

Original Research Article

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Effect of *Glomus mosseae* Inoculation on the Growth of *Dalbergia sissoo* Seedlings with Normal, Stress and Re-watering Conditions

Amit Kumar¹, R. S. Beniwal^{1*}, M. K. Singh¹, Sandeep Arya¹ and Rakesh Kumar Chugh²

¹Department of Forestry, ²Department of Plant Pathology, CCS Haryana Agricultural University, Hisar, India

*Corresponding author

ABSTRACT

Keywords

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Pots experiment was conducted in screen house condition of Forestry Department, CCSHAU, Hisar. In this experiment *Dalbergia sissoo* seeds were sown in both the conditions of soils *i.e.* inoculated and un-inoculated with *Glomus mosseae* in different water treatments *i.e.*, normal, drought and re-watering conditions to evaluate the performance of growth characters (collar diameter, shoot and root length, fresh and dry weight, no. of leaves and leaf area index) of seedlings. It was observed that seedlings which were grown in the pots containing AM fungi performed significantly better in terms of growth parameters than the seedlings which were grown in the pots with un-inoculated soils in all water treatments under studied.

Introduction

Dalbergia sissoo Roxb. commonly known as Shisham belong to the family Fabaceae, is a multipurpose tree species grown well in India, Nepal and Pakistan. It grows gregariously in alluvial soil and on freshly exposed soils along with roads, streams and for rehabilitation of degraded lands. Forest plantation sites in India are usually located on poor soils in terms of water holding capacity, fertility or drainage and also many locations of our tropical region experience frequent drought of varying intensity and duration due

to erratic or scarce amount of rainfall; thus maintaining sustainable forest productivity is a major problem of prime concern. Drought is one of the most important abiotic stress factors limiting plant growth and productivity (Polle and Rennenberg, 2006). During water stress, soil water is more strongly retained and solute transfer to plants is less efficient and may not meet nutrient demands of the plants. Arbuscular mycorrhizal fungi are obligate biotrophs that forms symbiotic or mutualistic association with roots of 80 percent plants species (Cekic *et al.*, 2012). The interaction between mycorrhizal fungi and host plants

under different conditions has received greater attention because of their mutual association (Miransari, 2009) especially when they are subjected to stresses. Without their fungal symbionts, most trees cannot acquire enough soil resources to grow or reproduce; however, it is also true that without the trees, the fungi have insufficient energy to carry out their life cycle. Because of this obligatory exchange, mycorrhizal fungi are considered the primary vectors for plant carbon to soils (Talbot *et al.*, 2008) and, conversely, the primary vectors of soil nutrients to plants (Hobbie and Hobbie, 2006). Application of mineral solubilizers to the seedlings helps in the early establishment of the seedlings in problematic soils (Gupta *et al.*, 2007). The process of inoculating microbes to the soil in a forest nursery could be an effective method to achieve higher growth (Beniwal and Toky, 1993) and establishment of tree species on water deficient sites. Mycorrhizal association at the nursery stage has been reported to enhance the productivity of forest trees like *Populus x Canescens* (Beniwal *et al.*, 2010). Inoculation of plant roots with mycorrhizal fungi may be effective in improving growth under water stress conditions.

Materials and Methods

To conduct the experiment *Dalbergia sissoo* seedlings were maintained in the screen house of Department of Forestry, CCSHAU, Hisar situated at 29°10'N latitude and 75°46'E longitude at an elevation of 215.2m above mean sea level.

The soil was taken from Balsamand Research Farm, CCSHAU, Hisar and mixed well of well rotted farm yard manure. Soil mixture was autoclaved at 121.4°C for 1 hour for two consequent days. In non-AM inoculated treatments, *Dalbergia sissoo* seeds were sown in pots containing of untreated sterilized soil, whereas, in AM-inoculated treatments, seeds

were sown in pots containing of untreated sterilized soil with mixing of 50g/kg of sterilized soil of *Glomus mosseae* inoculums in upper 7-10 cm pot soil.

Uniform seeds of *Dalbergia sissoo* were collected from plus tree and six seeds were sown in pots; however, after germination three seedlings were maintained in each pot. Each pot received normal watering upto 90 days of growth. For the whole cycle of water stress period, normal watering (NW) plant were supplied with tap water on alternate days in the morning and rest of the seedlings were maintained without water application to observe the water stress effects on Shisham seedlings. Seedlings were subjected to drought for 25 days by withholding the water till the drooping of leaves and the plants which were maintained to determine the after effects of drought were harvested. Growth parameters under studied were recorded after each stage of water stress.

Results and Discussion

Water stress caused decreases in collar diameter, shoot and root length compared to well water plants (Table 1). These decreases were, however, less in AM-inoculated plants. A significant increment in radial growth was observed among the AM-inoculated plants compared to non-inoculated plants and it was found that the collar diameter of AM fungal inoculated plants were significantly higher than those of their respective non-inoculated counterparts at three stages of watering conditions *i.e.* Normal watering (NW), Drought (D), Re-watering (RW). Similarly, shoot and root lengths of AM-inoculated plants were also significantly higher than non-AM plants. Water deficit has profound effects on plant growth, even plants with an optimum water supply experience transient water-shortage periods, where water absorption cannot compensate for water loss by

transpiration. Arbuscular mycorrhizal symbiosis has been shown to increase plant tolerance to water deficit, although the exact mechanisms involved are still a matter of debate (Ruiz-lozano, 2003). The effects of *Glomus mosseae* on plant water status have been associated with enhanced host nutrition, especially phosphorus (P) nutrition (Almagrabi and Abdelmoneim, 2012).

In this experiment, *Dalbergia sissoo* seedlings were subjected to water stress for 25 days by withholding the water till the drooping of leaves. AM fungi inoculated seedlings had significantly more increased in whole plant fresh and dry weights as compared to the non-AM plants during the period of water stress (Table 2). Mean fresh weight and dry weight of plant were recorded significantly higher in AM-plants as compared with non-AM plants under normal watering, water stress and re-watering conditions. Stressed AM-inoculated seedlings could maintain a significantly higher dry weight than non AM-plants may be due to extra radical fungus mycelia, which extend the root surface area and improved the uptake of water and nutrients by the roots (Dickie, 2000). Singh and Chugh, (2019) found that seedlings of plants *i.e.*, *P.cineraia*, *D. sissoo*, *E. tereticornis*, *A. indica* and *A. excelsa* grown in pots inoculated with mycorrhizae fungi performed significantly better than uninoculated soils. Further, Kaushik *et al.*, (2003) reported that VAM inoculation increased N, P and K concentration in roots and shoot in *A. nilotica* and *D. sissoo*.

Water stress also led to significant decrease in number of leaves and specific leaf area under both AM-inoculated and non-inoculated plants (Table 3). This loss was mainly caused to the seedlings as a result of wilting and leaf shedding, but wilting and leaf shedding was comparatively more in control stressed plants. Number of leaves and leaf area was found

higher, in AM-plants compared to their non-AM counterparts. During recovery from water stress, all the stressed re-watered seedlings recovered well their drooped leaves; however, the non-AM plants could not recover their drooped leaves and remained wilted till the harvest of the seedlings. Water stress caused a significant reduction in the AM abundance in the roots because of shrinking and loss of the hyphal mantle compared to well-irrigated plants. Reduction in mycorrhizal colonization owing to water stress conditions (Beniwal *et al.*, 2010) in poplar. Kaushik *et al.*, (2000) found that *Glomus mosseae* inoculation on root pathogens in *Acacia nilotica* and *Dalbergia sissoo* seedlings decreased the disease intensity and increased the N, P, K content in roots and shoots. The mycorrhizal percentage of re-watered plants was similar to that of well-watered individuals. Drought had promoted leaf shedding in *Dalbergia sissoo*, in all the stressed conditions of mycorrhizal and non-mycorrhizal seedlings. But leaf shedding was comparatively more in non-AM water stressed seedlings. Increased leaf shedding in moderately and severely stressed poplar in response to drought acclimation (Beniwal *et al.*, 2010).

The periodic increment in collar diameter and shoot length from initiation of drought to drought harvest stage (25 days) and drought harvest stage to re-watering harvest stage (7 days) was also determined. In the present study, inoculation of shisham seedlings with AM-fungi resulted in the significantly more periodic increment in root collar diameter and shoot length (Table 4). It has been shown that mycorrhizal plants absorb water more efficiently under water deficit environment (Khalvati *et al.*, 2005) that might be due to modification in root architecture which results in better root growth due to numerous branched roots (Berta *et al.*, 2005). The findings of this work showed that the symbiotic effectiveness of *Glomus mosseae*

enhanced plant total fresh and dry weight, number of leaves and leaf area in plants of *Dalbergia sissoo* subjected to water stress and re-watering conditions, which may be because mycorrhizal colonization caused a proportionally greater allocation of

carbohydrates to the shoots and root tissues, indicating that AM-plants protected themselves against drought successfully than non AM-plants. Similar benefits of mycorrhization (Jesus *et al.*, 2004) in *Rosmarinus officinalis* also observed.

Table.1 Effect of water stress on the collar diameter, shoot and root length of AM-inoculated *Dalbergia sissoo* seedlings maintained at three watering regimes

Treatment		Collar diameter (mm)				Shoot length (cm)				Root length (cm)			
		NW	D	RW	Mean	NW	D	RW	Mean	NW	D	RW	Mean
Inoculated		4.81	4.25	4.36	4.47	51.16	43.47	46.13	46.92	28.62	24.28	25.95	26.28
Non-Inoculated		4.23	3.53	3.60	3.78	40.62	28.64	30.28	33.18	22.83	17.77	19.12	19.90
	Mean	4.52	3.89	3.98		45.89	36.05	38.81		25.72	21.02	22.53	
CD at 5%	T	0.145				1.35				0.92			
	W	0.177				1.65				1.13			
	T x W	NS				2.34				NS			

Values presented are means. Treatments: NW= Normal watering, D= Drought, RW= Re-watering, T= Treatment of soil, W= Watering conditions, NS= Non significant.

Table.2 Effect of water stress on the whole plant fresh and dry weight of AM-inoculated *Dalbergia sissoo* seedlings maintained at three watering regimes

Treatment		Fresh weight of plant (g)				Dry weight of plant (g)			
		NW	D	RW	Mean	NW	D	RW	Mean
Inoculated		17.25	12.12	13.85	14.40	11.18	7.99	9.11	9.42
Non-Inoculated		12.79	6.59	8.12	9.56	7.68	4.19	6.11	5.60
	Mean	15.02	9.35	10.98		9.43	6.09	7.02	
CD at 5%	T	0.41				0.246			
	W	0.05				0.468			
	T x W	0.71				0.945			

Values presented are means. Treatments: NW= Normal watering, D= Drought, RW= Re-watering, T= Treatment of soil, W= Watering conditions.

Table.3 Effect of water stress on the number of leaves and leaf area of AM-inoculated *Dalbergia sissoo* seedlings maintained at three watering regimes

Treatment		No. of leaves				Leaf area (cm ²)			
		NW	D	RW	Mean	NW	D	RW	Mean
Inoculated		48.20	30.21	32.05	36.81	35.69	25.44	26.19	29.10
Non-Inoculated		36.67	15.73	16.57	22.99	24.54	19.79	20.60	22.31
	Mean	42.43	22.97	24.31		31.12	22.61	23.39	
CD at 5%	T	0.47				0.65			
	W	0.59				0.80			
	T x W	0.73				1.13			

Values presented are means. Treatments: NW= Normal watering, D= Drought, RW= Re-watering, T= Treatment of soil, W= Watering conditions.

Table.4 Effect of water stress on the periodic increment in collar diameter and shoot length of AM-inoculated *Dalbergia sissoo* seedlings

Treatment		Collar diameter (mm)			Shoot length (cm)		
		NW-D (25 d)	D-RW (7 d)	Mean	RW-D (25 d)	D-RW (7 d)	Mean
Inoculated		0.68	0.21	0.45	8.54	2.32	5.43
Non-Inoculated		0.31	0.14	0.23	4.34	1.78	3.06
	Mean	0.50	0.16		6.44	2.05	
CD at 5%	T	0.012			0.99		
	W	0.015			0.13		
	T x W	NS			1.12		

Values presented are means. Treatments: NW= Normal watering, D= Drought, RW= Re-watering, T= Treatment of soil, W= Watering conditions, NS= Non significant, d= days

The AM contribution to plant drought tolerance might also have occurred through drought avoidance mechanisms such as hyphal water uptake (Marulanda *et al.*, 2003) or increased water uptake related to mycorrhizal changes in root morphology or soil structure.

It may be concluded that AM-inoculation substantially protected plant growth performance compared with non-AM shisham. Upon microscopically looking the AM-inoculated shisham root segments, it was found that *Glomus mosseae* penetrates the cortical cells of feeder roots intra-cellularly and make large vesicles and arbuscules in root cells and thus, helped the shisham plants in increasing the effective absorption root surface area, which cause the flow of moisture and nutrient to plants that ultimately increase the plant growth by increasing relative water content. It is suggested that using the arbuscular mycorrhizal fungi in water stressed *Dalbergia sissoo* seedlings, we can achieve the increased growth in AM shisham plants and thus the adverse effect of water stress on them can be minimized.

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