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# Effect of Different Temperature and Culture Media on the Mycelia Growth of *Trichoderma viride* Isolates

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

Effect of different temperature and culture media was observed on all ten *Trichoderma viride* isolates. The excellent growth of *Trichoderma* was found at temperature range of 25–30˚C. All the 10 *Trichoderma* isolates were evaluated on four different liquid media for assessing growth and sporulation. The maximum mycelial weight was found in potato dextrose broth (276.00 mg) and the lowest Czapek Dox broth (96.00 mg).

## Introduction

*Trichoderma* sp. are free-living filamentous fungi that are common in soil and root ecosystems. They are opportunistic, avirulent plant symbionts, as well as being parasites of other fungi (Harman *et al.,* 2004). They are highly interactive in root, soil and foliar environments. They release secondary metabolites that induce localized or systemic resistance responses in plants. Root colonization by *Trichoderma* strains frequently enhances root growth and development, crop productivity, resistance to abiotic stresses and the uptake and use of nutrients (Arora *et al.,* 1992). The microbial inoculants as biocontrol agents are effective and attractive alternatives to prevent the deficiencies brought about by the exclusive reliance on chemicals (Nakkeeran *et al.,* 2002).*Trichoderma* sp. can be identified based on the morphology and colour of the colonies obtained on the potato dextrose agar medium. Further identification can be confirmed on the basis of the morphology of the conidia and conidiophores of different *Trichoderma* spp. when viewed under a microscope. *Trichoderma* sp. has evolved numerous mechanisms that are involved in attacking other fungi. These mechanisms include competition for space and nutrients, mycoparasitism for production of inhibitory compounds, inactivation of the pathogen's enzymes and induced resistance. The potentiality of *Trichoderma* species has been greatly influenced by the physiological parameters like temperature, moisture, pH and nutrients. Temperature plays important role in

determining the effectiveness of *Trichoderma* against phytopathogens. Sharma *et al.*, (2005) also reported that media, pH and temperature showed profound effect on growth and sporulation of fungi. None of the *Trichoderma* species grew at 40˚C. Temperature has a significant influence on growth and sporulation of *Trichoderma* species. The most favorable temperature for *Trichoderma* species was found in between 25-30˚C where average growth recorded between (53-90mm diameter) (Singh and Kumar, 2009).

## Materials and Methods

An experiment was conducted in the Biocontrol Laboratory of department of plant pathology, C.S. Azad University of Agriculture and Technology Kanpur, to evaluate the performance of different pH, Temp and media for growth of *T. viride.*

### Effect of temperature on growth and sporulation of *T. viride*

Effect of temperatures on the mycelial growth of *Trichoderma viride* isolates was studied *in vitro* on autoclaved PDA medium (20ml) in 90mm Petri plates adjusted for each temperature. The actively 7 days old culture of *T. viride* isolates were inoculated by 5mm disc at the centre of the Petri plates with the help of sterile cork borer and three replications were taken for each treatment. The cultures were incubated at 20, 25, 30 and 35˚C in BOD incubator and daily observation on mycelial growth of *T. viride* was recorded at every 24 hours up to 7 days.

## Results and Discussion

### Effect of different temperatures on growth of *Trichoderma viride*

Present study was undertaken to find out the optimum as well as the best temperature for the growth of *Trichoderma viride* by growing the bio agent at different temperatures ($20^0$C, $25^0$C, $30^0$C and $35^0$C) on Potato Dextrose Agar medium. After 7 days of incubation the average radial growth (mm) was recorded and presented in table 1.

From the perusal of the data given in table 1 and it is evident that after 7 days of incubation at 20˚C, 25˚C and 30˚C temperature, average maximum radial growth 54.50 mm, 84.50 mm, 82.00 mm and 69.50mm, respectively was observed in case of 49CP isolate of *Trichoderma viride*. However, at 20˚C, 25˚C, 30˚C and 35˚C temperatures levels, the minimum radial growth was obtained as 41.00mm, 67.50mm, 65.00mm and 53.00mm respectively observed in case of isolate 117CP .Thus, it can also be concluded from table 1 that the growth of *Trichoderma viride* continues progressing up to 30˚C and thereafter the growth starts decreasing. The growth of the bio-agent significantly affected with either increase or decrease in temp. The statistical analysis shows that isolate (49CP) exhibited the highest growth which was significantly superior to other isolates. Rest of the isolates was at par with each other. However, the excellent growth was recorded at temperature range between 25-30˚C.

### Growth of *Trichoderma* isolates on different liquid media

Studies were carried out to determine the average weight of the dry mycelial mat produced by *T. viride* isolates on four different liquid media including Potato Dextrose Broth, Sabouraud Broth, Corn Meal Broth and Czapek Dox Broth in three replications. Average dry mycelial weight was taken as a standard value for comparison of growth for different treatments. The growth was observed and the data summarized in table 2. Among the all ten tested isolates of *Trichoderma viride*, 49CP isolate supported significant growth and the highest mycelial

weight was recorded in Potato Dextrose Broth (276.0mg) followed by Sabouraud Broth (236.0mg), Corn Meal Broth (156.5mg) and Czapek Dox Broh (96.0mg). The excellent sporulation was observed in PDB, good sporulation in Sabouraud Broth, fair sporulation was observed on Corn Meal Broth. However, poor on mycelial weight was observed in case of Czapek Dox Broth. The growth of *Trichoderma viride* was studies at four different temperatures such as 20˚C, 25˚C and 30˚C. The maximum radial growth and sporulation was found at 25˚C (84.50mm) and minimum at 20˚C (41.00mm) by 49CP and 117CP isolates respectively. Thus, it can also be concluded the growth of *Trichoderma viride* continues progressing up to 30˚C and thereafter the growth starts decreasing. However, the excellent growth was recorded at temperature range between 25-30˚C.

**Table.1** Growth of *Trichoderma viride* isolates at different temperature

| ***T.viride* isolates** | **Radial growth of mycelium (mm)** | | | |
|---|---|---|---|---|
| | **$20^0$C** | **$25^0$C** | **$30^0$C** | **$35^0$C** |
| **02CP** | 50.500 | 75.500 | 72.000 | 61.500 |
| **10CP** | 51.500 | 77.500 | 75.500 | 63.500 |
| **13CP** | 47.500 | 73.000 | 70.500 | 60.500 |
| **21CP** | 49.500 | 74.500 | 71.500 | 62.500 |
| **25CP** | 52.000 | 79.500 | 76.500 | 66.000 |
| **45CP** | 53.000 | 81.000 | 79.500 | 67.500 |
| **49CP** | **54.500** | **84.500** | **82.000** | **69.500** |
| **64CP** | 45.000 | 71.000 | 69.500 | 59.500 |
| **70CP** | 43.500 | 69.000 | 67.000 | 55.000 |
| **117CP** | 41.000 | 67.500 | 65.000 | 53.000 |
| **CD @ 5%** | **3.111** | **4.788** | **5.293** | **4.919** |

**Table.2** Effect of liquid media on growth and sporulation of *T. viride*

| ***T. viride* isolates** | **Dry weight of mycelium(mg)** | | | |
|---|---|---|---|---|
| | **PDB** | **SMB** | **CMB** | **CDB** |
| **02CP** | 267.00 | 222.00 | 141.00 | 92.50 |
| **10CP** | 267.50 | 223.50 | 144.00 | 90.00 |
| **13CP** | 258.00 | 217.50 | 139.50 | 50.00 |
| **21CP** | 262.50 | 218.00 | 145.50 | 85.50 |
| **25CP** | 269.00 | 228.50 | 148.00 | 94.50 |
| **45CP** | 273.50 | 230.50 | 151.50 | 55.50 |
| **49CP** | **276.00** | **236.00** | **156.50** | **96.00** |
| **64CP** | 258.00 | 214.50 | 136.00 | 50.50 |
| **70CP** | 253.50 | 213.00 | 138.50 | 54.50 |
| **117CP** | 248.00 | 210.00 | 131.00 | 37.50 |
| **CD @ 5%** | **7.620** | **5.269** | **4.761** | **8.059** |

The growth and sporulation of the bioagent *Trichoderma viride* was tested on four different liquid media such as Potato Dextrose Broth, Sabouraud Maltose Broth, Corn Meal Broth and

Czapek Dox Broh. The highest mycelial weight was recorded in Potato Dextrose Broth (276.0mg) and lowest Czapek Dox Broth mycelium dry weight (96.0mg). Potato dextrose broth was the best liquid media for growth and sporulation of *Trichoderma viride.* The experimental finding is well supported with Singh *et al.,* (2014) conducted experiments to determine the optimal parameters for the biomass production of *Trichoderma* sp. *in vitro* at different pH and temperatures. A significant difference in the biomass production was recorded among the species at tested pH levels i.e. 4.0, 4.5, 5.0, 5.5, 6.0, 6.5, 7.0, 7.5 and 8.0. The most favourable pH ranges between 5.5 and 7.5 in which total dry weight of mycelium varies between 1.41 and 1.35g. Although all the species of *Trichoderma* produced sufficient biomass at different temperatures viz. 20, 25, 30 and 35°C but they were found to be best grown at a temperature range of 25°C to 30°C. Srivastava *et al.,* (2014) conducted an experiment on physiological aspect of *Trichoderma sp.* against different liquid media, temp, pH, whereas PDB reveals the excellent average mycelium weight (190.9mg). The most favorable temperature for growth and sporulation of *Trichoderma harzianum* (*Th azad*) was found to be 30˚C followed by 25˚C. Similarly, most favorable pH 6.5-7.5 ranges was found in which total dry weight of mycelium also varies between 144.805-142.4mg. Maximum pH was found at 7.0 (162.5mg).

In conclusion, all the *Trichoderma* isolates were evaluated on four different liquid media for assessing growth and sporulation. The maximum mycelial weight was found in Potato Dextrose broth and the lowest Czapek Dox Broth.

Effect of different temperature and pH was observed on all ten *Trichoderma* isolates. The excellent growth of *Trichoderma* was found at temperature range of 25-30˚C, while 6.5 pH (Slightly acidic) was found the most favorable for the growth and sporulation of *Trichoderma* isolates.

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