

**“Identification of compatible trait of specific
Trichoderma strains for management of soil borne
diseases.”**



THESIS

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**Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya,
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by

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2021

CERTIFICATE - I

This is to certify that the thesis entitled “**Identification of compatible trait of specific Trichoderma strains for management of soil borne diseases.**” submitted in partial fulfillment of the Degree of **MASTER OF SCIENCE IN AGRICULTURE (Plant Pathology)** of Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior is a record of the bonafide research work carried out by **Ms. ANUJA PANDEY**, ID no. Awaited under my guidance and supervision. The subject of the thesis has been approved by the Student’s Advisory Committee and the Director of Instruction.

No part of the thesis has been submitted for any other degree or diploma or has been published. All the assistance and help received during the course of these investigations has been acknowledged by the scholar.

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CERTIFICATE – II

This is to certify that the thesis entitled “**Identification of compatible trait of specific *Trichoderma* strains for management of soil borne diseases.**” submitted by **Ms. ANUJA PANDEY** ID no. Awaited to the **Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior**, in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE IN AGRICULTURE** in the **Department of Plant Pathology** has been accepted after evaluation by the external examiner and approved by the Student’s Advisory Committee after on oral examination the same.

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Place: Indore

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Date:

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ABBREVIATION

| S.N | Legends | Description |
|-----|------------------|------------------------------------|
| 1 | S.N. | Serial number |
| 2 | CD | Critical difference |
| 3 | Cfu | Colony forming unit |
| 4 | Cm | Centimeter |
| 5 | d.f. | Degree of freedom |
| 6 | et al. | Allied (and other) |
| 7 | G | Gram |
| 8 | i.e. | That is(in reference to) |
| 9 | Kg | kilogram |
| 10 | mm. | Millimeter |
| 11 | Spp. & sp. | Species |
| 12 | Viz. | Wide list |
| 13 | ^o C | Degree Celsius |
| 14 | % | Percent |
| 15 | No. | Number |
| 16 | Fig. | Figure |
| 17 | PDA | Potato Dextrose Agar |
| 18 | TSM | <i>Trichoderma</i> specific medium |
| 19 | Ppm | Parts Per Million |
| 20 | gl ⁻¹ | Grams/liter |
| 21 | EC | Emulsifiable Concentrate |
| 22 | SL | Suspendable liquid |
| 23 | WG | Wetteble granules |
| 24 | WS | Water Soluble |
| 25 | MSS | Mean Sum of Square |
| 26 | SE(m)± | Standard error of mean |
| 27 | S.S. | Sum of Square |
| 28 | S.V. | Source of variance |
| 29 | CRD | Completely Randomized design |
| 30 | C.V. | Coefficient of variation |
| 31 | ANOVA | Analysis of variance |
| 32 | EMS | Error means sum of square |
| 33 | Cal. | Calculate |
| 34 | Tab. | Tabulated |

CHAPTER- I

INTRODUCTION

Pulses own significant value in the diet of the predominantly vegetarian population of India as they contain 2 to 3 times more protein than cereals. Chickpea (*Cicer arietinum*) family leguminaceae is the most important pulse crop of Indian sub-continent, is predominantly consumed as a pulse, dry chickpea and also used in preparation of variety of snacks, sweets and condiments and green fresh chickpea is commonly consumed as vegetable. It is grown especially in dry areas of the Indian subcontinent and Europe. Besides important as human food and animal feed, it also plays a major role in management of soil fertility. Nutritionally it is a rich source of protein 17-22%, carbohydrate 60-64% and contains 3-4% fat, vitamins and minerals (Anonymous, 2014). It's leaves possess mallic acid and citric acid which are very useful for stomach ailments and blood purification .India is the largest chickpea producer as well as consumer in the world, its distribution throughout the country, six states viz., Madhya Pradesh, Rajasthan, Maharashtra, Uttar Pradesh, Karnataka and Andhra Pradesh together contribute 92% of the production and 95% of the area in the country.

Chickpea (*Cicer arietinum* L.) is the third most important food crop occupying first rank in area as well as production amongst the pulses grown in India. It covers about 10.22 m ha area, 11.23 m tones annual production with per cent share in total production 44.51% in the country (Commodity Profile for Pulses-March, 2019). Madhya Pradesh is one of the major chickpea growing states the country, where it occupies an area of 3.48 m ha, 3.82 with m tones annual production (GOI, Anon, 2014).

Diseases are the most serious constraints to high chickpea productivity causing huge losses. Environmental factors and intensity of abiotic stresses are known to compound the occurrence and severity of diseases. Chickpea is subjected to infection by several fungi, among them Fusarium wilt incited by *Fusarium oxysporum* f. sp. *ciceri* (Padwick) is a more serious disease in most of chickpea growing countries with yield losses ranging from 10 to 90 % (Singh

and Dahiya, 1973). This disease was reported for the first time in India by Butler (1918). Role of *Fusarium oxysporum* f.sp. *ciceri* in causing vascular wilt has been reported from many countries of the world.

Biological control is one of the best low-cost and ecologically sustainable methods for managing plant diseases caused by soil-borne pathogens like *Fusarium*, *Rhizoctonia*, and *Pythium*. Among various biocontrol agents (BCAs) examined against the plant pathogenic fungi, *Trichoderma* spp. have been widely studied for their biocontrol ability (Abd El-Khairet *et al.*, 2010; Mohiddin *et al.*, 2010). However, their use as BCAs in agriculture is limited due to their unpredictable efficiency, which is affected by biotic and abiotic factors in the soil. *Trichoderma* spp. have demonstrated biocontrol potential greater than that of other BCAs because the success of *Trichoderma* as biocontrol agents (BCAs) is due to their high reproductive capacity, ability to survive under quite adverse conditions.

Trichoderma is a biocontrol agent incorporated into soil has to possess a high level of competence to survive in the soil and compete specially with soil inhabitant microorganisms. A high rate of proliferation and maintenance of viability for an extended period of time are characteristics that will contribute towards such competence. Various external factors such as temperature, pH, heavy metal ions and water availability are reported to have an influence on the growth, proliferation, and development as well as the biocontrol efficacy of *Trichoderma* spp. (Kredics *et al.* 2003).

Therefore, in the study, the effect of variable range of temperature and H ion concentration on sporulation and survival of *Trichoderma* sp., and compatibility of *Trichoderma* strains with pesticides was tested at Department of Plant Pathology laboratory and field, College of Agriculture, Indore (M.P.)

Objectives:-

1. Isolation of *Trichoderma* from the collected soil.

2. To assess the compatibility of *Trichoderma* strains in response of Fungicide, insecticide and weedicide.
3. To identified the *Trichoderma* strains tolerant to higher temperature and pH.
4. To evaluate the *Trichoderma* strains against soil borne diseases *in vitro* and *in vivo*.

CHAPTER- II

REVIEW OF LITERATURE

Studies on various isolates of *Trichoderma* spp. were undertaken on the aspects of their collection and isolation, compatibility with different agrochemicals, tolerance to higher temperature and H ion concentration and antagonistic potential against *Fusarium oxysporum* f. sp. *Ciceri* the pathogen of wilt of chickpea under *in vitro* and *in vivo* condition. Information on these aspects is reviewed and presented in this chapter.

The use of bio agents has opened a new avenue for management of soil and seed borne disease (Shivpuri et al., 1997, Sindhan et al., 1999) as a promising and possible alternative to the existing chemical control practices.

Trichoderma spp., are common inhabitant of soil and rhizosphere. They are highly interactive in root, soil and foliar environments. They produce or release a variety of compounds that induce localized or systemic resistance response in plants.

2.1 Isolation of *Trichoderma* strains from rhizospheric and non rhizospheric soil.

Sivakumar *et al.* (2000) isolated *Trichoderma* from soil samples collected from three Rambutan fields where leaf litter was allowed to accumulate. Soil suspensions were prepared by adding 1.0 g soil to 10 ml of sterile distilled water and shaking for 15 minutes, suspension was serially diluted to 10⁻⁵ and 0.1 ml of it was spread on a potato dextrose agar (PDA) plate which was then incubated at 28°C for 5 days.

Cigdem and Merih (2003) collected thirty-one soil samples from different agricultural fields and forests in Eskisehir and inoculated on potato dextrose agar, malt extract agar, rose bengal agar and oat flour agar and incubated at 28 °C for 5 days.

Mustafa *et al.* (2009) isolated three different *Trichoderma* species (*T. harzianum*, *T. viride*, and *T. longibrachiatum*) on five different culture media including Potato Dextrose Agar, Waksman agar, Agar-agar, Czepak's agar and Corn Meal agar. PDA was the best medium in terms of growth, spore production and biomass yield.

Rahman *et al.* (2011) isolated the *Trichoderma* species that were collected from different habitats and were quantified and characterised. The identified strains are *T. harzianum* (IMI-392432, 392433, 392434); *T. pseudokoningii* (IMI-392431) and *T. virens* (IMI-392430). Out of five strains, *T. harzianum* was the most common in all of the habitats. Colony forming units (cfu's) of *Trichoderma* species varied significantly ($P = 0.05$) in different habitats and were positively correlated with the physicochemical characteristics of the habitat. *Trichoderma* was found to have adapted to an acidic habitat.

Krishna Kumar *et al.* (2012) isolated twelve isolates of *Trichoderma* spp. isolated from different locations of South Andaman were characterized for their cultural, morphological and antagonistic activity against soil borne and foliar borne pathogens. The isolates revealed differential reaction patterns against the test pathogens viz., *Sclerotium rolfsii*, *Colletotrichum gloeosporioides* and *C. capsici*. However, the isolates, TND1, TWN1, TWC1, TGD1 and TSD1 were most effective in percentage inhibition of mycelial growth of test pathogens.

Pingolia *et al.* (2013) isolated *Trichoderma* spp. by use of different kind of agar medium like PDA and MEA for the growth of mycelium of *Trichoderma viride*. The best mycelium growth was observed in PDA with 2.5 gm glucose and 1.5gm lactose respectively. Similarly, while using MEA medium the best growth recorded is a good indicator of soil health.

2.2 Identified the *Trichoderma* strains tolerant to higher temperature and pH.

Jayaswal *et al* (2003) isolated the strains of *Trichoderma* and the strain that showed maximum growth and sporulation was of *T. viride* with pH range between 4.5 to 5.5. Temperature between 20°C and 37°C were good for both growth and sporulation of *T. viride*. At lower temperatures (i.e. below 20°C) growth and sporulation were inhibited.

Anuradha *et al.* (2014) isolated and studied seven species from the rhizospheric soils of chickpea; pigeon pea and lentil crops of different areas of an Indian State (Uttar Pradesh) and these were later tested *in vitro* at different pH, temperatures and varying agitation speed. The most favorable pH range was between 5.5 and 7.5 in which total dry weight of mycelium varies between 1.41 and 1.35 g. Although all the species of *Trichoderma* produced sufficient biomass at different temperatures viz. 20°C, 25°C, 30°C and 35°C.

Poosapati *et al.* (2014) isolated *Trichoderma* spp. that were collected from varied agro-climatic zones of India and screened for high temperature and salinity tolerance. Among all the isolates tested, *T. asperellum*, TaDOR673 was highly tolerant to heat shock of 52°C.

Domingues *et al.* (2016) conducted an experiment on the mycelial growth of the isolates of *T. asperellum* and *T. asperelloides*, as well as of *S. minor* and *S. Sclerotiorum*. The strains were evaluated at temperatures ranging from 7 to 42°C. The isolates of *Trichoderma* grew at temperatures ranging from 12 to 37°C with maximum growth at 27°C.

Petrisor *et al.* (2016) conducted an experiment to study the effect of temperature and pH on the radial growth rate and biomass yield of two strain of *Trichoderma* (Td85 and Td50) with high biocontrol potential. The most favourable pH range was between 4.5 and 5.5 in which biomass yield varies between 1.5 g and 2.46 g and both the both strains studied and grew better at 25-30°C and very slow at 15 °C.

Zehra *et al.* (2017) conducted an experiment on *Trichoderma* to study the effect of temperature and pH. The favourable temperature for growth and sporulation of *T. harzianum* and *T. Viride* was found in between 25-40°C, whereas for *T. asperellium* and *T.hamatum* was 25-35°C, Similarly the most favourable range of pH was in between 4.6-7.6 for all the four spp. of *Trichoderma*.

Abeyratne *et al.* (2018) studied the effect of pH on the biocontrol mechanisms of a *Trichoderma* sp. that were isolated from the soils. The rate of sporulation of the *Trichoderma* sp. in PDA plates with amended pH values (4, 5, 6 and 7) was also estimated and a significantly high ($p \leq 0.05$) rate of sporulation (5×10^7 spores ml⁻¹) was observed at pH 6 after 49 days.

Anwer *et al* (2020) conducted an experiment in order to identify *Trichoderma* strains that could be stable at adverse environmental conditions including high temperature, pH and high salinity. For this, several *Trichoderma* isolates were collected from agro ecological zone IIIA of Bihar, India. *In vitro* antagonistic potential of *Trichoderma* isolates against soil borne pathogens (*Sclerotium rolfsii*, *Rhizoctonia solani* and *Fusarium oxysporum*) were evaluated. In all *in vitro* tests, *Trichoderma* isolate Tvb1 (isolated from hot spring soil) was found to be the most efficient tolerating high temperature up to 45 °C for four days,

2.3 Compatibility of *Trichoderma* strains in response of fungicide, insecticide and weedicide.

Mundal *et al.* (1995) conducted an experiment on *Trichoderma* in *in vitro* condition and resulted that *Trichoderma harzianum* showed high compatibility even at higher concentration with insecticides like monocrotophos and chloropyriphos.

Gouder *et al.* (1999) studied and observed the compatibility of *Trichoderma harzianum* with monocrotophos under all the concentrations was from 1 to 100 ppm and moderate compatibility with Thimet and Chlorpyriphos.

Madhavi *et al.* (2011) studied the compatibility of *Trichoderma viride* with 25 pesticides that was evaluated *in vitro*. Among six seed-treatment chemicals tested, *T. viride* showed a high compatibility with the insecticide Imidacloprid (7.6cm mycelial growth), followed by Mancozeb (6.3cm) and Tebuconazole (3.7cm). Contact fungicides, viz., Pencycuron and Propineb were found to be fully compatible with *T. viride*. Among the 10 herbicides also tested, the fungus was highly compatible with Imazathafir (9.0cm) followed by 2,4-D Sodium salt (8.9cm) and Oxyfluforen (6.5cm) while being totally incompatible with systemic fungicides like Carbendazim, Hexaconazole, Tebuconazole and Propiconazole.

Vasundara *et al.* (2015) studied the compatibility with the fungicide mancozeb (3000 ppm) and this showed high compatibility with *Trichoderma viride*.

Vasundra *et al.* (2015) conducted an experiment to check the compatibility of two insecticides, three fungicides and their combinations on *Trichoderma Viridae*. It showed variable responses against the tested pesticides (fungicides and insecticides) and their combinations at recommended concentrations for field studies. The treatments of Mancozeb (3000 ppm), Imidacloprid (2000 ppm) and combination of Mancozeb (3000 ppm) + Imidacloprid (2000 ppm) showed high compatibility with *Trichoderma Viridae* by showing 7, 11 and 11 percent growth inhibition respectively. The treatments viz. Carbendazim (1000 ppm) + Chlorpyriphos (6000 ppm), Tebuconazole (1000 ppm) + Imidacloprid (2000 ppm) and Tebuconazole (1000 ppm) + Chlorpyriphos (6000 ppm) showed high incompatibility with 100 percent growth inhibition.

Khirallah *et al.* (2016) evaluated the compatibility of some active ingredients of fungicides *in vitro* on mycelia radial growth, conidia production and germination of three strains of *Trichoderma harzianum* and one strain of *T.*

viride. The compatibility estimated on the mycelial radial growth was moderate between the combination pyraclostrobin + boscalid and *Trichoderma* spp. in the first week and became very important from a second week. This compatibility was moderate with conidia germination and low with the conidia production.

Ashish *et al.* (2019) conducted an experiment to study about the compatibility of two isolates of *Trichoderma viride* against eight different fungicide at five different concentrations. *Trichoderma* spp with Mancozeb 75%WP revealed that this fungicide is compatible with *Trichoderma viride* upto 50 ppm. However, above 50 ppm concentration it started inhibiting the growth of both the isolates of *T. viride*. With thiram 75%WP, the maximum average per cent inhibition of 69.26 per cent was recorded at 1000 ppm which shows 30.74 per cent compatibility of *Trichoderma* with Thiram 75%WP at 1000 ppm concentration.

2.4 Antagonist effect of different *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri* under *In vitro* condition.

Dubey *et al.* (2007) evaluated 10 isolates belonging to three species of *Trichoderma* (*Trichoderma viride*, *Trichoderma harzianum*, and *Trichoderma virens*) against four isolates of *F. oxysporum* f. sp. *ciceri*. The isolates of *Trichoderma* species were evaluated against the pathogen in dual culture and through production of volatile and non-volatile inhibitors. They also enhanced seed germination, root and shoot length, and decreased wilt incidence under greenhouse condition. The isolates proved potential *in vitro* tests were evaluated along with other bioagents.

Mustafa *et al.* (2009) conducted an experiment under *in-vitro* on *Trichoderma* spp. and strongly antagonised six different seed borne pathogenic fungi viz. *Fusarium moniliforme*, *Fusarium oxysporum*, *Rhizoctonia solani*, *Fusarium solani*, *Botryodiplodia theobromae* and *Alternaria alternata* in dual culture assay. *Trichoderma harzianum* gave maximum inhibition of mycelia growth among all of the pathogenic fungi.

Reena *et al.* (2013) studied the antagonism of *Trichoderma viride* on pathogenic fungi, viz., *Fusarium* sp., *Curvularia* sp., *Rhizopus* sp., *Aspergillus niger*, *A. flavus* and *A. fumigatus* under *in vitro* condition. The results of the activity assay of crude extracts of the antagonistic fungi *Trichoderma viride* at different concentrations on the radial mycelial growth and percent radial mycelial growth inhibition of different pathogenic fungi significantly varied among crude extract concentrations of the antagonist fungi and in between the plant pathogens.

Patole *et al.* (2017) evaluated *T. viride* and *T. harzianum* against the *Fusarium* f. sp. *Ciceri*. Results showed that *T. viride* and *T. harzianum* alone or in combination significantly inhibited the mycelial growth of the pathogen. The combined effect of both antagonists (*T. viride*+ *T. harzianum*) was found to be most effective (86.57%) in inhibition of *Fusarium* mycelia growth as compared to the control followed significantly by *T. harzianum* (82.57%) and *T. viride* (80.24%).

Abhiram and Masih (2018) tested *T. viride* and *T. harzianum* for evaluating their efficacy against five *F. oxysporum* strains by dual culture technique. *T. harzianum* showed maximum inhibition of 77.77% over *F. oxysporum* strain (E), whereas in *T. viride* showed minimum inhibition of 71.00% over *F. oxysporum* strain (E) in dual culture plate technique.

2.5 Antagonist effect of different *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri* under *In vivo* condition.

Dubey *et al.* (2007) evaluated 10 isolates belonging to three species of *Trichoderma* (*Trichoderma viride*, *Trichoderma harzianum*, and *Trichoderma virens*) against four isolates of the pathogen representing four different races commonly prevalent in India. Dharwad (race 1), Kanpur (race 2), Ludhiana (race 3), and Delhi (race 4) isolates of *F. oxysporum* f. sp. *Ciceris*. *T. viride* isolated from Ranchi followed by *T. harzianum* (Ranchi) and *T. viride* isolated from Delhi inhibited maximum mycelial growth of the pathogen. They also

enhanced seed germination, root and shoot length, and decreased wilt incidence under greenhouse condition.

Khan *et al.* (2014) investigated the effects of *T. harzianum*, *T. hamatum*, *T. viride*, *T. polysporum*, and *T. koningii* on the wilt disease complex of chickpea caused by *Fusarium oxysporum* f. sp. *ciceri* and *Rhizoctonia solani* under field conditions during 2 consecutive years. Chickpea cultivar Avrodhi, grown in plots inoculated with *F. oxysporum* f. sp. *ciceri* and *R. solani*, showed chlorosis of leaves and wilting of foliage and exhibited a 22%-25% decrease in yield. Soil application of biocontrol agents (BCAs) checked the severity of wilt by 25%-56% and 39%-67% and increased the yield of chickpea by 12%-28% and 8%-24% in the 2 years, respectively.

Pandey *et al.* (2017) evaluated the effective management of diseases through seed biopriming and soil application of biocontrol agents. Seed biopriming checked the incidence of wilt and root rot in the range of 45%-60% and increased the yield of chickpea by 10%-20%. However, combined applications of seed biopriming as well as soil application significantly checked the disease incidence in the range of 46%-78%.

Sonkar *et al.* (2018) evaluated the efficacy of *Trichoderma* spp against chickpea and Tomato Fusarium wilt in field. *Fusarium oxysporum* f.sp.*Ciceri* and *Fusarium oxysporum* f.sp. *Lycopersici* was mixed in soil. However, plants did treated with nine isolate of *Trichoderma* spp. The production of microplot, T1, T2, and T3 was best for tomato production and only T4 was better in chickpea production then tomato product. T6 was represented very low efficacy in production with comparison to other treatment. Consistently, experiment was proved that different *Trichoderma* spp. have different efficacy and provide defence from wilt.

Suman *et al.* (2018) evaluated the antagonistic traits under greenhouse conditions to control *Fusarium* wilt in chickpea varieties GPF-2 and JG-41. Maximum reduction in disease incidence was recorded with Ps45 (74.48±0.67 %) and (70.32±1.00%) compared to fungicide treatment (61.53±0.89% and 58.69±0.33%) in chickpea genotypes GPF-2 and JG-41 respectively.

CHAPTER- III

MATERIAL AND METHODS

The present experiment was under taken on “**Identification of computable trait of specific *Trichoderma* strains for management of soil borne diseases**” the effect of variable range of temperature and H ion concentration on *Trichoderma* strains, Compatibility with pesticides and in vitro an in vivo evaluation against *Fusarium oxysporum* f. sp. *ciceri* were carried out in the laboratory, Department of Plant Pathology, College of Agriculture, Indore, M.P, India during the season 2019-2020. Details of the materials used, experimental procedures followed and techniques adopted are given as here under

3.1 Materials:

The materials used included soil samples, ingredients of culture media, diseased plant samples, glass wares, equipment's, chemicals and a few miscellaneous articles

3.1.1 Collection of soil samples:

Collection of soil samples from rhizospheric and non rhizospheric soil from different crops and places will be collected by roving survey from Nimar valleys namely, Khargone, Dhar, Khandwa, and Barwani of Madhya Pradesh. The details of different *Trichoderma* strains were collected from different rhizosphere mentioned under below the table (Table 3.1).

Table 3.1 Details of *Trichoderma* strains collection from different places

| S. No. | Trichoderma strains | District | Crop | Rhizospheric/ Non rhizospheric | Farmer name | Address |
|--------|---------------------|----------|---------------|-----------------------------------|----------------------|--|
| 1 | TKHR1 | Khargone | Cotton | Rhizospheric | Vijay Gurjar | Viil- Dhangaon, Teh. + Dist. Kharogone |
| 2 | TKHR2 | | Cotton | Non rhizospheric | Ramkrishan Patel | Viil- Dhangaon, Teh. + Dist. Kharogone |
| 3 | TKHR3 | | Cotton | Non rhizospheric | Beniram Choudhary | Viil- Dhangaon, Teh. + Dist. Kharogone |
| 4 | TKHR4 | | Pigeon pea | Rhizospheric | BabluChoudhary | Viil- Dhangaon, Teh. + Dist. Kharogone |
| 5 | TKHR5 | | Soybean | Non rhizospheric | Rahul Yadav | Viil- Dhangaon, Teh. + Dist. Kharogone |
| 6 | TKHR6 | | Guava orchard | Non rhizospheric | Ramesh Parmar | Viil- Baswa, Teh. + Dist. Kharogone |
| 7 | TKHR7 | | Soybean | Rhizospheric | Kewal Ram Patel | Viil- Baswa, Teh. + Dist. Kharogone |
| 8 | TKHR8 | | Chilli | Rhizospheric | Ram raj Sisodiya | Viil- Badud, Teh. + Dist. Kharogone |
| 9 | TKHR9 | | Cotton | Non rhizospheric | Sitaram Gurjar | Viil- Badud, Teh. + Dist. Kharogone |
| 10 | TKHR10 | | Cotton | Rhizospheric | Asharam Gurjar | Viil- Badud, Teh. + Dist. Kharogone |
| 11 | TDHR11 | Dhar | Cotton | Rhizospheric | Mangila Parte | Vill- Deshwaliya, Teh- Kukshi, Dist-Dhar |
| 12 | TDHR12 | | Cotton | Non rhizospheric | Bherulal Kanel | Vill-Deshwaliya, Teh. -Kukshi, Dist-Dhar |
| 13 | TDHR13 | | Soybean | Non rhizospheric | Bhanu Badole | Vill-Deshwaliya, Teh. -Kukshi, Dist-Dhar |
| 14 | TDHR14 | | Soybean | Non rhizospheric | Shankar Solanki | Vill-Deshwaliya, Teh. -Kukshi, Dist-Dhar |

| | | | | | | | |
|----|--------|------------|---------|------------------|-------------------|--|--|
| 15 | TDHR15 | | Maize | Rhizospheric | Rajendra Solanki | Vill-Deshwaliya, Teh. -Kukshi, Dist-Dhar | |
| 16 | TDHR16 | | Maize | Rhizospheric | Ramesh Ewane | Vill-Deshwaliya, Teh.- Kukshi, Dist-Dhar | |
| 17 | TDHR17 | | Bajra | Rhizospheric | Sependra Ewane | Vill-Deshwaliya, Teh. -Kukshi, Dist-Dhar | |
| 18 | TKHW18 | Khandwa | Soybean | Rhizospheric | Ganesh Gurjar | Vill - Deshgaon, Teh. + Dist.- Kandwa | |
| 19 | TKHW19 | | Cotton | Rhizospheric | Mukesh Sejgayan | Vill - Deshgaon, Teh. + Dist.- Kandwa | |
| 20 | TKHW20 | | Maize | Non rhizospheric | Parmanad Rathore | Vill - Deshgaon, Teh. + Dist.- Kandwa | |
| 21 | TKHW21 | | Soybean | Rhizospheric | Mahendra Gurjar | Vill - Deshgaon, Teh. + Dist.- Kandwa | |
| 22 | TKHW22 | | Cotton | Rhizospheric | Kailash Solanki | Vill - Deshgaon, Teh. + Dist.- Kandwa | |
| 23 | TBRW23 | | Soybean | Rhizospheric | Hiralal Patidar | Vill- Jholpipri, Teh.- Anjad, Dist.- Barwani | |
| 24 | TBRW24 | | Maize | Rhizospheric | Nathuram Badole | Vill- Jholpipri, Teh.- Anjad, Dist.- Barwani | |
| 25 | TBRW25 | | Cotton | Rhizospheric | Rahul Patidar | Vill- Jholpipri, Teh.- Anjad, Dist.- Barwani | |
| 26 | TBRW26 | | Barwani | Maize | Non rhizospheric | Kamal Mujalde | Vill- Jholpipri, Teh.- Anjad, Dist.- Barwani |
| 27 | TBRW27 | | | Pigeon pea | Non rhizospheric | Jagdish Shivhare | Vill-Golata, Teh.- Anjad, Dist.- Barwani |
| 28 | TBRW28 | Pigeon pea | | Non rhizospheric | Ashok Parte | Vill-Golata, Teh.- Anjad, Dist.- Barwani | |
| 29 | TBRW29 | Maize | | Non rhizospheric | Kailash Bamniya | Vill-Golata, Teh.- Anjad, Dist.- Barwani | |
| 30 | TBRW30 | Chilli | | Rhizospheric | Bhagirath Solanki | Vill- Chhapri, Teh.- Anjad, ist. Barwani | |

3.1.2 Collection of diseased specimens:

Diseased chickpea plants exhibited typical symptoms of wilt were collected from the wilt sick plot College of Agriculture, Indore, The diseased plants were brought to the laboratory. The specimens were critically examined for the symptoms and the pathogen was isolated from such plants to obtain the culture of *F. oxysporum* f. sp. *ciceri*.

3.1.3 Different strains of *Trichoderma* spp:

Thirty strains of *Trichoderma* spp. isolated from soil samples collected from four districts of M.P. were used for checking the compatibility with pesticides and antagonistic potential against *F. Oxysporum*. f.sp. *ciceri* under *in vitro* and *in vivo* condition.

3.1.4 Chemicals:

(a) Cleaning solution:

Cleaning solution containing potassium dichromate 80g, distilled water 300 ml and concentrated sulphuric acid 400 ml was used to clean the glass wares.

(b) Mercuric chloride solution:

A 1:1000 solution of $HgCl_2$ was prepared and used for surface sterilization of samples during isolation of the pathogen.

(c) Mounting medium:

Lacto phenol and cotton blue with the following composition were used as the working stain for studying the characteristics of the pathogen and the antagonists.

i. Lacto phenol:

| | |
|---|-------|
| Phenol | 20g |
| (Pure crystals liquefied by gentle heating on a water bath) | |
| Lactic acid | 20 ml |
| Glycerol | 40 ml |

| | |
|-----------------|-------|
| Distilled water | 20 ml |
|-----------------|-------|

ii. Cotton blue:

| | |
|------------------------|-------|
| Anhydrous lacto phenol | 67 ml |
| Distilled water | 20 ml |
| Cotton blue | 0.1 g |

3.1.5: Ingredient of culture media:

Three culture media, namely, potato dextrose agar, nutrient broth media and *Trichoderma* selective medium (TSM) with following ingredients were used during the course of investigation

(i) *Trichoderma* selective medium (TSM):

Selective isolations of *Trichoderma* spp. from soil were made on modified selective medium (Elad and Chet, 1982) having following ingredients

| Ingredients | - | quantity(g l⁻¹) |
|--------------------------------------|----------|-----------------------------------|
| MgSO ₄ .7H ₂ O | - | 0.2 g |
| K ₂ HPO ₄ | - | 0.9g |
| KCl | - | 0.15g |
| NH ₄ NO ₃ | - | 1.0 g |
| Glucose | - | 3.0g |
| Agar-agar | - | 20 g |
| Distilled water | - | 1000 ml |
| pH | - | 6.5 |

After autoclaving following components were added

| Ingredients | - | Quantity (g l⁻¹) |
|--------------------|---|------------------------------------|
| Chloramphenicol | - | 0.25g |
| Rose Bengal-150 | - | 0.15 g |
| Captan 50 WP | - | 0.15 g |
| Apron 35-SD | - | 0.025 g |

(ii) Potato dextrose agar (PDA) medium: (Rangaswami and Mahadeven, 2001)

| ingredient's | - | Quantity(g l⁻¹) |
|-----------------------------------|---|-----------------------------------|
| Peeled and sliced potato(extract) | - | 200g |
| Dextrose | - | 20 g |
| Agar-agar | - | 20 g |
| Distilled water | - | 1 ltr |
| pH (adjusted to) | - | 6.5 |

(iii) Nutrient broth medium:

| Ingredient's | - | Quantity (g/l) |
|------------------------|---|-----------------------|
| Peptone | - | 10 |
| Beef extract | - | 10 |
| Sodium chloride | - | 5 |
| pH after sterilization | - | 6, 7, and 8 (Adjust) |

3.1.6: Glass wares:

Standard "Borosil" make glass wares like Petridishes, beakers, funnels, pipettes, Erlenmeyer flask, culture tubes, measuring cylinder etc. were used during the course of study.

3.1.7: Equipments:

Equipment's used during the investigation included research microscope, refrigerator, autoclave, hot air oven, BOD incubator, laminar air flow, weighting balance, LPG gas burner, Bunsen burner, hot plate etc. Small instruments like Inoculation needle, scalpel, razor, glass cavity slides, polythene bags, desiccators, glass marking pencils, cover slips brush, dropper, match box etc. were also used during the study.

3.2: METHODS

3.2.1: Cleaning and sterilization of glass wares:-

The glass wares were cleaned by dipping them in cleaning solution for 5 minutes and finally rinsed with running tap water for 30 minutes. The Petri dishes were sterilized in a hot air oven at $180 \pm 2^\circ\text{C}$ for $1\frac{1}{2}$ to 2 hours. The inoculation needle, cork borer and other metallic instruments were sterilized by dipping them in alcohol and heating red hot over flame of the spirit lamp/Bunsen burner.

(i) Glass wares and their cleaning

Borosil and Corning glass wares were kept in the cleaning solution containing 60 g of potassium dichromate ($\text{K}_2\text{Cr}_2\text{O}_7$) and 60 ml of concentrated sulphuric acid (H_2SO_4) in one litre of water for whole day. Then they were cleaned by washing with detergent powder followed by rinsing several time in tap water and finally in distilled water.

(ii) Sterilization

. All the media were sterilized for 15 minutes at 1.1 kg per sq cm pressure, except those containing sugars and nitrogen sources which were sterilized at 0.7 kg per sq. cm pressure for 10 minutes and soil used for experiment was sterilized at 1.33 kg/sq cm pressure for two hours.

(iii) Surface sterilization of diseased plant parts

Plant materials were surface sterilized using 0.1 per cent mercuric chloride solution for 1 minute and then washing in sterile water thrice.

3.2.2 Preparation of media:

(i) Potato dextrose agar:

Peeled potatoes cut into pieces were boiled in ½ litre water in a sauce pan for 8-10 minutes. In another pan, agar-agar was boiled in ½ litre water separately so that it gets completely dissolved. Potato extract was collected after filtration on muslin cloth over a funnel and added to dissolve agar-agar along with dextrose. The medium was poured in conical flask of 250 ml, plugged with non-absorbent cotton and autoclaved at 121.6°C (15 lbs) for 25 minutes. Streptomycin sulphate @ 1.0 g/l was added to the medium before pouring into Petri dishes to avoid bacterial contamination. It was used during the investigation.

(ii) *Trichoderma* selective Medium (TSM):

All the ingredients except Apron 35-S D, chloramphenicol, rose bengal and captan were mixed in 1 litre distilled water, which was boiled to dissolve the agar-agar. The medium was transferred to 250 ml Erlenmeyer flask, plugged with cotton and autoclaved at 121.6°C (15 lbs) for 25 minutes. After cooling (55°C), the required quantity of Apron 35-S D, chloramphenicol, rose bengal and captan were added to the medium just before pouring into the sterilized Petri dishes.

(iv) Nutrient broth medium:-

Suspend 25 grams in 100 ml purified/distilled water. Heat if necessary to dissolve completely. Sterilise autoclaving at 10 lbs pressure (115 °C) for 30 minutes or alternatively at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Beef extract and peptone provide the necessary nitrogen compounds, carbon vitamins and also some trace ingredients to the non-fastidious organism. Sodium chloride maintains osmotic equilibrium of the medium.

3.2.3 Isolation of *Trichoderma* from rhizosphere and non-rhizosphere soil sample:

The soil samples were collected from the different location and season of different districts of Nimar valley of Madhya Pradesh at a depth ranging 5-

6 cm, by removing top 2 cm surface soil. The soil collected from rhizosphere and non-rhizosphere soil of healthy plants. All collected soil samples were mixed thoroughly to make a composite sample. A working samples 1 gram (dry weight basis) soil was taken from composite sample. 1 gram soil was mixed into 10 ml of sterile distilled water then 1 ml of suspension was taken into another tube containing 9 ml of sterile distilled water. This serial dilution technique was continued up to 10⁻² to 10⁻⁶. From the 10⁻⁶ dilution, 0.1 ml suspension was transferred to each of the three Petri dishes, 20 ml of melted *Trichoderma* Specific Medium (TSM) (Elad and Chet, 1983) was poured in each plate and mixed with the suspension by giving a gentle whirling motion to the plate and allowed them to incubate in room temperature (Islam *et al*, 2001; Bhagat, 2008, Bhagat and Pan, 2007). The probable colonies were observed closely and picked up from Petri plates and transferred to PDA slants and finally pure culture was obtained by repeated subculture. They were identified on the basis of their morphological characters (Bhagat and Pan, 2007).

3.2.4 Test organism/ Isolation and purification of the pathogen:

The pathogen *Fusarium oxysporum* f. sp. *ciceri* was isolated from root region of the diseased plants by time segment method (Rangaswami and Mahadevan, 2001) and purified by hyphal tip method and maintained on potato dextrose agar stands. The affected portions (roots) of the diseased plants were cut with the help of sharp razor and rinsed with sterilised water to remove traces of dirt. These were surface sterilised by dipping in 1:1000 mercuric chloride solution for one minute and washed twice with sterile water. These pieces were transferred aseptically to sterilised Petri dishes containing solidified PDA in a laminar air flow. The Petri dishes were incubated at 26±1°C. The appearing fungus was observed after 72 hours and isolations were made from developing colonies for further study.

3.2.5 Weight of colony radial growth

To check the tolerance level of different isolated strain of *Trichoderma* at variable range of temperature (20°C, 25°C and 30°C) and H ion concentration (6, 7 and 8pH) in nutrient broth medium. The data recording

after colony growth of different *Trichoderma* strains, Liquid+Mait+ (gm.), Wet Mait (gm) and Dry Mait (mg)

3.2.6 Maintenance of cultures

The fungi were sub-cultured on potato dextrose agar slants and allowed to grow at $27\pm 1^{\circ}\text{C}$ for one week. Such slants were preserved in a refrigerator at 5°C and renewed once in two months to maintain the virulence of the pathogen and antagonism to *Trichoderma* spp

3.2.7 Compatibility of *Trichoderma* strains tested against Fungicides, insecticides and weedicides under *in vitro* condition

Evaluate the compatibility of commonly used pesticides in agriculture at two different concentrations (500 and 1000 ppm) with *Trichoderma* strains that is being used as a bio control agent against soil borne plant pathogens. Three group commonly used pesticides such as Weedicides (Glyphosate 41% SL, Metsulfuron methyl 20%WP and Imazathypr 10%SL), Insecticides (Phorate 10 Cg, Chlorpyriphos 20 %EC and Quinalphos 25% EC) and Fungicides (Metiram 70% WG, Mancozeb 75% WP and Metalaxyl 8%+Mancozeb 64%WP).

The required amount of the pesticides was gently mixed with 60 ml melted but cooled PDA at the time of pouring. To avoid bacterial contamination streptomycin @ 1 g l^{-1} was added to each flask before pouring. Medium without any chemicals served as control. Three replications were maintained for each treatment. Mycelial discs (6 mm diameter) from seven days old culture of trichoderma spp. were placed aseptically in the centre of petri dish. The petri dish were then incubated at $26\pm 1^{\circ}\text{C}$. Observations for radial growth of fungal were recorded 48 hrs. 72hrs. and 96 hrs. after inoculation. Radial growth of pathogen isolates was measured and per cent inhibition of average radial growth was calculated in relation to mycelial growth of the controls as follows (Vincent 1947).

3.2.7 Evaluation of antagonistic potential of *Trichoderma* strains against *F. oxysporum f. sp. ciceri* by dual culture technique *in vitro*

The antagonistic potential of eight strains of *Trichoderma* spp. was evaluated against *F. oxysporum* (Denis and Webster 1971). A 6mm diameter mycelial disc from the margin of the 7 days-old culture of *Trichoderma* isolates were inoculated at periphery region opposite to each other at equal distance. The experimental design used was a completely randomized with three Petri dishes for each isolates. In control plates (without *Trichoderma*), a sterile agar disc was placed at opposite side of the soil borne inoculated plates. Inoculated plates were incubated at $28 \pm 1^{\circ}\text{C}$ for 7 days. 2-3 days after the incubation period, radial growth of pathogen isolates was measured and per cent inhibition of average radial growth was calculated in relation to mycelial growth of the controls as follows (Vincent 1947).

$$I = \frac{C - T}{C} \times 100$$

Where, I = Per cent inhibition of growth

C = Mycelial growth in control plate

T = Mycelial growth in test plate

All strains were tested in three replications and were carried out twice for each isolate.

3.2.8 Evaluation of *Trichoderma* strains against *F. oxysporum f. sp. ciceri* in the field condition:

A field experiment was conducted for measurement of antifungal activity and the ability of the *Trichoderma* stains to reduce the infestation of *F. oxysporum f. sp. ciceri* in the green house to chickpea plants, in this experiment. The pathogen was multiplied in the PDB medium and maintained in the soil @of 10^7 cfu/Kg soil, which was pre sterilized at 121°C and 15lb pressure for 15 minutes and filled in pots. Chickpea seeds were treated with different strains of *Trichoderma* spores obtained from the

cultures at the rate of 4g/Kg seed. The seeds were placed in a clean container, sprinkled with water until they were wet, adequate quantities of the bio-control agents available in a talc based dry powder (Devikarani *et al.*, 2009) was added and the container was agitated thoroughly until the seeds were uniformly coated. The seeds were sown in the field with were pre infested with the pathogen. 240 seeds per plot were sown at the 2-3 cm in three replications for each of the eight strains of *Trichoderma* and a control in which the seeds were not treated with the strains of *Trichoderma* only treated with water. Watering was done adequately and regular observations on germination percentage and disease incidence were recorded at 5 DAS interval.

Incidence of disease was calculated by the formula -

$$\text{Disease incidence (\%)} = \frac{\text{No. of plants exhibiting wilt}}{\text{Total number of plant observed}} \times 100$$

Location: The experiment was conducted in Research Field, Department of Plant Pathology, College of Agriculture, Indore (M.P.) 452001.

Experiment details:

| | | |
|-------------------------|---|-------------------------------------|
| Crop | - | Chickpea (<i>Cicer arietinum</i>) |
| Variety | - | JG-315 |
| Year | - | 2019-2020 |
| Treatments | - | 9 |
| Replications | - | 3 |
| Design | - | RBD (Randomized Block Design) |
| Date of sowing | - | 20/11/2018 |
| Plot size | - | 4×2m |
| Distance Plant to plant | - | 10 cm |
| Row to row | - | 40 cm |
| Net area | - | 352 m ² |

Different treatments:

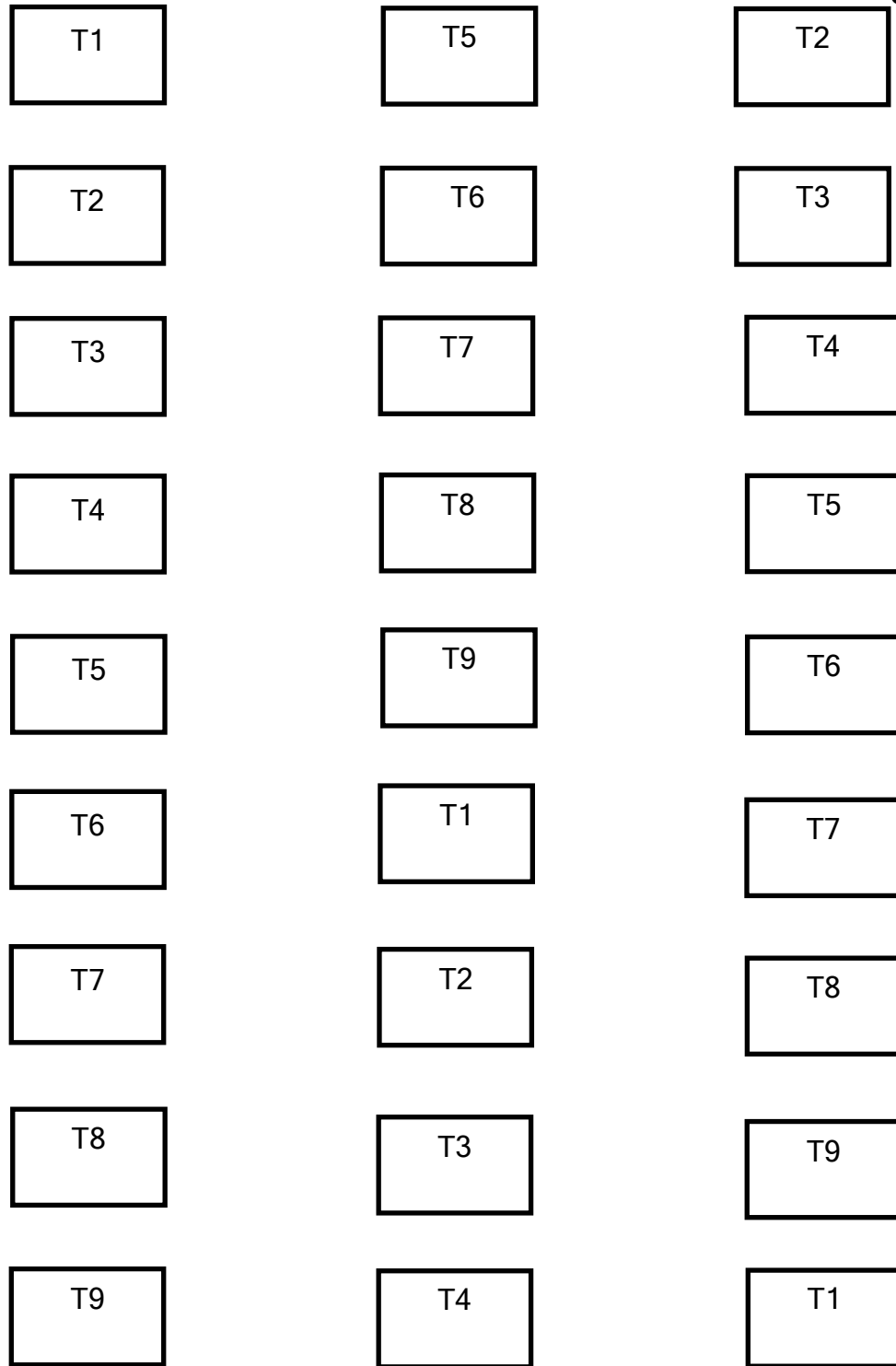
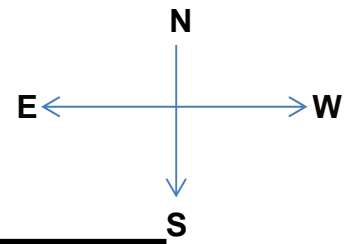
| S. No. | Treatment |
|---------------|------------------|
| T1 | TKHR1 |
| T2 | TKHR2 |
| T3 | TKHR6 |
| T4 | TKHR10 |
| T5 | TDHR12 |
| T6 | TDHR15 |
| T7 | TKHW18 |
| T8 | TKHW19 |
| T10 | Control |

3.2.10 Statistical analysis

Statistical analysis was done for the interpretation of the results obtained. Completely randomized design (CRD) (for *In vitro* experiments) and Randomize block design (RBD) (for *In vivo* experiments) were employed to analyses the data. The critical differences were worked out at 0.05% probability level. The value in percentage was transformed by angular transformation. The experiments were conducted in the PG laboratory, Department of Plant Pathology, College of Agriculture, Indore (M.P.)

Detailed two tier layout plan of the experiment for management of *F. oxysporum* f. sp. *Ciceri*.

Plot size:-
4m×2m



CHAPTER - IV

RESULTS

The present investigation on “**Identification of compatible trait of specific *Trichoderma* strains for management of soil borne disease**” were carried out in College of Agriculture Indore, Department of Plant Pathology laboratory under *in vitro* and *in vivo* condition during the season 2019-2020, on thirty isolates of *Trichoderma* on the identified the *Trichoderma* strains tolerant to higher temperature and variable range of pH, assessment the compatibility of *Trichoderma* strains in response of fungicide, insecticide and weedicide and evaluated the prominent strains of *Trichoderma* against soil born disease *in vitro* and *in vivo*. Results on these aspects have been reported here under in depicting the behavior of different isolates of the *Trichoderma* spp.

4.1 Collection of soil samples for isolation and purification of *Trichoderma* spp.

Total 30 soil samples collected from rhizospheric and non-rhizospheric soil from different crops and places of Nimar valleys zone of Madhya Pradesh. Four diverse districts of M. P. namely, Khargone (10) districts followed by Dhar (7), Khandwa (5), and Barwani (8). Isolation of *Trichoderma* strains from the soil by serial dilution method on *Trichoderma* selective medium (Eladet *al.* 1981) under *in vitro* condition and the purified strains was maintained on potato dextrose agar in petri plates and slants. Such petri plates and slants preserved in a refrigerator at 5⁰C and renewed once in two months to maintain the *Trichoderma* strains for physiological studies and antagonist effect with pesticides.

4.2 Collection of rhizosphere soil samples for isolation of *Trichoderma* spp.

Soil samples were collected from the rhizospheric and non-rhizospheric soil zone of different crops namely, Soybean, Cotton, Pigeon

pea, Maize, Chili, Guava orchard and Bajra. and different places and isolated the *Trichoderma* strains by serial dilution technique.

4.3 Recovery of *Trichoderma* spp. from rhizospheric and non rhizospheric soils of different crops.

Total 30 strains of *Trichoderma* were isolated from the collected soils and rhizospheric and non rhizospheric samples. The details of the districts and crop rhizospheres from which strains of *Trichoderma* isolated were given in detail in the table 4.1.

Table 4.1 Details of *Trichoderma* strains collection from different places

| S. No. | Trichoderma strains | District | Crop | Rhizospheric/ Non rhizospheric | Farmer name | Address |
|--------|---------------------|----------|---------------|-----------------------------------|---------------------|--|
| 1 | TKHR1 | Khargone | Cotton | Rhizospheric | Vijay Gurjar | Viil- Dhangaon, Teh. + Dist. Kharogone |
| 2 | TKHR2 | | Cotton | Non rhizospheric | Ramkrishan Patel | Viil- Dhangaon, Teh. + Dist. Kharogone |
| 3 | TKHR3 | | Cotton | Non rhizospheric | BeniramChoudhary | Viil- Dhangaon, Teh. + Dist. Kharogone |
| 4 | TKHR4 | | Pigeon pea | Rhizospheric | BabluChoudhary | Viil- Dhangaon, Teh. + Dist. Kharogone |
| 5 | TKHR5 | | Soybean | Non rhizospheric | Rahul Yadav | Viil- Dhangaon, Teh. + Dist. Kharogone |
| 6 | TKHR6 | | Guava orchard | Non rhizospheric | Ramesh Parmar | Viil- Baswa, Teh. + Dist. Kharogone |
| 7 | TKHR7 | | Soybean | Rhizospheric | Kewal Ram Patel | Viil- Baswa, Teh. + Dist. Kharogone |
| 8 | TKHR8 | | Chilli | Rhizospheric | Ram raj Sisodiya | Viil- Badud, Teh. + Dist. Kharogone |
| 9 | TKHR9 | | Cotton | Non rhizospheric | SitaramGurjar | Viil- Badud, Teh. + Dist. Kharogone |
| 10 | TKHR10 | | Cotton | Rhizospheric | AsharamGurjar | Viil- Badud, Teh. + Dist. Kharogone |
| 11 | TDHR11 | Dhar | Cotton | Rhizospheric | Mangila Parte | Vill- Deshwaliya, Teh- Kukshi, Dist-Dhar |
| 12 | TDHR12 | | Cotton | Non rhizospheric | BherulalKanel | Vill-Deshwaliya, Teh. -Kukshi, Dist-Dhar |
| 13 | TDHR13 | | Soybean | Non rhizospheric | BhanuBadole | Vill-Deshwaliya, Teh. -Kukshi, Dist-Dhar |
| 14 | TDHR14 | | Soybean | Non rhizospheric | Shankar Lal Solanki | Vill-Deshwaliya, Teh. -Kukshi, Dist-Dhar |
| 15 | TDHR15 | | Maize | Rhizospheric | Rajendra Solanki | Vill-Deshwaliya, Teh. -Kukshi, Dist-Dhar |

| | | | | | | | |
|----|--------|------------|---------|------------------|-------------------|--|--|
| 16 | TDHR16 | | Maize | Rhizospheric | Ramesh Ewane | Vill-Deshwaliya, Teh.- Kukshi, Dist-Dhar | |
| 17 | TDHR17 | | Bajra | Rhizospheric | SependraEwane | Vill-Deshwaliya, Teh. -Kukshi, Dist-Dhar | |
| 18 | TKHW18 | Khandwa | Soybean | Rhizospheric | Ganesh Gurjar | Vill - Deshgaon, Teh. + Dist.- Kandwa | |
| 19 | TKHW19 | | Cotton | Rhizospheric | MukeshSejgayan | Vill - Deshgaon, Teh. + Dist.- Kandwa | |
| 20 | TKHW20 | | Maize | Non rhizospheric | ParmanadRathore | Vill - Deshgaon, Teh. + Dist.- Kandwa | |
| 21 | TKHW21 | | Soybean | Rhizospheric | MahendraGurjar | Vill - Deshgaon, Teh. + Dist.- Kandwa | |
| 22 | TKHW22 | | Cotton | Rhizospheric | Kailash Solanki | Vill - Deshgaon, Teh. + Dist.- Kandwa | |
| 23 | TBRW23 | | Soybean | Rhizospheric | HiralalPatidar | Vill- Jholpipri, Teh.- Anjad, Dist.- Barwani | |
| 24 | TBRW24 | | Maize | Rhizospheric | NathuramBadole | Vill- Jholpipri, Teh.- Anjad, Dist.- Barwani | |
| 25 | TBRW25 | | Cotton | Rhizospheric | Rahul Patidar | Vill- Jholpipri, Teh.- Anjad, Dist.- Barwani | |
| 26 | TBRW26 | | Barwani | Maize | Non rhizospheric | Kamal Mujalde | Vill- Jholpipri, Teh.- Anjad, Dist.- Barwani |
| 27 | TBRW27 | | | Pigeon pea | Non rhizospheric | JagdishShivhare | Vill-Golata, Teh.- Anjad, Dist.- Barwani |
| 28 | TBRW28 | Pigeon pea | | Non rhizospheric | Ashok Parte | Vill-Golata, Teh.- Anjad, Dist.- Barwani | |
| 29 | TBRW29 | Maize | | Non rhizospheric | Kailash Bamniya | Vill-Golata, Teh.- Anjad, Dist.- Barwani | |
| 30 | TBRW30 | Chilli | | Rhizospheric | Bhagirath Solanki | Vill- Chhapri, Teh.-Anjad, ist. Barwani | |

4.4 Screening of *Trichoderma* strains tolerant to variable range of temperature.

Screening of 30 *Trichoderma* strains tolerant to variable range of temperature viz., 20°C, 25°C and 30°C tried for the growth of the *Trichoderma*. Influence of temperature regimes on the growth of *Trichoderma spp.* The variable range of temperature will be maintained on BOD incubator. In this experiment was PDA broth solution used for screening of strains performance by the means of colony diameter. Twenty ml of the medium was pipette out into each 100 ml conical flask. Each treatment was replicated thrice. The data on radial growth of colony, and dry weight of the mycelia mat were recorded and analyses statistically.(Table 4.2).

Among the maximum weight of dry mycelium (mg) in 20°C temperature with *Trichoderma* strain viz. TKHR6 (0.52), TKHW19 (0.50), TDHR15 (0.49) and TKHW18 (0.48). The lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TDHR16 (0.20).

Among the 25°C temperature maximum weight of dry mycelium (mg) of *Trichoderma* strain viz., TKHR6 (0.56), TDHR15 (0.54), TKHW19 (0.53), TKHW18 (0.52) and TBRW24 (0.52). The lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TDHR16 (0.28).

Among the 30°C temperature maximum weight of dry mycelium (mg) of *Trichoderma* strain viz., TKHR10 (0.46), TDHR15 (0.45), TKHW19 and TKHW18 (0.43). The lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TDHR16 (0.10).

Table-4.2 Screening of *Trichoderma* strains tolerant to variable range of temperature

| Trichoderma strains | 20°C Temperature | | | 25°C Temperature | | | 30°C Temperature | | |
|---------------------|----------------------|-----------------|-----------------|----------------------|-----------------|-----------------|----------------------|-----------------|-----------------|
| | Liquid+Mat+ (gm.) | Wet Mat (gm) | Dry Mat (mg) | Liquid+Mat+ (gm.) | Wet Mat (gm) | Dry Mat (mg) | Liquid+Mat+ (gm.) | Wet Mat (gm) | Dry Mat (mg) |
| TKHR1 | 98.32 | 6.80 | 0.45 | 99.43 | 6.86 | 0.49 | 97.73 | 6.72 | 0.37 |
| TKHR2 | 98.68 | 6.82 | 0.46 | 99.51 | 6.88 | 0.49 | 98.50 | 6.76 | 0.42 |
| TKHR3 | 95.82 | 6.62 | 0.32 | 97.09 | 6.73 | 0.44 | 95.19 | 6.53 | 0.25 |
| TKHR4 | 92.95 | 6.53 | 0.31 | 93.63 | 6.66 | 0.36 | 92.59 | 6.41 | 0.18 |
| TKHR5 | 92.11 | 6.52 | 0.30 | 93.25 | 6.64 | 0.35 | 91.33 | 6.40 | 0.17 |
| TKHR6 | 103.74 | 7.48 | 0.52 | 103.50 | 7.64 | 0.56 | 99.04 | 6.85 | 0.42 |
| TKHR7 | 90.41 | 6.48 | 0.29 | 91.83 | 6.55 | 0.34 | 89.98 | 6.11 | 0.15 |
| TKHR8 | 91.80 | 6.49 | 0.29 | 92.41 | 6.58 | 0.34 | 91.60 | 6.40 | 0.17 |
| TKHR9 | 95.00 | 6.58 | 0.31 | 95.17 | 6.67 | 0.39 | 94.37 | 6.48 | 0.23 |
| TKHR10 | 99.45 | 7.02 | 0.47 | 99.72 | 6.95 | 0.51 | 102.94 | 7.36 | 0.46 |
| TDHR11 | 96.31 | 6.64 | 0.36 | 96.94 | 6.72 | 0.44 | 95.20 | 6.53 | 0.26 |
| TDHR12 | 98.76 | 6.82 | 0.47 | 99.58 | 6.92 | 0.50 | 97.79 | 6.74 | 0.37 |
| TDHR13 | 95.97 | 6.64 | 0.34 | 96.89 | 6.71 | 0.43 | 95.55 | 6.58 | 0.28 |
| TDHR14 | 98.10 | 6.70 | 0.41 | 98.98 | 6.76 | 0.48 | 97.39 | 6.59 | 0.28 |
| TDHR15 | 101.32 | 7.19 | 0.49 | 102.01 | 7.44 | 0.54 | 101.12 | 7.19 | 0.45 |
| TDHR16 | 88.58 | 6.25 | 0.20 | 89.71 | 6.45 | 0.28 | 88.26 | 6.28 | 0.10 |

| | | | | | | | | | |
|------------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|
| TDHR17 | 96.01 | 6.64 | 0.35 | 96.87 | 6.71 | 0.43 | 95.33 | 6.57 | 0.26 |
| TKHW18 | 100.81 | 7.12 | 0.48 | 100.67 | 7.18 | 0.52 | 100.27 | 7.05 | 0.43 |
| TKHW19 | 101.63 | 7.28 | 0.50 | 100.69 | 7.22 | 0.53 | 100.93 | 7.12 | 0.43 |
| TKHW20 | 98.32 | 6.74 | 0.45 | 99.36 | 6.86 | 0.49 | 97.53 | 6.64 | 0.33 |
| TKHW21 | 96.89 | 6.67 | 0.38 | 97.93 | 6.74 | 0.45 | 96.38 | 6.58 | 0.28 |
| TKHW22 | 95.44 | 6.59 | 0.32 | 96.13 | 6.67 | 0.39 | 99.05 | 6.94 | 0.43 |
| TBRW23 | 89.25 | 6.42 | 0.22 | 90.01 | 6.53 | 0.30 | 88.74 | 6.32 | 0.12 |
| TBRW24 | 99.42 | 6.92 | 0.47 | 100.26 | 7.06 | 0.52 | 94.99 | 6.50 | 0.24 |
| TBRW25 | 92.98 | 6.56 | 0.31 | 93.82 | 6.67 | 0.38 | 92.78 | 6.44 | 0.20 |
| TBRW26 | 97.70 | 6.68 | 0.39 | 98.12 | 6.74 | 0.47 | 97.46 | 6.61 | 0.31 |
| TBRW27 | 89.54 | 6.45 | 0.24 | 90.18 | 6.53 | 0.32 | 89.23 | 6.32 | 0.14 |
| TBRW28 | 98.10 | 6.71 | 0.44 | 98.85 | 6.76 | 0.48 | 97.67 | 6.65 | 0.36 |
| TBRW29 | 89.77 | 6.47 | 0.25 | 93.65 | 6.67 | 0.37 | 89.37 | 6.33 | 0.14 |
| TBRW30 | 97.80 | 6.68 | 0.40 | 96.35 | 6.69 | 0.40 | 97.52 | 6.63 | 0.33 |
| SEm± | 1.02 | 0.16 | 0.02 | 1.08 | 0.15 | 0.01 | 1.04 | 0.17 | 0.01 |
| CD at 5 % | 2.89 | 0.46 | 0.04 | 3.05 | 0.45 | 0.05 | 2.96 | 0.48 | 0.04 |

* Mean of three replications

4.5 Screening of *Trichoderma* strains tolerant to variable range of H ion concentration.

Screening of 25 *Trichoderma* strains tolerant to variable range of H ion concentration viz., 6, 7 and 8. The variable range of H ion concentration will be maintained in liquid medium used in this experiment was PDA broth solution for screening of strains performance by the means of colony diameter. Adjustment of pH was done by adding 0.1 N alkalis (NaOH) or acid (HCl). Twenty ml of the medium was pipette out into each 100 ml conical flask. Each treatment was replicated thrice. The pH of the culture filtrate was determined by using Precision pH meter. The data on radial growth of colony, and dry weight of the mycelia mat were recorded and analyses statistically.(Table 4.3)

Among the maximum weight of dry mycelium (mg) in 6 pH with *Trichoderma* strain viz. TBRW24 (0.51), TDHR12 (0.50), TKHR1 (0.50), TBRW23 (0.49) and TKHW19 (0.48). The lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TBRW29 (0.17).

Among the 7 pH maximum weight of dry mycelium (mg) of *Trichoderma* strain viz., TKHR6 (0.56), TDHR15 (0.54), TKHW19 (0.53), TKHW18 (0.52) and TBRW24 (0.52). The lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TDHR16 (0.28).

Among the 8 pH maximum weight of dry mycelium (mg) of *Trichoderma* strain viz., TKHR2 (0.42), TBRW24 (0.41), TKHW19 and TKHW20 (0.40). The lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TKHW21 (0.11)

Table- 4.3 Screening of *Trichoderma* strains tolerant to variable range of H ion concentration.

| Trichoderma strains | 6 pH | | | 7 pH | | | 8 pH | | |
|---------------------|------------------|--------------|--------------|-------------------|--------------|--------------|------------------|--------------|--------------|
| | Liquid+Mat (gm.) | Wet Mat (gm) | Dry Mat (mg) | Liquid+Mat +(gm.) | Wet Mat (gm) | Dry Mat (mg) | Liquid+Mat (gm.) | Wet Mat (gm) | Dry Mat (mg) |
| TKHR1 | 100.54 | 6.96 | 0.49 | 96.64 | 6.75 | 0.40 | 92.22 | 6.38 | 0.22 |
| TKHR2 | 101.48 | 7.21 | 0.45 | 99.85 | 6.99 | 0.52 | 102.50 | 7.27 | 0.42 |
| TKHR3 | 90.21 | 6.33 | 0.30 | 94.04 | 6.70 | 0.38 | 94.85 | 6.44 | 0.27 |
| TKHR4 | 91.56 | 6.34 | 0.28 | 96.09 | 6.73 | 0.39 | 95.00 | 6.45 | 0.28 |
| TKHR5 | 88.96 | 6.21 | 0.28 | 94.01 | 6.68 | 0.36 | 94.00 | 6.43 | 0.24 |
| TKHR6 | 97.87 | 6.59 | 0.35 | 100.03 | 7.04 | 0.52 | 91.07 | 6.34 | 0.18 |
| TKHR7 | 95.14 | 6.43 | 0.44 | 93.90 | 6.67 | 0.35 | 92.30 | 6.38 | 0.23 |
| TKHR8 | 89.61 | 6.31 | 0.26 | 97.48 | 6.80 | 0.45 | 97.02 | 6.50 | 0.33 |
| TKHR9 | 94.81 | 6.41 | 0.25 | 93.70 | 6.64 | 0.33 | 91.27 | 6.34 | 0.21 |
| TKHR10 | 92.05 | 6.34 | 0.38 | 100.23 | 7.13 | 0.54 | 98.82 | 6.74 | 0.40 |
| TDHR11 | 89.31 | 6.31 | 0.23 | 99.24 | 6.87 | 0.47 | 88.51 | 6.18 | 0.13 |
| TDHR12 | 100.62 | 7.00 | 0.50 | 102.36 | 7.52 | 0.56 | 89.19 | 6.25 | 0.17 |
| TDHR13 | 96.85 | 6.54 | 0.37 | 99.90 | 7.04 | 0.52 | 97.29 | 6.56 | 0.36 |
| TDHR14 | 88.40 | 6.20 | 0.20 | 92.85 | 6.64 | 0.33 | 96.08 | 6.48 | 0.32 |
| TDHR15 | 99.22 | 6.93 | 0.42 | 100.07 | 7.08 | 0.53 | 97.58 | 6.62 | 0.38 |
| TDHR16 | 96.12 | 6.54 | 0.34 | 98.37 | 6.82 | 0.46 | 98.81 | 6.67 | 0.39 |

| | | | | | | | | | |
|------------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|
| TDHR17 | 99.18 | 6.78 | 0.46 | 94.23 | 6.73 | 0.39 | 98.33 | 6.65 | 0.39 |
| TKHW18 | 99.03 | 6.77 | 0.42 | 103.51 | 7.69 | 0.60 | 95.26 | 6.47 | 0.30 |
| TKHW19 | 98.13 | 6.71 | 0.48 | 99.74 | 6.97 | 0.50 | 100.57 | 6.93 | 0.40 |
| TKHW20 | 98.00 | 6.62 | 0.35 | 97.49 | 6.80 | 0.45 | 99.96 | 6.84 | 0.40 |
| TKHW21 | 92.56 | 6.35 | 0.19 | 96.74 | 6.76 | 0.42 | 88.13 | 6.18 | 0.11 |
| TKHW22 | 92.97 | 6.36 | 0.41 | 99.42 | 6.92 | 0.49 | 97.20 | 6.52 | 0.34 |
| TBRW23 | 95.74 | 6.44 | 0.49 | 98.90 | 6.84 | 0.47 | 95.37 | 6.48 | 0.32 |
| TBRW24 | 103.54 | 7.36 | 0.51 | 97.38 | 6.77 | 0.44 | 101.06 | 6.97 | 0.41 |
| TBRW25 | 97.57 | 6.55 | 0.31 | 97.43 | 6.77 | 0.44 | 89.01 | 6.22 | 0.15 |
| TBRW26 | 98.52 | 6.74 | 0.46 | 100.76 | 7.22 | 0.54 | 97.38 | 6.57 | 0.37 |
| TBRW27 | 95.90 | 6.52 | 0.32 | 92.26 | 6.62 | 0.30 | 94.76 | 6.43 | 0.26 |
| TBRW28 | 97.73 | 6.58 | 0.43 | 100.84 | 7.31 | 0.55 | 89.68 | 6.31 | 0.18 |
| TBRW29 | 95.80 | 6.47 | 0.17 | 91.07 | 6.58 | 0.28 | 94.54 | 6.43 | 0.25 |
| TBRW30 | 97.48 | 6.55 | 0.31 | 90.12 | 6.57 | 0.26 | 97.25 | 6.56 | 0.35 |
| SEm± | 1.03 | 0.16 | 0.01 | 1.11 | 0.16 | 0.01 | 1.08 | 0.14 | 0.01 |
| CD at 5 % | 2.91 | 0.45 | 0.05 | 3.16 | 0.45 | 0.05 | 3.07 | 0.40 | 0.04 |

*Mean of three replications

4.6 Assessment of the compatibility of *Trichoderma* strains in response of pesticides.

A present experiment was undertaken to evaluate the compatibility of commonly used pesticides in agriculture at two different concentrations (500 and 1000 ppm) with *Trichoderma* strains that is being used as a bio control agent against soil borne plant pathogens. Three group commonly used pesticides such as Weedicides (Glyphosate 41% SL, Metsulfuron methyl 20%WP and Imazathypr 10%SL), Insecticides (Phorate 10 Cg, Chlorpyriphos 20 %EC and Quinalphos 25% EC) and Fungicides (Metiram 70% WG, Mancozeb 75% WP and Metalaxyl 8%+Mancozeb 64%WP) were tested *in vitro* against eight selective strains of *Trichoderma* viz., TKHR1, TKHR2, TKHR6, TKHR10, TDHR12, TDHR15, TKHW1 and TKHW19, which are better performer against variable range of temperature and H ion concentration to see their tolerance levels. The procedure was explained in detail in the previous chapter materials and methods.

4.6.1 Weedicides

Three weedicides namely Glyphosate 41% SL, Metsulfuron methyl 20%WP and Imazathypr 10%SL at two different concentrations of 500 ppm and 1000 ppm were tested against 8 strains of *Trichoderma* to see their tolerance levels under *in vitro* condition. The data recoded per cent inhibition on the colony diameter of different strains of *Trichoderma* at 48 hrs. 72 hrs.and 96hrs. was studied.

4.6.1.1 Glyphosate 41 % SL

The effect of Glyphosate 41 % SL at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The mean of three replications at two different concentrations have been presented in Table 4.4, which revealed that the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW19 (23.87%) and minimum per cent inhibition with the *Trichoderma* strainTKHR6 (15.58%)at 500 ppm after 96 hrs.

Among the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW19 (29.95%) and minimum per cent inhibition with the *Trichoderma* strainTKHR6 (24.56%)at 1000 ppm after 96 hrs. All the strains of *Trichoderma* showed significant differences among variable concentration levels.

Table 4.4 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Glyphosate 41 % SL

| Treatments | Glyphosate 41 % SL | | | | | |
|------------|--------------------|--------------|--------------|--------------|--------------|--------------|
| | 500 ppm | | | 1000 ppm | | |
| | 48 hrs. | 72 hrs. | 96 hrs. | 48 hrs. | 72 hrs. | 96 hrs. |
| TKHR1 | 5.96(14.13) | 10.57(18.97) | 15.92(23.51) | 9.73(18.17) | 17.62(24.70) | 26.11(30.72) |
| TKHR2 | 8.20(16.64) | 12.28(20.52) | 22.64(28.40) | 11.19(19.53) | 20.85(27.16) | 28.74(32.41) |
| TKHR6 | 3.56(10.88) | 7.81(16.23) | 15.58(23.25) | 9.67(18.12) | 17.34(24.57) | 24.56(29.71) |
| TKHR10 | 6.29(14.52) | 11.15(19.50) | 16.03(23.60) | 10.56(18.96) | 18.05(25.13) | 26.49(30.97) |
| TDHR12 | 7.25(15.62) | 11.63(19.94) | 19.28(26.05) | 11.15(19.49) | 20.29(26.76) | 27.96(31.92) |
| TDHR15 | 7.46(15.85) | 11.74(20.04) | 21.16(27.38) | 11.16(19.50) | 20.60(26.99) | 28.06(31.97) |
| TKHW18 | 6.30(14.54) | 11.43(19.76) | 16.32(23.83) | 10.81(19.19) | 18.94(25.89) | 26.71(31.12) |
| TKHW19 | 9.43(17.88) | 12.40(20.62) | 23.87(29.24) | 12.81(20.95) | 19.66(26.32) | 29.95(35.18) |
| Mean | 6.81(15.01) | 11.13(19.45) | 18.85(25.66) | 10.89(19.24) | 19.17(25.93) | 27.32(31.50) |
| SEm± | 0.05 | 0.06 | 0.22 | 0.20 | 0.57 | 0.38 |
| CD at % | 0.16 | 0.17 | 0.66 | 0.60 | 1.72 | 1.16 |

* Mean value of three replication ** $\sqrt{\text{Arc sign transform value}}$

4.6.1.2 Metsulfuron methyl 20 % WP

The effect of Metsulfuron methyl 20 % WP at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The mean of three replications at two different concentrations have been presented in Table 4.5, which revealed that the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW18 (23.52%) and minimum per cent inhibition with the *Trichoderma* strainTKHR1 (16.77%)at 500 ppm after 96 hrs.

Among the maximum per cent inhibition recorded with the *Trichoderma* strain TKHW18 (27.65%) and minimum per cent inhibition with the *Trichoderma* strainTKHR1 (20.79%)at 1000 ppm after 96 hrs. All the strains of *Trichoderma* showed significant differences among variable concentration levels.

Table 4.5 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Metsulfuron methyl 20%

| Treatments | Metsulfuron methyl 20 % WP | | | | | |
|------------|----------------------------|--------------|--------------|--------------|--------------|--------------|
| | 500 ppm | | | 1000 ppm | | |
| | 48 hrs. | 72 hrs. | 96 hrs. | 48 hrs. | 72 hrs. | 96 hrs. |
| TKHR1 | 3.02(9.98) | 9.60(18.04) | 16.77(24.17) | 9.11(17.56) | 14.08(22.02) | 20.79(27.12) |
| TKHR2 | 3.88(11.33) | 9.78(18.21) | 19.55(26.24) | 9.41(17.85) | 15.96(23.54) | 21.22(27.43) |
| TKHR6 | 4.47(12.16) | 10.82(19.19) | 21.93(27.92) | 10.66(19.06) | 19.93(26.51) | 23.35(28.89) |
| TKHR10 | 4.40(11.97) | 10.68(19.07) | 21.78(27.81) | 10.04(18.48) | 19.41(26.09) | 22.87(28.54) |
| TDHR12 | 4.14(11.73) | 10.51(18.92) | 21.26(27.45) | 9.60(18.05) | 17.67(24.83) | 21.60(27.68) |
| TDHR15 | 3.98(11.51) | 10.29(18.69) | 20.63(27.01) | 9.58(18.03) | 16.00(23.58) | 21.53(27.63) |
| TKHW18 | 7.04(15.38) | 13.70(21.71) | 23.52(29.00) | 11.28(19.63) | 20.67(27.03) | 27.65(31.71) |
| TKHW19 | 4.78(12.60) | 11.22(19.56) | 22.41(28.24) | 11.49(19.81) | 20.49(26.90) | 23.59(29.05) |
| Mean | 4.46(12.08) | 10.82(19.17) | 20.98(27.23) | 10.15(18.56) | 18.03(25.06) | 22.83(28.51) |
| SEm± | 0.21 | 0.57 | 0.33 | 0.11 | 0.43 | 0.44 |
| CD at % | 0.63 | 1.71 | 1.00 | 0.34 | 1.31 | 1.33 |

* Mean value of three replication ** $\sqrt{\text{Arc sign transform value}}$

4.6.1.3 Imazathypr 10 %SL

The effect of Imazathypr 10 %SL at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The mean of three replications at two different concentrations have been presented in Table 4.6, which revealed that the maximum per cent inhibition recorded with the *Trichoderma* strain TKHW18 (28.11%) and minimum per cent inhibition with the *Trichoderma* strain TKHR6 (22.16%) at 500 ppm after 96 hrs.

Among the maximum per cent inhibition recorded with the *Trichoderma* strain TKHW18 (34.88%) and minimum per cent inhibition with the *Trichoderma* strain TKHR6 (26.01%) at 1000 ppm after 96 hrs. All the strains of *Trichoderma* showed significant differences among variable concentration levels.

Table 4.6 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Imazathyr 10 %SL

| Treatments | Imazathyr 10 %SL | | | | | |
|----------------|------------------|--------------|--------------|--------------|--------------|--------------|
| | 500 ppm | | | 1000 ppm | | |
| | 48 hrs | 72 hrs | 96 hrs | 48 hrs | 72 hrs | 96 hrs |
| TKHR1 | 4.47(12.15) | 13.24(21.33) | 24.89(25.92) | 8.45(16.89) | 19.24(26.01) | 30.19(33.29) |
| TKHR2 | 4.64(12.42) | 14.52(22.39) | 25.17(30.09) | 9.26(17.71) | 20.23(26.72) | 32.91(35.01) |
| TKHR6 | 3.88(11.35) | 11.45(19.77) | 22.16(28.07) | 6.02(14.20) | 17.77(24.92) | 26.01(30.62) |
| TKHR10 | 5.95(14.05) | 14.94(22.71) | 26.40(30.91) | 9.71(18.16) | 20.98(27.25) | 33.73(35.50) |
| TDHR12 | 6.71(14.98) | 16.01(23.57) | 26.53(30.98) | 9.86(18.28) | 21.01(27.26) | 34.79(36.14) |
| TDHR15 | 4.45(12.15) | 13.17(21.26) | 23.81(29.18) | 7.39(15.78) | 17.84(24.97) | 26.20(30.78) |
| TKHW18 | 6.87(15.15) | 17.06(24.39) | 28.11(32.01) | 10.23(18.84) | 22.49(28.28) | 34.88(36.19) |
| TKHW19 | 5.10(13.04) | 14.81(22.63) | 25.56(30.37) | 9.37(17.81) | 20.47(26.88) | 33.19(35.16) |
| Mean | 5.26(13.16) | 14.40(22.26) | 25.33(30.19) | 8.79(17.18) | 20.00(26.54) | 31.49(34.08) |
| SEm± | 0.24 | 0.30 | 0.55 | 0.17 | 0.41 | 0.74 |
| CD at % | 0.72 | 0.92 | 1.67 | 0.53 | 1.25 | 2.22 |

* Mean value of three replication ** $\sqrt{\text{Arc sign transform value}}$

4.6.2 Insecticide

Three insecticides namely Phorate 10 Cg, Chlorpyrifos 20 %EC and Quinalphos 25% EC at two different concentrations of 500 ppm and 1000 ppm were tested against 8 strains of *Trichoderma* to see their tolerance levels under *in vitro* condition. The data recorded per cent inhibition on the colony diameter of different strains of *Trichoderma* at 48 hrs. 72 hrs and 96hrs was studied.

4.6.2.1 Phorate 10 Cg

The effect of Phorate 10 Cg at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The mean of three replications at two different concentrations have been presented in Table 4.7, which revealed that the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW19 (23.88%) and minimum per cent inhibition with the *Trichoderma* strainTKHR1 (15.17%)at 500 ppm after 96 hrs.

Among the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW19 (31.54%) and minimum per cent inhibition with the *Trichoderma* strainTKHR1 (15.17%)at 1000 ppm after 96 hrs. All the strains of *Trichoderma* showed significant differences among variable concentration levels.

Table 4.7 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Phorate 10 Cg

| Treatments | Phorate 10 Cg | | | | | |
|----------------|---------------|--------------|--------------|--------------|--------------|--------------|
| | 500 ppm | | | 1000 ppm | | |
| | | | | | | |
| TKHR1 | 4.90(12.78) | 9.33(17.78) | 15.17(22.92) | 6.55(14.76) | 14.72(22.51) | 23.77(29.17) |
| TKHR2 | 5.29(13.29) | 10.86(19.23) | 16.54(23.84) | 7.71(16.11) | 18.46(25.44) | 25.78(30.51) |
| TKHR6 | 5.45(13.49) | 11.12(19.46) | 18.26(25.28) | 8.67(17.12) | 20.22(26.72) | 26.76(31.13) |
| TKHR10 | 5.80(13.87) | 11.15(19.49) | 18.46(20.40) | 9.06(17.42) | 20.90(27.18) | 28.95(32.54) |
| TDHR12 | 5.22(13.17) | 10.59(18.98) | 16.51(23.91) | 6.64(14.89) | 18.34(25.34) | 24.07(29.37) |
| TDHR15 | 5.15(13.09) | 10.13(18.55) | 15.93(23.49) | 6.63(14.88) | 17.92(25.01) | 24.00(29.33) |
| TKHW18 | 5.84(13.97) | 11.40(19.72) | 19.37(26.09) | 9.62(18.07) | 21.07(27.28) | 30.92(33.78) |
| TKHW19 | 6.60(14.87) | 12.71(20.80) | 23.88(29.25) | 10.07(18.50) | 21.37(27.52) | 31.54(34.16) |
| Mean | 5.53(13.57) | 10.91(19.26) | 18.02(25.02) | 8.12(16.47) | 19.12(25.88) | 26.97(31.25) |
| SEm± | 0.20 | 0.27 | 0.68 | 0.30 | 0.51 | 0.42 |
| CD at % | 0.61 | 0.82 | 2.04 | 0.92 | 1.54 | 1.28 |

* Mean value of three replication ** $\sqrt{\text{Arc sign transform value}}$

4.6.2.2 Chlorpyrifos 20 %EC

The effect of Chlorpyrifos 20 %EC at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The mean of three replications at two different concentrations have been presented in Table 4.8, which revealed that the maximum per cent inhibition recorded with the *Trichoderma* strain TDHR15 (26.03%) and minimum per cent inhibition with the *Trichoderma* strain TKHR1 (20.53%) at 500 ppm after 96 hrs.

Among the maximum per cent inhibition recorded with the *Trichoderma* strain TDHR15 (28.52%) and minimum per cent inhibition with the *Trichoderma* strain TKHR1 (24.74%) at 1000 ppm after 96 hrs. All the strains of *Trichoderma* showed significant differences among variable concentration levels.

Table 4.8 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Chlorpyriphos 20 %EC

| Treatments | Chlorpyriphos 20 %EC | | | | | |
|----------------|----------------------|--------------|--------------|--------------|--------------|--------------|
| | 500 ppm | | | 1000 ppm | | |
| | 48 hrs | 72 hrs | 96 hrs | 48 hrs | 72 hrs | 96 hrs |
| TKHR1 | 4.00(10.72) | 9.49(17.94) | 20.53(26.93) | 8.25(16.67) | 13.84(21.83) | 24.74(29.83) |
| TKHR2 | 5.97(14.12) | 19.26(26.00) | 22.58(28.35) | 9.64(18.09) | 19.89(26.48) | 27.05(31.34) |
| TKHR6 | 5.15(13.10) | 13.71(21.73) | 21.92(27.90) | 8.64(17.08) | 14.76(22.59) | 25.60(30.40) |
| TKHR10 | 5.83(13.93) | 18.19(25.24) | 22.52(28.31) | 9.22(17.64) | 18.23(25.26) | 26.60(31.05) |
| TDHR12 | 5.52(13.54) | 18.04(25.12) | 22.08(28.02) | 8.98(17.43) | 16.13(23.68) | 26.23(30.80) |
| TDHR15 | 6.53(14.75) | 21.04(27.26) | 26.03(30.67) | 10.81(19.18) | 20.58(26.97) | 28.52(32.24) |
| TKHW18 | 5.00(12.91) | 12.79(20.95) | 20.91(27.20) | 8.40(16.83) | 14.61(22.47) | 25.56(30.37) |
| TKHW19 | 6.34(14.56) | 19.74(26.37) | 22.86(28.54) | 10.41(18.82) | 20.53(26.94) | 27.12(31.38) |
| Mean | 5.54(13.46) | 16.53(23.83) | 22.43(28.24) | 9.29(17.72) | 17.32(24.53) | 26.43(30.93) |
| SEm± | 0.34 | 0.42 | 0.51 | 0.22 | 0.30 | 0.26 |
| CD at % | 1.04 | 1.27 | 1.53 | 0.67 | 0.90 | 0.79 |

* Mean value of three replication ** $\sqrt{\text{Arc sign transform value}}$

4.6.2.3 Quinalphos 25% EC

The effect of Quinalphos 25% EC at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The mean of three replications at two different concentrations have been presented in Table 4.9, which revealed that the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW19 (20.15%) and minimum per cent inhibition with the *Trichoderma* strainTKHR10 (14.44%)at 500 ppm after 96 hrs.

Among the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW19 (24.52%) and minimum per cent inhibition with the *Trichoderma* strainTKHR10 (19.50%)at 1000 ppm after 96 hrs. All the strains of *Trichoderma* showed significant differences among variable concentration levels.

Table 4.9 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Quinalphos 25% EC

| Treatments | Quinalphos 25% EC | | | | | |
|----------------|-------------------|--------------|--------------|--------------|--------------|--------------|
| | 500 ppm | | | 1000 ppm | | |
| | 48 hrs. | 72 hrs. | 96 hrs. | 48 hrs. | 72 hrs. | 96 hrs. |
| TKHR1 | 6.78(14.99) | 11.82(26.06) | 17.33(24.59) | 10.01(18.44) | 17.67(24.84) | 23.87(29.24) |
| TKHR2 | 4.98(12.81) | 10.03(18.47) | 14.72(22.55) | 8.80(17.23) | 15.04(22.82) | 20.33(26.78) |
| TKHR6 | 6.56(14.58) | 11.77(20.06) | 17.26(24.52) | 9.51(17.93) | 15.65(23.26) | 23.53(29.01) |
| TKHR10 | 4.52(12.22) | 9.85(18.29) | 14.44(22.32) | 8.46(16.91) | 12.10(20.33) | 19.50(26.20) |
| TDHR12 | 5.67(13.77) | 10.08(18.50) | 15.15(22.91) | 8.94(17.39) | 15.35(23.06) | 21.71(27.76) |
| TDHR15 | 5.82(13.91) | 10.16(18.57) | 16.45(23.92) | 9.45(17.89) | 15.52(23.14) | 21.87(27.88) |
| TKHW18 | 7.45(15.52) | 12.56(20.69) | 18.55(25.50) | 10.25(18.62) | 17.79(24.87) | 24.07(29.31) |
| TKHW19 | 7.49(15.85) | 13.03(21.15) | 20.15(26.67) | 11.94(20.20) | 18.72(25.62) | 24.52(29.67) |
| Mean | 6.16(14.21) | 11.16(19.47) | 16.76(24.12) | 9.67(18.08) | 15.98(23.49) | 22.43(28.23) |
| SEm± | 0.52 | 0.32 | 0.33 | 0.29 | 0.55 | 0.57 |
| CD at % | 1.56 | 0.97 | 0.99 | 0.86 | 1.66 | 1.72 |

* Mean value of three replication ** $\sqrt{\text{Arc sign transform value}}$

4.6.3 Fungicides

Three fungicides namely, Metiram 70% WG, Mancozeb 75% WP and Metalaxyl 8%+Mancozeb 64% WP at two different concentrations of 500 ppm and 1000 ppm were tested against 8 strains of *Trichoderma* to see their tolerance levels under *in vitro* condition. The data recorded per cent inhibition on the colony diameter of different strains of *Trichoderma* at 48 hrs. 72 hrs and 96hrs.was studied.

4.6.3.1 Metiram 70% WG

The effect of Metiram 70% WG at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The mean of three replications at two different concentrations have been presented in Table 4.10, which revealed the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW18 (23.56%) and minimum per cent inhibition with the *Trichoderma* strainTKHR1 (13.46%)at 500 ppm after 96 hrs.

Among the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW18 (32.04%) and minimum per cent inhibition with the *Trichoderma* strainTKHR1 (26.36%)at 1000 ppm after 96 hrs. All the strains of *Trichoderma* showed significant differences among variable concentration levels.

Table 4.10 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Metiram 70% WG

| Treatments | Metiram 70% WG | | | | | |
|------------|----------------|--------------|--------------|--------------|--------------|--------------|
| | 500 ppm | | | 1000 ppm | | |
| | 48 hrs. | 72 hrs. | 96 hrs. | 48 hrs. | 72 hrs. | 96 hrs. |
| TKHR1 | 5.33(13.35) | 12.85(21.00) | 13.46(21.49) | 8.85(17.26) | 19.57(26.26) | 26.36(30.89) |
| TKHR2 | 6.63(13.98) | 13.19(21.2) | 19.05(25.87) | 9.98(18.41) | 20.86(27.17) | 27.00(31.30) |
| TKHR6 | 7.23(15.58) | 18.69(25.60) | 20.04(26.59) | 10.33(18.71) | 23.86(29.23) | 27.93(31.89) |
| TKHR10 | 8.15(16.59) | 20.12(26.63) | 20.72(27.06) | 10.56(18.95) | 23.94(29.28) | 28.33(32.16) |
| TDHR12 | 6.59(14.85) | 14.48(22.37) | 19.89(26.47) | 10.28(18.70) | 21.63(27.71) | 27.06(31.34) |
| TDHR15 | 8.45(14.90) | 20.92(22.21) | 22.19(28.11) | 11.08(19.39) | 24.48(29.65) | 30.12(33.28) |
| TKHW18 | 8.84(17.28) | 21.37(27.53) | 23.56(28.97) | 11.40(19.71) | 26.18(30.77) | 32.04(34.47) |
| TKHW19 | 6.86(15.15) | 15.37(23.07) | 19.97(26.53) | 10.29(18.70) | 23.00(28.64) | 27.35(31.53) |
| Mean | 7.26(15.46) | 17.12(24.34) | 19.86(26.39) | 10.35(18.73) | 22.94(28.59) | 28.27(32.11) |
| SEm± | 0.45 | 0.33 | 0.54 | 0.32 | 0.41 | 0.36 |
| CD at % | 1.36 | 1.01 | 1.62 | 0.96 | 1.25 | 1.10 |

* Mean value of three replication ** $\sqrt{\text{Arc sign transform value}}$

4.6.3.2 Mancozeb 75% WP

The effect of Mancozeb 75% WP at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The mean of three replications at two different concentrations have been presented in Table 4.11, which revealed that the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW18 (23.03%) and minimum per cent inhibition with the *Trichoderma* strainTKHR1 (16.67%)at 500 ppm after 96 hrs.

Among the maximum per cent inhibition recorded with the *Trichoderma* strainTKHW18 (30.67%) and minimum per cent inhibition with the *Trichoderma* strainTKHR1 (23.93%)at 1000 ppm after 96 hrs. All the strains of *Trichoderma* showed significant differences among variable concentration levels.

Table 4.11 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Mancozeb 75% WP

| Treatments | Mancozeb 75% WP | | | | | |
|------------|-----------------|--------------|--------------|--------------|--------------|--------------|
| | 500 ppm | | | 1000 ppm | | |
| | 48 hrs | 72 hrs | 96 hrs | 48 hrs | 72 hrs | 96 hrs |
| TKHR1 | 5.36(13.34) | 9.53(17.96) | 16.67(24.08) | 9.79(18.23) | 18.90(25.76) | 23.93(29.28) |
| TKHR2 | 7.04(15.38) | 11.08(19.43) | 20.47(26.90) | 10.04(18.48) | 20.07(26.61) | 25.39(30.26) |
| TKHR6 | 5.88(14.02) | 11.00(19.37) | 18.59(25.53) | 9.86(18.29) | 19.48(26.18) | 24.98(29.99) |
| TKHR10 | 9.19(17.65) | 17.22(24.51) | 22.56(28.33) | 11.93(20.18) | 22.05(28.00) | 28.82(32.46) |
| TDHR12 | 8.93(17.37) | 16.35(23.80) | 21.72(27.76) | 11.36(19.68) | 21.90(27.90) | 27.10(31.37) |
| TDHR15 | 8.33(16.74) | 11.35(19.68) | 20.55(26.95) | 10.42(18.79) | 20.28(26.75) | 25.79(30.52) |
| TKHW18 | 10.31(18.73) | 18.60(25.52) | 23.03(28.66) | 12.25(20.44) | 23.60(29.04) | 30.67(33.63) |
| TKHW19 | 8.51(16.93) | 14.45(22.33) | 21.64(27.71) | 10.89(19.27) | 21.36(27.52) | 26.74(31.13) |
| Mean | 7.94(16.27) | 13.70(21.57) | 20.65(26.99) | 10.82(19.17) | 20.95(27.22) | 26.68(31.08) |
| SEm± | 0.21 | 0.39 | 0.45 | 0.31 | 0.36 | 0.32 |
| CD at % | 0.64 | 1.17 | 1.35 | 0.93 | 1.08 | 0.96 |

* Mean value of three replication ** $\sqrt{\text{Arc sign transform value}}$

4.6.3.2 Metalaxyl 8%+Mancozeb 64% Wp

The effect of Metalaxyl 8%+Mancozeb 64% Wp at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The mean of three replications at two different concentrations have been presented in Table 4.12, which revealed that the maximum per cent inhibition recorded with the *Trichoderma* strain TKHW18 (25.48%) and minimum per cent inhibition with the *Trichoderma* strain TKHR6 (19.87%) at 500 ppm after 96 hrs.

Among the maximum per cent inhibition recorded with the *Trichoderma* strain TKHW18 (32.82%) and minimum per cent inhibition with the *Trichoderma* strain TKHR6 (24.23%) at 1000 ppm after 96 hrs. All the strains of *Trichoderma* showed significant differences among variable concentration levels.

Table 4.12 Per cent inhibition on the colony diameter of different strains of *Trichoderma* against Metalaxyl 8%+Mancozeb 64% WP

| Treatments | Metalaxyl 8%+Mancozeb 64% Wp | | | | | |
|----------------|------------------------------|--------------|--------------|--------------|--------------|--------------|
| | 500 ppm | | | 1000 ppm | | |
| | 48 hrs | 72 hrs | 96 hrs | 48 hrs | 72 hrs | 96 hrs |
| TKHR1 | 6.22(14.44) | 11.72(19.98) | 20.53(26.93) | 9.55(17.99) | 18.82(25.70) | 25.32(30.21) |
| TKHR2 | 6.55(14.82) | 12.30(20.48) | 20.89(27.19) | 10.50(18.90) | 20.27(26.75) | 28.03(31.96) |
| TKHR6 | 5.15(13.11) | 11.36(19.68) | 19.87(26.47) | 8.67(17.13) | 17.29(24.55) | 24.23(29.49) |
| TKHR10 | 6.22(14.44) | 11.44(19.75) | 19.95(26.52) | 9.45(17.90) | 17.73(24.90) | 25.31(30.20) |
| TDHR12 | 8.73(17.18) | 16.60(24.03) | 21.26(27.44) | 11.21(19.53) | 21.46(27.59) | 32.75(34.91) |
| TDHR15 | 6.38(14.62) | 12.29(20.52) | 20.76(27.10) | 9.71(18.15) | 19.21(25.99) | 26.33(30.87) |
| TKHW18 | 9.13(17.58) | 18.62(25.55) | 25.48(30.31) | 11.68(19.98) | 23.26(28.83) | 32.82(34.94) |
| TKHW19 | 7.67(16.04) | 15.23(22.97) | 21.11(27.34) | 10.54(18.94) | 20.83(27.15) | 28.24(32.10) |
| Mean | 7.01(15.28) | 13.70(21.62) | 21.23(27.41) | 10.17(18.56) | 19.86(26.43) | 27.88(31.83) |
| SEm± | 0.15 | 0.36 | 0.33 | 0.23 | 0.28 | 0.29 |
| CD at % | 0.46 | 1.09 | 1.00 | 0.70 | 0.86 | 0.87 |

4.7 Evaluation the prominent strains of *Trichoderma* against soil borne diseases *in-vitro* and *in-vivo*

4.7.1 *In-vivo* evaluation of *Trichoderma* strains against wilt incidence of chickpea

A field experiment was entailed to evaluate the ability of the *Trichoderma* strains to control wilt incidence of chickpea plant under field condition (*In-vivo*) at college of Agriculture Indore during the season 2019-2020. Field conditions were set up using different strains of *Trichoderma* applied as seed treatment and by inoculating inoculums of the pathogen in the soil, the procedure of which was described early in the chapter Materials and methods.

The antagonistic potential of eight strains of *Trichoderma* was evaluated under field conditions for their effect against wilt incidence of chickpea. The results of per cent germination and incidence of disease were presented in table 4.13. The result indicated the highest per cent germination of chickpea seeds with the *Trichoderma* strain TKHR2 (99.03%) and the lowest per cent germination with the control (92.50%).

Among the antagonist potentiality of *Trichoderma* strains against incidence of wilt disease in chickpea. The data recorded after first disease symptom appearance of wilt incidence of chickpea when 25, 30, 35, 40, and 45 days after sowing.

The Minimum percent ofwilt incidence was recorded on chickpea with *Trichoderma* strain TKHR1 (2.08%) as compare to control (6.81%) after 45DAS. All the strains of *Trichoderma* significantly reduced the incidence of wilt disease of chickpea caused by wilt incidence of chickpea and increase the germination per cent of chickpea seed after *Trichoderma* product used ^sas a bio control agent.

Table-4.13 *In-vivo* evaluation of *Trichoderma* strains against wilt disease of chickpea

| Treatments | Germination % | Disease incidence % | | | | |
|----------------|------------------|---------------------|-----------------|-----------------|-----------------|-----------------|
| | | 25 DAS after | 30 DAS after | 35 DAS after | 40 DAS after | 45 DAS after |
| TKHR1 | 96.11 | 0.14 | 0.69 | 1.11 | 1.53 | 2.08 |
| TKHR2 | 99.03 | 0.83 | 1.25 | 1.53 | 2.22 | 3.19 |
| TKHR6 | 97.64 | 0.69 | 0.97 | 1.39 | 1.94 | 2.78 |
| TKHR10 | 97.36 | 0.69 | 0.83 | 1.25 | 1.94 | 2.92 |
| TDHR12 | 97.36 | 0.42 | 0.83 | 1.25 | 1.67 | 2.50 |
| TDHR15 | 97.92 | 0.69 | 1.11 | 1.39 | 1.94 | 3.06 |
| TKHW18 | 97.36 | 0.28 | 0.69 | 1.25 | 1.67 | 2.22 |
| TKHW19 | 98.06 | 0.69 | 1.11 | 1.53 | 2.22 | 3.19 |
| Conrol | 92.50 | 1.39 | 2.22 | 3.06 | 4.31 | 6.81 |
| SEm± | 1.15 | 0.96 | 0.57 | 0.54 | 0.49 | 0.83 |
| CD at % | 3.45 | 2.87 | 1.72 | 1.64 | 1.47 | 2.50 |

* Mean value of three replications

4.7.2 *In-vitro* evolution of *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri*

A experiment was conducted with eight *Trichoderma* isolated strain were screened for their biocontrol potential against *Fusarium oxysporum* f. sp. *Ciceri* under *in-vitro* conditions using dual culture technique as described in the previous chapter materials and methods. The result of mycelium inhibited area of fungal growth incubated at 24 ± 2 °C and data recording after 48 hrs., 72 hrs. and 96 hrs. in table-4.14.

The data indicated that all the strains of *Trichoderma* inhibited the colony growth of *Fusarium oxysporum* f. sp. *Ciceri*. The percentage inhibition on the growth of the pathogen was maximum with the strain TDHR15 (40.74%), which was found at par with all the other *Trichoderma* strains TKHW18 (35.07%), TKHR6 (35.01%), TKHW19 (34.43%), TKHR10 (32.61%), TKHR1 (28.64%), TDHR12 (22.02%) and TKHR2 (21.26%) at incubation after 96 hrs.

Table 4.14 *In-vitro* evolution of *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri*

| Treatments | Per cent inhibition of fungal pathogen | | |
|------------|--|--------------|--------------|
| | 48 hrs. | 72 hrs. | 96 hrs. |
| TKHR1 | 8.89(17.34) | 18.65(25.58) | 28.64(32.35) |
| TKHR2 | 7.14(15.49) | 13.36(21.43) | 21.26(27.45) |
| TKHR6 | 14.39(22.29) | 23.08(28.71) | 35.01(36.27) |
| TKHR10 | 10.96(19.32) | 20.10(26.62) | 32.61(34.81) |
| TDHR12 | 8.15(16.59) | 15.11(22.87) | 22.02(27.96) |
| TDHR15 | 98.33(85.69) | 27.02(31.31) | 40.74(39.66) |
| TKHW18 | 17.82(24.95) | 24.37(29.57) | 35.07(36.31) |
| TKHW19 | 13.81(21.80) | 22.82(28.51) | 34.43(35.93) |
| Mean | 22.44(27.93) | 20.56(26.83) | 31.22(33.84) |
| SEm± | 0.69 | 0.28 | 0.27 |
| CD at % | 2.07 | 0.84 | 0.83 |

* Mean value of three replication ** $\sqrt{\text{Arc sign transform value}}$

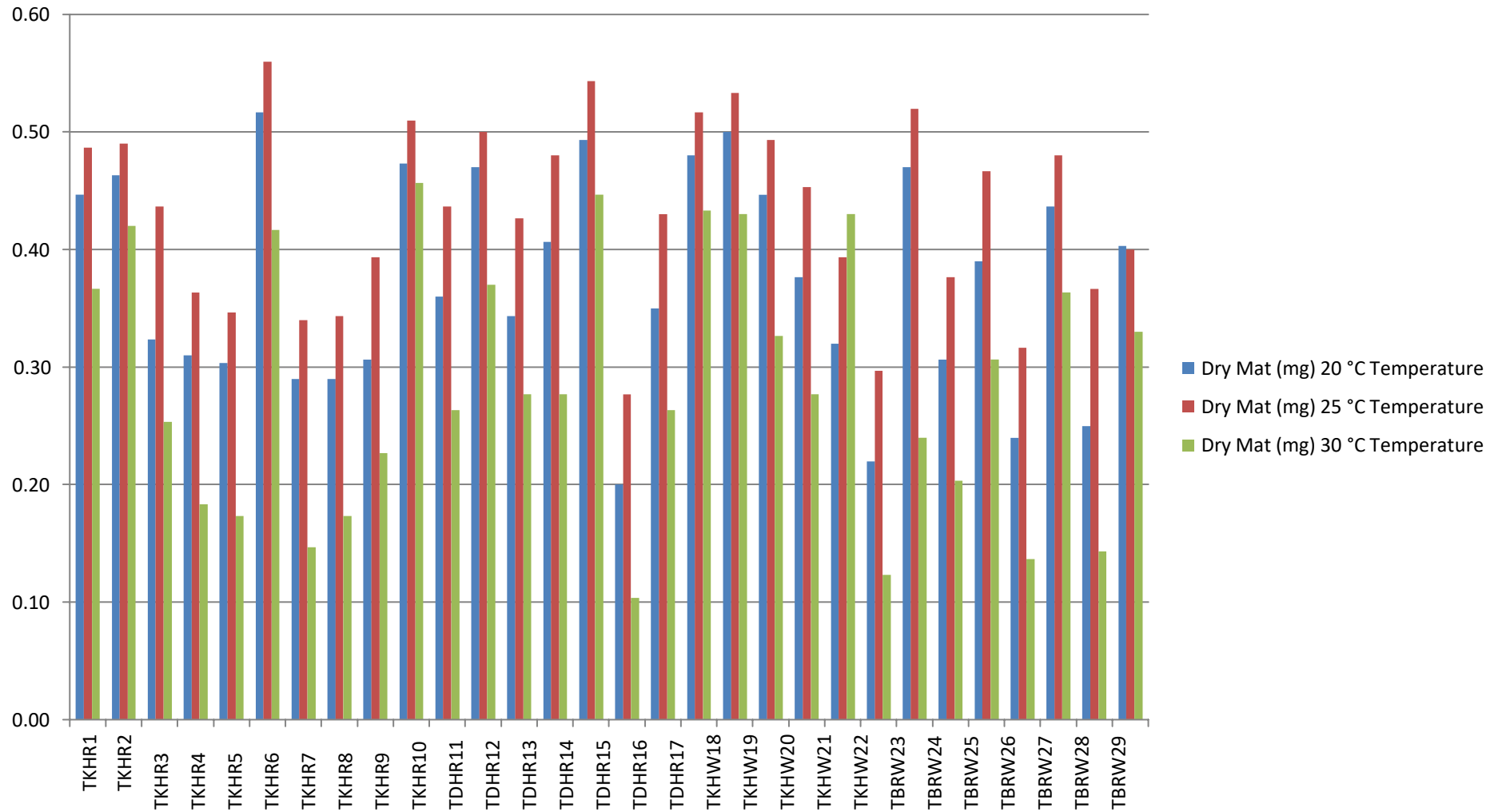
Figure-4.2 Screening of *Trichoderma* strains tolerant to variable range of temperature

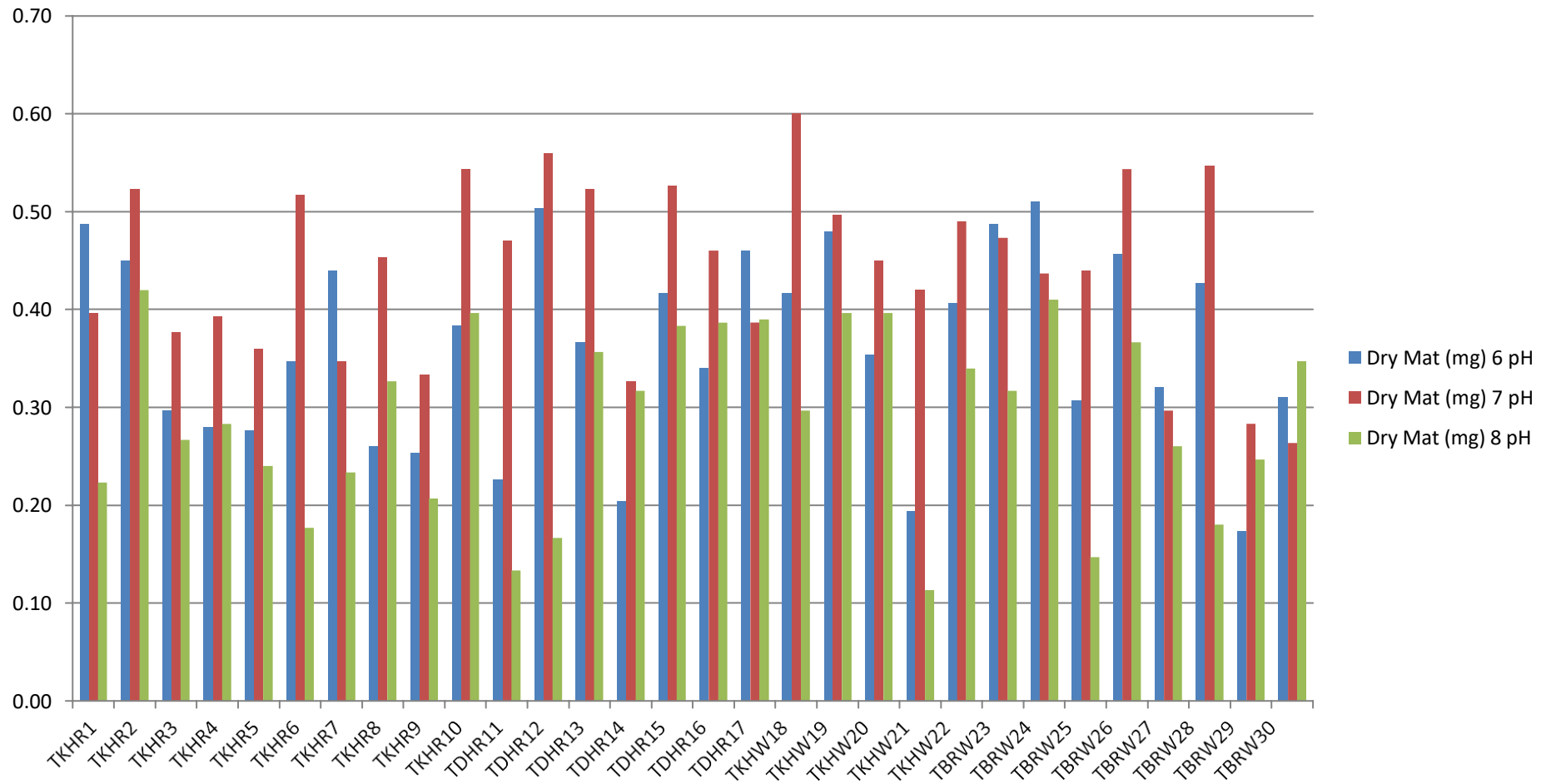
Figure-4.3 Screening of *Trichoderma* strains tolerant to variable range of H ion concentration.

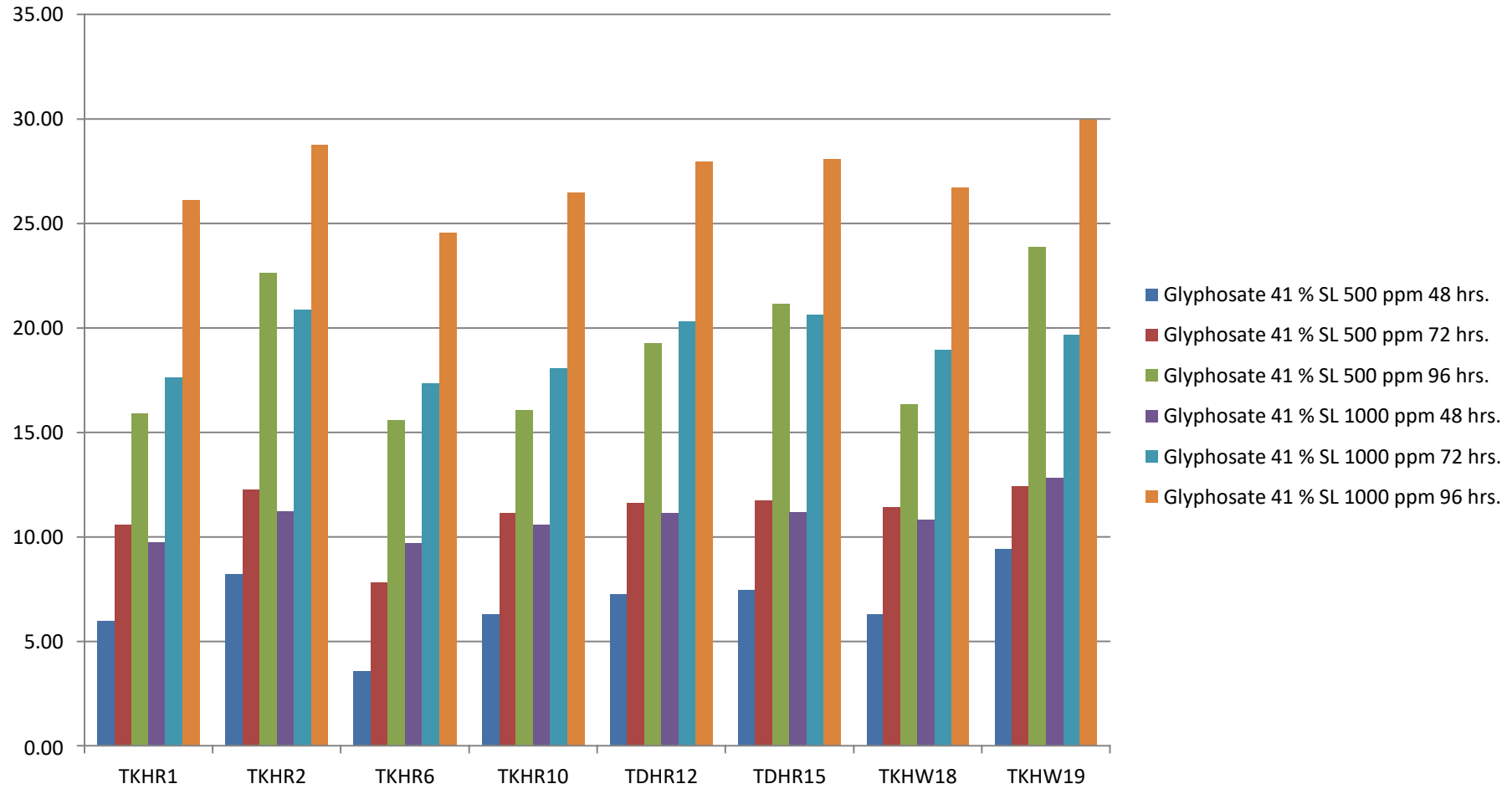
Figure- 4.4 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Glyphosate 41 % SL

Figure- 4.5 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Metsulfuron methyl 20% WP

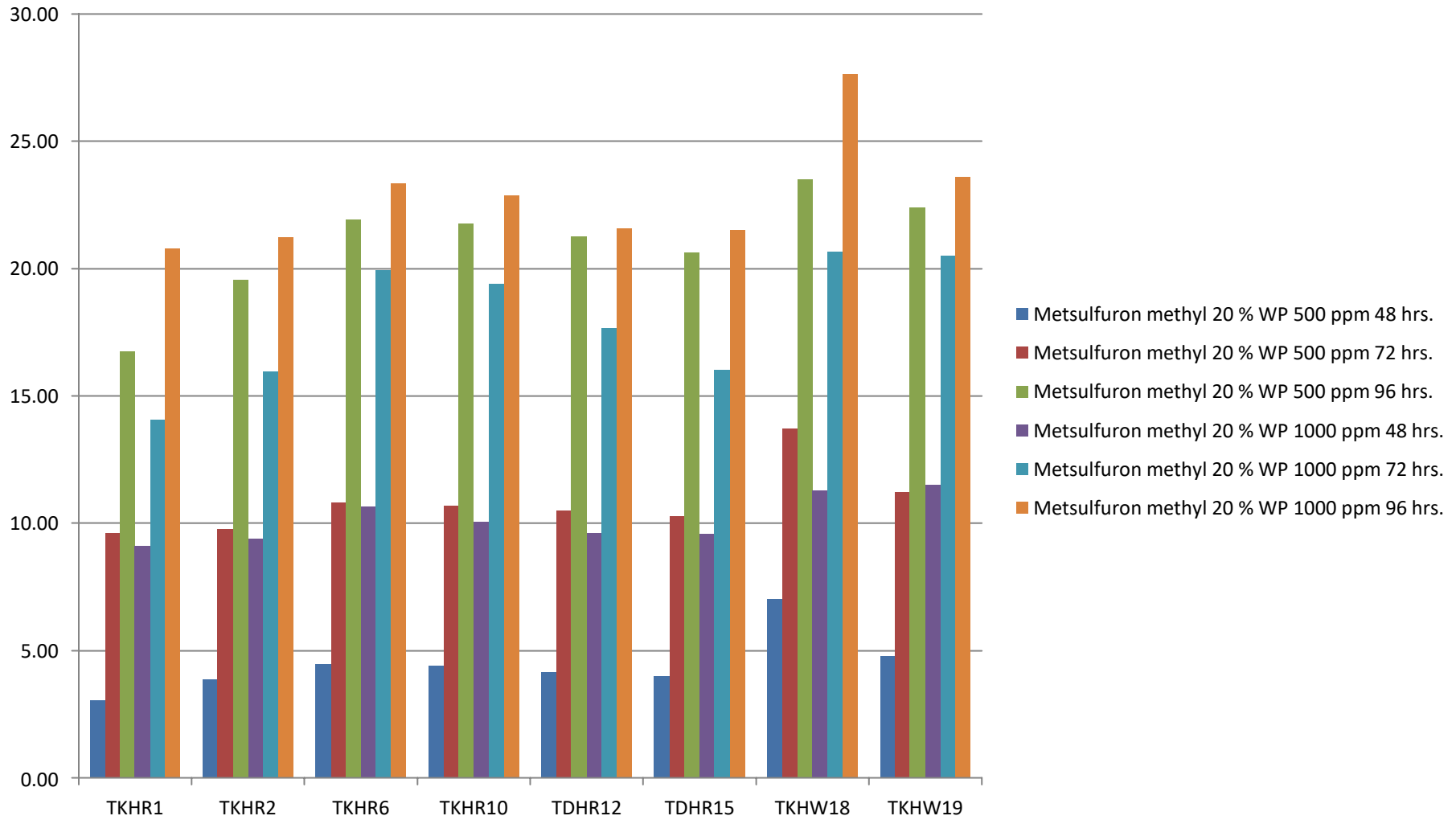


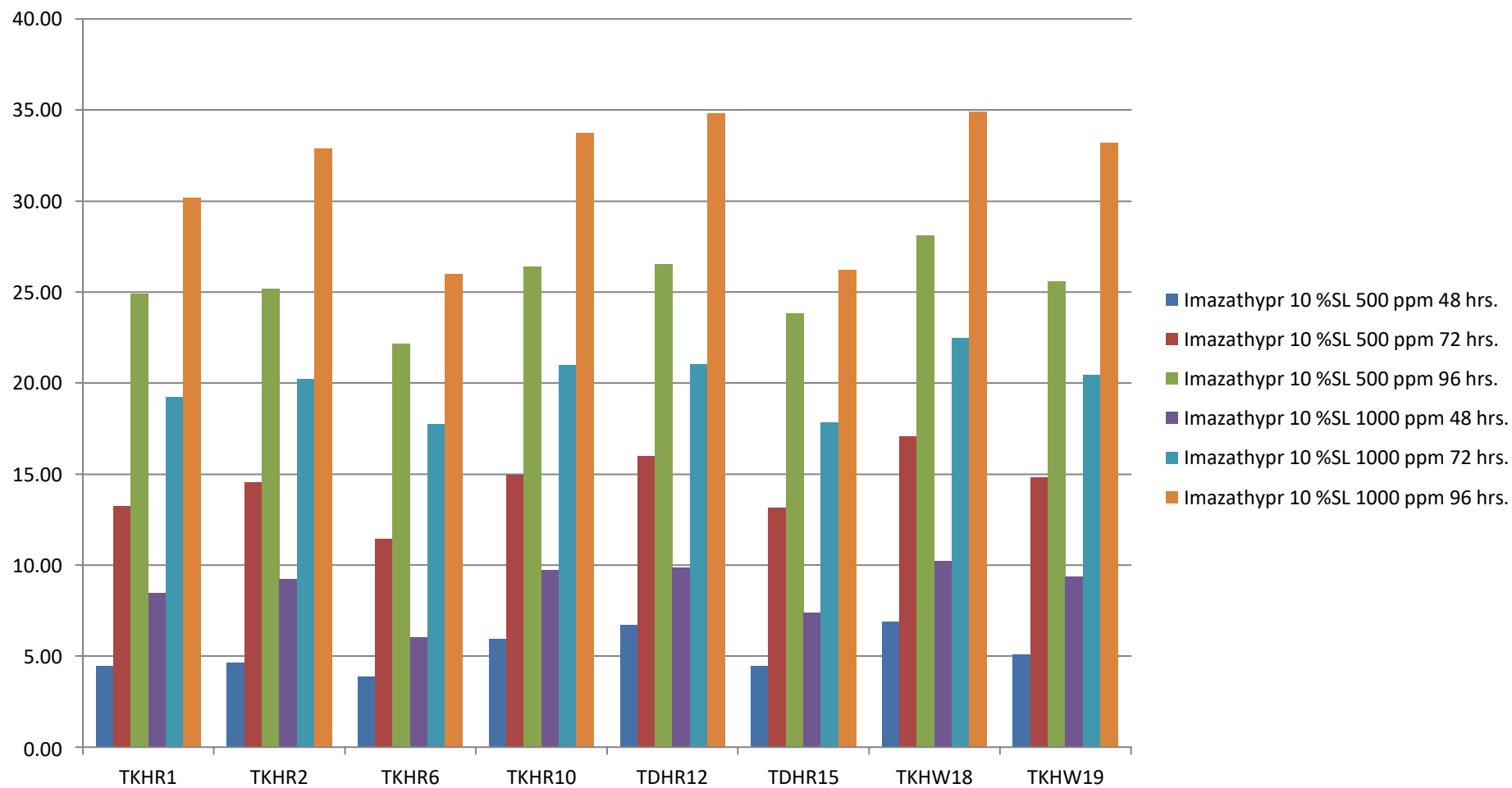
Figure- 4.6 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Imazathyr 10 %SL

Figure- 4.7 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Phorate 10 Cg

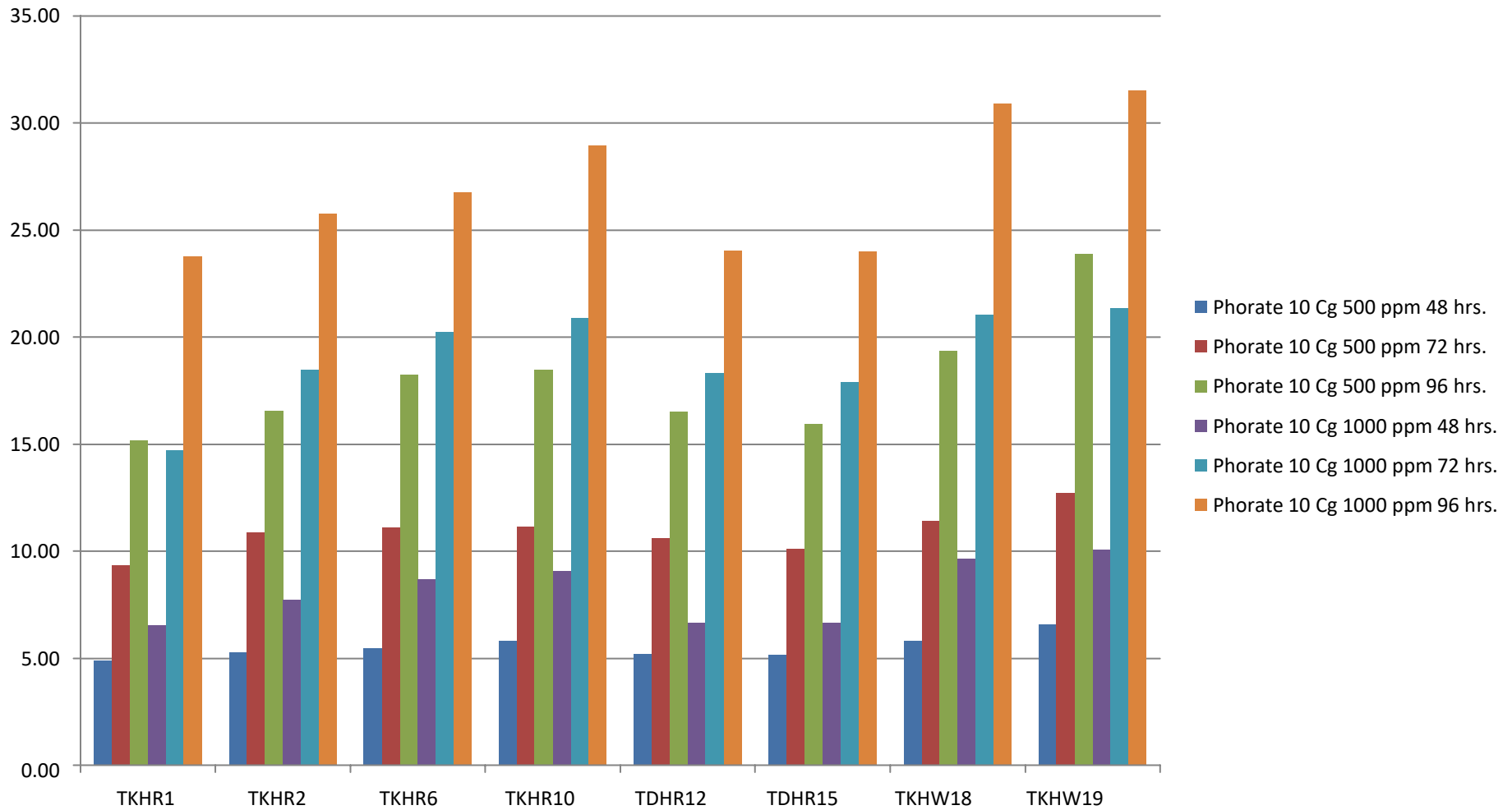


Figure- 4.8 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Chlorpyriphos 20 %EC

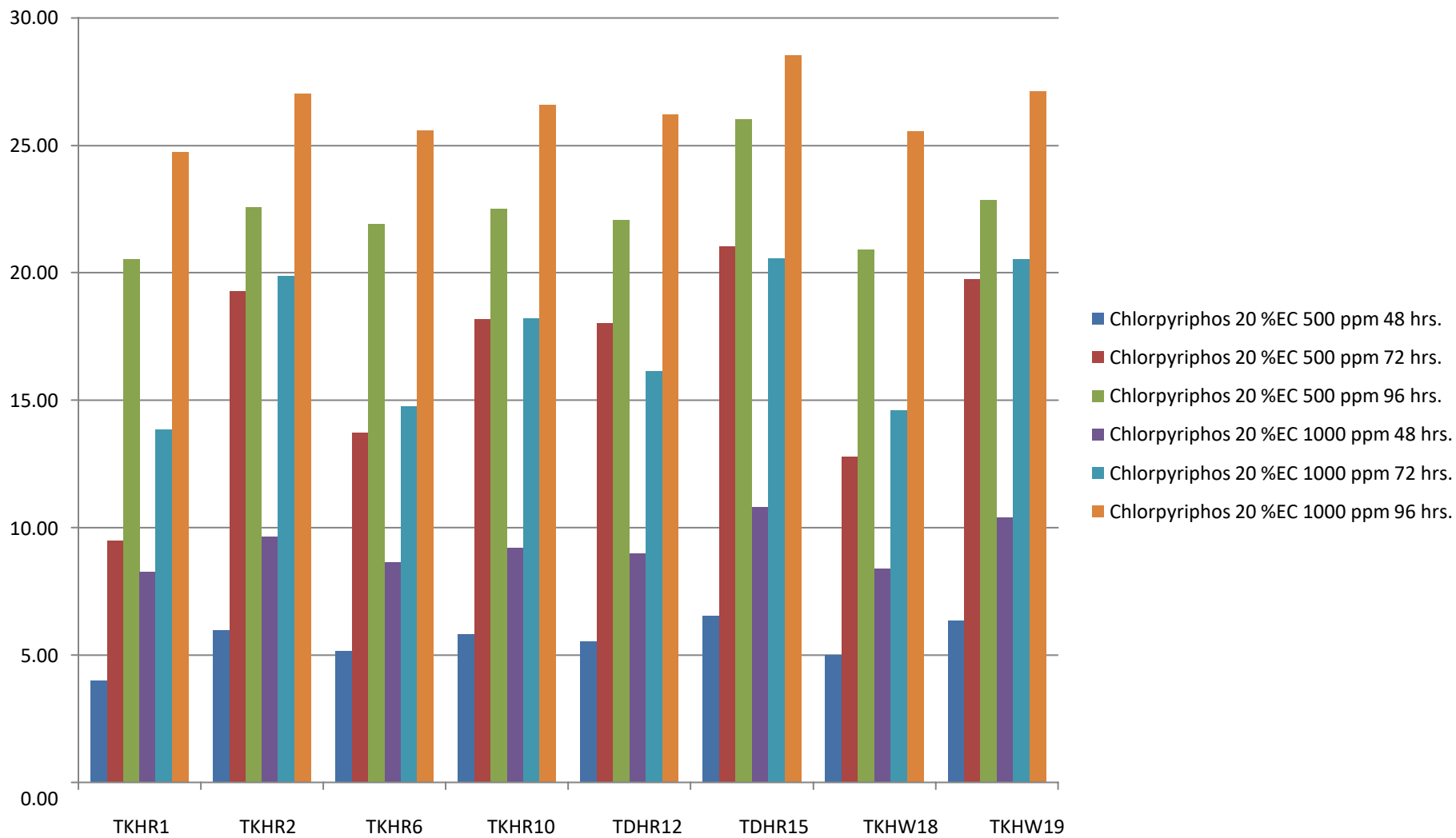


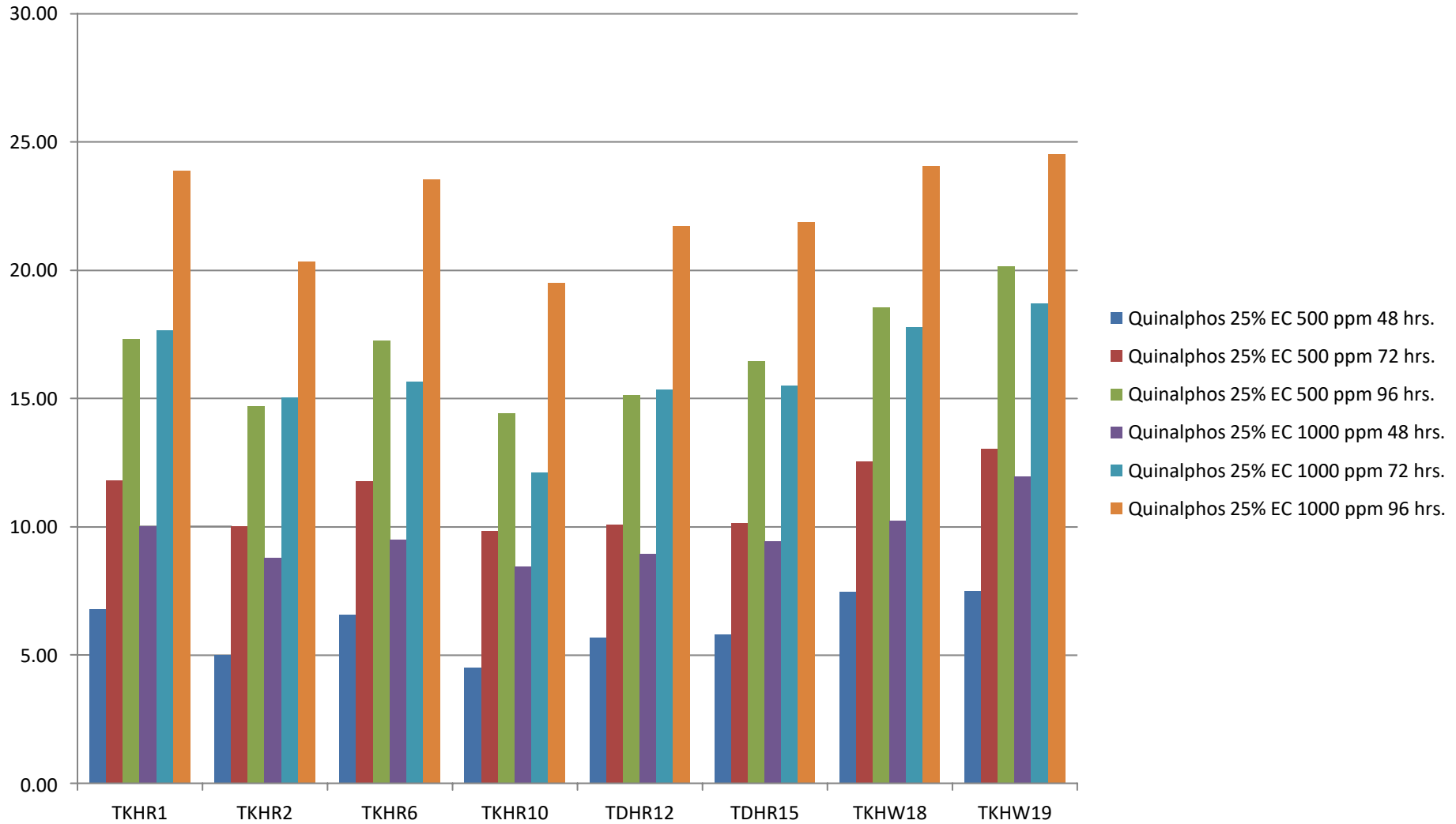
Figure- 4.9 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Quinalphos 25% EC

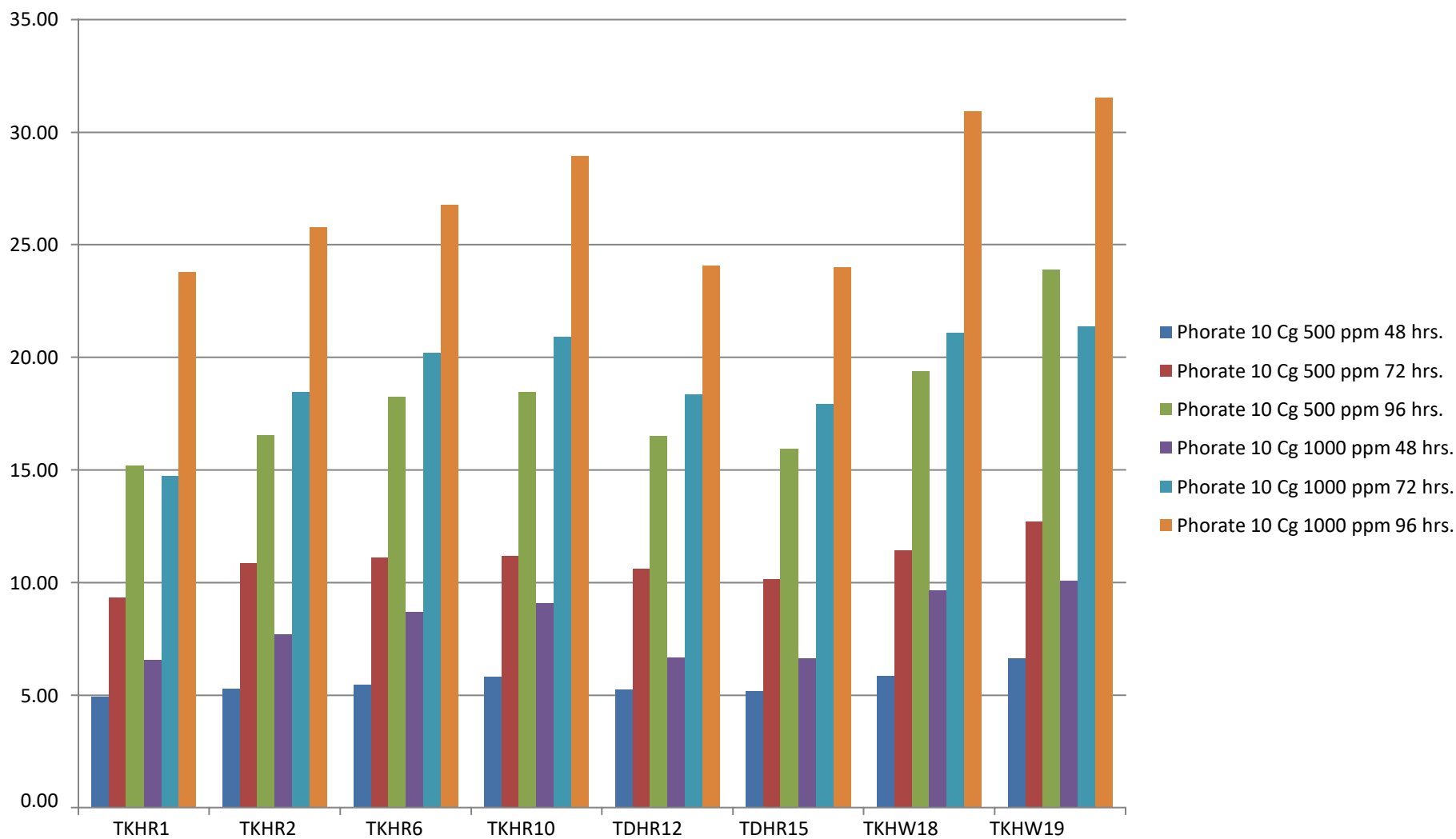
Figure- 4.10 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Metiram 70% WG

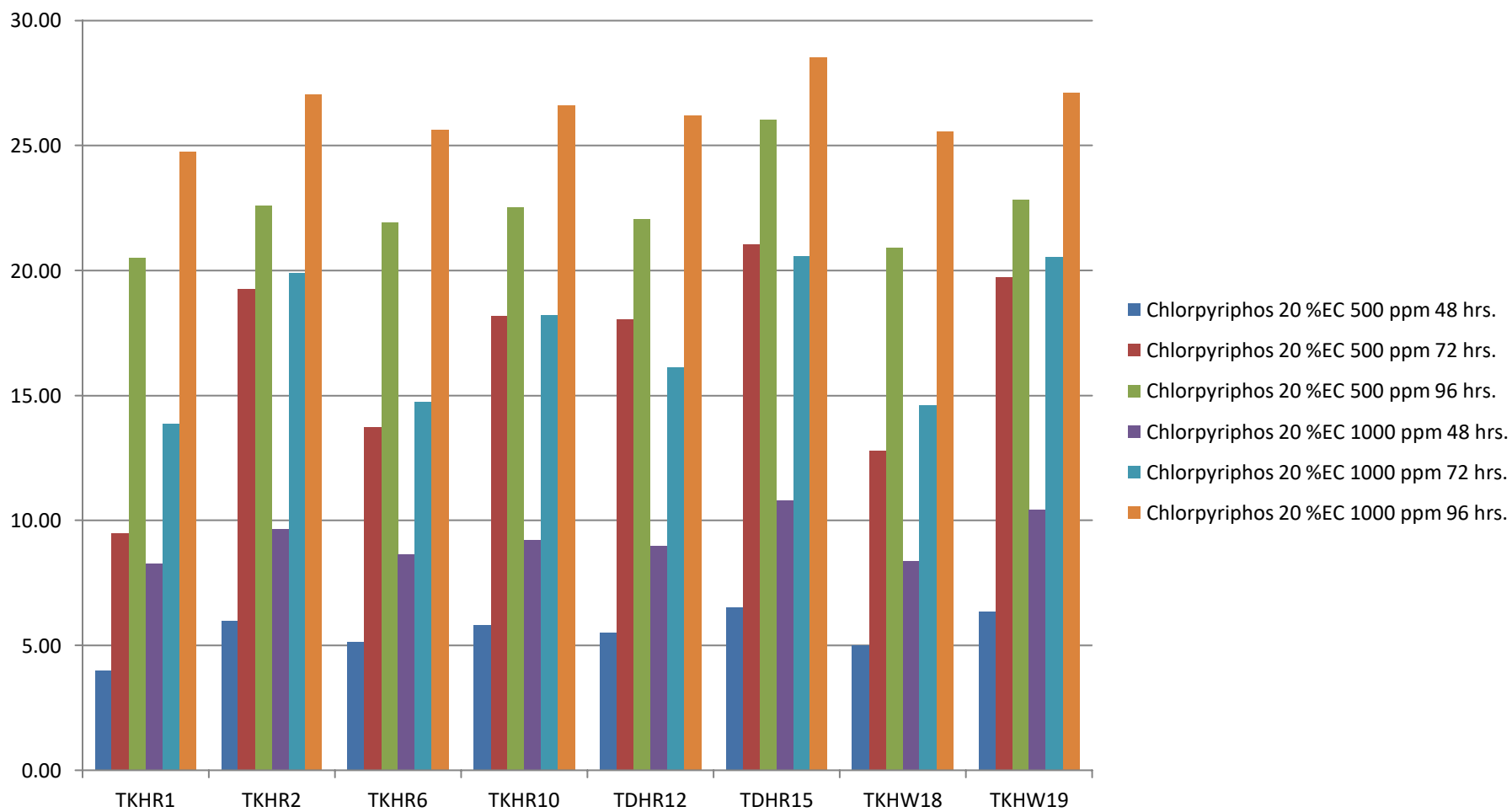
Figure- 4.11 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Mancozeb 75% WP

Figure- 4.12 per cent inhibition on the colony diameter of different strains of *Trichoderma* against Metalaxyl 8%+Mancozeb 64% WP

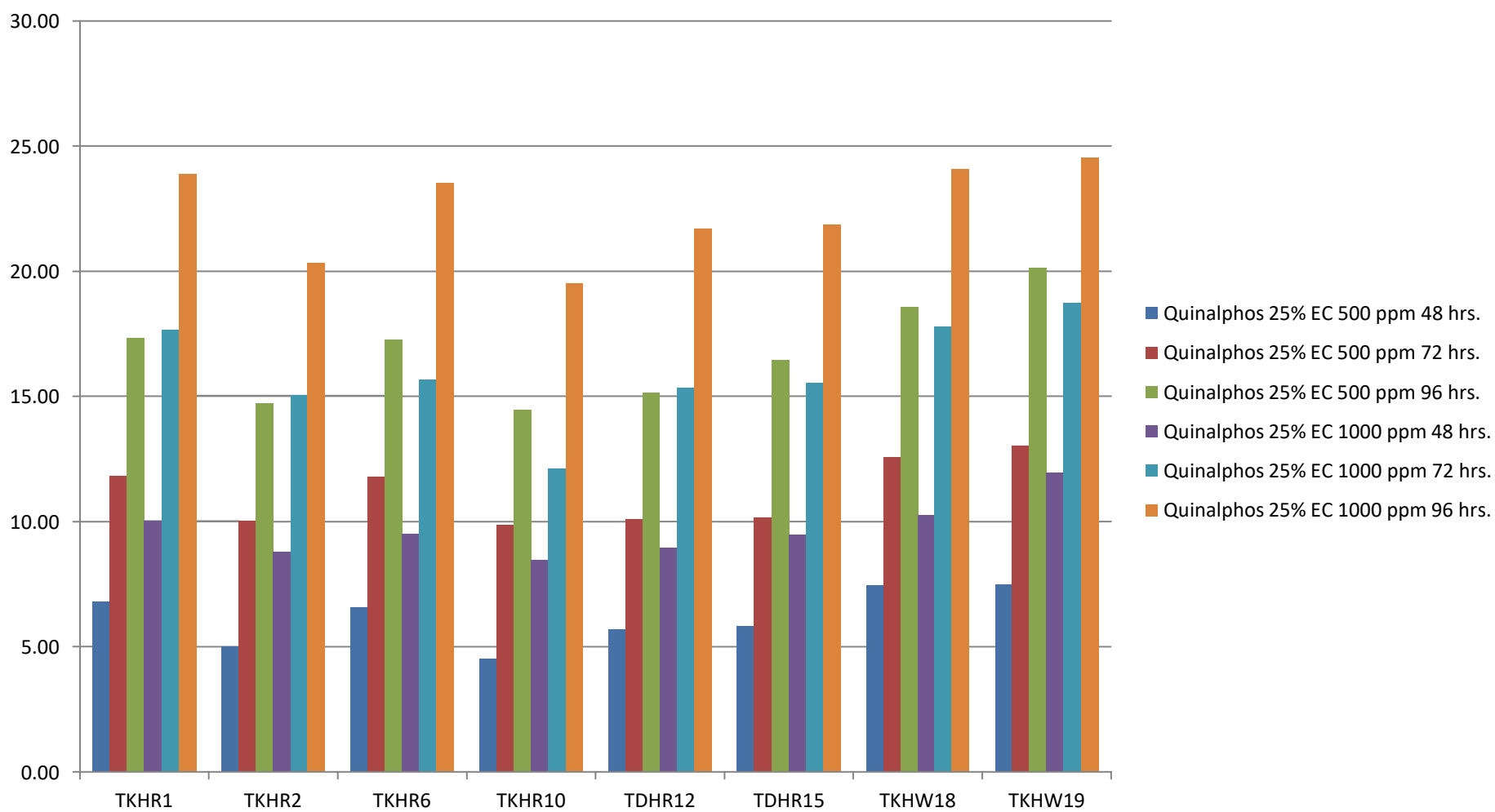


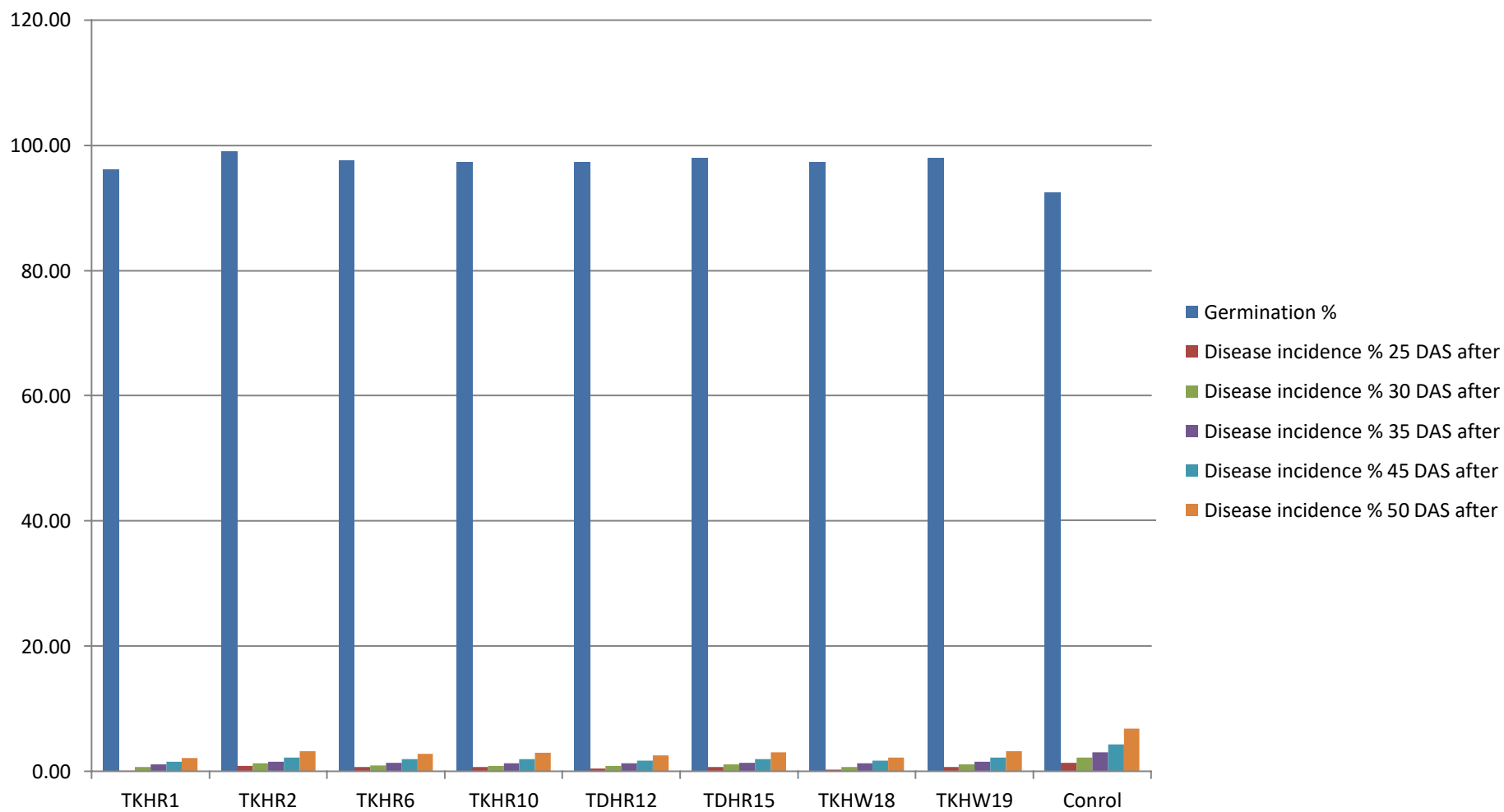
Figure- 4.13 *In-vivo* evaluation of *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri*

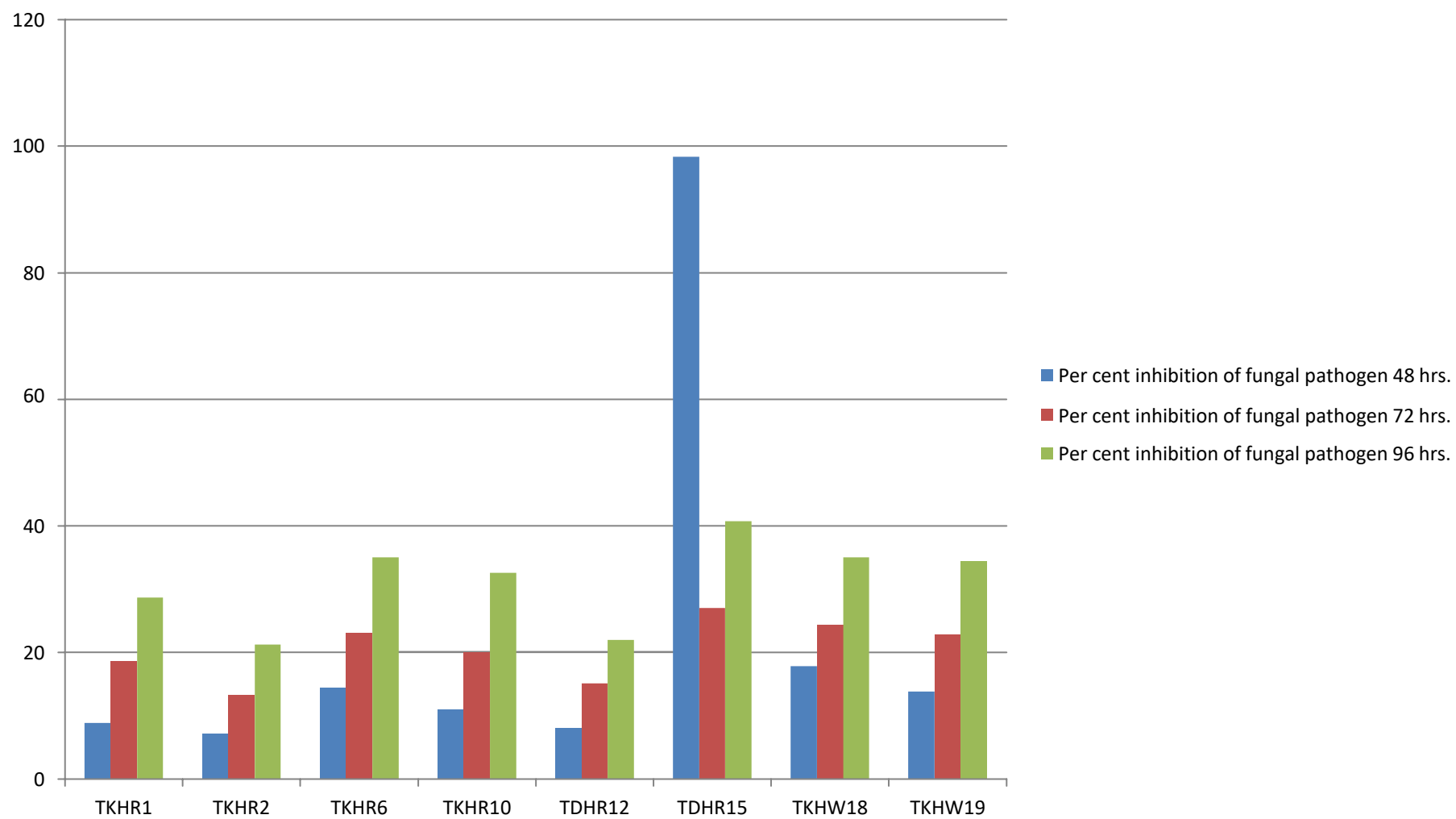
Figure- 4.14 *In-vitro* evolution of *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri*

Plate-1 Isolation of *Trichoderma* strains were collected different places or soil samples



Plate-2 Assessment of different *Trichoderma* strains at variable H ion concentration.



Plate-3 Assessment of different *Trichoderma* strains at variable temperature range



Plate-4 Compatibility of *Trichoderma* strains against different weedicides

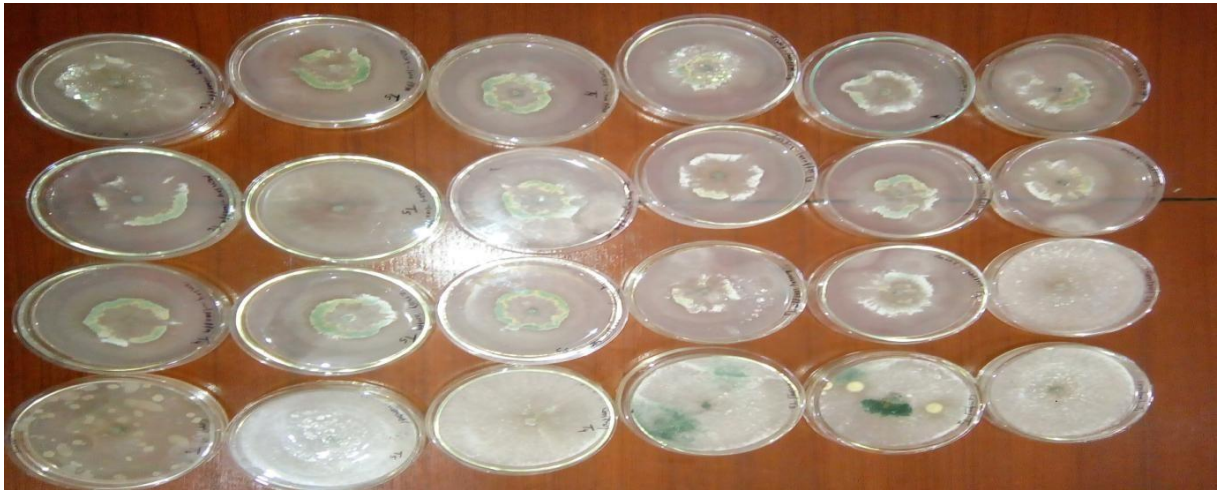


Plate-4 Compatibility of *Trichoderma* strains against different insecticides

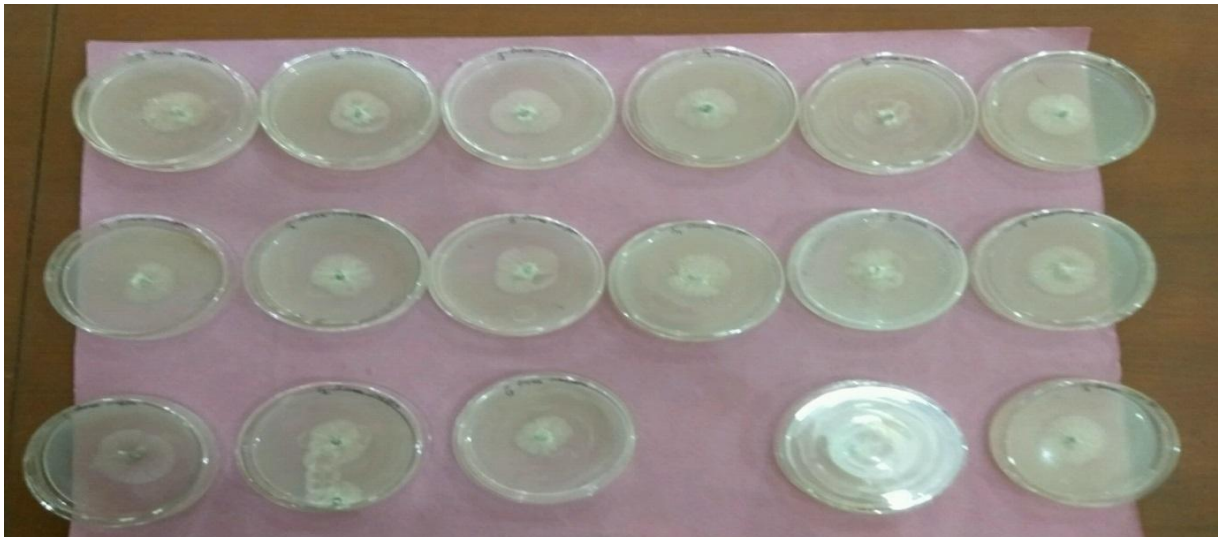
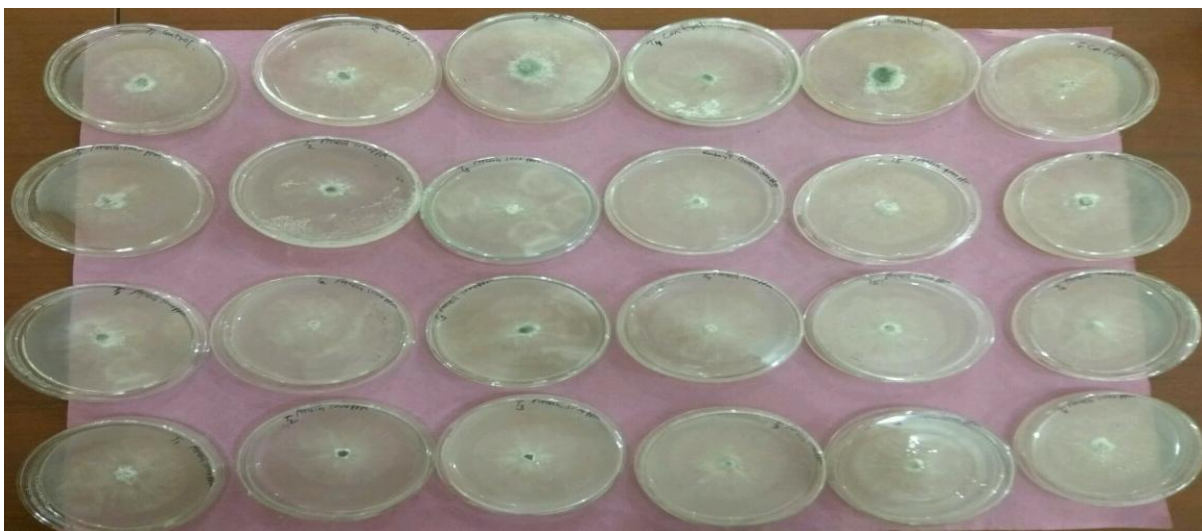


Plate-4 Compatibility of *Trichoderma* strains against different fungicides



CHAPTER- V

DISCUSSION

The present experiment was done on “**Identification of compatible trait of specific Trichoderma strains for management of soil borne diseases**”. Isolates of *Trichoderma* on the identified the *Trichoderma* strains tolerant to higher temperature and variable range of pH, assessment the compatibility of *Trichoderma* strains in response of fungicide, insecticide and weedicide and evaluated the prominent strains of *Trichoderma* against soil borne diseases *in vitro* and *in vivo*.

The isolated of thirty *Trichoderma* strains were carried out from rhizospheric and non-rhizospheric soil samples collected from cultivated fields around Nimar valley plateau zones of Madhya Pradesh, for the characterisation of the Isolation and identification of *Trichoderma* spp, *in vitro* evaluation of *Trichoderma* strains and findings on these aspects are being discussed in this chapter.

Trichoderma is the most widely studied mycoparasitic fungi. *Trichoderma* genus comprises a great number of fungal strains that act as biocontrol agent, the antagonistic properties of which are based on activation of multiple mechanisms. The direct and indirect mechanism may act co-orinatetly and their importance depends on *Trichoderma* strains, antagonized fungus, environmental conditions, pH and temperature and iron concentration. (Tahia *et al.*2004)

5.1 Collection and isolation of *Trichoderma* spp. from rhizospheric and non rhizospheric soil of different Crops.

In the present investigation 30 soil samples were collected from rhizospheric and non-rhizospheric soil of different crops and from different districts namely, Khargone, Dhar, Khandwa and Barwani of Nimar valley zones of M.P. All the strains of *Trichoderma* spp. were isolated through serial dilution technique. The result indicated that *Trichoderma* spp. could grow and survive in

various kinds of soil conditions. This evidence agrees with the report of Sivakumar *et al.* (2000) who had isolated the *Trichoderma* strains from different soil samples in PDA medium.

Similarly previously studies on the topic many scientist and gave suggestions, isolated *Trichoderma* from soil samples collected from three Rambutan fields where leaf litter was allowed to accumulate. Soil suspensions were prepared by adding 1.0 g soil to 10 ml of sterile distilled water and shaking for 15 minutes, suspension was serially diluted to 10⁻⁵ and 0.1 ml of it was spread on a potato dextrose agar (PDA) plate which was then incubated at 28°C for 5 days.

5.2 Identified the *Trichoderma* strains tolerant to different range of temperature.

In the present investigation was on thirty strain of *Trichoderma* tolerant to different range of temperature *viz.*, 20°C, 25°C and 30°C tried for the growth of the *Trichoderma*. Influence of temperature regimes on the growth of *Trichoderma spp.* The different range of temperature will be maintained on BOD incubator. The data on radial growth of colony, and dry weight of the mycelia mat were recorded and analyses statistically.

The maximum weight of dry mycelium (mg) in 20°C temperature with *Trichoderma* strain *viz.* TKHR6 (0.52), TKHW19 (0.50), TDHR15 (0.49) and TKHW18 (0.48). The lowest weight of dry mycelium (mg) with *Trichoderma* strains *viz.*, TDHR16 (0.20).

The maximum weight of dry mycelium (mg) of *Trichoderma* strain at 25°C *viz.*, TKHR6 (0.56), TDHR15 (0.54), TKHW19 (0.53), TKHW18 (0.52) and TBRW24 (0.52). The lowest weight of dry mycelium (mg) with *Trichoderma* strains *viz.*, TDHR16 (0.28).

The maximum weight of dry mycelium (mg) of *Trichoderma* strain at 30°C *viz.*, TKHR10 (0.46), TDHR15 (0.45), TKHW19 and TKHW18 (0.43). The

lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TDHR16 (0.10).

5.3 Identified the *Trichoderma* strains tolerant to Variable range of H ion concentration.

The present investigation was conducted on thirty strain of *Trichoderma* tolerant Variable range of H ion concentration viz., 6, 7 and 8. The variable range of H ion concentration will be maintained in liquid medium used in this experiment was PDA broth solution for assessment of strains performance by the means of colony diameter. The data on radial growth of colony, and dry weight of the mycelia mat were recorded and analyses statistically.

The maximum weight of dry mycelium (mg) in 6 pH with *Trichoderma* strain viz. TBRW24 (0.51), TDHR12 (0.50), TKHR1 (0.50), TBRW23 (0.49) and TKHW19 (0.48). The lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TBRW29 (0.17).

The maximum weight of dry mycelium (mg) of *Trichoderma* strain at 7 pH viz., TKHW18 (0.60), TDHR12 (0.56), TBRW28 (0.55), TBRW26 and TKHR10 (0.54). The lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TBRW29 (0.26).

The maximum weight of dry mycelium (mg) of *Trichoderma* strains 8 pH viz., TKHR2 (0.42), TBRW24 (0.41), TKHW19 and TKHW20 (0.40). The lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TKHW21 (0.11)

Similarly studied the isolated strains of *Trichoderma* and resulted maximum growth and sporulation of *T. viride* was between pH 4.5 to 5.5. Temperatures between 20°C and 37°C were good for both growth and sporulation of *T. viride*. At lower temperatures (i.e. below 20°C) growth and sporulation were inhibited. Jayaswal *et al* (2003). 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 1.35 g. Although all the species of *Trichoderma* produced sufficient biomass at different temperatures viz. 20°C, 25°C, 30°C and 35°C. Anuradha *et al.* (2014).

5.4 Assessment of the compatibility of *Trichoderma* strains in response of pesticides.

Evaluation of the compatibility of commonly used pesticides in agriculture at two different concentrations (500 and 1000 ppm) with *Trichoderma* strains that is being used as a bio control agent against soil borne plant pathogens. Three group commonly used pesticides such as Weedicides (Glyphosate 41% SL, Metsulfuron methyl 20%WP and Imazathypr 10%SL), Insecticides (Phorate 10 Cg, Chlorpyriphos 20 %EC and Quinalphos 25% EC) and Fungicides (Metiram 70% WG, Mancozeb 75% WP and Metalaxyl 8%+Mancozeb 64%WP) were tested *in vitro* against eight selective strains of *Trichoderma* viz., TKHR1, TKHR2, TKHR6, TKHR10, TDHR12, TDHR15, TKHW1 and TKHW19.

5.4.1. Compatibility of *Trichoderma* strains against weedicides.

In our present investigation four weedicide namely, Glyphosate 41% SL, Metsulfuron methyl 20%WP and Imazathypr 10%SL at two different concentrations of 500 ppm and 1000 ppm were tested against 8 strains of *Trichoderma* with control.

The effect of Glyphosate 41% SL at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW19 (23.87%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR6 (15.58%) after inoculation of 96 hrs. at 500 ppm. Among the maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW19 (29.95%) and minimum

percent inhibition of strains of *Trichoderma* was recorded in TKHR6 (24.56%) after inoculation of 96 hrs. at 1000 ppm.

The effect of Metsulfuron methyl 20 % WP at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* were evaluated. The maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW18 (23.52%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR1 (16.77%) after inoculation of 96 hrs. at 500 ppm. Among the maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW18 (27.65%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR1 (20.79%) after inoculation of 96 hrs. at 1000 ppm.

The effect of Imazathypr 10 %SL at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* were evaluated. The maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW18 (28.11%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR6 (22.16%) after inoculation of 96 hrs. at 500 ppm. Among the maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW18 (34.88%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR6 (26.01%) after inoculation of 96 hrs. at 1000 ppm.

5.4.2. Compatibility of *Trichoderma* strains against insecticides.

In our present investigation four weedicide namely, namely Phorate 10 Cg, Chlorpyriphos 20 %EC and Quinalphos 25% EC at two different concentrations of 500 ppm and 1000 ppm were tested against 8 strains of *Trichoderma* with control.

The effect of Phorate 10 Cg at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma*. was studied. The maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW19 (23.88%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR1 (15.17%) after inoculation of 96 hrs. at 500 ppm. Among the maximum per cent inhibition of

strains of *Trichoderma* was recorded in TKHW19 (31.54%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR1 (15.17%) after inoculation of 96 hrs. at 100.

The effect of Chlorpyrifos 20 %EC at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The maximum per cent inhibition of strains of *Trichoderma* was recorded in TDHR15 (26.03%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR1 (20.53%) after inoculation of 96 hrs. at 500 ppm. Among the maximum per cent inhibition of strains of *Trichoderma* was recorded in TDHR15 (28.52%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR1 (24.74%) after inoculation of 96 hrs. at 1000 ppm.

The effect of Quinalphos 25% EC at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW19 (20.15%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR10 (14.44%) after inoculation of 96 hrs. at 500 ppm. Among the maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW19 (24.52%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR10 (19.50%) after inoculation of 96 hrs. at 1000 ppm.

5.4.3. Compatibility of *Trichoderma* strains against fungicides.

Our present investigation four weedicide namely, Metiram 70% WG, Mancozeb 75% WP and Metalaxyl 8%+Mancozeb 64% WP at two different concentrations of 500 ppm and 1000 ppm were tested against 8 strains of *Trichoderma* with control.

The effect of Metiram 70% WG at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW18 (23.56%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR1 (13.46%) after inoculation of 96 hrs. at 500 ppm. Among the maximum per cent inhibition of

strains of *Trichoderma* was recorded in TKHW18 (32.04%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR1 (26.36%) after inoculation of 96 hrs. at 1000 ppm.

The effect of Mancozeb 75% WP at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied. The maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW18 (23.03%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR1 (16.67%) after inoculation of 96 hrs. at 500 ppm. Among the maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW18 (30.67%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR1 (23.93%) after inoculation of 96 hrs. at 1000 ppm.

The effect of Metalaxyl 8%+Mancozeb 64% WP at different concentrations (500 ppm and 1000 ppm) on the per cent inhibition on the colony diameter of different strains of *Trichoderma* was studied . The maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW18 (25.48%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR6 (19.87%) after inoculation of 96 hrs. at 500 ppm. Among the maximum per cent inhibition of strains of *Trichoderma* was recorded in TKHW18 (32.82%) and minimum percent inhibition of strains of *Trichoderma* was recorded in TKHR6 (24.23%) after inoculation of 96 hrs. at 1000 ppm.

Studies of many scientist viz. Vasundra *et al.* (2015) conduct the experiment to check the compatibility of two insecticides, three fungicides and their combinations on *Trichoderma Viridae*. The treatments of Mancozeb (3000 ppm), Imidacloprid (2000 ppm) and combination of Mancozeb (3000 ppm) + Imidacloprid (2000 ppm) showed high compatibility with *Trichoderma Viridae* by showing 7, 11 and 11 percent growth inhibition respective-ly. The treatments viz. Carbendazim (1000 ppm) + Chlorpyriphos (6000 ppm), Tebuconazole (1000 ppm) + Imidacloprid (2000 ppm) and Tebucon-azole (1000 ppm) + Chlorpyriphos (6000 ppm) showed high incompatibility with 100 percent growth inhibition. *Trichoderma harzianum* showed high compatibility even at higher

concentration with insecticides like monocrotophos and chloropyriphos. (Mundal *et al.*,1995).

5.5 *In-vivo* evaluation of *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri*.

Evaluation of the ability of *Trichoderma* strains to control *Fusarium oxysporum* f.sp. *ciceri* of chickpea plant was conducted under field condition (*In-vivo*). The antagonistic potential of eight strains of *Trichoderma* namely, TKHR1, TKHR2, TKHR6, TKHR10, TDHR12, TDHR15, TKHW18, TKHW19 and with control was evaluated under field conditions for their effect against *Fusarium oxysporum* f.sp. *ciceri*.

The result indicated the highest per cent germination of chickpea seeds with the *Trichoderma* strain TKHR2 (99.03%) and the lowest per cent germination with the control (92.50%).

The data was recorded during incidence of *Fusarium oxysporum* f.sp. *ciceri*. on chickpea under field condition at 35, 40, 45, 50, and 55 days after sowing. Minimum percent recorded incidence of *Fusarium oxysporum* f.sp. *ciceri* with *Trichoderma* strain TKHR1 (2.08%) as compare to control (6.81%).

Sonkar *et al.* (2018) evaluated the efficacy of *Trichoderma* spp against *Fusarium oxysporum* f.sp.*Ciceri* on chickpea. The treatment T₄ gave the best result to control the wilt incidence of chickpea

5.6 *In-vitro* evolution of *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri*.

Eight *Trichoderma* isolated strains were screened for their biocontrol potential against *Fusarium oxysporum* f. sp. *Ciceri* under *in-vitro* conditions and data recording after 48 hrs, 72 hrs. and 96 hrs.

The strains of *Trichoderma* inhibited the colony growth of *Fusarium oxysporum* f. sp. *Ciceri*. The percentage inhibition on the growth of the pathogen was maximum with the strain TDHR15 (40.74%), which was found at par with all the other *Trichoderma* strains TKHW18 (35.07%), TKHR6 (35.01%)

and minimum per cent inhibition with the *Trichoderma* strain TKHR2 (21.26%) at incubation after 96 hrs.

Similarly Dubey *et al.* (2007) evaluated 10 isolates belonging to three species of *Trichoderma* (*Trichoderma viride*, *Trichoderma harzianum*, and *Trichoderma virens*) against four isolates of *F. oxysporum* f. sp. *Ciceri*. They also enhanced seed germination, root and shoot length, and decreased wilt incidence under in vitro condition.

CHAPTER – VI

SUMMARY, CONCLUSION AND SUGGESTIONS

FOR FURTHER WORK

6.1 Summary

Our present investigation several species of *Trichoderma* have been extensively studied for their ability to grow on different temperature and different concentration of H ion and compatibility with agrochemicals and antagonist effect of *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri* under *in vitro* and *in vivo* condition. *Trichoderma* strains need proper abiotic environmental condition for the good radial growth. Thirty strains of *Trichoderma* isolated from the rhizospheric and non rhizospheric soil from different crops and places. The results obtained are summarized below.

6.2 Location and crop were isolated *Trichoderma* strains

Total 30 strains of *Trichoderma* collected from four districts of Nimar Valley zones of M.P. namely, Khargone, Dhar, Khandwa and Barwani randomly and 6 crops namely, Soybean, Cotton, Pigeon pea, Maize, Chili, Guava orchard and Bajra.

6.3 Identified the *Trichoderma* strains tolerant to different range of temperature.

Assessment of comparative performance of 30 *Trichoderma* strains in variable range of temperature *viz.*, 20°C, 25°C and 30°C tried for the growth of the *Trichoderma*.

The highest dry weight of mycelium (mg) observed with the *Trichoderma* strain at 25°C *viz.*, TKHR6 (0.56), TDHR15 (0.54), TKHW19 (0.53), TKHW18 (0.52) and TBRW24 (0.52). The lowest weight of dry mycelium (mg) with *Trichoderma* strains *viz.*, TDHR16 (0.28) as compare to 20°C or 30°C

6.4 Identification of the *Trichoderma* strains tolerant to Variable range of H ion concentration.

Identified the tolerant level of 30 *Trichoderma* strains at variable range of H ion concentration viz., 6, 7 and 8.

The highest dry weight of mycelium (mg) observed with the *Trichoderma* strain at 7 pH viz. TKHR6 (0.56), TDHR15 (0.54), TKHW19 (0.53), TKHW18 (0.52) and TBRW24 (0.52). The lowest weight of dry mycelium (mg) with *Trichoderma* strains viz., TDHR16 (0.28) as compare to 6 pH or 8 pH.

6.5 Assessment of the compatibility of *Trichoderma* strains in response of pesticides.

Compatibility of isolated *Trichoderma* strains (TKHR1, TKHR2, TKHR6, TKHR10, TDHR12, TDHR15, TKHW1 and TKHW19) against three group of pesticides, weedicides (Glyphosate 41% SL, Metsulfuron methyl 20%WP and Imazathypr 10%SL), Insecticides (Phorate 10 Cg, Chlorpyriphos 20 %EC and Quinalphos 25% EC) and Fungicides (Metiram 70% WG, Mancozeb 75% WP and Metalaxyl 8%+Mancozeb 64%WP) at two concentration 500 ppm or 1000ppm.

The fungicides significantly inhibited the mycelial growth of different *Trichoderma* strains after inoculating of 42 hrs., 72 hrs. and 96 hrs. as compare to use of weedicides and insecticides

6.6 *In-vivo* evaluation of *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri*.

Evaluate the different *Trichoderma* strains to control *Fusarium oxysporum* f.sp. *ciceri* of chickpea plant under field condition (*In-vivo*). The antagonistic potential of eight strains of *Trichoderma* namely, TKHR1, TKHR2, TKHR6, TKHR10, TDHR12, TDHR15, TKHW18, TKHW19 and with control was evaluated under field conditions.

The maximum percent germination of chickpea with the *Trichoderma* strain TKHR2 (99.03%) and the lowest per cent germination with the control (92.50%).

Incidence of *Fusarium oxysporum* f.sp. *ciceri*. on chickpea under field condition at 35, 40, 45, 50, and 55 days after sowing. Minimum percent recorded incidence of *Fusarium oxysporum* f.sp. *ciceri* with *Trichoderma* strain TKHR1 (2.08%) as compare to control (6.81%)

6.7 In-vitro evaluation of *Trichoderma* strains against *Fusarium oxysporum* f. sp. *Ciceri*.

Eight *Trichoderma* isolated strains were screened for their biocontrol potential against *Fusarium oxysporum* f. sp. *Ciceri* under *in-vitro* conditions. all the strains of *Trichoderma* inhibited the colony growth of *Fusarium oxysporum* f. sp. *Ciceri*. The percentage inhibition on the growth of the pathogen was maximum with the strain TDHR15 (40.74%) and minimum per cent inhibition with the strain TKHR2 (21.26%) at incubation after 96 hrs.

6.8 Conclusion and suggestions for further work

1. Different *Trichoderma* strains was found to be most efficient tolerant to variable range of temperature and H ion concentration. Hence the effect of various abiotic environmental factor on the sporulation of *Trichoderma*.
2. Eight promising strains viz., TKHR1, TKHR2, TKHR6, TKHR10, TDHR12, TDHR15, TKHW18, and TKHW19 of *Trichoderma* were compatibility test against *agrochemicals* in dual culture. These strains of *Trichoderma* compatible of different *agrochemicals* at different concentration.
3. The control of mycelial growth and incidence of *Fusarium oxysporum* f. sp. *ciceri* significantly inhibited with *Trichoderma* strains viz., TKHR1, TKHR2, TKHR6, TKHR10, TDHR12, TDHR15, TKHW18, TKHW19 under *in vitro* and *in vivo* condition.
4. Total thirty *Trichoderma* strains isolated from four district and six crops..
5. Abiotic environment factor affect the sporulation of *Trichoderma*

6. More soil samples from different cropped areas need to be collected and tested for their bio-control potential against the major soil borne diseases.
7. Need to execute more number and diverse range of field experiments for evaluation of real effectiveness of selected strains of *Trichoderma*.
8. More studies such as *in vivo* and *in vitro* tests have to be conducted using these *Trichoderma* isolates before they can be used as a biological control agent in agricultural field.

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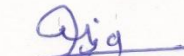
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