Effect of value added compost on suppression of wilt disease of Brinjal (*Solanum melongena*) caused by *Fusarium* oxysporum f. sp. melongenae

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INTRODUCTION

Compost is the product resulting from the controlled biological decomposition of organic material that has been sanitized through the generation of heat and stabilized to the point that it is beneficial to plant growth. Compost bears little physical resemblance to the raw material from which it originated. Compost is an organic matter resource that has the unique ability to improve the chemical, physical, and biological characteristics of soils or growing media. It contains plant nutrients but is typically not characterized as a fertilizer. Compost products contain a considerable variety of macro and micronutrients. Although often seen as a good source of nitrogen, phosphorous, and potassium, compost also contains micronutrients essential for plant growth. Since compost contains relatively stable sources of organic matter, these nutrients are supplied in a slow-release form. Therefore to improve the quality and nutrient contents, the enrichment of compost is essential. One of the best ways of increase the nutrient contents of the final compost is microbial enrichment technique with plant growth promoting bacteria. The addition of composts to soil has a positive effect on soil productivity and other soil characteristics as they increase organic matter and nutrient levels (Garcia et al. 1991). In addition, they activate the autochthonous microorganisms of the soil and, indirectly, the biogeochemical cycles therein (Pascual et al. 1997). Furthermore, when used in containers or as soil amendments, they may have highly

suppressive effects against disease caused by a variety of soil borne plant pathogens, such as Pythium sp.(Mandelbaum and Hadar 1990; Pascual et al. 2000), Phytophthora spp. (Hoitink and Boehm 1999; Widmer et al. 1999) and Rhizoctonia sp.(Kuter et al. 1983; Tuiter et al. 1998). Disease incidence on many plants may be influenced by the level and type of organic matter and microorganisms present in soils. Research has shown that increased population of certain microorganisms may suppress specific plant diseases such as Pythium and Fusarium as well as nematodes. Suppression of various soil-borne diseases by traditional thermophilic composts has been reported (Chung, *et al*., 1988). Disease suppression by composts has been attributed to the activities of competitive or antagonistic microorganisms. The biological control of soil borne plant pathogens in compost amended container media has received much attention in the last decade. Among the organic materials used for the preparation of potting media, some were suppressive against Fusarium wilts (Chef et al., 1872; Hoitink and Fahy, 1875; Serra-Wittling et al., 1996). Brinjal is one of the most popular and important commercial crops grown through out the world. Many diseases and disorders can affect Brinjals during the growing seasons. *Fusarium oxysporum* f. sp. *Melongenae* is a highly destructive pathogen of Brinjals. The disease caused by this fungus is characterized by wilted plants, yellowed leaves and minimal or absent crop yield. Therefore the main objective of our experiments was to determine the potential of value added compost to suppress the incidence of the plant diseases *Fusarium oxysporum* L in Brinjal

MATERIAL AND METHODS

Plant pathogen isolation

Fusarium oxysporum was isolated from infected brinjal plants; a piece of an infected part was cut and disinfected with sodium hypochlorite (1%) for 5 min. The piece was put on specific *F. oxysporum* medium [peptone pentachloronitrobenzene agar (PPA) modified by Nash and Snyder (1962)]. The PPA contained the following: agar (15 g $l^{-}1$), peptone (15 g $l^{-}1$), KH_2PO4 (1 g $l^{-}1$), $MgSO_4.7H_2O$ (0.5 g $l^{-}1$) autoclaved at 120 ºC for 20 min. After 5 days at 28 ºC, F. oxysporum growth around the part was developed. A section from the margin of the developed mycelium was subculture for three times on potato dextrose agar (PDA) at 25 ºC for 7 days to be sure that only F. oxysporum mycelium was on it. A piece of isolated F. oxysporum mycelium growth on PDA, was suspended in a flask containing 250 ml of potato dextrose broth at 28 ºC for 5 days till the liquid medium was brown-pink in colour, the number of spores was counted by haemocytometer and diluted to reach $3.3X10^5$ CFU $ml^{-}1$. The *F. oxysporum* suspension was used to infect the melon plants

Enrichment of compost

Different organic waste materials were collected and allowed to decompose in heap. The composted materials were air dried, sieved and used for analysis of various chemical contents. The enrichment of compost was done by addition of PGPMOs and VAM spores. *Azatobacter choroccum, Pseudomonas florescence, Azospirillum sp, Trichoderma harzianum* and *Glomus mosseae* were isolated from different rhizospheric soil. These microorganisms were grown in broth culture and apply at the rate of 10 ml / 200 gm of compost for enrichment of compost. The chemical characteristics of the compost are given in the Table-1.

Table-1: Chemical characteristics of the compost

C	N	C : N	P	K	Ca	Mg
38.68	1.57	24.48	0.07	0.81	0.93	**0.36**

Invitro test of the suppressive effect of enriched compost

Thirty millilitres of sterile water were poured into a Petri plate with 7 day old mycelium of F. oxysporum on PDA. The fungus was suspended in water by stirring with a sterile string. Then 0.1 ml of the suspension was spread on plates with fresh PDA. After 24 hr of incubation at 28º C, samples (0.5g) of enriched compost (compost + PGPRs) placed in the centre of each plate. Plates containing samples of enriched compost autoclaved for 30 min and prepared as described above, served as a control. There were 3 replicates per treatment. The plates were incubated at 25ºC for 20 days and the development of fungus was observed. The suppressiveness of the enriched compost was estimated by the presence of an inhibitory zone around the samples.

Pot Experiment

Brinjal seeds were surface-disinfected by immersing them first in 95 % ethanol for 30 s and then in 0.2 % solution of HgCl2 for 3 min, following several rinses with sterilized water to remove disinfectant (Russell et al., 1982). Brinjal seedlings were planted in the pots containing 5 Kg of sterilized soil which was inoculated by adding 10 ml of *Fusarium oxysporum* spore suspension (3.3 x 10^{-5} CFU ml^{-1}) per pot two weeks before planting. After planting soil was treated with compost enriched with PGPMOs as per the treatment given below-

T_1 : Control (soil only)

T_2 : *Fusarium oxysporum*

T_3 : Normal compost + *F .oxysporum*

T_4 : Enriched compost (+*Azatobacter choroccum*+ *F .oxysporum*)

T_5 : Enriched compost (+*Pseudomonas fluorescence* + *F .oxysporum*)

T_6 : Enriched compost (+*Azospirillum* sp + *F .oxysporum*)

T_7 : Enriched compost (+*Trichoderma harzianum* + *F .oxysporum*)

T_8 : Enriched compost (+*Glomus mosseae* + *A. choroccum*+ *F .oxysporum*)

T_9 : Enriched compost (+*Glomus mosseae* + *P. fluorescence* + *F .oxysporum*)

T_{10} : Enriched compost (+*Glomus mosseae* + *Azospirillum* sp + *F .oxysporum*)

T_{11} : Enriched compost (+*Glomus mosseae* + *T. harzianum* + *F .oxysporum*)

T_{12} : Enriched compost (+*A. choroccum*+ *P. fluorescence*+ *Azospirillum* sp+ *G. mosseae* +*T. harzianum* + *F .oxysporum*

Plant height and number of leaves were recorded in 15 days of interval and diseases index was recorded at 30 days interval. Post harvest parameters such as biomass both root & shoot were recorded after the harvest. Mycorrhizal spore count and percent of mycorrhizal colonization were calculated.

Statistical analysis

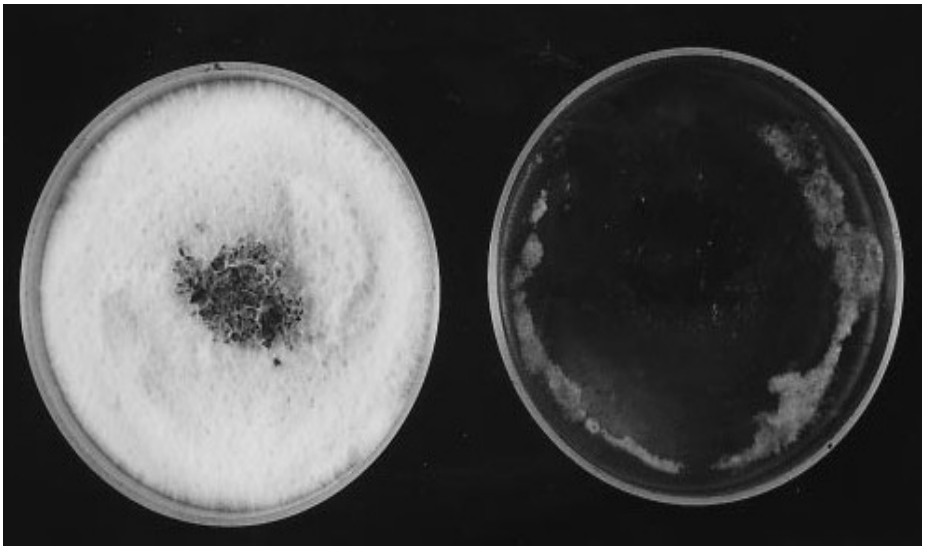
All data were subjected to analysis of variance and the Newman_Keuls test was used to estimate the significance of differences between means (P=0.05).

RESULTS & DISCUSSION

Invitro test of the suppressive effect of enriched compost

Unsterilized enriched compost induced a strong inhibition of growth of *F.oxysporum* on agar plates (Fig.1). For the sterilized enriched compost no suppressive effect was observed and even samples of this substrate were covered with mycelium. The suppressiveness of enriched compost toward soil- borne fungal plant diseases has been described in only a few papers (Szczech et al., 1993; Szczech and Brzeski, 1994, Szczech, 1995; Kostecka et al., 1996). Similar results were obtained by other authors using various composts (Hoitink and Fahy, 1986; Hadar et al., 1982; Hoitink et al., 1995). In most cases, the soil suppressiveness to *F.oxysporum* has been eliminated after heating, indicating the biotic nature of the disease suppression (Hoitink and Fahy, 1986). Enriched compost strongly inhibited the growth of F. oxysporum on agar medium, while after sterilization of this substrate, mycelium of the pathogen covered all of the plate. Microscopic observations showed that hyphae removed from plates with unsterilized vermicompost were completely destroyed and colonized by microbes. On plates with sterilized substrate or peat the hyphae were unaffected.

Fig. 1: Influence of unsterile and sterile enriched compost on growth of *F. oxysporum* mycelium on agar medium.



Pot experiment