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CORRELATION BETWEEN PLANT GROWTH AND ARBUSCULAR MYCORRHIZAL COLONIZATION IN SOME RAINY SEASON GRASSES

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Abstract

Mycorrhizal status and correlation between plant growth and mycorrhizal colonization of 13 rainy season grasses of Punjab, Pakistan viz. *Brachiaria ramosa* (L.) Stapf., *Brachiaria reptans* (L.) Gard. & Hubb., *Dactyloctenium aegyptium* Beauv., *Digitaria timorensis* (Kunth) Balansa, *Echinochloa colonum* (L.) Link, *Eleusine indica* Gaetn., *Eragrostis poaeoides* Beauv., *E. tenella* Roem. & Schult., *Leptochloa chinensis* Nees, *Paspalidium flavidum* (Retz.) A. Camus, *Setaria glauca* Beauv., *Setaria verticillata* Beauv., *Urochloa panicoides* Beauv., were studied. A great variation in different root and shoot growth parameters was recorded in different grasses. All the test grass species were found to be mycorrhizal. However, there was a great variation in degree of mycorrhizal colonization in different grass species. Mycelial, arbuscular and vesicular infections ranged from 21–82%, 0–56% and 4–42% in various test grasses. *D. timorensis, B. ramose, B. reptans* and *E. tenella* were found to be the more densely colonized grasses than rest of the test species. Mycelial and vesicular infections were negatively correlated with different root and shoot growth parameters. However, the correlations between different mycorrhizal and root/shoot growth parameters were insignificant.

Introduction

The grass family is one of the largest and important family of the plant kingdom consisting of about 600 genera and over 10,000 species, widely distributed all over the world. Grasses show great adaptability and are essential component of all type of ecosystems.

The arbuscular mycorrhizal (AM) status of grasses has been studied with great contradictory results. Since grasses have fibrous and highly branched root systems and are relatively efficient in nutrient absorption, hence are generally considered to be only weakly dependent on mycorrhizal symbiosis for nutrient acquisition (Baylis, 1974; Janos, 1980). In contrasts grasses were regarded as heavily mycorrhizal ones by Nicolson (1960), Nicolson & Johnston (1979) and de la Pena *et al*., (2006). Furthermore, Boullard (1963) and Selivanor & Utemove (1968) have reported that most of the forage grasses form mycorrhizae. Clark (2002) reported that mycorrhizal inoculation increased mineral uptake in switchgrass (*Panicum virgatum* L.). Hetrick *et al*., (1988) investigated that cool season C_3 grasses with highly fibrous root system, were only weakly dependent on mycorrhizal symbiosis, while the warm season C_4 grasses with course root system were obligate mycotrophs. In contrast, the results of researches conducted by other workers contradict the generalization that warm season grasses are highly dependent on mycorrhizal symbiosis while cool season grasses are not (Cerlingion *et al.*, 1988; Anderson & Liberta, 1989; Nasim & Zahoor, 1994). According to Javaid *et al.*, (1995), allelopathic grasses are generally lower in mycorrhizal colonization status than non-allelopathic grasses. Present research work aimed to study the mycorrhizal colonization status of some rainy season grasses of Punjab and to investigate if there is any correlation between root/shoot growth and mycorrhizal colonization.

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Materials and Methods

Thirteen rainy season grasses viz., *Brachiaria ramosa*., *Brachiaria reptans*, *Dactyloctenium aegyptium, Digitaria timorensis, Echinochloa colonum, Eleusine indica, Eragrostis poaeoides*, *E. tenella, Leptochloa chinensis*, *Paspalidium flavidum, Setaria glauca*, *Setaria verticillata*, *Urochloa panicoides* were selected for mycorrhizal studies. Seven plants of each grass species, at flowering stage, were randomly collected from different undisturbed localities of the Punjab University, Quaid-e-Azam Campus, Lahore, Pakistan during the rainy months of July and August 2004. Plants were very carefully uprooted and washed under tap water. Roots were separated from shoots. Data regarding shoot length, maximum and total root length, number of roots per plant, and root and shoot dry biomass were recorded. Data were analyzed statistically by applying Duncan's Multiple Range Test (Steel & Torrie, 1980).

A part of fresh root system of each grass species was cut into 1-cm pieces. After careful rinsing with tap water, the root samples were cleared and stained for analysis of colonization by mycorrhizal fungi using a modified Phillips & Hayman (1970) procedure. The roots were cleared for about 30 minutes in 10% KOH solution at 100°C, placed in 10% HCl for 10 minutes for neutralization and then stained with glycerol-trypan blue solution (0.05%) at 100°C for 20 minutes. Twenty root pieces of each sample were studied under compound microscope. Each root piece was observed at five points and data regarding the presence and/or absence of mycorrhizal structures viz., mycelium, arbuscules and vesicles were recorded and the percentage occurrence of these structures was calculated. Number of arbuscules and vesicles per centimeter of root length was also recorded. Finally the correlations between different mycorrhizal structures and plant growth parameters were calculated on computer software Microsoft Excel.

Results and Discussion

Root and shoot growth of grasses: A great variation in different root and shoot growth parameters was recorded in 13 test grass species. Shoot length ranged from 29–87 cm in different grasses. *E. colonum, S. glauca, L. chinensis* and *D. timorensis* were found to be the taller grasses exhibiting a shoot length greater than 70 cm. The highest shoot length was recorded in *E. colonum* that was significantly greater than all other test grasses. The minimum shoot length of 29 cm was recorded in *E. tenella* (Table 1). *E. colonum* also exhibited highest and significantly greater shoot biomass than all other grasses. *S. glauca* was found to be the second highest biomass producing grass. Minimum biomass production was recorded in *E. tenella* followed by *L. chinensis*. Rest of the species exhibited shoot dry biomass ranging from 3.2–6.4 g plant^{-1} (Table 1).

The data regarding various root growth parameters is presented in Table 1. The maximum and significantly greater number of 166 roots per plant was recorded in *E. colonum. E. tenella, L. chinensis and S. verticillata* exhibited less than 30 roots per plant. However, in most of the test species number of roots per plant ranged from 40–60. The maximum root length showed a narrow range of 10–21 cm. However, there was a great variation in total root length in different test grass species. The maximum total root length of 2075 cm was recorded in *E. colonum* followed by 1537 and 1162 cm in *S. glauca* and *D. timorensis*, respectively. *E. tenella, L. chinensis* and *S. verticillata* exhibited very low total root length ranging from 116–199 cm. The rest of the test grasses showed an intermediate total root length ranging between 356 and 622 cm. The highest root fresh and dry biomass was recorded in *E. colonum* that was significantly greater than root biomass of all other test grasses. *E. tenella* and *L. chinensis* exhibited the lowest root biomass that was significantly lower than most of the test species. Most of the test grasses showed a root dry biomass ranging from 0.30 to 0.80 g.

Table 1. Root and shoot growth of different rainy season grasses.

Grass species	Shoot length (cm)	Shoot fresh biomass (g)	Shoot dry biomass (g)	No. of roots/ plant	Max. root length (cm)	Total root length (cm)	Root fresh biomass (g)	Root dry biomass (g)
Brachiaria ramosa	61cd	30 cd	5.2 cd	34 cd	17 b-d	356 d-f	0.53 e-g	0.19 de
Brachiaria reptans	51d-f	17 ef	5.1 cd	55 bc	11 fg	505 de	0.57 e-g	0.32 c-e
Dactyloctenium aegyptium	56 de	14 ef	4.2 c-e	40 b-d	14 c-e	542 d	0.24 fg	0.16 de
Digitaria timorensis	71 bc	17 ef	6.4 bc	43 bc	18 b	1162 c	0.63 e-g	0.34 c-e
Echinochloa colonum	87 a	84 a	18.7 a	166 a	21 a	2075 a	4.90 a	3.15 a
Eleusine indica	41 f	17 ef	3.2 c-e	56 bc	14 c-e	487 de	1.40 cd	0.77 b
Eragrostis poaeoides	43 f	12 e-g	6.2 bc	53 bc	12 fg	465 de	0.80 d-f	0.62 bc
E. tenella	29 g	1 g	0.44 e	25 d	10 h	116 f	0.10 g	0.06 e
Leptochloa chinensis	76 ab	6 fg	1.82 de	20 d	10 gh	121 f	0.15 fg	0.09 e
Paspalidium flavidum	56 de	17 ef	4.2 c-e	54 d	13 b-d	580 d	2.02 b	0.70 bc
Setaria glauca	80 ab	49 b	9.60 b	63 b	14 d-f	1537 b	1.72 bc	0.71 bc
Setaria verticillata	60 cd	34 c	5.5 b-d	24 d	15 b-d	199 ef	1.04 de	0.37 b-d
Urochloa panicoides	47 ef	21 de	4.1 c-e	54 bc	17 bc	622 d	1.16 c-e	0.50 b-d

Values with different letters in a column show significant difference as determined by DMR Test.

Mycorrhizal colonization status of grasses*:* Data regarding the mycorrhizal colonization of the test grasses is presented in Table 2. All the 13 test grass species were found to be colonized by arbuscular mycorrhizal fungi. However, there was a great variation in degree of mycorrhizal colonization in various test species. In contrast to the present study, Muthukumar & Udaiya (2000) studied 53 grass species collected from Western Ghats region, Southern India and reported that only 18 species were colonized with arbuscular mycorrhizal fungi.

Percentage mycelial infection was fairly high ranging from 21–82% in different grasses. *B. ramose, E. tenella, D. timorensis, E. indica, S. glauca* and *U. panicoides* showed above 60% mycelial infection (Table 2). Earlier Nicolson (1960) and Nicolson & Johnston (1979) regarded the grasses as being heavily mycorrhizal ones. Percentage arbuscular infection also showed a very wide range of 0–56% in different grasses. *D. timorensis* exhibited the highest arbuscular infection of 56% with 51 arbuscules cm^{-1} followed by *B. ramosa* and *D. aegyptium*, respectively. *L. chinensis* and *S. glauca* exhibited the lowest arbuscular infection of 3 and 6%, respectively. The rest of the test grasses showed an intermediate arbuscular infection of 11–29%. Arbuscules were absent in *P. flavidum* (Table 2). Absence of arbuscules in other grasses like *Chrysopogon echinulatum, E. frumentacea* and *Oplismenus compositus* has also been reported by Nasim & Zahoor (1994). Highest vesicular infection in terms of percentage infection and number of vesicles per cm of root was recorded in *E. tenella* followed by *D. timorensis* and *U. panicoides.* The percentage vesicular infection and number of vesicles ranged from 23–42% and 8–44 vesicles cm^{-1} in these grasses. In contrast, *E. colonum, E. poaeoides. L. chinensis* and *P. flavidum* exhibited very low vesicular infections of 3 – 6% with 1–10 vesicles cm^{-1}. The rest of the test grasses showed an intermediate vesicular infection of 11–19% with 2–12 vesicles cm^{-1} (Table 2). The results of the present study are in line with the findings of Hetrick *et al*., (1989) who reported that warm season grasses are obligate mycorrhizal symbionts.

Correlation studies: Percentage mycelial and vesicular infections as well as number of vesicles were negatively correlated with all the studied root and shoot growth parameters. Similarly arbuscular infection showed a negative correlation with number of roots and shoot biomass. The rest of the root growth parameters exhibited a positive correlation with arbuscular infection. Number of arbuscules were negatively correlated with shoot biomass while the association between number of arbuscules and various root growth parameters was positive. However, all the correlations between various mycorrhizal and root/shoot growth parameters were insignificant statistically (Table 3). The negative associations of mycelial and vesicular infection with various parameter of root growth indicates that in the presence of fibrous and highly branched root system grasses are less dependant on mycorrhizal association for nutrient acquisition. Earlier Baylis (1974) and Janos (1980) have reported similar findings. These workers suggested that development of dependence on mycorrhizae and development of highly branched root systems are generally regarded as alternatively evolutionary strategies for nutrient acquisition by plants. Corkidi *et al*., (2002) reported that arbuscular mycorrhizal inoculation enhanced length, biomass and relative growth rate of grasses over non-mycorrhizal treatments in two semiarid grasslands in Mexico. However, percent root length colonized by mycorrhizal fungi was not directly related to plant performance. In the present study, the negative correlation between different mycorrhizal parameters and shoot biomass production of the test grasses indicates that plant growth in these grasses may be more dependant on nutrient acquisition by roots rather than mycorrhizal symbiosis. It is suggested that similar studies should be carried out on winter grasses in order to unearth the more hidden factors regarding the role of mycorrhizal fungi in this grass family. Furthermore, studies should also be extended to forage grasses.

Table 2. Mycorrhizal colonization in 13 annual rainy season grasses.

Grass species	% Mycorrhizal colonization			No. of arbuscules /cm	No. of vesicles /cm
	Mycelium	Arbuscules	Vesicles		
Brachiaria ramosa	67 b	52 a	12 e	16 b	2 ef
B. reptans	51 c	44 b	15 de	15 b	5 d-f
Dactyloctenium aegyptium	50 c	19 de	11 e	5 de	3 ef
Digitaria timorensis	80 a	56 a	27 b	51 a	13 b
Echinochloa colonum	43 c	25 cd	4 f	13 bc	1 f
Eleusine indica	68 b	29 c	12 e	13 bc	6 de
Eragrostis poaeoides	43 c	26 c	5 f	6 d	2 ef
E. tenella	82 a	16 ef	42 a	10 c	44 a
Leptochloa chinensis	43 c	3 h	6 f	1 ef	1 f
Paspalidium flavidum	21 d	0 h	5 f	0 f	10 b-d
Setaria glauca	64 b	6 gh	13 e	4 def	8 cd
S. verticillata	48 c	11 fg	19 cd	5 de	12 bc
Urochloa panicoides	68 b	11 fg	23 bc	4 def	8 cd

Values with different letters in a column show significant difference as determined by DMR Test.

Table 3. Correlation between mycorrhizal colonization and different parameters of root and shoot growth of rainy season grasses.

	NR	MRL	TRL	RFB	RDB	SL	SFB	SDB
%MI	-0.25	-0.17	-0.22	-0.24	-0.13	-0.01	-0.35	-0.28
%AI	-0.02	0.01	0.12	0.06	0.34	0.11	-0.13	-0.03
%VI	-0.47	-0.35	-0.41	-0.40	-0.12	-0.24	-0.42	-0.40
NOA	0.15	0.01	0.14	0.05	0.05	0.28	-0.06	-0.01
NOV	-0.51	-0.34	-0.40	-0.31	-0.32	-0.26	-0.27	-0.28

NR=No. of roots per plant, **SDB=**Shoot dry biomass, **MRL=**Maximum root length, **TRL=**Total root length, **RFB=**Root fresh biomass, **MI=**Mycelial infection, **RDB=**Root dry biomass, **AI=**Arbuscular infection, **SL=**Shoot length, **VI=**Vesicular infection, **SL=**Shoot length, **NOA=**Number of arbuscules, **SFB=**Shoot fresh biomass, **NOV=**Number of vesicles
All the correlations were insignificant.

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