$) of surface sterilized and unsterilized tissues and seeds were calculated:

Frequency of occurrence (%) = [(Number of segments colonized) ÷ (Total segments screened)] ×100

Mean % frequency of each fungus = (Total % frequency on all tissues) ÷ (Total tissues screened)

Mean % frequency/fungus = (Total % frequency of fungi) ÷ (Total fungi)

Mean % frequency/core-group fungus = (Total % frequency of core-group fungi) ÷ (Total fungi)

The Simpson and Shannon diversities (Magurran, 1988) and evenness (Pielou, 1975) were estimated. Jaccard's index of similarity was calculated pair-wise among different tissues and seeds based on the presence or absence of each fungal species (Kenkel and Booth, 1992). Student's *t*-test was employed to assess the difference in frequency of occurrence between sterilized and unsterilized tissues (StatSoft Inc., 1995).

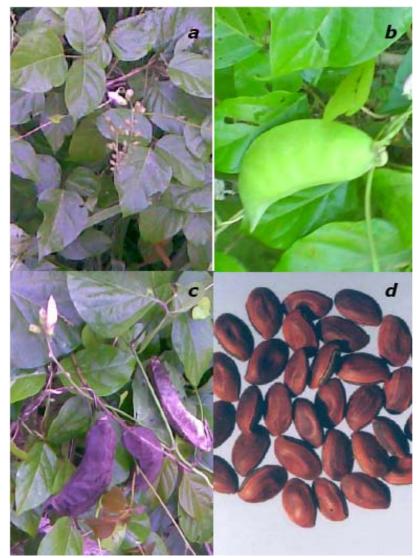


Figure 1. *Canavalia cathartica* grown in wetlands of Nethravathi mangrove showing inflorescence (a), tender pod (b), dry pods (c) and seeds (d).

RESULTS

Sterilized tissues

Five surface sterilized tissues yielded 36 endophytic fungi (Table 1). A maximum of 15 species was recovered in stem followed by 14 species in root and pod tissues(Fig. 2a). Aspergillus niger was the most dominant fungus in all tissues (29.3%) followed by *Penicillium chrysogenum* (24.6%) and Aspergillus flavus (19.1%), Fusarium oxysporum (16.9%) and a non-sporulating sp. 1 (16.5%). Six fungi belonged to

core-group (frequency of occurrence $\geq 10\%$) in at least one of the tissues. Except for stem (2 species), rest of the tissues consist of four species of core-group fungi. Five fungi were confined to root/leaf/seed, while four and three species to stem and pod respectively. The mean frequency of occurrence per fungus was highest in pods (11.6%) followed by leaves (9.8%) (Fig. 2b). The contribution of core-group fungi was ranged between 49 and 72%. There was a steep increase in species accumulation curve of total fungi than coregroup fungi from root to seed (Fig. 2c).

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Table 1. Frequency of occurrence (%) of fungi in surface sterilized tissues of *Canavalia cathartica* of Nethravathi mangroves (MFO, mean % frequency of occurrence) (*also found in unsterilized tissues).

Fungus	Root	Stem	Leaf	Pod	Seed	MFO
*Aspergillus niger Tiegh.	28.9	28.9	42.2	31.1	15.6	29.3
*Penicillium chrysogenum Thom	4.5		6.7	6.7	6.7	24.6
*Aspergillus flavus Link	11.1	11.1	8.9	46.7	17.8	19.1
*Fusarium oxysporum E.F. Sm. & Swingle	20.0	26.7	8.9	15.6	13.5	16.9
Non sporulating sp. 1	15.6	8.9	20	22.3	15.6	16.5
*Aspergillus tamarii Kita	6.7	4.5	4.5	2.2	4.5	4.5
*Trichoderma harzianum Rifai	8.9	8.9				3.6
*Penicillium sp.1		11.1		2.2		2.7
*Alternaria alternata (Fr.) Keissl.		2.2		8.9		2.2
Cladosporium sp.				6.7	4.5	2.2
*Penicillium italicum Stoll	8.9			2.2		2.2
*Aspergillus fumigatus Fresen.				8.9		1.8
*Aspergillus sp. 5			8.9			1.8
Chaetomium sp.		2.2	6.7			1.8
Nigrospora sp.			8.9			1.8
Trichoderma hamatum (Bonord.) Bainier	6.7					1.3
Trichoderma pseudokoningi Rifai	2.2	4.5				1.3
Yeast sp. 1				6.7		1.3
Alternaria sp.		2.2				0.9
*Aspergillus ochraceus G. Wilh.				4.5		0.9
*Aspergillus parasiticus Speare		2.2		2.2		0.9
*Cladosporium oxysporum Berk. & M.A.			4.5			0.9
Curtis						
Drechslera halodes (Drechsler) Subram. &	4.5					0.9
B.L. Jain						
Penicillium sp.2					4.5	0.9
*Scytalidium lignicola Pesante		4.5				0.9
Aspergillus sp. 1					2.2	0.4
Aspergillus sp. 2					2.2	0.4
Aspergillus sp. 3			2.2			0.4
Aspergillus sp. 4	2.2					0.4
Curvularia clavata B.L. Jain	2.2					0.4
Curvularia eragrostidis (Henn.) J.A. Mey.	2.2	2.2				0.4
Curvularia prasadii R.L. Mathur & B.L.			2.2			0.4
Mathur						0.1
*Eurotium chevalieri L. Mangin					2.2	0.4
Mucor sp.		2.2			4.4	0.4
Phytophthora sp.		<i></i>			2.2	0.4
Rhizopus sp.	2.2				2.2	0.4
Total fungi	14	15	13	14	12	0.7
Total core-group fungi	4	15 4	13	4	4	

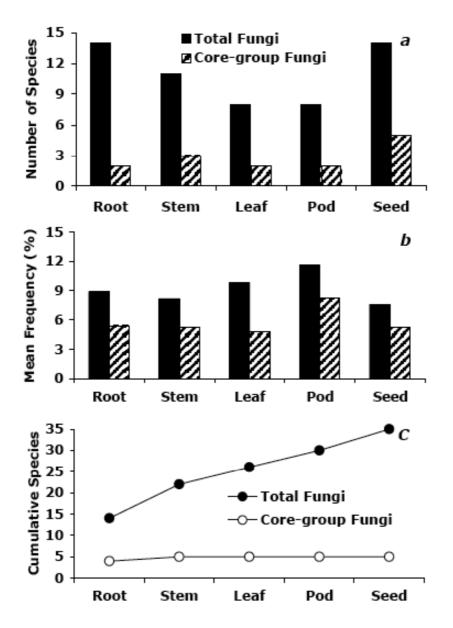


Figure 2. Number of fungi (a), mean percent frequency of occurrence (b) and species accumulation curve (c) in surface sterilized tissues of *Canavalia cathartica*.

Unsterilized tissues

Unsterilized tissues yielded 40 fungi (Table 2) with a maximum of 25 species in leaf followed by pod (23), root (19) and stem (18) tissue (Fig. 3a). Aspergillus niger was most dominant (48.5%) followed by *Fusarium oxysporum* (33.4%). Nine fungi belonged to core-group fungi and dominant at least in one of the tissues. Leaves consist of a maximum of six core-

group fungi (Fig. 3b). Eighteen fungi were confined at least to one of the tissues. The mean frequency of occurrence per fungus was highest in seed followed by pod and root (Fig. 3b). The contribution of core-group fungi was ranged between 70.7% (stem) and 84.4% (seed). There was a steep increase in the species accumulation curve of total fungi from root to seed (Fig. 3c). Table 2. Frequency of occurrence (%) of fungi in unsterilized tissues of mature plants of *Canavalia cathartica* of Nethravathi mangroves (MFO, mean % frequency of occurrence) (*also found in sterilized tissues).

Fungus	Root	Stem	Leaf	Pod	Seed	MFO
*Aspergillus niger Tiegh.	80	53.4		84.5	66.7	48.5
*Fusarium oxysporum E.F. Sm. & Swingle	11.1	46.7	44.5	64.5		33.4
*Aspergillus flavus Link	6.7	8.9	2.2	71.1		17.8
Rhizopus sp.	13.4				64.5	15.0
Non sporulating sp. 2	15.6	22.2	31.1	2.2		14.2
Penicillium citrinum Sopp		17.8	33.4	4.5	4.5	12.0
*Penicillium chrysogenum Thom	6.7	13.4	15.6	2.2	2.2	8.0
*Aspergillus ochraceus G. Wilh.		2.2		26.7	2.2	6.
*Aspergillus tamarii Kita	4.5	8.9	8.9	2.2		4.9
Penicillium glabrum (Wehmer) Westling	2.2		4.5	2.2	8.9	3.
*Penicillium italicum Stoll	2.2	6.7	6.7	2.2		3.0
Yeast sp. 2	2.2	4.5	6.7	2.2		3.
*Trichoderma harzianum Rifai	11.1		2.2			2.7
Mucor microsporus Bonord.		4.5	2.2	4.5		2.2
Nigrospora sp.			8.9	2.2		2.1
<i>Cladosporium cladosporioides</i> (Fresen.)		2.2	2.2	4.5		1.8
G.A. de Vries						1.
Alternaria sp.		6.7				1.
* <i>Cladosporium oxysporum</i> Berk. & M.A.		0.7	6.7			1.
Curtis			0.7			1
Penicillium sp.3	4.5	2.2				1.
Philophora agaricina Wallr.	1.5	2.2	2.2	4.5		1.
Syncephalastrum sp.	6.7		2.2	т.Ј		1.
*Aspergillus fumigatus Fresen.	0.7			4.5		0.9
*Aspergillus parasiticus Speare		2.2		2.2		0.9
Curvularia eragrostidis (Henn.) J.A. Mey.		2.2		2.2	2.2	0.9
Curvularia sp.		2.2	2.2	2.2	2.2	0.
Periconia sp.		2.2	2.2			0.9
*Alternaria alternata (Fr.) Keissl.		2.2	2.2	2.2		0.4
Alternaria dianthi J.V. Almeida & Sousa da				2.2		0.4
Câmara				2.2		0.4
<i>Alternaria tenuissima</i> (Kunze) Wiltshire			2.2			0.4
			2.2			0.4
*Aspergillus sp. 5		2.2	2.2			0.4
Codinea sp.		2.2		2.2		
Colletotrichum lindemuthianum (Sacc. &				2.2		0.4
Magnus) Briosi & Cavara			2.2			0
Curvularia pallescens Boedijn			2.2		2.2	0.4
* <i>Eurotium chevalieri</i> L. Mangin					2.2	0.4
<i>Kallichroma tethys</i> (Kohlm. & Kohlm.)					2.2	0.4
Kohlm. & VolkmKohlm.						<u>_</u>
Mucor plumbeus Bonord.			2.2	• •		0.4
*Penicillium sp.1				2.2		0.4
Phoma sp.			. -	2.2		0.4
*Scytalidium lignicola Pesante			2.2			0.4
Sporotrichum sp.			2.2			0.4
Total fungi	13	18	25	23	9	
Total core-group fungi	5	5	6	4	2	

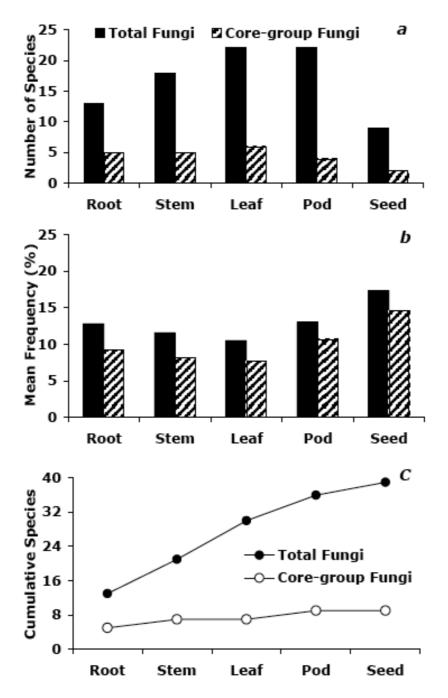


Figure 3. Number of fungi (a), mean percent frequency of occurrence (b) and species accumulation curve (c) in surface unsterilized tissues of *Canavalia cathartica*.

Diversity and similarity

In surface sterilized tissues, the Simpson and Shannon diversities and evenness were highest in root (Table 3). The highest similarity of fungi was seen between pod and seed (41%) followed by root vs. pod and stem vs. pod (40%), while it was least between root and leaf (22%) (Table 4). In unsterilized tissues, the diversity was highest in leaf (Table 3). The similarities ranged

between 16% (stem vs. seed) and 50% (stem vs. leaf). Sixteen fungi were common in surface sterilized and unsterilized tissues (Table 1, 2). Aspergillus flavus, A. niger, Fusarium oxysporum and Penicillium chrysogenum were the top four core-group fungi common in sterilized and unsterilized tissues. The percent frequency of occurrence of fungi did not differ significantly between surface sterilized and unsterilized tissues (p > 0.05).

Table 3. Diversity of fungi in surface sterilized and unsterlized tissues (in parenthesis) of *Canavalia cathartica* of Nethravathi mangroves.

	Root	Stem	Leaf	Pod	Seed
Simpson diversity	0.863	0.848	0.817	0.835	0.850
	(0.723)	(0.843)	(0.891)	(0.799)	(0.622)
Shannon diversity	2.398	2.358	2.226	2.236	2.273
	(1.919)	(2.349)	(2.673)	(2.089)	(1.302)
Evenness	0.786	0.705	0.717	0.668	0.809
	(0.524)	(0.582)	(0.580)	(0.351)	(0.409)

Table 4. Jaccard's percent similarity index of surface sterilized and unsterilized (in parenthesis) tissues of *Canavalia cathartica* of Nethravathi mangroves.

	Stem	Leaf	Pod	Seed
Root	26	22	40	30
	(36)	(32)	(29)	(29)
	Stem	33	40	26
		(50)	(43)	(16)
		Leaf	27	32
			(36)	(17)
			Pod	41
				(23)

DISCUSSION

Estuarine and mangrove regions of the southwest coast of India encompass diverse natural vegetation (mangroves, mangrove associates and salt-tolerant plants) (Rao and Suresh, 2001). Among mangrove tree species (54) and mangrove associates (60), about 50% have been investigated for their fungal association (Hyde and Lee, 1995; Jones and Alias, 1996). Among wild legumes found in mangrove and estuarine habitats of Nethravathi, *C. cathartica* is of immense value to farmers for improvement of soil fertility as it serves as a good cover crop, green manure and fix atmospheric nitrogen. It is a perennial creeper and conserves the soil through its soil binding ability by nodal roots especially in coastal sand dunes (CSD) of the southwest coast of India (Arun *et al.*, 1999).

Endophytes are generally defined as the mutualistic microorganisms associated with healthy tissues without exhibiting disease symptoms in host. Besides arbusuclar mycorrhizal fungi (Kulkarni *et al.*, 1997; Bhagya *et al.*, 2005; Arun and Sridhar, 2006), only one study is available on fungal endophytes of *C. cathartica* growing on the CSD of southwest coast of India (Seena and Sridhar, 2004). The percent frequency of occurrence of endophytic fungi increased between seed and root/stem/leaf segments in CSD *C. cathartica* (Seena and Sridhar, 2004). In the current study, the mean percent frequency of endophytic fungi

was highest in pod (11.6%), followed by leaf (9.8%) and root (8.9%). In unsterilized tissues, seed showed the highest mean frequency of fungi (17.3%) followed by pod (13%) and root (12.8%). In C. cathartica of CSD, Chaetomium globosum was the dominant endophytic fungus and root showed its dominance as single species (C. globosum) (Seena and Sridhar, 2004). But in mangrove C. cathartica, multispecies dominance was seen in sterilized tissues (Aspergillus flavus, A. niger, Fusarium oxysporum and nonsporulating sp. 1). Acremonium, Colletotrichum and Fusarium were common endophytes in halophytes of CSD (Fisher and Petrini, 1987; Beena et al., 2000), mangroves (Kumaresan and Suryanarayanan, 2001; Ananda and Sridhar, 2002; Maria and Sridhar, 2003) and sea grass (Devarajan et al., 2002). In our study, F. oxysporum was a dominant endophyte in root, stem, pod and seed, while Colletotrichum lindemuthianum confined to unsterilized pod segments (2.2%). However, unsterilized seed showed single species dominance by A. niger (66.7%). Many endophytic fungi did not establish on the PDA medium without surface sterilization due to suppression by the epiphytic/saprophytic fungi. For instance, seeds without surface sterilization yielded only nine fungi, while 12 fungi in surface sterilized seeds (see Table 1, 2). As a highly dominant fungus, it is likely A. niger (66.7%) has suppressed the growth of other fungi in unsterilized seeds. Tissue specificity of endophytic fungi in whole-stem and xylem has been reported in tree species (Pinus and Fagus; Petrini and Fisher, 1988). In our study, some fungi were restricted to specific tissue in both treatments (e.g. leaf: Cladosporium oxysporum, Aspergillus sp. 5; pod: Aspergillus fumigatus; seed: Eurotium chevalieri).

Usually ascomycetes are the dominant decomposers of plant detritus in mangrove and marine habitats (Kohlmeyer and Volkmann-Kohlmeyer, 1991). However, endophytic fungi were dominated by mitosporic fungi in mangroves (Ananda and Sridhar, 2002; Suryanarayanan and Kumaresan, 2000; Maria and Sridhar, 2003) and CSD halophytes (Beena *et al.*, 2000; Seena and Sridhar, 2004). Although mangrove detritus consists of marine fungi, endophytes of Tropical and Subtropical Agroecosystems, 10 (2009): 225 - 235

mangrove plant species represent a few marine fungi. In our study, none of the typical marine fungi found on the plant detritus were endophytic, but unsterilized seeds consist of one marine fungus, Kallichroma tethys (2.5%). These findings corroborate with earlier studies on C. cathartica in CSD (Halosarpheia sp., 3%) (Seena and Sridhar, 2004) and Acanthus ilicifolius in Nethravathi mangrove habitats (Cumulospora marina, 4%) (Maria and Sridhar, 2003). However, up to 13% of root endophytes belonged to marine fungi (Monodictys pelagica, Periconia prolifica, Verruculina enalia and Zalerion maritimum) in roots of three non-leguminous CSD plant species (Beena et al., 2000).

Importance of endophytic fungi in grasses has been understood better than non-grass endophytic fungi (Hyde and Soytong, 2009). The endophytic fungi of non-grass endophytes are important because of deterring or decreasing insect herbivory, enhancing drought/disease resistance in plants and increasing plant growth (Fröhlich et al., 2000; Sieber, 2007). Besides these facts, Schulz et al. (1999) opined that production of herbicidally active metabolites by endophytic fungi is higher than phytopathogenic and soil fungi. A recent study revealed that many endophytic fungi (Aspergillus spp., Pestalotiopsis sp., non-sporulating fungus) isolated from the mangrove associate plant species (Acanthus ilicifolius and produce Acrostichum aureum) metabolites antagonistic to bacteria (Gr+ve and Gr-ve) and yeast (Candida albicans) (Maria et al., 2005). Being a perennial plant species, mangrove C. cathartica may be a good candidate for exploitation of its endophytic fungi in biological control. Further studies need to focus on the role of endophytic fungi of mangrove C. cathartica in production of novel metabolites and prevention of herbivory.

CONCLUSIONS

Canavalia cathartica is a perennial wild legume growing in mangrove wetlands of the southwest coast of India. It is used as cover crop, mulch, green manure for plantations and agricultural crops. Surface sterilized tissues (root, stem, leaf, pod and seed) of C. fungi cathartica yielded 36 endophytic and unsterilized tissues yielded 40 species of epiphytic or saprophytic fungi. The frequency of occurrence of fungi did not differ significantly between surface sterilized and unsterilized tissues. Aspergillus niger was most dominant in sterilized (29.3%) and unsterilized (48.5%) tissues followed by A. flavus, Fusarium oxysporum and Penicillium crysogenum. A typical marine fungus, Kallichroma tethys was found on unsterilized seeds (2.2%). The endophytic fungal assemblage resembles more of terrestrial mitosporic fungi than marine fungi corroborate the earlier studies

on plant species of mangroves, mangrove associates and halophytes. Future studies needs to exploit endophytic fungi of mangrove *C. cathartica* for their novel metabolites.

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