Growth Response of Buffel Grass (*Cenchrus ciliaris*) to Phosphorus and Mycorrhizal Inoculation

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Summary

Arbuscular-mycorrhizal symbiosis confers numerous benefits to host plants including improved tolerance to abiotic and biotic stresses. Although the majority of grasses form an arbuscular-mycorrhizal symbiosis, little is known of the mycorrhization of Buffel grass (*Cenchrus ciliaris*). A pot study was conducted in sterilized soil to determine the effect of mycorrhizal inoculation and phosphorus amendment on the biomass production in *C. ciliaris*. Mycorrhizal fungi used were *Gigaspora rosea, Glomus intraradices* and *Glomus etunicatum*. Inoculation with *Gigaspora rosea* alone, and combined incoculation with *Glomus intraradices* + *Gigaspora rosea* and *Glomus intraradices* + *Glomus etunicatum* significantly ($P<0.05$) increased dry biomass in unamended and 25 kg P_2O_5 ha^{-1} treatments. Combined inoculation with *Glomus intraradices* + *Gigaspora rosea* and *Glomus intraradices* + *Glomus etunicatum* showed pronounced ($P<0.05$) effect on dry biomass compared to inoculation with *Gigaspora rosea* alone in unamended and 25 kg P_2O_5 ha^{-1} treatments. Combined inoculation with *Glomus intraradices* + *Glomus etunicatum* resulted in significantly ($P<0.05$) higher dry biomass campared to the combined inoculation with *Glomus intraradices* + *Gigaspora rosea* and inoculation with *Gigaspora rosea* alone in unamended and 25 kg P_2O_5 ha^{-1} treatments. The results clearly show that inoculation of *C. ciliaris* plants with mycorrhizal fungi *Gigaspora rosea, Glomus intraradices* and *Glomus etunicatum* is highly beneficial for the growth and biomass production in the absence or presence of P_2O_5 under sterile soil conditions. Inoculation of *C. ciliaris* plants with these mycorrizal fungi may help in forage production in marginal and shallow soils of the rangelands of Pakistan.

Key words

Cenchrus ciliaris, phosphorus, mycorrhizal inoculation, biomass, forage production

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Introduction

Arbuscular mycorrhizal fungi form a widespread and ecologically important symbiosis with plants in the land ecosystem (Schreiner et al. 2003). Arbuscular mycorrhizal strains occurring under field conditions are still poorly defined and their root colonization and effectiveness in terms of contribution to nutrient acquisition differs markedly between various species, isolates and host plant genotypes (Ahiabor and Hirata 1995, Raja 2006). Colonization of plant roots by arbuscular mycorrhizal fungi greatly increases the uptake of phosphorus and nitrogen by the plants (Chen et al. 2005, Duponnois 2006, George et al. 1995). Many tested fungal isolates increased phosphorus and nitrogen uptake of the plant by absorbing phosphate, ammonium and nitrate from the soil. However, the contribution of arbuscular mycorrhizal fungi to plant phosphorus uptake is usually much larger than the contribution to plant nitrogen uptake. The most prominent contribution of arbuscular mycorrhizal fungi is increased uptake of nutrients by extra radical mycorrhizal hyphae (Ruiz-Lozano 2006).

Arbuscular mycorrhizal fungi occur widely under various environmental conditions and are found in associations of forage crops (Carrenho et al. 2001, Chen et al. 2005, Souchie et al. 2006). The beneficial effect of mycorrhizal fungi on the phosphate nutrition of crop plants in soils low in phosphorus has been reported by different workers (Chen et al. 2005, Duponnois 2006, Toro et al. 1997). Mycorrhizal inoculation has proved better and more beneficial in some unsterile soils than in sterile soils due to the presence of native endophytes and other soil microorganisms (Oehl et al. 2003, Van der Heijden et al. 2003). In addition, plant hormones produced by soil bacteria have been reported to stimulate mycorrhizal formation (Azcón et al. 1978). Pelletier and Dionne (2004) reported that turfgrass inoculated with *Glomus intraradices* at rates between 40 and 60 mL m^{-2} established more quickly than the turfgrass inoculated with *Glomus etunicatum* at the time of seeding, with no irrigation or fertilization inputs.

The rangelands of Pakistan are deficient in phosphorus, bacause soils are calcareous, alkaline, and are dominated by mica mineralogy. Phosphorus deficiency has been observed in 90% of the soils of the Pakistan (Athar 2005). The efficiency of the phosphorus use in rangelands is, therefore, very low and it needs to be increased to a considerable extent for boosting forage production. Use of phosphorus fertilizers to improve phosphorus deficiency of rangeland soils is not feasible due to economical returns. Moreover, fertilizers are unavailable in many developing countries or are beyond the reach of subsistence ranchers. Rangeland productivity may be increased by introducing the nitrogen-fixing legumes and mycorrhizal plants to the rangelands.

Buffel grass (*Cenchrus ciliaris* L.) is highly nutritious and is considered excellent for pasture in hot, dry areas and is valued for its production of palatable forage and intermittent grazing during drought periods in the tropics (Quraishi et al. 1993). Buffel grass thrives from sea level to 2000 m, in dry sandy regions, with rainfall 250-750 mm annually (but tolerates much higher rainfall), on shallow soils of marginal fertility. Such characteristics extend its production range and increase its value for pasturage. Yield of some strains makes it good for forage during the wet season also. Buffel grass contains 11.0% proteins, 2.6% fat, 73.2% total carbohydrate, 31.9% fiber and 13.2% ash on a zero-moisture basis (Gohl 1981). The grass, fed green, turned into silage or made into hay, is said to increase flow of milk in cattle and impart a sleek and glossy appearance. The present study was conducted to determine the effect of mycorrhizal inoculation and phosphorus amendment on biomass production in *C. ciliaris* plants grown under sterile soil conditions.

Materials and nethods

A pot experiment was conducted in sterilized soil at University of Arid Agriculture, Rawalpindi, Pakistan to study the effect of mycorrhizal inoculation and P_2O_5 amendment on biomass production in *Cenchrus ciliaris*. Mycorrhizal fungi tested were supplied by the Department of Biological Sciences, Quaid-e-Azam University, Islamabad and included *Gigaspora rosea, Glomus intraradices*, and *Glomus etunicatum*. Seeds of *C. ciliaris* were obtained from the National Agricultural Research Center, Islamabad. Autoclaved garden soil weighing about 8 kg was used in 16 cm diameter earthen pots. Half of the soil was ameliorated with 25 kg P_2O_5 ha^{-1}. Chemical characteristics of soil are provided in Table 1.

Table 1. Chemical characteristics of soil used in the experiments

pH	7.4
Moisture	32%
Total organic carbon	0.6%
Total nitrogen	16 mg kg^{-1}
Phosphorus	5.3 mg kg^{-1}
Potassium	140 mg kg^{-1}

Earthen pots were filled with phosphorus-amended and unamened soil. Twenty seeds were sown in each pot, that later maintained four plants. *Gigaspora rosea, Glomus intraradices* + *Gigaspora rosea* and *Glomus intraradices* + *Glomus etunicatum* were used for inoculation. Inoculation with mycorrhizal fungi was done by layering method (Menge and Timmer 1982). Non-inoculated plants served as control. There were three inoculated and

Table 2. Effect of mycorrhizal inoculation on dry weight of the roots and the shoots of *Cenchrus ciliaris* grown in phosphorus amended and unamended sterile soil

Treatment	Root Weight (g plant^{-1})		Shoot Weight (g plant^{-1})	
	0 kg P_2O_5 ha^{-1}	25 kg P_2O_5 ha^{-1}	0 kg P_2O_5 ha^{-1}	25 kg P_2O_5 ha^{-1}
Control	4.08 h	4.64 f	8.10 h	9.20 f
Gigaspora rosea	4.31 g	5.46 d	8.82 g	11.17 d
Glomus intraradices + Gigaspora rosea	4.95 e	6.32 b	9.90 e	12.40 b
Glomus intraradices + Glomus etunicatum	5.71 c	6.72 a	11.42 c	13.44 a

Means in each column differ significantly at P< 0.05 for roots and shoots dry weight.

a non-inoculated treatment with and without phosphorus amendment. Each treatment was replicated three times and pots were arranged in completely randomized design. Pots were kept in open air under natural field conditions and were watered daily with tap water. Plants were harvested after 10 weeks just at seed formation. The extractable phosphorus was estimated by the method of Olsen and Sommers (1982). Extractable potassium was estimated by using method of Richards (1954). The data were subjected to statistical analysis comparing means by LSD multiple mean comparison test.

Results and discussion

The results on dry biomass production in *C. ciliaris* in response to mycorrhizal inoculation and phosphorus amendment are presented in Table 2. The shoot dry weight of plants in non-inoculated and 25 kg P_2O_5 ha^{-1} treatment was significantly (P<0.05) higher than the plants in non-inoculated and unamended treatment (Table 2). Inoculation with *Gigaspora rosea* alone, *Glomus intraradices + Gigaspora rosea*, and *Glomus intraradices + Glomus etunicatum* significantly (P<0.05) increased the shoot dry weight in unamended and 25 kg P_2O_5 ha^{-1} treatments (Table 2). Combined inoculation with *Glomus intraradices + Gigaspora rosea* and *Glomus intraradices + Glomus etunicatum* showed pronounced (P<0.05) effect on shoot dry weight compared to inoculation with *Gigaspora rosea* alone in unamended and 25 kg P_2O_5 ha^{-1} treatments (Table 2). Combined inoculation with *Glomus intraradices + Glomus etunicatum* resulted in a significantly (P<0.05) higher shoot dry weight campared to the combined inoculation with *Glomus intraradices + Gigaspora rosea* and inoculation with *Gigaspora rosea* alone in unamended and 25 kg P_2O_5 ha^{-1} treatments (Table 2). This may be because of the beneficial response from mycorrhizal inoculation at moderate fertility. These results corroborate previous studies which report that the plants benefit from mycorrhizal inculation alone and in the presence of additional phosphorus at various rates (Duponnois et al. 2005, Oehl et al. 2003, Toro et al. 1997). Duponnois et al. (2005) reported that *Glomus intraradices* inoculation and rock phosphate amendment influenced plant growth and microbial activity in the rhizosphere of *Acacia holosericea* by increasing plant height and dry matter production of root and shoot.

The root dry weight was significantly (P<0.05) higher in non-inoculated treatment at 25 kg P_2O_5 ha^{-1} than in non-inoculated and unamended treatment (Table 2). Root dry weight was significantly (P<0.05) higher due to combined inoculation. Root weight increased significantly (P<0.05) in plants inoculated with *Gigaspora rosea* alone, *Glomus intraradices + Gigaspora rosea*, and *Glomus intraradices + Glomus etunicatum*, both in unamended and 25 kg P_2O_5 ha^{-1} treatments (Table 2). Combined inoculation with *Glomus intraradices + Gigaspora rosea* and *Glomus intraradices + Glomus etunicatum* showed significantly (P<0.05) increased effect on root dry weight compared to inoculation with *Gigaspora rosea* alone in unamended and 25 kg P_2O_5 ha^{-1} treatments (Table 2). Combined inoculation with *Glomus intraradices + Glomus etunicatum* also resulted in significantly (P<0.05) higher root dry weight compared to the combined inoculation with *Glomus intraradices + Gigaspora rosea* and inoculation with *Gigaspora rosea* alone in unamended and 25 kg P_2O_5 ha^{-1} treatments (Table 2). This is in conformity with the previous results which showed that mycorrhizal inoculation significantly increased the weight of root and shoot of plants both in absence and presence of different levels of phosphorus (Ahiabor and Hirata 1995, Bolan 1991, Raja 2006, George et al. 1995, Pelletier et al. 2004). Fay et al. (1996) investigated the effects of arbuscular mycorrhizal infection by *Glomus mosseae* on growth and photosynthesis of barley *(Hordeum vulgare* L. cv. Manitou) in sand culture at five levels of calcium phosphate. Mycorrhizal infection was low and varied with phosphorus supply. It was at the lowest phosphorus supply that vesicular arbuscular mycorrhizal plants had higher rates of photosynthesis and greater phosphorus and nitrogen efficiency.

Phosphorus is an essential element for plant nutrition and it can be only assimilated as soluble phosphate. Plants inoculated with arbuscular mycorrhizal fungi utilize more soluble phosphorus from soil mineral phosphate than non-inoculated plants (Duponnoi et al. 2005). It has been reported that arbuscular mycorrhizal fungi

Gigaspora rosea and *Glomus etunicatum* + *Glomus* ·*intraradices* affect plant growth, plant-available phosphate and soil microbial activity with and without added rock phosphate (Bolan 1991, Chen et al. 2005, Duponnoi et al. 2005, Souchie et al. 2006, Toro et al. 1997). Mycorrhizal inoculation improves the absoption of phosphorus and other nutrients by plants increasing the contact surface and explored soil volume (Clark and Zeto 2000) and possibly facilitating nutrient transport among plants (Chen et al. 2005). Van der Heijden (2003) reported that the composition of mycorrhizal communities determines how plant species coexist and to which plant species nutrients are allocated, affecting the plant succession. The reduction of mycorrhizal infection under field conditions can decrease plant diversity affecting the secondary succession process (Fischer et al. 1994). Therefore, the mycorrhizal communities in the areas with rapid succession, as grass pasture and secondary forest need to be explored. Generally grass pasture presents greater response to mycorrhizal inoculation than the secondary forests.

Our results clearly show that inoculation of mycorrhizal fungi *Gigaspora rosea, Glomus intraradices* and *Glomus etunicatum* is highly beneficial for the growth and dry biomass production in *C. ciliaris* plants in the absence or presence of P_2O_5 under sterile soil conditions. Additional research is needed under field conditions to elucidate the response of these mycorrhizal fungi before they are included in forage production in the rangelands of Pakistan with shallow soils of marginal fertility.

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