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# Evaluation of Biocontrol Agents in Management of Wilt Disease of Gladiolus Caused by *Fusarium oxysporum* F. Sp. *Gladioli*

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**A B S T R A C T**

*Fusarium oxysporum* f. sp. *gladioli* cause wilt in gladiolus. *Fusarium* wilt is a major constraint to flower industry with respect to good quality and yield. Use of fungicides for the management of *Fusarium* wilt has been found to be inconsistent. Application of biocontrol agents constitutes an effective option for the management of *Fusarium* wilt. The present study was carried out to analyze the efficacy of biocontrol agents against *Fusarium* wilt. Twenty eight *Trichoderma* isolates and one fluorescent pseudomonad isolate were tested *in vitro* for their efficacy against *Fusarium* wilt pathogen of gladiolus. Isolates of *T. harzianum* (IIHR09, GJ16B, TN2A, HAR4B) and fluorescent pseudomonad (IIHRFP1) were found effective and further evaluated *in vivo*. All *Trichoderma* isolates were found effective in controlling *Fusarium* wilt of gladiolus in pot experiment while, 77.77 and 44.44 per cent of wilt incidence was observed in case of control and fluorescent pseudomonad isolate. Field trial for the management of *Fusarium* wilt of gladiolus using carbendazim tolerant *Trichoderma* isolate GJ16B was found effective that reduced wilt incidence from 18 to 4 per cent. The biocontrol mechanism studies revealed that selected *Trichoderma* isolates were possessed good competitive saprophytic ability. The isolate HAR4B was found to be the best in production of volatile compounds, while isolate GJ16B was the best in non-volatile compounds production. Besides GJ16B possessed strong mycoparasitic ability against *Fusarium oxysporum* f. sp. *gladioli*.

## Introduction

Gladiolus (*Gladiolus hybridus* Hort.) belongs to the family Iridaceae. It is one of the important ornamental bulbous flower crops. It is mainly grown for its beautiful flowers. Owing to their different sizes, shades and excellent vase life, gladiolus hybrids are one among the preferred cut flowers (Bose *et al.*, 2003; Riaz *et al.*, 2008).

The *Fusarium* wilt of gladiolus was first reported by Pryal (1909) and the pathogen was identified as *Fusarium oxysporum* f. sp. *gladioli* (Massey) Snyd. & Hans. In India it was first reported by Singh (1969). Tomar (1997) reported *F. moniliforme* causing wilt in gladiolus from Himachal Pradesh of India.

*Fusarium* wilt of gladiolus considerably reduces the flower and corm production. It rapidly spreads through infected corms and

soil from one region to another and at times from one country to another. It becomes necessary to manage the disease both in field and storage conditions. Management strategies for *Fusarium* wilt were sanitation measures, using healthy corms, soil fumigation and using chemicals. Due to high cost of chemicals, loss of soil microflora and environmental pollution by using chemicals for management of disease, the use of other cultural practices and introduction of biological control agents may be an alternative for managing the disease (Chandel and Deepika, 2010). In the present work the evaluation for efficacy of *Trichoderma* isolates in management of *Fusarium* wilt of gladiolus was studied.

## Materials and Methods

### Isolation of the pathogen

The wilted gladiolus plants along with rotten corms were collected from Solan, Himachal Pradesh. Infected rotted corms of gladiolus were used for the isolation of *Fusarium* pathogen. Tissue isolation technique was followed for the isolation of the pathogen by placing infected bits on Petri plates containing potato dextrose agar media and incubated at ambient temperature (27 ± $2^0$C) and observed periodically for the growth of pure colonies. The colonies were developed from the bits transferred to PDA plates to get pure colonies and incubated at 27 ± $1^0$C for 7 days. The *Fusarium* plates were used to study the cultural characters in the laboratory based on the morphological and cultural characters by Booth (1971).

### Isolation of antagonist from the rhizosphere region

The *Trichoderma* was isolated from soil by using *Trichoderma* selective medium (TSM) developed by Elad and Chet (1983). Standard serial dilution technique was adopted for the isolation of antagonist from the rhizosphere region soil. Finally the dilutions of $10^{-5}$ and $10^{-6}$ were plated and plates were incubated at room temperature (28 ± $2^0$C) for five days and observed daily for appearance of *Trichoderma* colonies. The pure culture obtained was maintained in silica gel in refrigerator. Twenty one isolates of *Trichoderma* were obtained from the National Bureau of Agricultural important Insects (NBAII), Bengaluru to test their efficacy against the *F*. *oxysporum* f. sp. *gladioli* (FOG).

### Screening of the fungal antagonists against *f. oxysporum* f. sp. *gladioli in vitro*

Twenty eight *Trichoderma* isolates and one fluorescent pseudomonad isolate were screened against *F. oxysporum* f. sp. *gladioli* by following dual culture method (Dennis and Webster, 1971a). The radial growth of the pathogen was measured at fifth and seventh day and per cent inhibition over control was worked out according to the equation given by Vincent (1927).

### *In vivo* evaluation of bioagents against *fusarium oxysporum* f. sp. *gladioli*

The pots were filled with sterilized soil. Talc formulations of bioagents were delivered as seedling dip at the rate of 10 g per litre with spore count 4.1 x $10^5$ cfu/g (GJ16B), 4.8 x $10^5$cfu/g (IIHR 9), 4.7x$10^5$ cfu/g (HAR4B), 4.5 x $10^5$ cfu/g (IIHR FP1) and 4.9 x $10^5$ cfu/g (TN2A) of the formulation used.

The corms were planted in respective assigned pots. Each pot was planted with three corms of gladiolus. After 15 days of planting the mass multiplied pathogen was inoculated at rate of 2.6 x $10^6$ spores/ml to root zone and properly mixed. Regular observations were recorded.

### Trial on the management of gladiolus wilt with carbendazim tolerant *trichoderma harzianum* isolates

A preliminary trial with carbendazim tolerant *Trichoderma harzianum* isolate (NBAIR, GJ16B) on the management of gladiolus wilt was carried out. The isolate was developed at NBAIR and the same was tested as soil application and corm treatment. The experiment was carried under natural field conditions. Three replications were maintained for each treatment. Talc formulation of the bioagents was used for treating the corms and soil application. Germination percentage and per cent wilt incidence was recorded.

### Biocontrol mechanism studies

### Competition

The dual culture method described earlier was followed to study the competition between selected *Trichoderma* isolates (GJ16B, IIHR 9, HAR 4B and TN2A) against FOG and the data was recorded at fifth and seventh day and the per cent inhibition was calculated and expressed in percentage.

### Antibiosis production of volatile inhibitory compounds by *trichoderma* against *fusarium oxysporum* f. sp. *gladioli*

The test was carried out by slightly modifying the sealed Petri plate technique described by Dennis and Webster (1971b). For this experiment two Petri dish bases (90 mm) containing 20 ml of solidified two per cent malt agar medium were sealed together by an adhesive tape and the candidate fungus (antagonistic fungus) was grown in the bottom plate corresponding upper plate was inoculated with the pathogen and incubated at 25 ± $2^0$C, allowing the medium on the upper plate to absorb any volatile compounds produced by the candidate fungus and radial growth of pathogen was recorded after third, fifth and seventh day compared with the growth in control plates. Based on the result, percentage inhibition of pathogen was calculated.

### Non-volatile compound estimation

The production of non-volatile substances by *Trichoderma* isolates against FOG isolate was studied using the method described by Dennis and Webster (1971b). *Trichoderma* isolates were inoculated in double strength 100 ml sterilized potato dextrose broth (PDB) in 250 ml conical flasks and incubated at 26 ± 2 $^0$C for ten days. The control flasks with single strength of potato dextrose broth were not inoculated with any of the culture. The *Trichoderma* culture was filtered through Whatmann filter paper for removing mycelial mat after ten days and sterilized by passing through 0.2 µm pore biological membrane filter.

Later the filtered potato dextrose broth was inoculated with the FOG isolate and control flask were inoculated with the pathogen and incubated at 26±2 $^0$C for seven days. The mycelium mats of all the flasks were collected separately and the dry mycelial weight of all were taken to know the per cent reduction of growth of the pathogen over control.

### Mycoparasitism of *Trichoderma* on *F. oxysporum* f. sp. *gladioli*

Dual culture method was followed by placing the selected isolates of *Trichoderma* on one end of the petri plate and the other end of the plate was inoculated with *Fusarium oxysporum* f. sp. *gladioli*. Mycelial samples were carefully taken from the interaction region of both the fungi, were fixed on the slides very carefully without disturbing the mycelial mat and observed under microscope

for the presence of coiling structures and they were photographed (Watanabe, 1984).

## Results and Discussion

Total of nine isolates of *Trichoderma* spp. and one fluorescent pseudomonad were isolated from the rhizosphere region of soil. From dual culture method it was found that the *Trichoderma harzianum* isolates IIHR9, TN2A, GJ16B and HAR4B, IIHRFP1 fluorescent pseudomonad isolate were found to be best, they were further evaluated i*n vivo* against FOG.

### *In vivo* evaluation of bioagents against *F. oxysporum* f. sp. *gladioli*

The experimental results of evaluation of bioagents against *F. oxysporum* f. sp. *gladioli* revealed there was significant effect of bioagents in controlling of *Fusarium* wilt. No wilting symptoms was observed in any plants treated with *Trichoderma* isolates while wilt incidence was 44.44 per cent in fluorescent pseudomonad treated plants with pathogen inoculation and 77.77 per cent in control inoculated only with pathogen. Similarly, *Fusarium* crown rot of tomatoes was reduced by 80 per cent when *T. harzianum* was applied as seed coating which also resulted in enhanced yield (Sivan *et al.,* 1987). *T. viride* (T-1) strain was used by Marois and Locke (1985) for controlling of the *Fusarium* wilt of chrysanthemum. The result indicated 50 per cent reduction in wilt disease caused by *F. oxysporum* f. sp. *chrysanthemi.*

Maximum plant height was observed in *Trichoderma* isolate GJ16B (109.91 cm) which was significantly superior over all treatments in experiment of without pathogen inoculation. Maximum plant height in experiments inoculated with pathogens was observed in isolate GJ16B (109.20 cm) which was statistically on par with isolate TN2A (108.44 cm). The least plant height was observed in isolate IIHR FP 1 (105.11 cm). The increased growth response induced by *Trichoderma* spp. is not fully understood. However, several possible mechanisms have been suggested to explain the phenomenon increase of plant growth. These factors may include (i) control of deleterious root microorganisms those were not causing obvious diseases (ii) direct production of growth stimulating factors, (iii) increased nutrient uptake through enhanced root growth or promoted availability of necessary nutrients, (iv) reduction in the concentrations of substances in soil that are inhibitory to plant growth (Sivan and Harman, 1991).

Spike length was highest in isolate GJ16B (107 cm) which was statistically on par with control plants (106.5 cm) without inoculation of pathogen and carbendazim treated plants (106.4 cm). The lowest stalk length was observed in isolate IIHR FP1 (101.25 cm). The maximum stalk length was observed in treatment GJ16B with pathogen inoculation (106.23 cm) and the least stalk length was observed in control plants (100.15 cm) with only pathogen. There was no effect of bioagents on floral characters like number of florets per spike, floret diameter and floret length and were found to be non-significant in both pathogen inoculated treatment and without any inoculation of pathogen. The details of parameters of different treatments were presented in table 1 and 2.

### Trial on the management of gladiolus wilt with carbendazim tolerant *Trichoderma harzianum* isolate

The results of trial on management of FOG revealed that there was significant difference observed between the treatments for wilt incidence and data is given in table 3. The maximum germination percentage was observed in *T. harzianum* isolate GJ16B

(96%), which was statistically on par with treatments of carbendazim + *Trichoderma* (94%), carbendazim (92%) and hot water treatment (92%). The least germination percentage was observed in treatment control (84%) which was statistically on par with treatment of hot water + carbendazim (85%).

Wilt incidence due to *Fusarium* was ranging from 4 to 18 per cent under field condition in the preliminary trial and wilt incidence varied significantly among the treatments. The highest wilt incidence was observed in control (18%), and lowest wilt incidence was observed in treatment of isolate GJ16B (4.1%) which was statistically on par with treatment of hot water + *Trichoderma* (4.3%) and carbendazim + *Trichoderma* (5.7%).

## Biocontrol mechanism studies

## Computation

Four isolates of *T. harzianum*, were tested for their antifungal activity against *F. oxysporum* f. sp. *gladioli*. There was significant variation among the isolates was observed. The highest per cent inhibition of radial mycelial growth was observed in isolate GJ16B (65.83%) which was statistically on par with isolate TN2A (65.24%), followed by isolate IIHR 9 (58.63%) and HAR4B (55.04%). The per cent inhibition ranged 55.04 to 68.83 per cent and data is presented table 4.

The results were in agreement with that of Mishra *et al.,* 2004 were they studied, *T. virens* isolates against *Fusarium oxysporum* f. sp. *gladioli* were analysed by dual plate method, the growth of *Trichoderma* was significantly faster than FOG and it was capable of growing over FOG colonies by its competitive nature. Isolate GJ16B showed highest inhibitory effect on FOG by inhibiting the radial mycelial growth. The rapid growing ability of *Trichoderma* may probably provide an advantage in competing for nutrients and space utilization. The role of competition for nutrient and space in suppressing the populations of *Fusarium* species has been explained by other researchers (Cugudda and Garibaldi, 1987; Widden and Scattolin, 1988).

## Efficacy of volatile compounds produced by *Trichoderma* spp. against *F. oxysporum* f. sp. *gladioli*

Volatile compounds released from *T. virens* were studied by Mishra *et al.,* (2004), where significant inhibition of colony growth of the *F. oxysporum* f. sp. *gladioli* on PDA was observed upto a tune of 53 per cent. A comparison between the inhibitory effects of volatile metabolites in the present study revealed that the maximum inhibition of *F. oxysporum* f. sp. *gladioli* by volatile compound production was observed by isolate GJ16B (40.21%) and was statistically on par with isolate HAR4B (37.50%). The minimum inhibition of 23.91 per cent was observed with isolate IIHR 9 on third day. On fifth day the isolate GJ16B was reduced the pathogen growth by 36.61 per cent and was statistically superior over other isolates. Isolate TN2A was the least effective with 21.28 per cent inhibition. On seventh day results showed that the isolate HAR4B was found most effective with 55.20 per cent reduction in the mycelial growth and was statistically on par with isolate GJ16B (52.80%). Isolate TN2A showed 48.00 percent reduction which was statistically on par with IIHR 9 isolate (47.20%) and the data were presented in table 5.

The results indicated that the volatile metabolites seemed to be more effective in the antagonistic mechanism. Such findings were also reported by El-Katatny *et al.,* (2001) who explained the biocontrol efficacy of *T. harzianum* (T24) against *Sclerotium rolfsii* due to over production of volatile

metabolites. Inhibitory effects through volatile compounds were evident with *T. harzianum*, *T. viride* and *T. virens* against soil borne fungal pathogens identified by Mukherjee and Tripathi (2000).

**Effect of non-volatile metabolites produced by selected *trichoderma* isolates against *F. oxysporum* f. sp. *gladioli***

The selected isolates of *Trichoderma* were evaluated for the production of non-volatile compound production and its effect on the FOG growth was done has described earlier. The dry mycelial weight was taken and compared with the control. The data obtained on weight of dry mycelial growth of FOG was presented in the table 6.

All the selected isolates produced non-volatile compounds and inhibited the growth of gladiolus *Fusarium*. The maximum dry mycelial weight was observed in control. The maximum dry mycelial weight of *F. oxysporum* f. sp. *gladioli* (1.096 g) was observed in control and minimum mycelial weight was observed in isolate GJ16B (0.4790 g) with 56.29 per cent reduction of mycelial weight over control and was statistically on par with isolates IIHR 9 (0.5744 g) and TN2A (0.6870 g). Dennis and Webster (1971b) showed the effect of diffusible antibiotic produced by *Trichoderma* spp. against a wide range of fungal pathogens. The antibiotics produced by different *Trichoderma* spp. varied from isolate to isolate, which were toxic to pathogen and inhibited the growth of pathogen. Kamala and Indra (2011) showed the inhibitory effect of non-volatile compounds produced by indigenous *Trichoderma* isolates, and observed that out of 20 isolates used for study only five isolates were able to produce non-volatile compounds and significantly inhibited the growth of pathogen.

**Table.1** Effect of talc formulated *Trichoderma* isolates and fluorescent pseudomonad isolate on plant growth parameters in gladiolus under pot culture conditions

| Treatment | Plant height at harvest (cm) | Days to bud emergence | Days taken for first floret opening | Spike length (cm) | No. of florets per spike | Floret diameter (cm) | Floret length (cm) |
|---|---|---|---|---|---|---|---|
| ***Trichoderma harzianum* isolate GJ16B** | 109.91$^{a}$ | 71.13$^{d}$ | 83.00$^{de}$ | 107.00$^{a}$ | 18.29 | 10.61 | 10.36 |
| ***Trichoderma harzianum* isolate IIHR 9** | 106.81$^{d}$ | 74.85$^{a}$ | 84.77$^{b}$ | 104.28$^{b}$ | 18.08 | 10.50 | 10.30 |
| ***Trichoderma harzianum* isolate HAR4B** | 107.63$^{c}$ | 74.50$^{ab}$ | 84.35$^{bc}$ | 104.57$^{b}$ | 18.16 | 10.53 | 10.33 |
| **fluorescent pseudomonad isolate IIHR FP1** | 107.00$^{d}$ | 74.00$^{b}$ | 86.50$^{a}$ | 101.25$^{c}$ | 18.04 | 10.34 | 10.35 |
| ***Trichoderma harzianum* isolate TN2A** | 108.75$^{b}$ | 73.00$^{c}$ | 84.15$^{bc}$ | 106.51$^{a}$ | 18.10 | 10.67 | 10.32 |
| **Carbendazim @ 0.1%** | 108.41$^{b}$ | 72.43$^{c}$ | 83.74$^{cd}$ | 106.40$^{a}$ | 18.00 | 10.44 | 10.32 |
| **Thiram + Carboxin @ 0.1%** | 108.45$^{b}$ | 72.86$^{c}$ | 84.82$^{b}$ | 106.58$^{a}$ | 18.05 | 10.50 | 10.35 |
| **Control** | 108.33$^{b}$ | 71.00$^{d}$ | 82.67$^{e}$ | 106.50$^{a}$ | 18.58 | 10.33 | 10.39 |
| **SEm ±** | 0.20 | 0.23 | 0.26 | 0.23 | 0.56 | 0.39 | 0.39 |
| **CD @ 5%** | 0.59 | 0.66 | 0.76 | 0.68 | NS | NS | NS |

**Table.2** Effect of talc formulated *Trichoderma* isolates and fluorescent pseudomonad isolate on plant growth parameters in gladiolus under pot culture conditions in presence of virulent pathogen

| Treatment | Plant height at harvest (cm) | Days to bud emergence | Days taken for first floret opening | Spike length (cm) | No. of florets per spike | Floret diameter (cm) | Floret length (cm) | % Wilt incidence |
|---|---|---|---|---|---|---|---|---|
| ***Trichoderma harzianum* isolate GJ16B + FOG SOLAN** | 109.20$^{a}$ | 70.20$^{b}$ | 82.20$^{d}$ | 106.00$^{ab}$ | 18.27 | 10.56 | 10.27 | 0 (0.28) |
| ***Trichoderma harzianum* isolate IIHR 9 + FOG SOLAN** | 106.13$^{cd}$ | 71.80$^{a}$ | 83.00$^{b}$ | 104.18$^{c}$ | 18.04 | 10.30 | 10.18 | 0 (0.28) |
| ***Trichoderma harzianum* isolate HAR4B + FOG SOLAN** | 106.71bcd | 71.80$^{a}$ | 82.30$^{cd}$ | 103.74$^{c}$ | 18.09 | 10.31 | 10.22 | 0 (0.28) |
| **Fluorescent pseudomonad isolate IIHR FP1 + FOG SOLAN** | 105.11$^{d}$ | 71.00$^{ab}$ | 82.88$^{bc}$ | 100.15$^{d}$ | 18.02 | 10.40 | 10.26 | 44.44 (41.80) |
| ***Trichoderma harzianum* isolate TN2A + FOG SOLAN** | 108.44$^{a}$ | 71.80$^{a}$ | 84.00$^{a}$ | 105.58$^{ab}$ | 18.10 | 10.60 | 10.20 | 0 (0.28) |
| **Carbendazim @ 0.1% + FOG SOLAN** | 107.99$^{ab}$ | 71.00$^{ab}$ | 83.40$^{ab}$ | 105.60$^{ab}$ | 18.00 | 10.56 | 10.20 | 0 (0.28) |
| **Thiram + Carboxin @ 0.1% + FOG SOLAN** | 107.71$^{abc}$ | 71.00$^{ab}$ | 83.20$^{b}$ | 105.48$^{b}$ | 18.03 | 10.50 | 10.25 | 0 (0.28) |
| **FOG SOLAN** | 108.11$^{ab}$ | 68.6$^{c}$ | 80.40$^{e}$ | 106.23$^{a}$ | 18.44 | 10.39 | 10.32 | 77.77 (61.87) |
| **SEm ±** | 0.59 | 0.39 | 0.23 | 0.25 | 0.55 | 0.39 | 0.47 | 0.11 |
| **CD @ 5%** | 1.70 | 1.12 | 0.65 | 0.72 | NS | NS | NS | 0.35 |

**Table.3** Field trial data of carbendazim tolerant *Trichoderma harzianum* isolate GJ16B

| Treatment | % germination | % wilt incidence |
|---|---|---|
| **Control** | 84.00[a] | 18.00[c] (25.09) |
| **Hot water treatment** | 92.00[b] | 9.60[b] (18.04) |
| **Carbendazim** | 92.00[b] | 7.60[b] (15.99) |
| **Trichoderma harzianum** | 96.00[b] | 4.10[a] (11.68) |
| **Hot water + Trichoderma** | 88.00[ab] | 4.30[a] (11.96) |
| **Carbendazim + Trichoderma** | 94.00[b] | 5.70[ab] (13.80) |
| **Hotwater + carbendazim** | 85.00[a] | 8.60[b] (17.05) |
| **SEm±** | **1.72** | **1.99** |
| **CD @ 5%** | **5.20** | **6.01** |

Values following same letter don't differ significantly at (P = 0.05) by DMRT

**Table.4** Growth of *F. oxysporum* f. sp. *gladioli* and bioagents in dual culture

| ***Trichoderma* isolates** | **5th Day** | | | **7th Day** | | |
|---|---|---|---|---|---|---|
| | ***Trichoderma* (cm)** | **FOG (cm)** | **Per cent reduction in FOG over control** | ***Trichoderma* (cm)** | **FOG (cm)** | **Per cent reduction in FOG over control** |
| **GJ 16 B** | 7.15[a] | 1.44[c] | 59.10 | 8.25[a] | 1.90[d] | 65.83 |
| **TN 2 A** | 6.85[b] | 1.50[c] | 57.39 | 8.00[b] | 2.10[cd] | 62.24 |
| **HAR 4 B** | 6.70[b] | 1.98[b] | 43.75 | 7.80[c] | 2.50[b] | 55.04 |
| **IIHR9** | 6.24[c] | 2.20[b] | 37.50 | 7.50[d] | 2.30[bc] | 58.63 |
| **Control** | - | 3.52[a] | - | - | 5.56[a] | - |
| **SEm ±** | **0.09** | **0.08** | | **0.05** | **0.08** | |
| **CD at 1%** | **0.38** | **0.37** | | **0.27** | **0.36** | |

Values following same letter don't differ significantly at (P = 0.01)

**Table.5** Influence of volatile compounds released by selected *Trichoderma* isolates *in vitro* on the growth of *F. oxysporum* f. sp. *gladioli*

| ***Trichoderma* isolate** | **Radial growth of *F. oxysporum* f. sp. *Gladioli*** | | | | | |
|---|---|---|---|---|---|---|
| | **3rd day** | | **5th day** | | **7th day** | |
| | **Growth (cm)** | **% reduction over control** | **Growth (cm)** | **% reduction over control** | **Growth (cm)** | **% reduction over control** |
| ***Trichoderma harzianum* GJ16B isolate (NBAIR)** | 1.10[d] | 40.21 | 2.25[d] | 36.61 | 2.95[c] | 52.80 |
| ***Trichoderma harzianum* TN2A isolate (NBAIR)** | 1.3[bc] | 29.34 | 2.80[b] | 21.12 | 3.25[b] | 48.00 |
| ***Trichoderma harzianum* HAR 4B isolate (NBAIR)** | 1.15[cd] | 37.5 | 2.5[c] | 29.57 | 2.8[c] | 55.20 |
| ***Trichoderma harzianum* IIHR 9 isolate (native isolate)** | 1.4[b] | 23.91 | 2.65[bc] | 25.35 | 3.3[b] | 47.20 |
| **Control** | 1.84[a] | - | 3.55[a] | - | 6.25[a] | - |
| **SEm ±** | **0.06** | | **0.08** | | **0.05** | |
| **CD at 1%** | **0.21** | | **0.34** | | **0.26** | |

Values following same letter don't differ significantly at (P = 0.01)

**Table.6** Effect of non-volatile compounds produced by selected isolates of *Trichoderma* on growth of *F. oxysporum* f. sp. *gladioli*

| *Trichoderma* isolates | Dry mycelial weight of FOG (g/100 ml) | Per cent reduction over control |
|---|---|---|
| ***Trichoderma harzianum* GJ16B isolate (NBAIR)** | 0.4790 | 56.29 |
| ***Trichoderma harzianum* TN2A isolate (NBAIR)** | 0.6870 | 37.31 |
| ***Trichoderma harzianum* HAR4B isolate (NBAIR)** | 0.7220 | 34.12 |
| ***Trichoderma harzianum* IIHR9 isolate (native isolate)** | 0.5744 | 47.59 |
| **Control** | 1.096 | - |
| **SD** | **0.232** | - |

**Studies on mycoparasitism of selected isolates of *Trichoderma* on *F. oxysporum* f. sp. *dianthi* and *F. oxysporum* f. sp. *gladioli***

The control of *Fusarium* wilt by *Trichoderma* isolates might be attributed to the pronounced colonization of rhizosphere by *Trichoderma* spp. in advance of the pathogen and also by mycoparasitism (Papavizas and Lewis, 1989). Similarly in the present investigation a strong coiling of FOG isolate by *Trichoderma* isolate GJ16B was observed. *Trichoderma* penetrated into *Fusarium* mycelium with its mycoparasitic ability and no coiling was observed by other *Trichoderma* isolates around FOG mycelium. The mycelia interaction and coiling between FOG mycelium and GJ16B was shown in plate 1.

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