



ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; 11(12): 994-1004
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www.thepharmajournal.com

Received: 08-09-2022

Accepted: 11-10-2022

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Antagonistic potential of *trichoderma* isolates from Konkan soils

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Abstract

The current investigation was carried out during 2020-21 where in twenty-seven isolates of *Trichoderma* were obtained on TSM from sixty seven soil samples collected from various places in Konkan region of Maharashtra. All the isolates were found effective against the five test pathogens *Fusarium* spp., *Rhizoctonia* spp., *Sclerotium* spp., *Colletotrichum* spp. and *Alternaria* spp. when tested by dual culture technique. Among them seven isolates viz., the isolates 11, 3, 23, 5, 14, 25 and 24 showed better inhibition. 11 was the most effective against *Fusarium* spp. (82.22% inhibition), 14 against *Rhizoctonia* spp. (81.11%), 23 against *Sclerotium* spp. (86.11%), 5 against *Colletotrichum* spp. (81.33%), 25 against *Alternaria* spp. (86.66%). In terms of fungicide compatibility, all the seven isolates were extremely sensitive to Carbendazim and five isolates were fairly compatible with Sulphur.

Keywords: *Trichoderma*, antagonism, compatibility, promising isolates

Introduction

Indiscriminate use of chemicals in management of plant diseases has caused everlasting damage to the crop ecosystem. In such a situation, use of effective bio-control agents against the pathogens is an eco-friendly and affordable strategy to manage the diseases of crop plants. In Konkan conditions anthracnose of mango incited by *Colletotrichum gloeosporioides* is one of the most damaging factor which causes huge losses in pre and post-harvest conditions. *Alternaria* spp. are also of common occurrence on solanaceous and cruciferous vegetable crops and flowering plants like marigold which are cultivated as subsidiary crops in many pockets of Konkan region. Like most of the soils all over the country, Konkan soils also harbour the notable amount of inoculum of *Fusarium* spp. It is, actually, a seed and soil borne fungus having a wide host range. *Sclerotium* spp. is another soil borne pathogen which is forceful to cause giant wounded in agricultural produce. Rice is the major cereal crop of Konkan. At present, sheath bight of rice is an emerging threat of this crop. The pathogen-*Rhizoctonia solani* was reported earlier as the causal agent of leaf blight of cardamom. Management of this pathogen by soil application of *Trichoderma* will be advisable. Hence in present study investigated about antagonistic ability and compatibility of the potential *Trichoderma* isolates.

Materials and Methods

Sixty-seven rhizospheric soil samples were collected from different soils of Konkan region out of which twenty-seven showed presence of *Trichoderma* spp. on both TSM and PDA medium at different dilutions viz., 10^{-5} , 10^{-6} , 10^{-7} , 10^{-8} and 10^{-9} . The antagonistic potential of these was tested against three soil borne (*Fusarium* spp., *Rhizoctonia* spp. and *Sclerotium* spp.) and two aerial (*Colletotrichum* spp., *Alternaria* spp.) plant pathogens by dual culture technique. Promising seven isolates were selected for compatibility with fungicides like Carbendazim, Hexaconazole, Thiophanate-methyl Copper-oxy-chloride, Sulphur and Mancozeb by poisoned food method.

Results

1.1. Antagonistic potential of local *Trichoderma* isolates against *Fusarium* spp., *Rhizoctonia* spp., *Sclerotium* spp., *Colletotrichum* spp. and *Alternaria* spp.

It was revealed from the data presented in table 1 that all the isolates were effective in inhibiting the growth of *Fusarium* spp.

None of the isolates recorded less the 50% inhibition. The maximum inhibition (82.22%) was recorded by the isolate 11 (Tas-Areca nut) and it was significantly superior to rest of the isolates. It was followed by 14 (Tgpal-Guava) which recorded 81.33% inhibition of the pathogen. These two isolates were followed by 3 (Tmnrij-Mango) (- inhibition -79.22%) and 5 (Tkorr-Rice-79.22%) which were at par with each other. Among the remaining isolates, 23 (Tbk-Brinjal-77.77%) was followed by 25 (Tckr-Cabbage 74.44%), 24 (Tbpal-Brinjal-69.11%), 26 (Tcaupal-Cauliflower-68.33%), 1 (Tmed Mango-65.22%). The isolates 6 (Trm-Rice-64.77) and 7 (Tralr-Rice-64.77) were at par with each other. So also the isolate 22 (Tbgu-Bottle gourd-64.44%) and 12 (Tcnv-Cashewnut-64.22%) were statistically at par with each other. They were followed by 27 (Tmcpal-Champak (Sonchafa)-64.11%) and 21(Tchupal-Chilli-62.77) and 8 (Tcojrr-Rice-60.88%). Eleven isolates recorded less than 60% inhibition. Isolate 13 (Tbw-Banana) recorded 59.22% inhibition while 4 (Tojrr2 -Rice-58.88%) was at par with 16 (Thm-Horse gram-58.88%) and 17 (Tlbhar-lablab bean- 58.66%). The lowest inhibition (51.66%) was recorded by 19 (Tefym-Elephant foot yam)

Data depicted in table 2 that, all the isolates were effective in inhibiting the growth of *Rhizoctonia* spp. None of the isolates recorded less the 58% inhibition. The maximum inhibition (81.11%) was recorded by the isolate 14 (Tgpal-Guava-Palghar) and it was significantly superior to rest of the isolates. It was followed by 22 (Tbgu-Bottle gourd – Unhavare) which recorded 77.77% inhibition of the pathogen. These isolates were followed by 11 (Tas- Arecanut inhibition -75.55%), 24(Tbpal-Brinjal-74.11%), 3 (Tmnrij-Mango-71.11%) and 7(Tralr-Rice-70.55%) which were at similitude. Among the remaining isolates, 5 (Tkorr- Rice- 69.56%) was followed by 26 (Tcaupal-Cauliflower-68.33%) and 16(Thm-Horse gram- 68.00%) which were at par with each other. They were followed by 6 (Trm-Rice-67.55%) and 17 (Tlbhar-lablab bean-67.00%) which were at par, followed by 15 (Tsptpal-Sapota-66.11%). 27 (Tmcpal-Champak (Sonchafa)-65.33%) and 21 (Tchupal-Chilli-65.33%) at par with each other. 25 (Tckr- Cabbage-64.77%) and 4 (Tojrr-Rice-64.44%) were also at par with each other. Remaining ten isolates recorded less than 63% inhibition.

The data in Table-3 revealed that, among these isolate 23 (Tbk) from brinjal rhizosphere showed lowest colony diameter (12.50 mm) with 86.11% inhibition and it was significantly superior to rest of the isolates. It was followed by 3 (Tmnrij-Mango) which recorded 80.55% inhibition of the pathogen. These isolates were followed by 11 (Tas-Arecanut-74.22%), 5 (Tkorr-Rice-71.66%) and 14 (Tgpal-Guava-52.55%). Rest of the twenty-two isolates showed below 50% inhibition.

In case of *Colletotrichum* spp. it was found that all the species of *Trichoderma* reduced the mycelial growth of *Colletotrichum* spp. The isolates 5 (Tkorr-rice) and 14 (Tgpal-guava) from Palghar gave the best results with minimum radial growth of *Colletotrichum* spp(16.80 mm) and maximum growth inhibition (81.33%). Both these isolates were numerically at par and significantly superior to all the treatments. These isolates were followed by 3 (Tmnrij-Mango-Lanja-80.22%), 24 (Tbpal-Brinjal-78.33%), 23 (Tbk-Brinjal-77.77%), 11(Tas-Arecanut-71.11%), 27 (Tmcpal-Champak (Sonchafa) 68.00%) whereas, 25 (Tckr-Cabbage-64.44%) and 12 (Tcnv-Cashewnut-64.11%) were at par with each other. 18 (Tgkh2020s-Groundnut-63.66%), 4(Tojrr2-Rice-63.55%) and 13 (Tbw-Banana-63.11%) were also at par to each other and followed by 21(Tchupal (T₂₁-60.33%). Remaining fourteen isolates showed below 59.00% inhibition.

Data presented in Table 5 revealed that the *Trichoderma* isolate *i.e.* 25 (Tckr -Cabbage-86.66%) was found significantly superior to all treatments against *Alternaria* spp. and followed by 11 (Tas-Arecanut-77.55%), 23 (Tbk-Brinjal-69.66), 5(Tkorr-Rice-66.44) and 3 (Tmnrij-Mango-66.11%) were at par to each other, followed by 14 (Tgpal-Guava-64.11%), 24 (Tbpal-Brinjal-63.66), 16 (Thm-Horse gram-62.77%). 15 (Tsptpal-Sapota-61.88%), 26 (Tcaupal-Cauliflower-61.66%) and 1 (Tmed-Mango-61.44%) were at par to each other. These isolate followed by 22 (Tbgu-Bottle gourd-61.33%). 6 (Trm -Rice-60.55%) and 21(Tchupal-Chilli-60.22%) were at par to each other. The isolates T7 (Tralr-Rice- 59.22%), 13 (Tbw- Banana- 59.22%) and 27 (Tmcpal-Champak (Sonchafa) 59.11%) were at par with each other. Rest of the seven isolates showed above 50% inhibition.

Table 1: *In vitro* efficacy of local *Trichoderma* isolates against *Fusarium* spp

Sr. No.	Isolates code	Colony diameter (mm)	Percent Growth Inhibition
1	Tmed	31.30	65.22
2	Tamsakh	41.50	53.88
3	Tmnrij	18.70	79.22
4	Tojrr2	37.00	58.88
5	Tkorr	18.70	79.22
6	Trm	31.70	64.77
7	Tralr	31.70	64.77
8	Tcojrr2	35.20	60.88
9	Tcbfn	38.20	57.55
10	Tcwki	37.50	58.33
11	Tas	16.00	82.22

12	Tcnv	32.20	64.22
13	Tbw	36.70	59.22
14	Tgpal	16.80	81.33
15	Tsptpal	39.30	56.33
16	Thm	37.00	58.88
17	Tlbhar	37.20	58.66
18	Tgkh2020s	39.20	56.44
19	Tefym	43.50	51.66
20	Tchal	38.30	57.44
21	Tchipal	33.50	62.77
22	Tgbu	32.00	64.44
23	Tbk	20.00	77.77
24	Tbpal	27.80	69.11
25	Tckr	23.00	74.44
26	Tcaupal	28.50	68.33
27	Tmcpal	32.30	64.11
28	Control	90.00	-
SE±		0.09	
CD 1%		0.33	

*Presented data in table is average of three replications

Table 2: *In vitro* efficacy of local *Trichoderma* isolates against *Rhizoctonia* spp

Isolates code	<i>Trichoderma</i> isolates	Colonydiameter (mm)	Percent Growth Inhibition
1	Tmed	36.50	59.44
2	Tamsakh	33.20	63.11
3	Tmnrj	26.00	71.11
4	Tojrr2	32.00	64.44
5	Tkorr	27.30	69.56
6	Trm	29.20	67.55
7	Trlar	26.50	70.55
8	Tcojrr2	34.00	62.22
9	Tcnfn	33.20	63.11
10	Tcwiki	33.50	62.77
11	Tas	22.00	75.55
12	Tcnv	37.20	58.66
13	Tbw	35.70	60.33
14	Tgpal	17.00	81.11
15	Tsptpal	30.50	66.11
16	Thm	28.80	68.00
17	Tlbhar	29.70	67.00
18	Tgkh2020s	35.70	60.33
19	Tefym	33.30	63.00
20	Tchal	33.80	62.44
21	Tchipal	31.20	65.33
22	Tgbu	20.00	77.77
23	Tbk	36.50	59.44
24	Tbpal	23.30	74.11
25	Tckr	31.70	64.77
26	Tcaupal	28.50	68.33
27	Tmcpal	31.20	65.33
28	Control	90.00	00.00
SE (M) ±		0.09	
Cd 1%		0.33	

*Presented data in table is average of three replications

Table 3: *In vitro* efficacy of local *Trichoderma* isolates against *Sclerotium* spp

Isolates code	<i>Trichoderma</i> isolates	Colonydia meter (mm)	Percent Growth Inhibition
1	Tmed	62.80	30.22
2	Tamsakh	73.30	18.55
3	Tmnrj	17.50	80.55
4	Tojrr2	58.80	34.66
5	Tkorr	25.50	71.66
6	Trm	64.30	28.55
7	Tralr	74.50	17.22
8	Tcojrr2	63.50	29.44
9	Tcbfn	52.00	42.22

10	Tcwki	68.70	23.66
11	Tas	23.20	74.22
12	Tcnv	75.20	16.44
13	Tbw	62.70	30.33
14	Tgpal	42.70	52.55
15	Tsptpal	74.20	17.55
16	Thm	58.20	35.33
17	Tlbhar	72.80	19.11
18	Tgkh2020s	54.30	39.66
19	Tefym	76.20	15.33
20	Tchal	70.80	21.33
21	Tchipal	71.30	20.77
22	Tbgu	65.00	27.77
23	Tbk	12.50	86.11
24	Tbpal	51.50	42.77
25	Tckr	51.80	42.77
26	Tcaupal	61.70	31.44
27	Tmcpal	73.80	18.00
28	Control	90.00	-
SE (M) ±		0.11	
Cd 1%		0.43	

*Presented data in table is average of three replications

Table 4: *In vitro* efficacy of local *Trichoderma* isolates against *Colletotrichum* spp

Isolates code	<i>Trichoderma</i> isolates	Colony diameter (mm)	Percent Growth Inhibition
1	Tmed	40.70	54.77
2	Tamsakh	43.70	51.44
3	Tmnrj	17.80	80.22
4	Tojrr2	32.80	63.55
5	Tkorr	16.80	81.33
6	Trm	37.80	58.00
7	Trlar	37.20	58.66
8	Tcojrr2	42.80	52.44
9	Tcbfn	36.30	59.66
10	Tcwki	37.70	58.11
11	Tas	26.00	71.11
12	Tcnv	32.30	64.11
13	Tbw	33.20	63.11
14	Tgpal	16.80	81.33
15	Tsptpal	45.30	49.66
16	Thm	39.50	56.11
17	Tlbhar	37.70	58.11
18	Tgkh2020s	32.70	63.66
19	Tefym	42.20	53.11
20	Tchal	40.20	55.33
21	Tchipal	35.70	60.33
22	Tbgu	41.00	54.44
23	Tbk	20.00	77.77
24	Tbpal	19.50	78.33
25	Tckr	32.00	64.44
26	Tcaupal	37.80	58.00
27	Tmcpal	28.80	68.00
28	Control	90.00	00.00
SE (M) ±		0.11	
Cd 1%		0.43	

*Presented data in table is average of three replications

Table 5: *In vitro* efficacy of local *Trichoderma* isolates against *Alternaria* spp

Isolates code	<i>Trichoderma</i> isolates	Colony diameter (mm)	Percent Growth Inhibition
1	Tmed	34.70	61.44
2	Tamsakh	39.50	56.11
3	Tmnrj	30.50	66.11
4	Tojrr2	37.70	58.11
5	Tkorr	30.20	66.44
6	Trm	35.50	60.55
7	Tralr	36.70	59.22

8	Tcojrr2	39.00	56.66
9	Tcbfn	38.30	57.44
10	Tcwki	37.00	58.88
11	Tas	20.20	77.55
12	Tcnv	41.50	53.88
13	Tbw	36.70	59.22
14	Tgpal	32.30	64.11
15	Tsptpal	34.30	61.88
16	Thm	33.50	62.77
17	Tlbhar	43.70	51.44
18	Tgkh2020s	38.30	57.44
19	Tefym	37.50	58.33
20	Tchal	40.00	55.55
21	Tchipal	35.80	60.22
22	Tbgu	34.80	61.33
23	Tbk	27.30	69.66
24	Tbpal	32.70	63.66
25	Tckr	12.00	86.66
26	Tcaupal	34.50	61.66
27	Tmcpal	36.80	59.11
28	Control	90.00	00.00
	SE (M) ±	0.12	
	Cd 1%	0.44	

*Presented data in table is average of three replications

1.2. Antagonistic potential of local *Trichoderma* isolates against five pathogens

It was visible from the results presented in table 6 that, among all the 27 isolates, 7 isolates 11 (Tas-Areca nut-Shriwardhan), 14 (Tgpal-Guava- kelwePalghar), 3 (Tmnrj-Mango –Lanja), 5 (Tkorr-Rice – Kolambe), 23 (Tbk-Brinjal – Karjat), 25 (Tckr-Cabbage-Karjat), 24 (Tbpal-Brinjal- Mahim) were very effective against all the five pathogens under study in below table as they showed as per percent inhibition sequentially. Due to best performance cultures were sent for molecular and morphological identification at AGI (Agarkar Research Institute), Pune. First three for molecular and identified isolate 11 as *Trichoderma asperellum*, 3 as *Trichoderma harzianum*, 23 as *Trichoderma asperellum* remaining four for morphological and identified 5 as *Trichoderma* sp. aff. *T. koningii*, 14 as *Trichoderma* sp. aff. *T. koningii*, 25 as *Trichoderma* sp. aff. *T. longibrachiatum*, 24 as *Trichoderma* sp. aff. *T. koningii*.

Out of these, three isolates were from Raigad, 2 from Palghar and 2 from Ratnagiri district of Konkan region. Isolate 11 was the most effective against *Fusarium*, 14 against *Rhizoctonia*, 23 against *Sclerotium*, 5 against *Colletotrichum*, 25 against *Alternaria*. The isolate 3 ranked second in antagonism against *Sclerotium*, third in control of *Fusarium* and *Colletotrichum* and fifth in *Rhizoctonia* and *Alternaria*. As far as the antagonism performance of the isolate 24 is concerned it ranked fourth against *Rhizoctonia* and *Colletotrichum*, sixth against *Sclerotium*, seventh against *Fusarium* and *Alternaria*.

All the isolates recorded 50% inhibition of all the pathogens except *Sclerotium*. In case of this pathogen, most of the isolates recorded growth inhibition within a range of 15- 42% and only 5 isolates recorded more than 50% inhibition.

2. Compatibility of promising isolates with fungicides.

Occasionally use of a combination of bio-agent and fungicide also facilitates the management strategy. It is, therefore, necessary to test the compatibility of the bio-agent with the recommended fungicides hence the seven promising isolates of *Trichoderma* were cultured in fungicide fortified medium. Three systemic fungicides and three contact fungicides were used in this experiment.

It was clear from the results in the table that carbendazim was the most detrimental for all the isolates as it completely inhibited the mycelial growth of these isolates. It was followed by hexaconazole which completely inhibited the mycelium of the isolate Tgpal. The growth inhibition by this fungicide in case of Tkorr was 90% followed by Tbpal (87.77%), Tas (86.11%), Tbk (85.77%), Tckr (84.11%) and that of Tmnrj (63.33%). The third systemic fungicide thiophanate methyl caused maximum inhibition (86.33) of Tbk, followed by Tgpal (80.22%), Tckr (80.00%), Tkorr (79.77%), Tbpal (75.77%), Tmnrj (68.33%) and Tas (58.00%).

Among the three contact fungicides, sulphur was the most compatible fungicide as the highest inhibition recorded by it was of Tkorr (23.11%), which was subsequently followed by Tbk (20.00%), Tckr (9.60%), Tbpal (5.88%), Tgpal (4.22%), Tas (3.66%) and the least inhibition (0.33%) of Tmnrj. These results suggest that at a slightly lower concentration this fungicide may not be inhibitory to the test isolates. Copper oxychloride recorded the least inhibition of (43.66%) of Tas and the maximum (84.44%) of Tkorr while mancozeb recorded the least inhibition (59.77%) of Tas and the highest (84.22%) of Tkorr.

Table 6: Comparative antagonistic potential of the local *Trichoderma* isolates against five pathogens

Sr. no	<i>Fusarium</i> spp.		<i>Rhizoctonia</i> spp.		<i>Sclerotium</i> spp.		<i>Colletotrichum</i> spp.		<i>Alternaria</i> spp.	
	Isolate code	Inhibition (%)	Isolate code	Inhibition (%)	Isolate code	Inhibition (%)	Isolate code	Inhibition (%)	Isolate code	Inhibition (%)
1	Tas	82.22	Tgpal	81.11	Tbk	86.11	Tkorr	81.33	Tckr	86.66
2	Tgpal	81.33	Tbgu	77.77	Tmnrj	80.55	Tgpal	81.33	Tas	77.55
3	Tmnrj	79.22	Tas	75.55	Tas	74.22	Tmnrj	80.22	Tbk	69.66
4	Tkorr	79.22	Tbpal	74.11	Tkorr	71.66	Tbpal	78.33	Tkorr	66.44
5	Tbk	77.77	Tmnrj	71.11	Tgpal	52.55	Tbk	77.77	Tmnrj	66.11
6	Tckr	74.44	Tralr	70.55	Tbpal	42.77	Tas	71.11	Tgpal	64.11
7	Tbpal	69.11	Tkorr	69.56	Tckr	42.77	Tmcpal	68.00	Tbpal	63.66
8	Tcaupal	68.33	Tcaupal	68.33	Tcbfn	42.22	Tckr	64.44	Thm	62.77
9	Tmed	65.22	Thm	68.00	Tgkh2020s	39.66	Tcnv	64.11	Tsptpal	61.88
10	Trm	64.77	Trm	67.55	Thm	35.33	Tgkh2020s	63.66	Tcaupal	61.66
11	Tralr	64.77	Tlbhar	67.00	Tojrr2	34.66	Tojrr2	63.55	Tmed	61.44
12	Tbgu	64.44	Tsptpal	66.11	Tcaupal	31.44	Tbw	63.11	Tbgu	61.33
13	Tcnv	64.22	Tchipal	65.33	Tbw	30.33	Tchipal	60.33	Trm	60.55
14	Tmcpal	64.11	Tmcpal	65.33	Tmed	30.22	Tcbfn	59.66	Tchipal	60.22
15	Tchipal	62.67	Tckr	64.77	Tcojrr2	29.44	Tralr	58.66	Tralr	59.22
16	Tcojrr2	60.88	Tojrr2	64.44	Trm	28.55	Tcwki	58.11	Tbw	59.22
17	Tbw	59.22	Tamsakh	63.11	Tbgu	27.77	Tlbhar	58.11	Tmcpal	59.11
18	Tojrr2	58.88	Tcbfn	63.11	Tcwki	23.66	Trm	58.00	Tcwki	58.88
19	Thm	58.88	Tefym	63.00	Tchal	21.33	Tcaupal	58.00	Tefym	58.33
20	Tlbhar	58.66	Tcwki	62.77	Tchipal	20.77	Thm	56.11	Tojrr2	58.11
21	Tcwki	58.33	Tchal	62.44	Tlbhar	19.11	Tchal	55.33	Tcbfn	57.44
22	Tcbfn	57.55	Tcojrr2	62.22	Tamsakh	18.55	Tmed	54.77	Tgkh2020s	57.44
23	Tchal	57.44	Tbw	60.33	Tmcpal	18.00	Tbgu	54.44	Tcojrr2	56.66
24	Tgkh2020s	56.44	Tgkh2020s	60.33	Tsptpal	17.55	Tefym	54.11	Tamsakh	56.11
25	Tsptpal	56.33	Tmed	59.44	Tralr	17.22	Tcojrr2	52.44	Tchal	55.55
26	Tamsakh	53.88	Tbk	59.44	Tcnv	16.44	Tamsakh	51.44	Tcnv	53.88
27	Tefym	51.66	Tcnv	58.66	Tefym	15.33	Tsptpal	49.66	Tlbhar	51.44

Table 7: Compatibility of promising isolates with fungicides

Colony diameter of <i>Trichoderma</i> isolates (mm)															
Tr. No	Fungicide Concentration	<i>Trichoderma</i> isolates													
		1		2		3		4		5		6		7	
		Tas (11)		Tmnrj (3)		Tbk (23)		Tkorr (5)		Tgpal (14)		Tckr (25)		Tbpal (24)	
		Colony diameter	% inhibition	Colony diameter	% inhibition	Colony diameter	% inhibition	Colony diameter	% inhibition	Colony diameter	% inhibition	Colony diameter	% inhibition	Colony diameter	% inhibition
T1	Carbendazim (1000 ppm)	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
T2	Hexaconazole (500 ppm)	12.50	86.11	33.00	63.33	12.80	85.77	9.00	90.00	0.00	100.00	14.30	84.11	11.00	87.77
T3	Thiophenate-methyl (500 ppm)	37.80	58.00	28.50	68.33	12.30	86.33	18.20	79.77	17.80	80.22	18.00	80.00	21.80	75.77
T4	Copper-oxychloride (2500 ppm)	50.70	43.66	28.20	68.66	23.50	73.88	14.00	84.44	21.00	76.66	15.20	83.11	22.20	75.33
T5	Sulphur (2500 ppm)	86.70	3.66	89.70	0.33	72.00	20.00	69.20	23.11	86.20	4.22	81.30	9.60	84.70	5.88
T6	Mancozeb(2500 ppm)	36.20	59.77	19.20	78.66	32.50	63.88	14.20	84.22	20.00	77.77	17.80	80.22	19.20	78.66
T7	Control	90.00	00.00	90.00	00.00	90.00	00.00	90.00	00.00	90.00	00.00	90.00	00.00	90.00	00.00
	Ftest	Sig	-	Sig	-	Sig	-	Sig	-	Sig	-	Sig	-	Sig	-
	SE(m)±	0.12	-	0.08	-	0.10	-	0.19	-	0.04	-	0.08	-	0.11	-
	CD(P=0.01)	0.51	-	0.32	-	0.43	-	0.78	-	0.19	-	0.32	-	0.46	-

Discussion

In the current study it was found that among the promising isolates *T. asperellum* (Isolate 11) was the most effective against *Fusarium* (82.22% inhibition), *T. koningii* (Isolate 14) against *Rhizoctonia* (81.11%), *T. asperellum* (Isolate 23) against *Sclerotium* (86.11%), *T. koningii* (Isolate 5) against *Colletotrichum* (81.33%), *T. longibrachiatum* (Isolate 25) against *Alternaria* (86.66%). The isolate *T. harzianum* (Isolate 3) ranked second in antagonism against *Sclerotium* (80.54%), third in control of *Fusarium* (79.22%) and *Colletotrichum* (80.22%) and fifth in *Rhizoctonia* (71.11%) and *Alternaria* (66.11%). *T. asperellum* (Isolate 11) isolate recorded 82.22% inhibition of *Fusarium* but the isolate *T. asperellum* (Isolate 23) recorded only 77.77% inhibition against the same pathogen. This difference in the performance of two isolates may be attributed to the difference in the two strains of the same fungus. In the findings of Komy *et al* (2015) they screened 30 isolates of *T. asperellum* against *F. oxysporum* causing wilt of tomato and reported that 6 isolates recorded the highest inhibition of the pathogen which ranged between 68 and 71%. Among remaining isolates most of the isolates recorded moderate inhibition (61-65%) and seven isolates recorded minimum inhibition (32-36%). The results of present study are in agreement with these results. Naher *et al* (2019) recorded 74.16% inhibition by *T. asperellum* against *F. oxysporum*. Rai and Maurya (2021) evaluated local strains of *T. asperellum* against *F. oxysporum* f.sp. *lycopersici* and recorded 73.91% inhibition and 64.49% inhibition against *F. oxysporum* f.sp. *cubense*. An isolate of *T. asperellum* caused 69.50% inhibition of *Rhizoctonia* spp (Restrepo *et al*, 2022). Asad *et al* (2014) recorded 74.4% inhibition of *R. solani* by *T. asperellum* after 72 hrs of inoculation. These results are in accordance with the results of present study and this suggests that *T. asperellum* is very effective against *Fusarium* species. Among the 11 isolates of *T. asperellum* tested by Sharma and Prasad (2018) against *S. sclerotiorum* the isolate T21 recorded 93% inhibition of the pathogen while the same isolate recorded 85.18% inhibition of *Colletotrichum asianum* causing anthracnose of *Tabernaemontana divericata*. The two isolates *T. asperellum* obtained in the current research (Isolate 23 and 11) were very effective against *C. gloeosporioides* as they recorded 77.77 and 71.11% inhibition respectively. Even though most of the workers have reported more than 50% inhibition potential of *T. asperellum* against many pathogens, Quiroz *et al* (2018) reported 14.971 and 22.50% inhibition against *C. gloeosporioides*. These results are contradictory to the present findings. *T. asperellum* isolates were very effective against *Alternaria* spp. as the isolate 11 recorded 75.55% inhibition of *Alternaria* spp while isolate 23 recorded 69.66% inhibition of the same pathogen. The results of Pradeep *et al* (2022) are in congruence with these findings as the reported 73.33% inhibition of *A. alternata* with an isolate of *T. asperellum*. But Reddy *et al.*, (2018) reported that T4 (*T. asperellum* isolate) was 35.50% effective against the same pathogen. Their results differ with the present findings. Among the four morphologically identified isolates three isolates (5, 14 and 24) were of *T. koningii*. The isolate 14 was superior to other isolates against *Fusarium* spp. (81.33% inhibition) and *Rhizoctonia* (81.11%). The isolate 5 was superior in case of *Sclerotium* (71.66%) and *Alternaria* (66.44%). It was at par with 14 (81.33%) in controlling *Colletotrichum*. The remaining isolate 24 was inferior two former two isolates in

case of all the pathogens under study. Mamtha and Yashoda (2006) recorded 77.43% inhibition of *Colletotrichum* by *T. koningii*. While against *A. alternata* the inhibition was 80.00%. Honmane (2007) recorded 83.33% inhibition of *F. moniliforme* and 80.74% inhibition of *C. gloeosporioides* with *T. koningii*. As per the results of Febrilia *et al* (2013), *T. koningii* recorded 83% inhibition *C. gloeosporioides*. These results are in conformity with present findings. Farah and Nasreen (2013) recorded 32.14, 79.45, 85.32 and 91.09% inhibition of *F. oxysporum*, *A. solani*, *F. solani* and *R. solani* with *T. koningii*. These results are contradictory to present findings in case of *Fusarium*. Bhale and Rajkonda (2015) observed about 50% inhibition of *R. solani* and *F. oxysporum* also 75 percent reduction in the growth of *A. alternata*. Musheer and Ashraf (2017) noted that *T. koningii* caused 52.46% inhibition of *C. gloeosporioides*. Rajkonda and Bhale (2018) tested the antagonism of *T. koningii* against the *A. alternata*, *A. tenuissima*, *R. solani*, *F. oxysporum* f.sp. *spinaceae* and *F. proliferatum*. They noted 61 -71% inhibition of all the five pathogens. The results of Reddy *et al.*, (2018) suggest that, *T. koningii* is not very effective against *A. alternata* as there was only 24.99% inhibition in pathogen growth. Naher *et al.*, (2019) recorded 71.40% inhibition of *F. oxysporum* by an isolate of *T. koningii*

During the present investigation only one isolate of *T. harzianum* was obtained. It caused 80.54% inhibition in the mycelial growth of *Sclerotium* spp. The results of Jana and Mandal (2017) indicated that the three isolates of *T. harzianum* viz. T3, T4 and T11 recorded 52.17, 48.91 and 46.20% inhibition of *S. rolfisii*. This indicates that even though all the isolates were identified as *T. harzianum* their antagonism potential varies depending upon the ability of the isolate to secrete metabolites which are detrimental to the pathogens. So, the antagonistic potential is a genetic character and therefore different isolates of the same bio-control agent perform differently against the same pathogen. *T. harzianum* isolates used by Kushwaha (2018) recorded 63.60% inhibition of *S. rolfisii* while Singh *et al.*, (2018) recorded 50.67% inhibition of the same pathogen causing collar rot of chickpea. *T. harzianum* isolate TspT recorded 81.27% inhibition of *S. rolfisii*, (Priyadharcini *et al.*, 2018). Amin *et al.*, (2010) reported that, Th-1 and Th-2 isolates of *T. harzianum* recorded 75.92 and 71.26% inhibition while Kumar *et al* (2011) recorded 80% and 72.1% inhibition of *S. rolfisii* by TWN1 and TWC2 isolates of *T. harzianum*. Goudar and Kulkarni (2000) recorded 85.40% inhibition of *F. udum* by *T. harzianum*. Jat and Agalave (2013) recorded 47.50 and 50.00% inhibition of *T. harzianum* isolate against *F. oxysporum* and *F. moniliforme* and 48.33% against *A. alternata*. Elshahawy (2016) noticed that the three isolates of *T. harzianum* (Th1, Th2 and Th3) recorded 59.2, 66.7 and 61.5% inhibition of *F. Solani* and 58.2, 52.2 and 56.3 of *F. oxysporum* sequentially. They also observed 38.2, 42.6 and 48.2% inhibition of *R. solani* by these isolates. Sangle and Bambawale (2004) recorded 79.54% inhibition of *F. oxysporum* f. sp *sesame*. Yadav *et al.*, (2005) recorded 62.5% inhibition against *F. udum* with same antagonist and also in another research in same year recorded 86.85% inhibition but against *F. moniliforme*. Honmane (2007) recorded 75.19% inhibition of *C. gloeosporioides* by *T. harzianum*. Raul (2007) recorded 86.11% against the same pathogen while Tapwalet *al.*, (2015) recorded the least inhibition i.e 15.00% of *C. gloeosporioides*. There are very few reports wherein such a

low inhibition of the mycelial growth has been recorded. Amin *et al.*, (2010) reported that isolate Th1 was capable to inhibit the growth of *R.solani* by 60.51% and in another research of Amin *et al.*, (2010) recorded 77.81 and 70.25% inhibition by Th-1 and Th-2. Tapwal *et al.*, (2015) reported that *T. harzianum* was least effective against *R. solani*(5.10% inhibition) but it performed moderately against *A. altarnata* (34.20% inhibition). Many researchers have reported the effective antagonism of *T. longibrachiatum* against different fungal pathogens. *T.longibrachiatum* is effective against *S. Rolfsii* (Shaigan *et al*, 2008; Shewarega *et al.*, 2019). It is also effective against *Fusarium*, *Rhizoctonia* (Shewarega *et al.*, 2019); *Colletotrichum* (Quiroz *et al.*, 2018) *Alternaria* (Elyousr *et al.*,2013; Prabhakaranet al 2015; Reddy *et al.*,2018)

In vitro evaluation of fungicides studies shown that amongst the systemic fungicides, Carbendazim was the most detrimental at 1000 ppm for all the isolates, as it entirely inhibited the mycelial growth of isolates followed by Hexaconazole which absolutely inhibited the mycelium of the isolate 14 at 500 ppm. The growth inhibition by that fungicide in case of isolate 5 (*T.koningii* Oudem) was 90% followed by isolate 24(*T. koningii* Oudem) (87.77%), 11 (86.11%), 23 (85.77%), 25(*T. longibrachiatum* Rifai) (84.11%) and up to 3 (*T. harzianum*Rifai.) (63.33%). The last systemic fungicide *i.e.*, Thiophanate methyl caused maximum inhibition (86.33) of isolate 23 at 500 ppm, followed by isolate 14(*T. koningii*Oudem- 80.22%), 25 (*T. longibrachiatum* Rifai)(80.00%), 5 (79.77%), 24 (75.77%), 3 (*T. harzianum* Rifai.) (68.33%) and 11 (58.00%). Rest of the three contact fungicides, Sulphur at 2500 ppm was the most compatible fungicide as the highest inhibition recorded by isolate 5 (23.11%), which was followed by isolate 23(20.00%), 25 (*T. longibrachiatum* Rifai) (9. 60%), 24 (5.88%), 14 (4.22%), 11(3.66%) and the least inhibition observed in isolate 3 (*T. harzianum* Rifai.) (0.33%). It was recorded that at a slightly lower concentration of this fungicide may not be inhibitory to the test isolates. But in case of Sulphur all the seven isolates were the most compatible and to some extent with Copper oxychloride. Copper oxychloride recorded the slightest inhibition of (43.66%) of isolate 11 at 2500 ppm and the maximum (84.44%) of isolate 5 although mancozeb recorded the slightest inhibition (59.77%) of isolate 11 at 2500 ppm and the highest (84.22%) of isolate 5. Bhat and Srivastava (2003) revealed that the triazole group fungicide Hexaconazole was detrimental to *T. harzianum* strain used in their study. Islam *et al.*, (2008) found that the growth of *Trichoderma* was very much inhibited in presence of Carbendazim 50 wp whereas, normal growth was observed in medium containing Copper oxychloride. Madhavi *et al.*, (2008) tested the compatibility of a mutant of *T. harzianum*(ThM₁) with Carbendazim (0.1%). The results indicated that the mutant was fairly compatible with Carbendazim but Mancozeb(0.25) was found inhibitory. The findings of Sarkar *et al.*, (2010) revealed that, Hexaconazole recorded cent% inhibition of *T. harzianum* at 10 ppm and above concentrations while Copper oxychloride was tolerable upto 100 ppm concentration. Ranganathaswamy *et al.*, (2012) assessed the compatibility of *T. harzianum* with fungicides and concluded that Sulphur and mancozeb were less toxic. Saxena *et al.*, (2014) stated that *T. harzianum*strain PBT23 was compatible with mancozeb up to 250 ppm. Bhale and Rajkonda (2015) checked the compatibility of *T. harzianum* and *T. koningii* with Mancozeb at 8 different

concentrations and revealed that the growth of *T. harzianum*was satisfactory up to 3000 ppm whilst that of *T. koningii* up to 1000 ppm. The results of Sharma *et al.*, (2016) revealed that *T.harzianum* strain TCMS-14 was exceptionally compatible with Sulphur at 2500 ppm where as it was compatible with Mancozeb upto 625 ppm only. Carbendazim was the most detrimental to *T. asperellum* at 100 ppm whereas, Mancozeb at the same concentration recorded the least (23.30%) mycelial inhibition (Kumar *et al.*, 2017). Mohamed and Radwan (2017) tested the compatibility of a local strain of *T. harzianum* with Copper oxychloride and sulphur at seven concentrations such as 1, 5, 10, 50, 100, 500 and 1000 ppm. None of the concentration of these two fungicides was inhibitory to the strain under study but mancozeb exhibited suppression of mycelial growth at the lowest concentration and at 100 ppm it recorded total inhibition. Sonavane and Venkataravanappa in 2017 assessed compatibility of a local strain of *T. harzianum*with contact fungicides COC, Sulphur and Mancozeb at 500, 1000, 1500 and 2000 ppm concentrations and systemic fungicides Carbendazim, Thiophanate methyl and Hexaconazole at 250, 500, 750 and 1000 ppm concentration and found that the isolate was compatible with all the three contact fungicides at 2000 ppm. But it was not compatible at all with the three systemic fungicides. All the three isolates of *T. harzianum* were incompatible with Carbendazim. Isolate Th1 was compatible with Mancozeb up to 600 pm and with Thiophanate methyl up to 500 ppm. The isolate Th2 was compatible with Mancozeb up to 500 and with Thiophanate methyl up to 700 ppm while Th3 was also compatible with Thiophanate methyl up to 700 ppm (Elshahawy *et al.*, 2016). Similar results of carbendazim were reported by Dwivedi and Vishunavat (2018) in case of *T. asperellum* and *T. harzianum*. Most of the workers have reported that Carbendazim completely inhibits the growth of *Trichoderma* species (Kumar *et al.*, 2019; Shashikumar *et al.*, 2019; Shrivastava, 2019). Tomar *et al.*,(2018) tested Mancozeb at selected concentration (25, 50, 75 and 100 ppm) for its compatibility with *T. harzianum* and observed that the Mancozeb was slightly inhibitory at 75 and 100 ppm (inhibition 5.19% and 7.03%, respectively) and Shashikumar *et al.*, (2019) concluded that Mancozeb was the least inhibitory (1.48%) at 0.15% concentration. Bagwan (2010) found Copper oxychloride at 2000ppm and Mancozeb at 2000ppm is safer against *T. harzianum* (Shrivastava, 2019; Maheshwary *et al.* 2020). Khan and Shahzad (2015) checked the tolerance level of two species *viz.* *T. harzianum* and *T. Longibrachiatum* against Topsin- M (Thiophanate methyl) and Carbendazim, at different concentrations (1, 10, 100, 1000 and 10,000 ppm) and reported that Topsin-Mand Carbendazim completely suppressed the growth of both species. Kumar *et al.*, (2017) revealed that all the four concentrations (10, 20, 40 and 80 ppm) of Hexaconazole 5% WP were totally detrimental to *T. asperellum*. Singh *et al.*, (2021) also stated that *T. harzianum* is highly incompatible with the same fungicide. Kumar *et al.*, (2017) revealed that *T. asperellum* can tolerate Mancozeb up to 100 ppm but the higher concentrations are highly injurious.The results of the studies revealed that Hexaconazole completely inhibited the growth of *Trichoderma* species(Kiran *et al.*, 2018;Singh *et al.*, 2021)Shrivastava (2019) tested the compatibility of *T. harzianum* with Carbendazim, Thiophanate methyl, Mancozeb, and Wet table sulphurat 500, 1000 and 1500 ppm

concentrations and reported that the mycelial growth of the bio-agent was above 70% in Mancozeb and Wettable sulphur while Carbendazim and Thiophanate methyl were detrimental. These results are contradictory to present findings in terms of mancozeb. Maheshwary *et al.* (2020) concluded that COC and Mancozeb at 500 ppm, favour the growth of *T. asperellum* but the higher concentrations (1000, 1500 and 2000) of both the fungicides are slightly injurious to this fungus. Carbendazim is inhibitory at 5 ppm. In the findings of Vyas *et al.*, (2020) resulted that both COC and Carbendazim found equally hazardous to *T. harzianum*. These results are in disagreement with present conclusions in context with COC.

Acknowledgement

I consider myself fortunate and greatly privileged in availing this golden opportunity to express my deepest sense of gratitude and humble indebtedness towards my chairman Dr. P. G. Borkar, Associate Professor of Plant Pathology, Dr. BSKKV, Dapoli Dist. Ratnagiri MH-415712 for his kind, generous and valuable guidance, also express my sincere thanks to Dr. M. S. Joshi, Head, Dept. of Plant Pathology, for providing all necessary facilities during the course of investigation, intellectual stimulation, kind suggestions and comments during investigation and completion of Research work. I wish to record again my genial thanks to Dr. Sudhir Navathe, Scientist from Agarkar Research Institute, Pune.

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