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Ecological significance of mycotrohpy in some tropical weeds

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Abstract: Weeds, which are considered to be unwanted plants, can play an important role in maintaining mycorrhizal inoculum during winter. To study this role, a survey was conducted on the mycorrhizal status of common weeds (*Ageratum conyzoides, Capsella bursa-pastoris*, *Lantana camara, Malvastrum coromandelianum*, *Medicago hispida, Parthenium hysterophorus*, *Reinwardtia indica, Solanum nigrum*, *Stellaria media* and *Vicia sativa*) at five locations in the campus of the Forest Research Institute, Dehradun, India. The highest root colonization was observed in *R. indica* (79.6 %) and the lowest root colonization (56.1 %) was quantified in roots of *S. nigrum*. Maximum mycorrhizal spore population in soil was found in *M. coromandelianum* (528.5/50 ml), while it remained minimum with *S. nigrum* (240.9). *C. bursa-pastoris*, in the family *Brassicaceae* and thought to be non-mycorrhizal, was found to be mycorrhizal. Weeds seem to support mycorrhizal communities during winter (off-season) that may serve as reservoir of inoculum of natural endophytes for other hosts such as trees during spring-summer (on-season) in tropical north India.

Resumen: Las malezas, consideradas como plantas no deseadas, pueden jugar un papel importante en el mantenimiento del inóculo micorrícico durante el invierno. Para estudiar este papel, se llevó a cabo un muestreo del estado micorrícico de malezas comunes (*Ageratum conyzoides, Capsella bursa-pastoris, Lantana camara, Malvastrum coromandelianum, Medicago hispida, Parthenium hysterophorus, Reinwardtia indica, Solanum nigrum, Stellaria media* y *Vicia sativa*) en cinco localidades en el campus del Instituto de Investigación Forestal, Dehradun, India. La mayor colonización de raíces fue observada en *R. indica* (79.6 %) y la colonización más baja (56.1 %) fue cuantificada en raíces de *S. nigrum*. La máxima población de esporas micorrícicas en el suelo fue hallada en *M. coromandelianum* (528.5/50 ml), mientras que tuvo su mínimo en *S. nigrum* (240.9). Se encontró que *C. bursa-pastoris*, perteneciente a la familia *Brassicaceae* y de la que pensaba que no era micorrícica, sí lo es. Las malezas parecen soportar comunidades micorrícicas durante el invierno (fuera de temporada) que puede servir como reservorio del inóculo de endofitos naturales para otros hospederos tales como árboles durante la primavera-verano (en temporada) en el norte tropical de la India.

Resumo: Infestantes, que são consideradas plantas indesejadas, podem jogar um papel importante na manutenção do inócuo micorrízico durante o inverno. Para estudar esse papel, conduziu-se um inventário sobre o status micorrízico de infestantes comuns (*Ageratum conyzoides, Capsella bursa-pastoris, Lantana camara, Malvastrum coromandelianum, Medicago hispida, Parthenium hysterophorus, Reinwardtia indica, Solanum nigrum, Stellaria media* e *Vicia sativa*) em cinco localizações no campus do Instituto de Investigação Florestal de Dehradun, Índia. A mais elevada colonização de raízes foi observada em *R. indica* (79.6 %), e a

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menor (56,1 %), foi quantificada nas raízes de *S. nigrum*. A máxima população de esporos micorrízicos foi encontrada em *M. coromandelianum* (528.5/50ml), enquanto permaneceu mínima com a *S. nigrum* (240.9. A *C. bursa-pastoris*, da família *Brassicaceae*, e que se pensava não ser micorrízica, revelou-o ser. As infestantes parecem suportar as comunidades micorrízicas durante o inverno (fora da estação), podendo assim servir de reservatórios de inócuo de endofíticas naturais para outros hospedeiros como sejam árvores durante a primavera-verão (durante a estação) no norte tropical da Índia.

Key words: Arbuscular mycorrhiza, field spore population, phosphorus, weeds.

Introduction

The term "weed" is been defined variously as a plant out of place, an unwanted plant, or a plant that is a pest in that it interferes with crop or livestock production. In a broad sense, weeds are plants which over dominate others by abundance, growth and coverage, thus disturbing the balance of ecosystems (Wittenberg & Cock 2001). Due to their inherent efficiency in nutrient uptake and use, weeds easily invade disturbed lands, adversely affecting ecosystems. Weeds, thus, can pose a serious challenge to sustainable management of forests, plantations, agricultural ecosystems and conservation of biodiversity.

In India, a wide spectrum of exotic plants has been introduced for agricultural, ornamental and other purposes, dating back to the earliest visits of Arab, Roman and Greek traders to the country (Gupta & Lamba 1978). According to Saxena (1991), 40 % of the plant species recorded in India are alien. Some of the most important alien weed species found in India include *Chromolaena odorata*, *Lantana camara*, *Mikania micrantha*, *Parthenium hysterophorus*, *Ageratum conyzoides*, *Ageratina adenophora*, *Eichhornia crassipes* and *Salvinia molesta*. In the Doon valley of Uttarakhand State, some weeds, namely, *L. camara*, *P. hysterophorus*, and *A. conyzoides*, have expanded at alarming rate in agricultural and abandoned fields, forests, railway tracts and road sides, becoming dangerous invaders of grasses and other herbaceous native flora (Negi & Hajra 2007; Sreenivasan 2003).

Arbuscular mycorrhizal (AM) fungi are common symbionts in terrestrial ecosystems, associating with about 80 % of plant families worldwide. They are recognized as an important, widespread component of most terrestrial ecosystems, benefiting plant establishment by enhancing plant nutrient acquisition, improving soil quality, increasing resistance to environmental stresses (Smith & Read 1997) and playing an important role in plant biodiversity, ecosystem variability and productivity. A substantial amount of information is available on mycorrhizal associations of obligate and facultative weeds. Bloss (1979) isolated *Glomus fasciculates* from soil and roots of the indigenous desert plants, *Parthenium incanum* (mariola). A survey of endogonaceous spores associated with xerophytic plants in Northern India was conducted by Singh & Verma (1981). Spores production by these endophytes was related to seasonal growth of their host. Mycorrhizal formation in weeds growing in the vicinity of crop plants was only promoted by crops strongly dependent on mycotrophic nutrition; the infection intensity of these crops themselves also increased. With facultatively mycotrophic crops, mycorrhizal infection can be negatively affected by weed species (Stejskalova 1990).

The distribution and diversity of AM fungi in different weed species of a particular agro-ecological zone are important to know in order to evaluate the natural status of AM fungi in that region and to understand their seasonal variations. It is now widely accepted that climatic and edaphic factors can influence directly or indirectly AM fungi spore numbers and their populations, which affects their association with hosts (Abbott & Robson 1991). The pressent study focuses on the role of common weeds of Doon valley in maintaining the mycorrhizal populations (based on their mycotrophy) in the ecosystem during the off-season (winter-December to February), that may, in turn, act as reservoir of fungal inoculum for companion host plants after growing season (spring to monsoon-April to July) sets in.

Table 1. Physico-chemical properties of soils.

Site	Soil property								
	Clay (%)	Silt (%)	Fine sand (%)	Coarse sand (%)	pH	Organic carbon (g kg^{-1})	Nitrogen (g kg^{-1})	Phosphorus (g kg^{-1})	Potassium (g kg^{-1})
Howard Road	16	18	18	38.5	7.3	14.1	1.5	0.016	0.120
Tireman Road	16	18	18	38.0	7.5	17.0	1.5	0.017	0.096
Pearson Road I	21	23	23	15.0	6.3	17.2	1.6	0.017	0.130
Pearson Road II	19	19	19	24.0	7.0	8.4	1.0	0.014	0.096
Travor Road	16	22	22	27.5	7.3	20.5	2.2	0.013	0.090

Materials and methods

A survey was conducted in February 2001 (winter) of the mycorrhizal status of commonly occurring weeds (*Ageratum conyzoides* Linn., *Capsella bursa-pastoris* Linn., *Lantana camara* Linn., *Malvastrum coromandelianum* Garcke., *Medicago hispida* Gaertn., *Parthenium hysterophorus* Linn., *Reinwardtia indica* Dum., *Solanum nigrum* Linn., *Stellaria media* Linn. and *Vicia sativa* Linn.) at the Forest Research Institute in Dehradun, Uttarakhand State of India. The Institute is situated at latitude 30º 20'15" N and longitude 77º 57'40" E. Dehradun receives annual rainfall of about 1,400 mm, most of which is concentrated during the monsoon months (July - September). The average temperature lies between 10.2 - 24.3 ºC during winter.

Five sampling sites were selected in the campus namely, (1) Howard Road, (2) Tireman Road, (3) Pearson Road (near library, I), (4) Pearson Road (paper plant, II) and (5) Trevor Road. Ten replicates of each weed were sampled. Rhizosphere soil was collected from a depth of 2 - 15 cm after scrapping away the top 1 - 2 cm. Soil samples were air-dried and stored at 4 ºC for further analysis. Soil texture was determined by the hydrometer method using the USDA textural classification chart. Soil pH was determined as described by Schofield & Taylor (1955) and soil organic C quantified according to Walkley & Black (1934). The total nitrogen content of soil samples was determined by the Kjeldahl method (Bremner 1965). Available P was determined by Bray's-I (Bray & Krutz 1945) and Bray's-II method (Olsen *et al.* 1954). Available K was quantified by ammonium acetate extraction followed by flame photometric determination. Soil properties are listed in Table 1.

Spore populations of mycorrhizal fungi was determined using the wet sieving and decanting technique of Gerdemann & Nicholson (1963) and the sugar density gradient centrifugation method of Daniel & Skipper (1982). Spores were counted using Doncaster's counting disc. Roots were stained using acid fuschin stain (Phillips & Hayman 1970). Quantification of mycorrhizal infection was measured by the grid line-intersect method (Giovanneti & Mosse 1980).

A one-way analysis of variance was performed for leaf phosphorus, and two-way analysis of variance was used for root colonization and soil spore population. For root infection, percent values were transformed into angular values before statistical analysis. The significance of differences between treatments means was tested at the 5 % level.

Results

Ten weeds (representing 8 families) were examined for their mycorrhizal status and types. Except for *L. camara* and *R. indica*, which are shrub/under-shrub, all other weeds are herbs. Most of them are annual, except for *C. bursa-pastoris* (perennial) and *M. coromandelianum* and *V. sativa* (biennial). With the exception of *S. media*, all weeds had one or the other mycorrhizal structures (Table 2). The mycorrhizal associations are characterized by presence of arbuscules only. Excluding *V. sativa,* all weeds had arbuscules in their root cortex and vesicles were absent in *C. bursa-pastoris* and *R. indica*. Extramatrical hyphae was absent in *V. sativa*.

The phosphorus content of leaves was maximum and significantly higher in *M. coromandelianum* (0.640 %), followed by *C. bursa-pastoris* (0.400 %). The lowest leaf phosphorus content was quantified in *L. camara* (0.256 %). Five weed

Table 2. Life history attributes, incidence and infection types, tentative root categorization and phosphorus content of leaves of weed plants of the Forest Research Institute, Dehradun.

Host	Family	Life form	Life cycle	Infection type			Tentative root category	Leaf phosphorus (%)
				AC	VC	EH		
Ageratum conyzoides	Asteraceae	Herb	Annual	+	+	+	Coarse	0.267
Capsella bursa-pastoris	Brassicaceae	Herb	Annual/ Perennial	+	-	+	Fibrous	0.400
Lantana camara	Verbenaceae	Shrub	Perennial	+	+	+	Coarse	0.256
Malvastrum coromandelianum	Malvaceae	Herb	Annual/ Biennial	+	+	+	Coarse	0.640
Medicago hispida	Papilionaceae	Herb	Annual	+	+	+	Coarse	0.346
Parthenium hysterophorus	Asteraceae	Herb	Annual	+	+	+	Coarse	0.377
Reinwardtia indica	Linaceae	Under shrub	Annual	+	-	+	Fibrous	0.307
Solanum nigrum	Solanaceae	Herb	Annual	+	+	+	Fibrous	0.320
Stellaria media	Caryophyllaceae	Herb	Annual	-	-	-	Fibrous	0.365
Vicia sativa	Papilionaceae	Herb	Annual/ Biennial	-	+	-	Coarse	0.377
							CD (0.05 %)	0.061

AC: Arbuscules, VC: Vesicle, EH: Extramatrical hyphae.

Table 3. Endomycorrhizal root infection (%) of various weed hosts at different sites at Forest Research Institute, Dehradun.

Host	Howard Road	Tireman Road	Pearson Road I	Pearson Road II	Trevor Road	Mean
Ageratum conyzoides	63.4	71.5	77.0	79.8	71.6	72.7
	(53.1)*	(57.8)	(61.5)	(63.3)	(57.8)	(58.7)
Capsella	49.8	75.0	76.1	76.0	74.8	70.3
bursa-pastoris	(44.8)	(60.1)	(60.9)	(60.7)	(59.9)	(57.3)
Lantana camara	63.8	77.6	76.1	50.8	71.6	68.0
	(54.6)	(62.9)	(62.2)	(45.4)	(58.0)	(56.6)
Malvastrum	88.0	75.6	73.7	68.1	72.9	75.7
coromandelianum	(69.7)	(60.5)	(59.1)	(55.9)	(58.9)	(60.8)
Medicago hispida	83.7	77.4	73.5	72.3	73.8	76.1
	(66.1)	(61.7)	(59.3)	(58.4)	(58.4)	(60.9)
Parthenium	72.9	76.1	81.9	85.6	73.8	78.0
hysterophorus	(58.8)	(61.3)	(64.8)	(67.8)	(59.2)	(62.4)
Reinwardtia indica	83.1	76.4	88.0	78.3	81.3	79.6
	(65.7)	(55.5)	(69.7)	(62.4)	(64.3)	(63.5)
Solanum nigrum	35.7	71.3	75.4	47.9	50.4	56.1
	(28.8)	(57.6)	(60.2)	(43.7)	(45.2)	(47.1)
Stellaria media	00.0	00.0	00.0	00.0	00.0	00.0
	(00.0)	(00.0)	(00.0)	(00.0)	(00.0)	(00.0)
Vicia sativa	72.3	86.5	78.2	71.8	76.1	77.0
	(58.3)	(68.7)	(62.8)	(58.3)	(60.3)	(61.7)
Mean	61.3	67.8	70.0	63.1	64.6	
	(50.00)	(54.6)	(56.1)	(51.6)	(52.4)	
CD (0.05 %)	Host (H)7.9	Site (S) 5.6			Interaction (H x S)17.6	

*Figures in parentheses are original values.

Table 4. Endomycorrhizal spore population (no./50 ml soil) of various weed hosts at different sites at Forest Research Institute, Dehradun.

Host	Howard Road	Tireman Road	Pearson Road I	Pearson Road II	Trevor Road	Mean
Ageratum conyzoides	562.5	366.7	400.0	525.0	662.5	503.3
Capsella bursa-pastoris	238.0	404.2	366.7	195.8	283.3	297.6
Lantana camara	233.3	195.8	279.2	295.9	329.2	266.7
Malvastrum coromandelianum	808.4	545.9	508.4	200.0	579.9	528.5
Medicago hispida	354.2	329.2	379.2	416.7	278.8	351.6
Parthenium hysterophorus	245.8	325.0	550.0	233.4	325.2	335.9
Reinwardtia indica	337.5	245.9	233.4	337.5	254.2	281.7
Solanum nigrum	145.9	208.4	250.0	212.5	387.5	240.9
Stellaria media	362.5	375.0	366.7	279.2	366.7	350.0
Vicia sativa	604.2	354.2	325.0	137.5	366.7	357.5
Mean	389.2	335.0	365.9	283.4	383.4	
	Host(H)		Site (S)		Interaction (H x S)	
CD (0.05 %)	61.5		43.5		137.5	

species, viz. *M. hispida* (0.346 %), *S. media* (0.365 %), *P. hysterophorus* (0.377 %), *V. sativa* (0.377 %) and *C. bursa-pastoris* (0.400 %), had similar phosphorus content in their leaves.

Different weeds supported different mycorrhizal colonization in their root systems. For example, the highest root colonization was observed in *R. indica* (79.6 %), while minimum root infection was found in roots of *S. nigrum* (56.1 %) (Table 3), irrespective of sampling sites. The root colonization was similar among all other weeds. A close look at mycorrhizal status within different sites revealed that weeds at Howard Road had the lowest root colonization (61.3 %), whereas weeds at Pearson Road I had the highest root colonization (70.0 %). No definite pattern of root infection emerged in terms of host × site interactions, although they were statistically significant. With respect to host site interaction, it is interesting to note that the highest root colonization was observed in the roots of *R. indica* at Pearson Road I (88.0 %), whereas *S. nigrum* at Howard Road had the lowest root colonization of 35.7 % (Table 3).

The highest mycorrhizal spore population in soil was noticed in *M. coromandelianum* (528.5/50 ml), which was similar to *A. conyzoides* (503.3) and was lowest for *S. nigrum* (240.9) (Table 4), irrespective of site. *P. hysterophorus* (335.9), *M. hispida* (351.6) and *V. sativa* (357.6) supported spore population second to *M. coromandelianum* and *A. conyzoides*. It is worth mentioning that *S. media,* which otherwise did not support mycorrhizal root colonization, had a spore population similar in size to *Parthenium, Medicago* and *Vicia* in its root zone (350.0). Irrespective of weed species, different sites also supported a spectrum of spore populations sizes. For example, the maximum number of mycorrhizal spores was isolated from Howard Road (389.2), although it was statistically similar to the spore population at Trevor Road (383.4) and Pearson Road I (365.9). The lowest number of spores in soil was observed at Pearson Road II (283.4). Similar to root colonization, no defined pattern of spore populations with regards to host x site interactions could be discerned, although these were statistically significant. The soil at Howard Road had the maximum number of spores in the rhizosphere of *M. coromandelianum* (808.4), while the lowest value was observed from the roots of *V. sativa* (137.5) at Pearson Road II.

Discussion

All weeds, except *S. media*, were found to be mycorrhizal in the survey. Berch *et al.* (1988) also reported that two species of *Stellaria* - viz. *S. samooei* and *S. salycantha* - are non-mycorrhizal. This is further supported by the observation of Brundrett (2009) that the plants of the family Caryophyllaceae, to which *S. media* belongs, are non-mycorrhizal. However, *S. media* supported a good number of spores (350.0/50 ml of soil) in its root zone. Its very shallow roots, coupled with presence of many mycorrhizal weeds (*Medicago* sp.

and *Oxalis* sp.) in its vicinity, may contribute to the large spore count in the soil around the roots of *S. media.* In addition, host/non-host succession under field conditions may add left over resting structures like chlamydospores.

Contrary to the studies of MuthuKumar & Udaiyan (2000), mycorrhizal infections were observed in the root of *L. camara*. Mycorrhizal associations of many weeds - viz. *A. conyzoides, M. coromandelianum, P. hysterophorus* - were also reported earlier by Singh & Verma (1981), Barthakur *et al.* (1989) and Rathi & Singh (1990) in different ecosystems. We observed that *C. bursa-pastoris,* in the family Brassicaceae, is mycorrhizal, with arbuscules and extra-matrical hyphae, and no vesicular structure (Table 2). This is in contrast to the study of Brundrett (2009), who indicated that plants (herbs habitat) in the family Brassicaceae are non-mycorrhizal.

Soil pH may affect the development and functioning of arbuscular mycorrhizae (Hayman & Tavares 1985) by altering the concentration of nutrients and toxic ions in the soil solution, as well as hydrogen ion concentration. The response of AM fungi to soil pH may depend on the species and strains constituting the indigenous AM flora (Robson & Abbott 1989). Van Aarle *et al.* (2002) reported that soil pH of ~6 positively influences AM root colonization as well as the growth and phosphatase activities of extraradical mycelium of two mycorrhizal species (*Scutellospora calospora* and *Glomus intraradices*). In the present study the soil pH of 6.3 at Pearson Road I likely supported the higher root infection (70.0 %; Table 3) observed for all the weeds.

John (1980) re-examined the Baylis hypothesis about root characteristics and mycorrhizal condition of 89 tropical American plants of 23 orders. He found that trees of the order Asterales were heavily mycorrhizal, Malvales were moderate, and Linales were lightly to moderate. In the present study, we found that *R. indica,* in the family Linaceae (with fibrous roots) had 79.6 % mycorrhizal infection, *M. coromandelianum* (Malvales, with coarse roots) had 75.7 % infection, and *A. conyzoides* (Asterales, with coarse roots) had 72.7 % infection (Table 3). This trend is opposite of that described by John (1980), although the differences in root colonization were not statistically significant (Table 3) and moderate in roots of these hosts. It may be due to winter season in tropical north India that supports low microbial activity fading the well defined categorization of root colonization proposed by John (1980).

A significant relationship emanated out of physical expression of symbiosis (in terms of root colonization and soil spore population) and root geometry, on the one hand and physiological expression (in terms of phosphorus content of leaves) on the other. Two weeds, viz. *M. coromandelianum* and *V. sativa*, stand out of the rest in relation to percent root colonization (75.7 and 77.0 %, respectively) and soil population of mycorrhizal spores in their rhizosphere (528.5 and 357.6 spores/50 ml of soil, respectively) (Tables 3 & 4). Interestingly, both have coarse root systems (Table 2), supporting the Baylis hypothesis (1975) that such plants are more dependent on symbiotic fungi and, in turn, more mycotrophic. This relationship is further supported by Hayman's (1982) theory that mycorrhizal dependence of a plant is primarily governed by its rate of growth and phosphorus required for growth of healthy foliage. The more mycotrophic *M. coromandelianum* and *V. sativa* seem to have a higher phosphorus content in their leaves (0.640 and 0.377 %, respectively) (Table 2).

Common weeds anchor the soil and serve as a source of organic matter that feeds the soil food-web. These fast-growing annuals also provide shade, hold moisture, and moderate soil temperatures that allow other plants, such as biennials and perennials (including grasses), to initiate growth. Weeds interact with crops and trees indirectly through their effects on soil microbial communities. One plant species may harbour particular soil microorganisms that either favour or hinder other plants to varying degrees. For example, the majority of crops and some weeds form symbiotic associations with mycorrhizal fungi that greatly benefit the plant by enhancing nutrient and moisture uptake, and protecting against soil-borne diseases. Jordan *et al.* (2000) suggested that mycorrhizae can change the functioning of weed communities so that the net effect of weeds becomes more beneficial to crops. This may happen if weeds promote the growth of mycorrhizae that later colonize the crop. This may explain why some crops grow better following some weed communities. In the present case, weeds seem to maintain the field populations of the endomycorrhiza during the off-season (winter). During the following spring and summer they may serve as reservoir of inoculum of the natural mycorrhizal communities for other plants, influencing companion crops and trees, as well as ecosystem health.

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