EFFECT OF VOLATILE AND NON VOLATILE COMPOUNDS OF TRICHODERMA SPP. AGAINST FUSARIUM ISOLATES CAUSING CHICKPEA WILT IN PUNJAB

Mohit Kumar, Vipul Kumar, Meenakshi Rana and Seweta Srivastava*

School of Agriculture, Lovely Professional University, Phagwara - 144 411, Punjab, India

Abstract

Background and Objective: Antagonistic strains belonging to the Trichoderma genera were able to produce various secondary metabolites which can play a role in the mechanism of their biological activity. So, the aim of this study was to assess the potential of volatile and non-volatile metabolites released from three selected species of Trichoderma viz. Trichoderma viride, Trichoderma harzianum and Trichoderma konigii isolates against three isolates of Fusarium oxysporum f. sp. ciceri which caused wilting of chickpea.

Materials and Methods: After isolation and identification of the experimental microbes, the effect of selected antagonists was checked against the test pathogen. In vitro bio-efficacy test of antagonists’ viz. Trichoderma viride, Trichoderma harzianum and Trichoderma konigii had been done against Fusarium oxysporum f. sp. ciceri with the help of dual culture and their effects were studied by Zone of Inhibition technique. The production of volatile compounds by the same isolates of antagonists against Fusarium oxysporum f. sp. ciceri was studied by using the inverted plate technique.

Results & Conclusion: Data proved that Trichoderma harzianum produced maximum inhibition zone (76.90%) against FOC strain of Fusarium oxysporum f. sp. ciceri followed by Trichoderma viridae (70.10%). In vitro studies have demonstrate that volatile compounds produced by Trichoderma harzianum showed strong inhibitory effect on the mycelial growth of all isolates of test pathogen but maximum against FOC2 isolate i.e., 79.25% followed by the Trichoderma viridae i.e., 64.16% against FOC1 isolate.

Key words: Antagonist, chickpea, Trichoderma, strain, metabolites, wilt

Introduction

Chickpea (Cicer arietinum L.) is an annual grain legume, grown mainly for human consumption. Susceptibility to several fungal, bacterial and viral diseases are the limiting factor contributing for the low yield of chickpea (Rehman et al., 2013). Among the economically important diseases, wilt (Fusarium oxysporum f. sp. ciceris), dry root rot (Rhizoctonia bataticola) and collar rot (Sclerotium rolfsii) are the major and widespread diseases affecting chickpea cultivation and production (Nene and Sheila 1999). Use of bio-control agents to manage the disease represents a viable alternative in place of chemical fungicides because it is considered to be a safe without having any residual effect, cost effective and eco-friendly method for plant disease management (Benitez et al., 2004).

Among all beneficial microbes, Trichoderma has long been considered as one of the most promising biocontrol agent uses to control several plant pathogens because it is produced many antifungal secondary metabolites that have an adverse affect on the growth of different fungal and bacterial phytopathogens (Barakat et al., 2014; Li et al., 2016). Production of antibiotic and cell wall degrading enzymes, competition for key nutrients, parasitism and stimulation of plant defense mechanisms are numerous modes of action have been proposed to explain the bio-control of plant pathogens by Trichoderma (Cook, 1985). Trichoderma spp. generally grows in its natural habit around the rhizosphere zone of host plant and therefore it controls root or soil borne diseases in particular (Faruk et al., 2002; Kamlesh and Gujar, 2002, Monte 2001). The species of Trichoderma have been evaluated against
the wilt pathogen and have exhibited greater potential in managing chickpea wilt under field condition (Podder et al., 2004).

*Fusarium* sp. is a soil borne fungal pathogen that attacks plants through roots at all stages of plant growth, is considered as one of the main soil-borne systemic diseases and the major limiting factor in the production of many crops both in greenhouse and field-grown (Srivastava et al., 2010; Borrero et al. 2004). In India, chickpea is ranked first in terms of production and consumption in the world (Patole et al., 2017). *Fusarium* wilt caused by the soilborne fungus *Fusarium oxysporum* f. sp. *ciceri*, has become a major limiting factor of chickpea production worldwide (Jiménez-Díaz et al., 2015).

Considering these points, the aim of this study was to assess the potential of volatile and non-volatile metabolites released from three selected species of *Trichoderma* viz. *Trichoderma viride*, *Trichoderma harzianum* and *Trichoderma konigii* isolates against three isolates of *Fusarium oxysporum* f. sp. *ciceri* which causes wilting of chickpea.

### Materials and Methods:

#### Cultural and morphological characters of *Fusarium* isolates

Isolations of *F. oxysporum* f. sp. *ciceri* was isolated from chickpea wilt samples collected from adjoining farmer’s field. Isolation was made from infected root samples and pure culture was maintained on Potato Dextrose Agar medium slants (Aneja, 2005).

#### Effect of non-volatile compounds produced by *Trichoderma* species on the mycelial growth of *Fusarium* isolates

Bio-efficacy test of antagonists has been done against *Fusarium oxysporum* f. sp. *ciceri* with the help of dual culture\(^1\). The effects of antagonists were studied by Zone of Inhibition technique (in vitro). The three antagonists were *Trichoderma viride*, *Trichoderma harzianum* and *Trichoderma konigii*.

#### Results

### Effect of Volatile compounds:

The results for volatile metabolites activity against *Fusarium* isolates were presented in table 1. After 7 days of incubation, the effect of volatile compounds was

| Table 1: Effect of different species of *Trichoderma* on the growth of *Fusarium oxysporum* f. sp. *ciceri* |
|-----------------|-----------------|-----------------|-----------------|-----------------|
| *Trichoderma* spp. | *Fusarium oxysporum* f. sp. *ciceri* | *FOC* | *FOC1* | *FOC2* |
| Radial average of growth pathogen | Per cent inhibitions of mycelial growth % | Radial average of growth pathogen | Percent inhibitions of mycelial growth % | Radial average of growth pathogen | Percent inhibitions of mycelial growth % |
| *Trichoderma viride* | 26.7 | 70.1 | 29.4 | 45.14 | 33.6 | 37.07 |
| *Trichoderma harzianum* | 20.6 | 76.9 | 23.6 | 55.97 | 25.2 | 52.80 |
| *Trichoderma konigii* | 36.7 | 58.9 | 27.3 | 49.06 | 27.2 | 49.06 |
| *Control* | 89.4 | 53.6 | 53.4 |
observed among three species of *Trichoderma* viz. *Trichoderma harzianum, Trichoderma viride* and *Trichoderma koningi*. *Trichoderma harzianum* exhibited maximum growth inhibition (79.25%) against the tested three isolates of *Fusarium* when compared to the others. The *Trichoderma viride* and *Trichoderma koningi* exhibited growth inhibition of 62.27% and 50%, respectively.

**Evaluation of antagonistic activity through production of antifungal non-volatile metabolites:**

The ability of *Trichoderma* species to produce the non-volatile substances was found most efficient in reducing the highest mycelial growth of tested *Fusarium* isolates by 76.9% and least recorded in *T. viride* (70.1%) followed by *T. koningi* (58.1%). From our results it is evident that among the three species of *Trichoderma, Trichoderma harzianum* is most potential species which could be better for soil borne pathogens for control the disease (table 2).

**Table 2:** To check the secondary metabolites of *Trichoderma* spp. against FOC, FOC 1, and FOC 2

<table>
<thead>
<tr>
<th>Trichoderma spp.</th>
<th>Growth inhibition (%) of <em>Fusarium oxysporum</em> f. sp. <em>ciceri</em></th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>FOC</td>
<td>FOC 1</td>
</tr>
<tr>
<td>Trichoderma viride</td>
<td>56.61</td>
<td>64.16</td>
</tr>
<tr>
<td>Trichoderma harzianum</td>
<td>64.16</td>
<td>67.93</td>
</tr>
<tr>
<td>Trichoderma koningi</td>
<td>44.23</td>
<td>51.93</td>
</tr>
</tbody>
</table>

The non-volatile substances secreted by *Trichoderma harzianum* during the experiment was found most efficient in reducing the highest mycelial growth of tested *Fusarium* isolates by 76.9%. Due to the presence of non-volatile substances, soil application with *T. hamatum, T. harzianum* or *T. viride* checked the severity of wilt and root rot disease as effectively as carbenazim (Khan et al., 2014). This *Trichoderma* spp. are active colonizers in soil (Akrami et al. 2009) and produce antibiotics like trichodermin, gliotoxins, viridin, cell wall-degrading enzymes (Bruckner and Przybylski 1984), and certain biologically active heat-stable metabolites like ethyl acetate (Mohiddin et al., 2010). These substances may inhibit the activity of soil-borne pathogens (Chet and Baker 1981; Khan et al., 2004; Khan et al., 2011).

Further, in addition to biological control of soil borne fungal pathogens seed inoculation of *Trichoderma* spp. also found to increase growth and yield of chickpea (*Cicer arietinum* L.) under greenhouse conditions (Rudresh et al., 2005).

**Conclusion**

As we know that use of bio-control in plant disease management is more effective, cheap without any residual effect. So, from the above findings it was concluded that non-volatile substances produced by the isolates of *Trichoderma* was found most efficient than volatile substances in reducing the highest mycelial growth of tested *Fusarium* isolates. It is also evident from above results that among the three species of *Trichoderma* used in present experiment; *Trichoderma harzianum* is most potential species which could be better for the management of soil borne pathogens like *Fusarium oxysporum* f. sp. *ciceri*. The non-volatile compounds of *Trichoderma harzianum* is found as effective as carbenazim or other systemic fungicide used to manage wilt.

**References**


