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Would *Trichoderma* Affect Seed Germination and Seedling Quality of Two Muskmelon Cultivars, Khatooni and Qasri and Increase Their Transplanting Success?

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ABSTRACT

In order to investigate effect of *Trichoderma harzianum Bi* on seed germination and seedling quality and field establishment of two muskmelon cultivars, *Khatooni* and *Qasri,* different tests was done at greenhouse and field. 4 concentration of trichoderma solution, control (0), 25%, 50% and 100% (Mazhabi 2010) in a factorial experiment with a completely randomized design and ten replications with 10 seeds in each replication placed in Petri-dishes to evaluate germination percentage and germination index. Emergence tests were carried out in seedling trays which were filled with enriched coco peat in 4 different concentrations, and then emergence percentage and index calculated during two weeks. Four weeks after emergence of first seedling, 5 seedlings per replication were selected randomly and shoot length and diameter, length of longest root and total dry weight of plugs measured. , successful transplantation percentage (STP) evaluated using two methods of trichoderma application in 4 different concentrations. Results show that trichoderma application significantly increased germination and emergence percentage and index and could help improving seedling health and vigor.STP was significantly affected by trichoderma application and 1st method of application had better results.

Keywords: priming, transplant production, field establishment, growth rate, successful transplantation percentage.

INTRODUCTION

Muskmelon (*Cucumis melo* L. var. Inodorus) is one of the major vegetable crops in the world. From seed sowing to emergence, excessive soil water can damage melon and cause problems and diseases (Sensoy *et al*., 2007). It is cultivated in all the temperate regions of the world due to its good adaptation to soil and climate. Fruits are consumed in the summer period and are popular because the pulp of the fruit is very refreshing and sweet, with a pleasant aroma (Villanueva *et al,* 2004). Using transplants instead of direct seed sowing will reduce risk of seedling death because of diseases. It also helps early crop production.

Seed priming allows for some of the metabolic processes necessary for germination to occur without actual germination. Primed seeds usually exhibit increased germination rate, greater germination uniformity, and sometimes greater total germination percentage (Basra *et al*., 2004). Increased germination rate and uniformity have been attributed to metabolic repair during imbibition (Bray *et al*., 1989), a buildup of germination-enhancing metabolites (Basra *et al*., 2005), osmotic adjustment (Bradford, 1986), and, for seeds that are not re-dried after treatment, a simple reduction in the lag time of imbibitions (Bradford, 1986). In muskmelon studies, seeds have been primed from 16 hours (Dhillon, 1995) to ten days (Yeoung *et al*., 1996). Adequate oxygen is required during seed priming (Heydecker *et al*., 1975; Bujalski *et al*., 1989).

Trichoderma spp. are being employed widely in plant agriculture, both for disease control and yield increases (Harman, 2006), even under axenic conditions (Lo *et al.,* 2000; Yedidia *et al.,* 2001). *Trichoderma* spp. have evolved multiple mechanisms that result in improvements in plant resistance to disease and plant growth and productivity (Harman *et al.,* 2004; Vinale *et al.,* 2008). It have been also shown to stimulate plant growth in cucumber, cabbage, lettuce, potato, tomato, carrot, beans and peas (ousley *et al*., 1994; Khan *et al*., 2004;rabeendran., 2000; Yossen *et al*,2003). Presumed mechanisms involved in the stimulation of plant growth by Trichoderma include interactions with plant roots similar to mycorhizae, in which Trichoderma penetrates and colonizes root tissues without eliciting specific defense responses against the colonizing strain (Yedidia *et al*., 1999).

Plants produce proteins in response to abiotic and biotic stress and many of these proteins are induced by phytohormones such as ABA (Jin *et al*., 2000) and salicylic acid (Hoyos & Zhang, 2000). Trichoderma can produce metabolites with activities analogous to plant hormones (Cutler *et al*., 1989, 1991) so it would affect plant protein production and act as a kind of seed priming.

IAA and its analogues have been shown to positively affect root growth and morphology. Trichoderma induce plant IAA production and so it helps root growth and seedling quality in different plants (Morales *et.al*, 2004; Gravel *et al*., 2006)

Sowing of infected seeds can reduce germination, vigour and potential yield by transmitting pathogen from seed to plants. The most adverse effect of seed-borne pathogen is contaminating disease free areas, thus

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seedborne pathogens acts as primary source of inoculum for disease development. Though, the level of seed-borne inoculum might be extremely low, the rate of its increase may be extremely high when exposed with favorable epidemiological factors, including local agricultural practices (Neergard, 1977). Trichoderma as had been reported by many authors (eg: Yedidia *et al.*, 1999; Howell, 2003; Hanson, 2004) can reduce seed infection and so prevent seed borne disease occurrence in the field.

This study was carried out with the objective to evaluate the influence of pre-sowing trichoderma seed treatments on the germination and seedling quality of two muskmelon cultivars.

MATERIALS AND METHODS

Trichoderma selection and preparation

As described before different trichoderma isolates have promotional effects on plant growth. *T.harzianum Bi* which had promoted growth parameters of polianthes, tulips, tagetes, zinnia and bell pepper seedlings (Mazhabi 2010) selected for this survey. Trichoderma solution and enriched coco peat supplied from Research department of Adonis Falat Sharq Co.

Seed preparation

Seeds of two generally cultivated cultivars of muskmelon in Iran, “Khatooni” and “Qasri” were extracted from totally ripened fruits using fermentation method. Then 600 uniform, intact, healthy seeds of any cultivar selected for trichoderma application.

Germination

4 concentration of trichoderma solution, control (0), 25%, 50% and 100% (Mazhabi 2010) in a factorial experiment with a completely randomized design and ten replications with 10 seeds in each replication placed in Petri-dishes to evaluate germination percentage and germination index (Mukhtar,2008). Seeds were considered germinated when the radical was at least 2 mm long (Al-harbi *et al*, 2008).

Germination percentage calculated as:

$$\text{Germination (\%)} = \frac{\text{Number of germinated seeds} \times 100}{\text{Total number of seeds}}$$

Germination Index calculated as:

$$\text{Germination Index} = \frac{\text{Number of germinated seed (first count)}}{\text{Days to first count}} + \frac{\text{Number of germinated seed (2}^{\text{nd}}\text{ count)}}{\text{Days to 2}^{\text{nd}}\text{ count}} + \frac{\text{Number of germinated seed (final count)}}{\text{Days to final count}}$$

Emergence and Seedling Growth

Emergence tests were carried out in seedling trays with 224 cells (4 ×4 × 6 cm), which were filled with sterilized coco peat[†] and sand 1:1 v/v and placed in a greenhouse at 25 ± 3°C 60% ± 15% relative humidity. Treatments applied using different volume of enriched coco peat, instead of normal coco peat in the seedling tray. 4 concentrations of enriched coco peat, 0 (control), 25, 50 and 100 percent of total coco peat were used. Factorial experiment with a completely randomized design with 10 replications and 10 seeds in each replication were exerted. A seedling was considered emerged when the hypocotyls hook was visible above the media surface (Al-harbi *et al*, 2008). For this test, emergence percentage and index calculated. Four

[†] Coir, coconut husks.

weeks after emergence of first seedling, 5 seedlings per replication were selected randomly and shoot length and diameter, length of longest root and total dry weight of plugs measured.

Transplanting

Two methods of treatment used. Half of selected seeds sawn in seedling trays as described in seedling test. The other half, first treated with trichoderma solutions as described in germination test, and then sawn in seedling tray containing coco peat and sand 1:1 v/v and put in the same condition described before. After 2 weeks, 240 plugs of each cultivar containing, 60 plugs for control, 180 plugs for 1st (60 for each treatment) and 180 plugs for 2nd method of treatment, selected and transplanted to field. Rows were 1m apart with in-row spacing of 50 cm in 6 replication and 10 transplants in each replication. 2 weeks later, successful transplantation percentage (STP) evaluated as:

$$STP = \frac{number\ of\ established\ transplants}{total\ transplanted\ plugs} \times 100$$

Data were analyzed using SPSS for Windows (SPSS Inc.) differences between treatments (multiple comparisons) were determined using least significant differences (LSD).

RESULTS AND DISCUSSION

For qasri cultivar germination percentage was effectively increased from 64 percent in control to 95 percent in 100% of trichoderma treatment. Khatooni had also increased from 60 percent in control to 96 percent in 100% of trichoderma treatment (Figure 1-A). Trichoderma treatments in both cultivars had significant effects on seed germination. 25 % treatment in qasri was significantly rise germination percentage to 82 %. There were no significant difference between 50 and 100 percent of trichoderma ($P \leq 1\%$) , although they were significantly higher than control and 25% ($P \leq 1\%$). 50 and 100% treatment of trichoderma are significantly different in ($P \leq 5\%$)

Germination index also increased in both cultivars while trichoderma concentration increased in treatments. Qasri and khatooni control value of GI was 2.05 and 1.95 respectively which had been significantly increased to 3.2 and 2.88 by applying 25% of trichoderma concentration (($P \leq 1\%$), Figure 1- B). Although there were not significant differences between 50 and 100% treatments of trichoderma for GI, they had significantly increased GI values of both cultivars in comparison to control and 25% treatment ($P \leq 1\%$).

Emergence percentage (EP) of qasri seeds increased significantly from 55% in control to 78, 89 and 93 % in 25, 50 and 100 % treatments, respectively (($P \leq 1\%$), Figure 1-C). There was a significant difference between control and all other treatments. Also 25% treatment had significant difference with 50 and 100% treatments. in khatooni either trichoderma treatments increased EP from 51 percent in control to 87 percent in 100 % treatment (($P \leq 1\%$), Figure 1-C).

Emergence index of both cultivars increased in a linear trend and control in both cultivars had the lowest value and 100% treatment shows the most value (Figure 1- D). difference between all treatments was significant in ($P \leq 1\%$) except 50 and 100% treatments which were significantly different in ($P \leq 5\%$).

There were no significant differences between two cultivars in the same treatment for GP and EP, but GI and EI had been significantly differ ($P<1\%$).

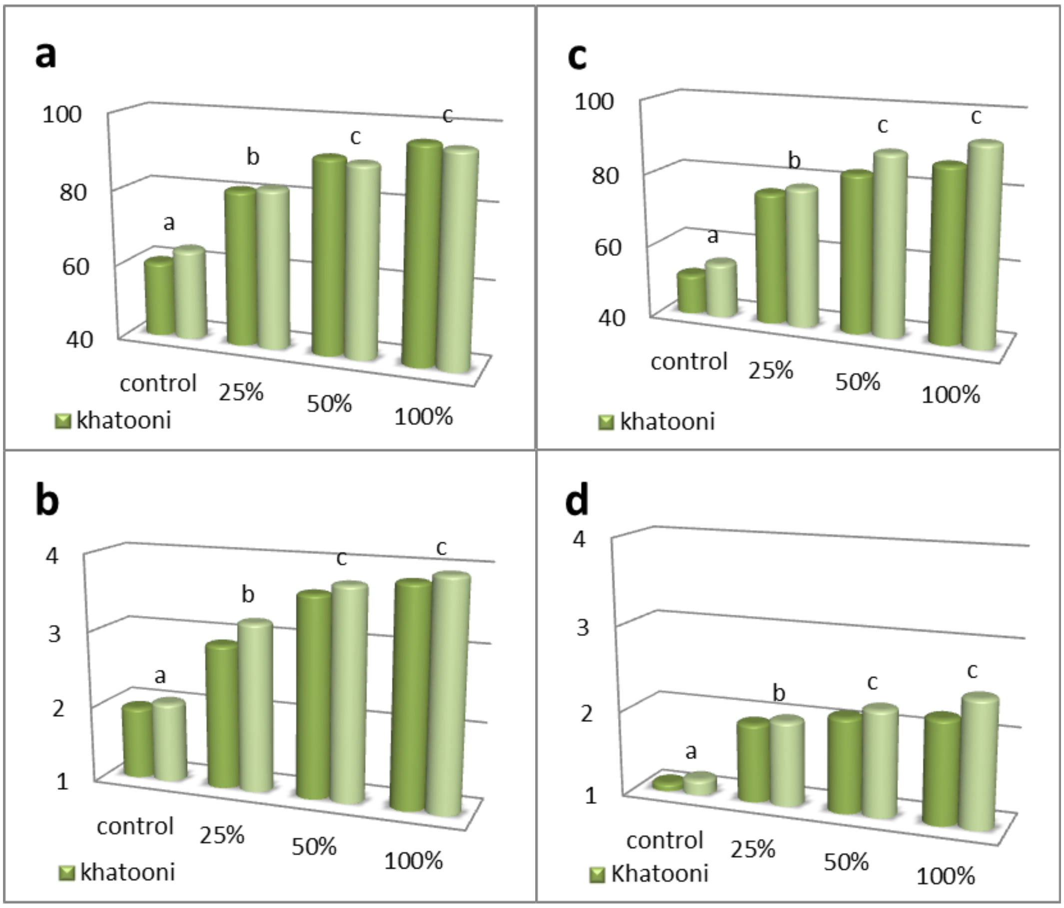

Figure1. Effect of trichoderma concentrations on khatooni and qasri cultivars. A- Germination percentage; B- Germination index; C- Emergence percentage; D- Emergence index. (Columns with different letters have significant difference in P≤ 1%.)

Stimulus effects of trichoderma application on transplanting potential of both studied muskmelon cultivars shown in result analysis. Although there were no significant difference in the same methods between khatooni and qasri but in each of named cultivars 1st method had more positive effects and was more effective to produce healthier transplant which could tolerate transplantation mechanism and it's induced stresses (e.g., mechanical, water and *etc*). Positive effects of trichoderma in 1st method in qasri increased STP from 41.66 % in control to 90% in 100% treatment. It also increased STP in 2nd method from 41.66% to 75% in 100% treatment (Figure 2). Khatooni else had such increscent in STP values. In the 1st method it increased to 88.33 % and in the 2nd method it increased to 75% in the highest level of trichoderma treatment (Figure 2). This shows that inoculation of trichoderma to culture medium would increase root colonization and may affect treated plants more than other method. Application of trichoderma solution on seeds may perform like other kind of seed priming's and it may affect germination more than later growth processes.

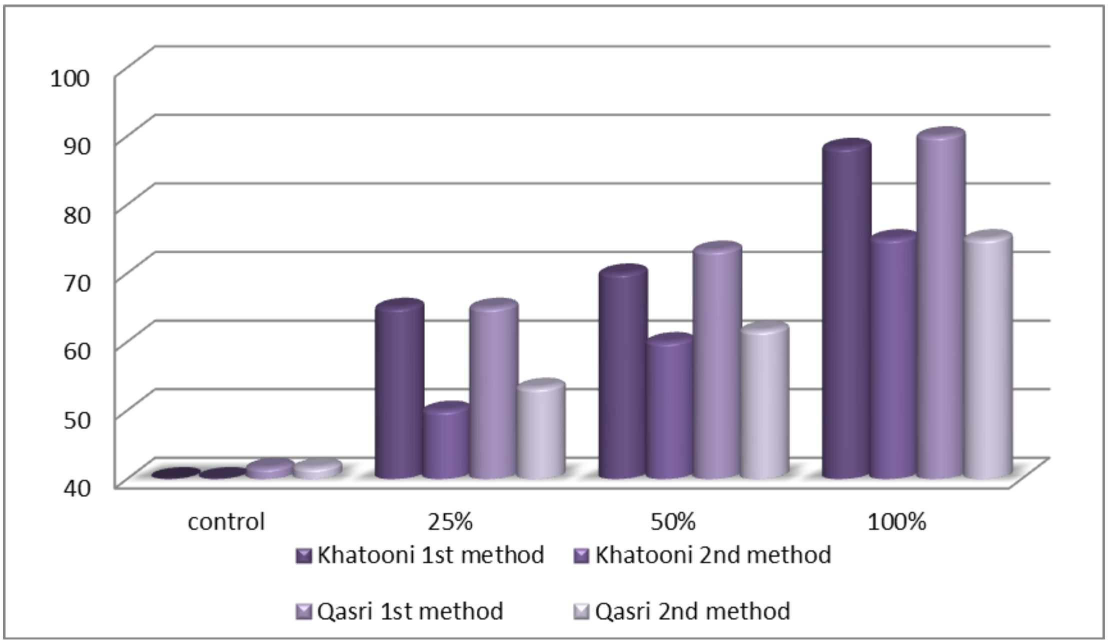

Figure 2. Successful transplantation percentage of two different methods of trichoderma application in 4 concentrations on Khatooni and Qasri muskmelon cultivars.

For shoot length (mm), shoot diameter (mm), root length (mm) and dry weight (mg) of both studied cultivars, all treatments were significant (P<1%). But there was not any difference between two cultivars at the same level of trichoderma application (Table 1).

Table 1. Effect of trichoderma inoculation to substrate during seed germination, emergence and seedling growth on seedling properties

	Shoot length (mm)		**Shoot diameter (mm)**		**Root length (mm)**		**Dry weight (mg)**	
	Qasri	Khatooni	Qasri	Khatooni	Qasri	Khatooni	Qasri	Khatooni
Control	44.3	42.8	6.54	6.4	3.8	3.68	177.94	171.02
25%	53.24	52.8	9.49	9.32	5.43	5.34	196.4	191.6
50%	79.74	79.18	12.33	11.97	6.46	6.43	209.58	206.72
100%	111.4	110.32	15.59	15.28	7.32	7.25	256.1	252.8
LSD value P≤1%	1.918	1.953	0.186	0.194	0.137	0.148	1.586	1.558
LSD value P≤ 5%	1.399	1.425	0.135	0.142	0.100	0.108	1.157	1.137

Production of plant growth regulators, solublization of insoluble minor nutrients in soil, increased availability of micronutrients, increase in nutrient transfer from soil to root, Induced resistance to different pathogens (mostly other fungus) are the main trichoderma mechanism of action in contact to plants(Ousley,1994; yedidia ,2001; Harman, 2006; Vinale, 2008) .

In this study application of trichoderma increased either germination and emergence percentage and index which means that seeds germinated better, seedlings emerged stronger and whole period occurred faster. This may be due to higher production of growth stimulators that been reported in plant-trichoderma interactions, like GA and IAA (Gravel, 2006).

The other possible mechanism which may cause production of vigorous and healthy seedlings is trichoderma pathogen interaction and induced overall tolerance in seedlings toward soil born diseases. This could help seedlings to grow easier.

Production of healthy and strong seedlings with enough root and shoot growth is a key factor in transplant establishment in the field. Trichoderma in this experiment increased dry matter accumulation in roots and shoots of seedlings and also made seedlings longer and branched roots. Maybe this is the main factor which caused that transplantation success increased to two fold in comparison to control.

CONCLUSIONS

According to the results of previous studies as well as (Harman, 2006; mazhabi 2010) and results of trichoderma application on growth stimulation of muskmelon cultivars in this experiment, it can be concluded that promotional effects of trichoderma on transplantation potential of qasri and khatooni muskmelons may be due to enhanced nutrient uptake during seedling growth, enhanced root development and increased root hair formation during seedling growth in trays and after transplantation that can occur because of an increase in hormone production. It is known that cucurbitaceous vegetables have a weak rooting power exactly after transplantation and so more than half of seedlings in control died after transplanting. Using trichoderma as a tool to increase rooting potentiality could help seedling to overcome these problem. Also more dry weight produced during seedling growth period in trichoderma treatments may be a beneficial point.

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