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RESEARCH OPINIONS IN ANIMAL & VETERINARY SCIENCES

Investigating of physical characteristics growth of corn (*Zea mays* L.) effect of inoculating fungi *Trichoderma spp* into compost

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Abstract

Evaluation of soil microorganisms and their beneficial relationships among ecosystem components is one of new topic in soil resource management for ensuring sustainable agriculture. In order to investigating effects of *Trichoderma spa* and compost on physical characteristics of growth of corn, an experiment was conducted at glasshouse in Sari Agricultural and Natural Resources Sciences University. The pot experiment was laid out factorial based on completely randomized design with four replications. Three seedbed materials consisted of field soil as check, compost (20 $Mg.ha^{-1}$) and (40 $Mg.ha^{-1}$) and two species of *Trichoderma* (*T. harzianum* and *T. Viride*) with a check were the treatments. According to results, using of compost (20 $Mg.ha^{-1}$), increased corn height, leaf area, SPAD, yield and harvest index compared to the check and compost (40 $Mg.ha^{-1}$) treatment. In addition, *T. harzianum* and *T. viridae* in 10gr rate, increased corn yield, leaf area and SPAD as much as 8.2, 5.8, 11.3 respectively, compared to check treatment. Leaf area and SPAD of corn significantly increased inoculation of *T. harzianum* in compost (20 $Mg.ha^{-1}$) treatment. Physical characteristics growth of corn was not significant effected in compost (40 $Mg.ha^{-1}$) treatment. Overall, inoculation of *T. harzianum* could improve physical characteristics growth of corn in compost (20 $Mg.ha^{-1}$) treatment.

Keywords: *Trichoderma*, Inoculate, Compost, Corn

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Introduction

There is a worldwide need to adopt the practice of sustainable agriculture, using strategies that are environment-friendly, less dependent on agricultural chemicals and less damaging to soil and water resources. The use of organic residues (compost and vermicompost) as a mean of maintaining and increasing soil fertility is a long-standing practice (Singh et al., 2007; Powlson et al., 2011). Farmers and scientists are now showing renewed interest in the proper and effective use of organic residues, compost, other recycled organic additives and green manures. In the recent years, the environmental contamination caused by excessive use of chemical pesticides increased the interest in integrated pest management, where chemical pesticides are substituted by biopesticides to control plant pests and plant diseases. Compost applied to soil improve its quality by altering the chemical and physical properties, increase organic matter content (Sivapalan et al., 2005; Wei and liu, 2005; Cherr et al., 2006), water holding capacity (Wilhelm et al., 2007), overall diversity of microbes (Singh et al., 2007), provide macro and micronutrients essential for plant growth and suppress diseases which indirectly contribute to plant growth enhancement (Cherr et al., 2006).

The fungal genus *Trichoderma* is cosmopolitan in soils, and the ecological adaptability of *Trichoderma* species is evidenced by their widespread distribution, including under different environmental conditions and on various substrates. Different species of *Trichoderma* have the potential to control soil borne plant pathogens more effectively than chemicals (Aneja et al., 2005; Bennet and Whipps, 2008; Ruano and Lopez, 2009; Rojan et al., 2010). Use of these fungi is not as harmful

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to the environment as chemical pesticides. In most instances, increased plant growth and yields were attributed to the reduction in plant disease by the biological control agents. *Trichoderma* spp are thought to promote plant growth by at least two different mechanisms: (i) - by controlling the population of pathogenic microorganisms in the rhizosphere (Brunner et al., 2005; Perazzolli et al., 2011), and (ii) - by influencing plant physiology through mineral solubilization or hormone secretion (Ainhoa et al., 2011). In vitro studies have shown that micronutrients and insoluble phosphates became soluble and available, therefore useful to the roots interacting with *T. harzianum* in the root zone (Bombiti et al., 2011). Species in the filamentous fungal genus *Trichoderma* are of great economic importance as sources of enzymes and antibiotics; plant growth promoters; degraders of xenobiotics, and most importantly, as commercial biofungicides. In addition to the above, various species of *Trichoderma* were effective in the promotion of growth and yield in various crops (Bal and Altintas, 2006). Cucumber, bell pepper and strawberry yields, were increased significantly whit application of *T. harzianum* in the root zone (Altintas and Bal, 2005; Bal and Altintas, 2006; Elad et al., 2006). Effective bio-inoculants should preferably penetrate the roots to not only directly antagonist root pathogens, but also benefit plant growth and vigor through various mechanisms such as nutrient mobilization and induction of host defenses (Rudresh et al., 2005). Therefore, it seemed important to analyze if commonly used plant growth regulators such as auxins, cytokinins and gibberelic acid, that are applied to different plant species or that are produced by soil microorganisms, could affect the biocontrol activity of *T. harzianum*.

Corn (*Zea mays* L.) is one of the main crops and is commonly employed in the human diet in its natural form as sweet corn or as a sub product in bread, flour and dough, among others (Regina et al., 2007). Corn also is an ideal crop for application of an inexpensive, long-term method for reducing disease via systemic resistance. Further, total control is not necessary; even a relatively modest decrease in disease would be a valuable addition to maize culture since foliar fungicides are generally too expensive for field maize. Subsequently, we considered that corn yield and quality characteristics may be enhanced using high dosages of a *Trichoderma* product, and therefore the present experiment was conducted.

Materials and Methods

An experiment was conducted at research farm of Sari Agricultural Sciences and Natural Resources University (Latitude 42.36N, longitude 13.53E and 16 m above mean sea level), Iran during 2009. Monthly distribution of total precipitation (135.6 mm), average temperature (26.5^{o}C) and relative humidity (65%) in the year 2009 are given in Table1. Experiment laid out as factorial plot based on randomized complete design. Three replicates were set up for each treatment. Three seedbed materials consisted of field soil as check, compost (20 Mg.ha^{-1}) and (40 Mg.ha^{-1}) and two species of *Trichoderma* with a check were the treatments. The compost was applied two weeks before swoing and mixed thoroughly with the soil of each pot.

Table 1: Monthly means of maximum (Max), minimum (Min) and average (Aver) air temperatures and rainfall in growth season in 2009 at studied area

Month	Max	Min	Aver	Rainfall (mm)	Relative humidity (%)
	(^{0}c)				
June	28.4	23.3	24.8	13.1	62
July	27.8	23.2	25.3	83.9	74
August	30.5	26.4	28.5	4.6	65
September	29.6	23.1	27.4	34.0	61

This work investigated the application of two selected beneficial microorganisms (*T. harzianum* and *T. viride*) to corn seed and their subsequent survival and establishment in the rhizosphere once the seed was planted. Fungi recovered from this system were subsequently grown on potato dextrose agar (PDA; Merck) (chacon et al., 2007) slopes for storage for 7 days at 30°C in a termostated incubator chamber with air circulation. This period showed to be sufficient for fungi sporulation. The substrates used were wheat bran. Wheat bran showed to be the most suitable substrate to produce Trichoderma spores for all strains that were evaluated (Cavalcante et al., 2008). The substrates were sterilized at 121°C for 15 min in a Phoenix autoclave model AV 50 and cooled down to room temperature before the inoculation, which was done until 24 h after sterilization. The dosages of two funguses applied were 5 and 10g per pot. Control plants were also available. A control plot in each block was left untreated and only water without the antagonistic fungus was provided. Corn seeds were sown in plastic pots on July 6, 2009.

Availability of water in the soil plays an important role in facilitating establishment and effectiveness of *Trichoderma* in the soil, Therefore irrigation was done so on a regular basis. Chlorophyll concentration can be measured in vivo by nondestructive spectroscopic techniques that do not require sample preparation. In this experiment, a commercial nondestructive dual-wavelength meter (model SPAD 502 Minolta crop) was used to directly estimate chlorophyll content. At harvest in the 2009 growth season, the plants removed the plot, Shoot dry weight was recorded after drying the samples at 70 for 48 h in an oven until they reached a constant weight, the grain was removed from the straw, and then

the straw and grain weights were recorded. Data were subjected to ANOVA using the SAS statistical software package using a GLM (SAS Institute, 2000) and Duncan's multiple range test was performed to compare the treatment means. The level of statistical significant was accepted as $P<0.05$, (Steel and Tore, 1960).

Results and Discussion

In our studies, Using of compost (20 $Mg.ha^{-1}$) increased corn height, SPAD reading, leaf area, yield and harvest index compared to control treatment. Plant dry matter yield decreased with increasing compost rate (40 $Mg.ha^{-1}$), this suggests that the lowest compost rate is adequate for corn production (Table 2). Using low rates would allow the compost to be spread over more acres of farmland, increasing food production. Different of *Trichoderma* species showed a various degree of increased corn plant response (Table 2). The results suggested that various unknown factors might interact to mediate responses. The factors might result in rlizosphers condition and survival ability of these species in different rate of compost. In addition, different amount of compost induced significant increases in dry matter, harvest index, yield, and leaf area as compared to control. No corn harvest index differences due to the rate of Compost (20 $Mg.ha^{-1}$) application compared the control, but all treatments receiving manure produced more corn grain than the unamended control. These results may be due to the higher levels of organic matter and nutrients in composts (Perez et al., 2006; Evanylo et al., 2008) than in control (without the compost). In this study a significant increase of plant growth treat with *T. harzianum* and *T. viridae* observed on the measured parameters (plant height, leaf area, plant dry weight) compared to the control (Table 2). Similarly, the chlorophyll content (SPAD) in leaves of corn treated by *Trichoderma* species was higher than that of untreated plant. Suggested that growth promotion effect in the presence of *T. harzianum* in the rhizosphere was due to increased root surface area allowing the roots to explore larger volumes of soil therefore more nutrients were made available to the plants especially under nutrient-limited soil environments.

The increased growth response induced by *Trichoderma* species is not fully understood. However, several possible mechanisms have been suggested do explain this phenomenon of increase plant growth. These factors may include 1) control of deleterious root microorganisms, those were not cussing obvious diseases, 2) direct production of growth stimulating hormones, 3) increased nutrients uptake through enhanced root growth. Biomass per plant was highest in combination treatments compared with individual inoculations, but biomass in individual inoculation treatments was greater than in the fertilized control. The lowest biomass was observed in the control. The increased growth response in inoculated treatments observed here may be due to a possible supply of soluble P and other nutrients by Trichoderma spp. in addition to production of growth promoting substances by these organisms (Bombit et al., 2011). The increased growth and biomass yield by the use of *Trichoderma* spp have been reported for various crops. For example Altintas and Bal (2005) and Bal and Altintas (2006) reported that *T. harzianum* increased plant vigor of Cucumber. Gravel et al. (2007) also reported that the fungus increased both root and shoot growth of tomato.

Inoculation of *Trichoderma* species (*T. viridae* and *T. harzianum*) significantly increased corn height, leaf area and dry matter compared with untreated plants. Type and species of *Trichoderma* may be an important factor in its effectiveness both in the control of phytopathogenic fungi and in the induction of enhanced plant growth and yield. Application of *T. harzianum* in 5gr treatment, increased corn yield and some quality characteristics however the increases were not statistically significant (Fig. 1). It seems the dosage of Trichoderma applied was 5gr in plot, which may have been too low a dosage for a Trichoderma product to be effective. But, *T. harzianum* increased corn yield in 10gr treatment. However, with the manipulation of dosage and frequency of Trichoderma application significant increases obtained in the characteristics studied.

Growth response to compost (20 $Mg.ha^{-1}$) with *T. harzianum* was greater than in the compost (40 $Mg.ha^{-1}$) treatment, with significant increases in corn yield and Harvest index as much as 8.2, 5.8 respectively (figur.1). Rudresh et al. (2005) reported that Inoculation of *Trichoderma* spp. showed increased yield of chickpea compared with the controls under both glasshouse and field conditions. However, application of Trichoderma was not conducive to increased yields in tomato (BAL and Altintas, 2008). In onion, yield and quality characteristics were not enhanced by the application of Trichoderma spp. (Latinas and Bal, 2008). In the work of Altintas and Bal (2008) The present authors previously obtained significant yield increases in cucumbers and bell peppers using a much higher dosage, 40 kg/ha (Latinas and Bal, 2005; Bal and Altintas, 2006). In addition, Yadav et al. (2009) reported that that application of *T. viride* enriched farmyard manure (FYM), however, brought economy in the use of fertilizer N by45.2 kg ha^{-1} and also increased the yield by 6.1 t ha^{-1}, compared to the control treatment. Leaf area and SPAD of corn significantly increased inoculation of *T. harzianum* in compost (20 $Mg.ha^{-1}$) treatment (figure 2, 3 and 4).

Table 2: Effect of *T. harzianum* and *T. viridae and* different rate of compost on physical Characteristics growth of corn

Treatments	Height	SPAD	Dry matter	Leaf Area	Yield (gr/plant)	Harvest index
Compost (20 Mg.ha^{-1})	238.8 a	44.12 a	182.0 a	0.49 a	101.91 a	52.19 a
Compost (40 Mg.ha^{-1})	222.7 b	41.40 b	181.2 a	0.44 b	89.32 b	49.13 b
Control	207.9 c	39.44 c	146.2 b	0.41 c	73.02 c	49.95 b
Trichoderma spp						
Control	206.7 c	37.88 d	159.0 d	0.40 c	77.56 c	48.47 c
T. harzianum 5gr	212.4 c	41.33 c	170.2 bc	0.46 ab	85.20 b	49.95 bc
T. harzianum 10gr	225.3 b	43.44 ab	175.5 ab	0.46 ab	95.14 a	52.22 ab
T. viridae 5gr	231.9 b	41.66 bc	164.7 cd	0.44 b	85.20 b	51.66 ab
*T. viridae*10gr	239.4 a	43.88 a	179.6 a	0.49 a	96.60 a	53.20 a
Significant						
A	**	**	**	**	**	**
B	**	**	**	**	**	*
A × B	ns	*	ns	*	*	*
CV	3.6	6.65	4.58	7.74	6.01	5.9

Levels of significant:* P< %5, ** P<%1, NS = not significant

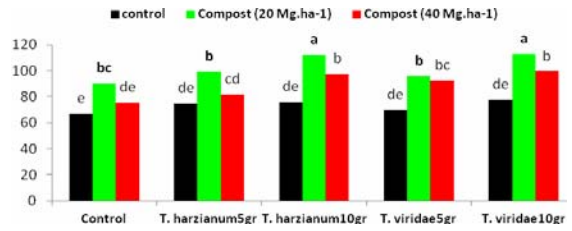

Fig. 1: Effect of *Trichoderma spp* and different rate of compost on yield corn

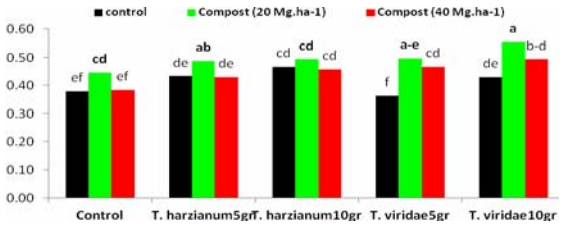

Fig. 3: Effect of *Trichoderma spp* and different rate of compost on LAI of corn

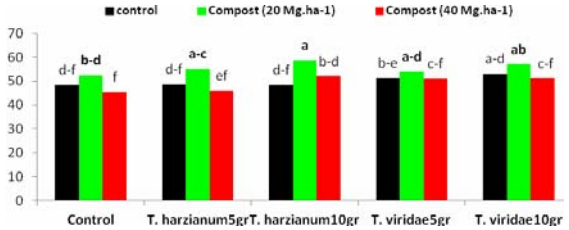

Fig. 2: Effect of *Trichoderma spp* and different rate of compost on harvest index of corn

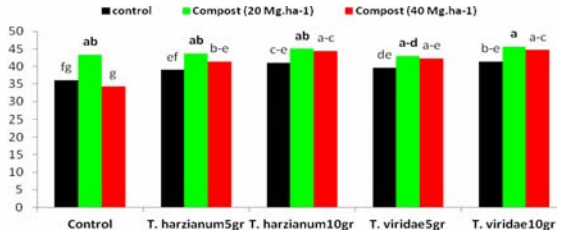

Fig. 4: Effect of *Trichoderma spp* and different rate of compost on SPAD of corn

Promotion of growth and yield by *Trichoderma spp*. is a result of increased root area allowing the roots to explore larger volumes of soil to access nutrients, and increased solubility of insoluble compounds as well as increased availability of micronutrients (Yedidia et al., 2004; Bal and Latinas, 2008; Perazzolli et al., 2011). *Trichoderma* spp are effective biocontrol agents for several soil-borne plant pathogens, and some are also known for their abilities to enhance systemic resistance to plant diseases and overall plant growth (Singh et al., 2007). The findings of the present study are in agreement with the findings of Rudresh et al. (2005) who reported that soil application of *Trichoderma spp* increased yield of chickpea. *Trichoderma harzianum* induces large changes in the proteome of shoots of maize (*Zea mays*) seedlings (Shoresh and harmar, 2008). Increased photosynthesis should have resulted in increased starch accumulation in seedlings and did indeed occur (Harman, 2005).

Conclusions

Our work has demonstrated that aborigine *Trichoderma* strains can be against towards. However, in this work, we found that, *T. spp* applied to soil was effective in the enhancement of yield and quality characteristics of corn. Physical characteristics growth of corn not significant effected in compost (40 ton/ha) treatment. Overall, inoculation of *T. harzianum* could improve physical characteristics growth of corn in compost (20 ton/ha) treatment. Certainly, other

mechanisms still need more studies for plant growth response of Trichoderma species in this works. However, further experiments under varying soil conditions, watering and fertilizer regimes and frequency of *Trichoderma* application may be necessary to fully judge on the effectiveness of *T. harzianum* on yield and quality characteristics of corn. The development of this isolate as a successful biostimulant is dependent upon understanding of the complex interactions of this organism and plants in the soil ecosystem.

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