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RESISTANCE AGAINST ROOT ROT AND STEM DECAY OF *AMARANTHUSHYBRIDUS* BY A DUAL TREATMENT WITH *GLOMUSFASCICULATUM* AND *TRICHODERMAMAHARZIANUM* VIS-À-VIS PRODUCTION

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ABSTRACT

Six accessions of *Amaranthus* sp. procured from National Bureau of Plant Genetic Resources (NBPGR), New Delhi. Among the six accessions, IC 95609 was highly susceptible to root rot and stem decay disease which causes loss in productivity. Root rot and stem decay disease caused by *Fusariumsolani* which affects different agro-morphological traits of the crop plant. *Amaranthus* is a leaf vegetable. In the present investigation, VAM fungus (*Glomusfasciculatum*) and antagonistic fungus (*Trichodermaharzianum*) combination used to improve the production and quality of different vegetative characters of the leafy vegetable. Dual inoculation of VAM and *Trichodermaharzianum* can enhance the productivity of the leafy vegetable and leads to sustainable agricultural practice.

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INTRODUCTION

Amaranthus spp. include 75 wild and weedy species in tropical and temperate region of the world (Sauer, 1993). Compared to other vegetable crop, this crop is rich in nutrition, minerals and amino acids with high level of sulphur (Prota, 2004). *Amaranthus* sp. is a herbaceous plant with erect stem and large inflorescence. *Amaranthus* sp. production has been severely reduced by several fungal pathogens. The most affected fungal diseases such as damping off, stem canker, wet rot (Prota 2004, Awurum and Uchegbu, 2013). Most of the people use different chemical fungicides to control disease of *Amaranthus* and these causes chemical hazard to the environment. Vesicular- arbuscular mycorrhizal fungi are recognized as biofertilizer because it provides advantages by increasing nutrients and water uptake to the host plant (Bohra, 2007). VAM fungi enhance plant growth and dry weight (Habibzadeh, 2015).

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Trichoderma spp. can stimulate mycorrhizae formation (Davey, 1971) and also growth of mycelium of *Glomus* sp. (Calvet et al., 1988). In the present study, it was aimed to increase productivity and disease resistance of the *Amaranthus* variety which is severely affected by root rot and stem decay disease.

MATERIALS AND METHODS

Six genotypes viz. IC 95609, IC 35482, IC 120617, IC 35626, IC 95595, and IC 95589 of *Amaranthus* sp. were procured from National Bureau of Plant Genetic Resources (NBPGR), New Delhi and were grown in Crop Research Farm (CRF), The University of Burdwan. Mature flowering twigs were preserved and dried for herbarium. Then the herbaria of the accessions were sent to the Botanical Survey of India (BSI), Shibpur, Howrah, for their proper identification. This identification was confirmed by BSI Shibpur, Howrah and all these six accessions were identified as *Amaranthushybridus* L. (Vide letter No. CNH/72/2013/Tech. II/82). Extensive field survey was conducted in the field of Crop Research Farm (CRF), The University of Burdwan for two consecutive years (2011 and 2012).

Among six accessions, only the accession IC 95609 was most susceptible to root rot and stem decay disease which is devastating in nature. The other accessions cropped in the field were noticed to be affected which were very negligible and not causing any significant damage to the crop such as leaf spot, leaf curl, leaf hole etc.

Isolation and identification of pathogen

In order to isolate the pathogen, diseased *Amaranthus* stem was collected from the experimental plot. Pathogen was isolated from diseased plant material. A white coloured fungal colony was developed in culture which was then sub-cultured in fresh PDA medium. The pathogenecity test was confirmed by Koch's postulates. The isolated fungus was studied in a phase contrast microscope. Then the isolate was sent to IARI for its proper identification which was later identified as *Fusarium solani* (Accession No. 9335.14). Root rot and stem decay disease causes damage to the production and quality of the particular variety IC 95609. To overcome this problem, different combinations of *Glomus fasciculatum* and *Trichoderma harzianum* were applied to the variety following the method of Ojha (2008).

Preparation of inoculums of VAM (*Glomus fasciculatum*) and seed bed to grow seedling of the accession-IC 95609

Soil culture of *Glomus fasciculatum* was collected from Bidhan Chandra Krishi Viswavidyalaya (BCKV), West Bengal. This VAM fungi was propagated with sterile soil and sand (pH 6.2; P45 ppm and organic carbon 0.6%) in 1:1 ratio in the root of *Zea mays* which is the suitable host. A thin layer of inoculums was placed 4 cm below the grains in the pot soil in which maize grains were sown. The rhizosphere soil was used as inoculums for the preparation of seed bed of Amaranth variety (IC 95609). These VAM containing soil apply to seedbed just before transplantation. Inoculation was conducted @ 1 kg soil inoculums of VAM with 5kg sterilized soil. On the other side only sterilized soil served as control. Seed of the accession were germinated in germination chamber and scattered on seedbeds. After 10 days, the seedling (both VAM infected and non VAM infected) were transplanted.

Preparation of mycelia mats and spore suspension

50 ml of PDA broth (pH 7.4) was autoclaved and prepared sterilized. Then test pathogen *F. solani* and *Trichoderma harzianum* were inoculated separately and incubated at 28±2°C for 15 days. After incubation, the mycelia mats and spores produced from culture were used to spore suspension.

Transplantation of seedlings and application of pathogen and antagonist

Transplantation was done in 5 sets including control set. To show interaction among *Glomus fasciculatum*, *Trichoderma harzianum* and *Fusarium solani*, five combinations (*F. solani*, *Glomus fasciculatum*+ *F. solani*, *Trichoderma harzianum* + *F. solani*, *F. solani* + *Glomus fasciculatum*+ *Trichoderma harzianum* and control set) were subjected for field trial performance to find out interaction between the microorganisms and their effects on host plants. Control set was transplanted with non infected seedling from non inoculated seedbed and served as control. Likewise, only *Fusarium solani* treated set was transplanted with non infected seedlings from non VAM treated but spore suspension of *F. solani* was added. Only VAM treated set, plants were raised with seedling from VAM treated seedbed. In, VAM and *Fusarium solani* treated set, VAM infected seedlings were transferred to non-sterile pots with added *F. solani* spore suspension. *Trichoderma harzianum* and *F. solani* treated set was transplanted with non infected seedlings from non VAM treated but spore suspension of *F. solani* and *Trichoderma harzianum* were added. The remaining set that contains *Trichoderma harzianum*, VAM and *F. solani*. Spore suspension and mycelial mats of *Trichoderma harzianum* were directly applied to another sets of VAM+ *Fusarium* treated pots by using previously sterilized wheat bran as substrate so as to get VAM+*Fusarium*+*Trichoderma harzianum* treated pot.

RESULTS AND DISCUSSION

Interaction effects of *Glomus fasciculatum*, *Trichoderma harzianum* and *Fusarium solani* was studied on the basis of productivity and resistance of host plant,

Table 1. Effect of interaction among *Glomus fasciculatum*, *Trichoderma harzianum* and *Fusarium solani*

Treatment	Days	Plant height (cm)	Leaves/ plant (number)	Leaf length (cm)	Leaf width (cm)	Basal diam. (cm)
<i>Trichoderma harzianum</i> + <i>Fusarium solani</i>	15	9.2±0.12	7.4±0.09	1.84±0.05	0.87±0.05	0.28±0.02
	30	15.4±0.09	10.2±0.11	2.11±0.08	1.16±0.09	0.37±0.01
	45	20.3±0.11	15.3±0.19	2.82±0.12	1.31±0.15	0.41±0.05
<i>Glomus fasciculatum</i> + <i>Fusarium solani</i>	15	9.2±0.12	8.1±0.22	1.98±0.06	1.16±0.11	0.32±0.07
	30	15.4±0.09	12.5±0.13	2.54±0.05	1.45±0.10	0.41±0.05
	45	20.3±0.11	15.4±0.25	3.27±0.11	1.72±0.20	0.50±0.09
<i>Glomus fasciculatum</i> + <i>Trichoderma harzianum</i> + <i>Fusarium solani</i>	15	9.5±0.15	20.1±0.34	4.10±0.15	2.13±0.06	0.72±0.03
	30	16.8±0.13	24.2±0.23	4.93±0.13	2.71±0.11	0.94±0.06
	45	24.6±0.11	31.7±0.45	6.12±0.18	3.56±0.15	1.10±0.12
<i>Fusarium solani</i>	15	28.4±0.14	4.3±0.13	1.12±0.11	0.54±0.13	0.14±0.01
	30	39.1±0.08	6.1±0.10	1.75±0.19	0.84±0.05	0.21±0.05
	45	58.6±0.23	10.4±0.20	2.20±0.14	1.16±0.06	0.32±0.08
Control	15	7.3±0.07	11.2±0.11	2.32±0.13	1.46±0.07	0.39±0.01
	30	12.4±0.06	16.7±0.13	2.98±0.16	1.83±0.05	0.45±0.04
	45	18.6±0.09	20.9±0.21	4.25±0.25	2.10±0.11	0.53±0.09

Amaranthushybridus. Effect of VAM and *Trichodermaharzianum* on disease plant. In case of only pathogen (*F.solani*) treated plants showed less productivity and less vigorous. When plants were treated with pathogen and VAM, it results better production than pathogen treated plants, and less productive than control set. Dual treatment of VAM and *Trichodermaharzianum* also results less productive than control set but better than *Fusariumsolani* treated plants. But combined inoculation of VAM and *Trichodermaharzianum* in *Fusariumsolani* treated plants exhibit effective resistance to plants and results high productivity among all treatments. All the phenotypic characters i.e. Plant height (cm), Leaves/ plant (number), Leaf length (cm), Leaf width (cm), and basal diameter showed more vigor and more herbage due to synergistic action of interactive microorganisms i.e. *Glomusfasciculatum*, *Trichodermaharzianum* and *Fusariumsolani*.

It is clear from the result (Table 1) that *Glomusfasciculatum* shows positive interaction in the root of *Amaranthus* sp. which is in line with the reports of the workers (Li *et al.*, 2007; Kakati and Mahanta, 2013; Srinivasa Murthy and Narayanappa, 2015). In the present investigation there are four treatments viz. *Fusariumsolani*, *Fusariumsolani* + *Glomusfasciculatum*, *Fusariumsolani* + *Trichodermaharzianum* and *Fusariumsolani* + *Glomusfasciculatum* + *Trichodermaharzianum* were undertaken. Single treatments of *Glomusfasciculatum* and *Trichodermaharzianum* in *Fusarium* inoculated plant exhibit less efficacy regarding different selected agro-morphological characters viz plant height, leaves per plant, leaf length, leaf width and basal diameter. But dual inoculation of *Glomusfasciculatum* and *Trichodermaharzianum* in diseased plant show better response in productivity parameters due to synergistic action of *Glomusfasciculatum* and *Trichodermaharzianum*. Previously, it was reported that *Sclerotiumrolfsii* was effectively reduced by combined inoculation of *Glomusfasciculatum* and *Trichodermaasperelloides* in ground nut plants (Doley *et al.*, 2014; Singha *et al.*, 2014). In the present study reveals that *Fusarium* treated plant become less vigorous and lost its productivity. Single inoculation of *Glomusfasciculatum* and *Trichodermaharzianum* in *F.solani* treated plant result less productive than control set but productive than only *Fusarium* treated plant. There is no doubt that dual inoculation shows best response in improvement vigor and productivity of the vegetable. Being a leafy vegetable, *Amaranthus* sp need to improve vegetative characters. *Fusarium* affected plant can also be made productive by combined inoculation of *Glomusfasciculatum* and *Trichodermaharzianum*. Root rot and stem decay disease of *Amaranthus* sp is a serious threat towards its production and economy. *Glomusfasciculatum* and *Trichodermaharzianum* treatment can increase productivity by improving different agro-morphological traits.

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