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Compatibility of *Rhizobium* spp. with agrochemicals used to control chickpea wilt incited by *Fusarium oxysporum* f. sp. *ciceri* (Padwick) Snyder and Hansen

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Abstract
The *Fusarium oxysporum* f. sp. *ciceri* was successfully isolated on PDA medium from naturally affected wilted chickpea and proved pathogenic on susceptible cultivar JG-62. Three isolates of *Rhizobium* spp. *viz.*, Rh1 Rh2 and Rh3 were isolated from chickpea root nodules collected from Latur, Osmanabad and Beed districts on YEMA medium and identified on the basis of biochemical characters. Among three isolates, Rh2 was found most effective in inhibition of mycelial growth of pathogen. Among seven systemic fungicides, Carbendazim was most compatible with *Rhizobium* spp. (Rh2) and Chlorothalonil and Thiophanate methyl + Pyraclostrobin were found compatible with *Rhizobium* spp. (Rh2) in contact and combi-product fungicides group. Emamectin Benzoate and Chlorantraniliprole were found compatible insecticides, whereas herbicides Imazethapyr was compatible with *Rhizobium* spp. (Rh2). Seed treatments of Carbendazim + Emamectin benzoate along Rh2 was superior in recording the least per cent disease incidence in Pot Culture experiment.

Keywords: *Rhizobium* spp., agrochemicals, chickpea wilt incited, *Fusarium oxysporum* f. sp. *ciceri*

1. Introduction
Chickpea (*Cicer arietinum* L.) belongs to family *Leguminosae* is called as King of Pulses and important pulse crop of India. Various fungal, bacterial, viral, nematode and abiotic stress has been reported as major production huddle. Among various fungal diseases, wilt of chickpea caused by *Fusarium oxysporum* f. sp. *ciceri* is mostly devastated, widespread and important throughout the world (Gupta *et al.* 1997) [7]. In India it is prevalent in all chickpea growing states and it causes 100% loss under specific condition (Jalali and Chand, 1992) [11] and at particular growth stages of crop like vegetative and reproductive (Halila and Strange, 1996) [8]. Current practices used for legume production include inoculation of seeds with rhizobia to ensure effective nodulation and subsequent Nitrogen (N) fixation along with treatment of fungicides to reduce seed rot, seedling damping-off and wilt resulting from infection by soil-borne pathogens. Fungicides affects microbial population as equally as pathogens present in the soil. The percent inhibition and its duration vary with chemical, environmental conditions and soil type. Fungicides differ in their effects on growth and survival of *Rhizobium* and *Bradyrhizobium* strains depending on the characteristics of that strain and concentration of fungicides (Hashem *et al.*, 1997) [9]. The agrochemicals applied to leguminous plants either as seed dressing or soil drenching may affect symbiotic relationship and may persist for longer time. However, the adverse effect of fungicides on agriculturally important microorganisms such as nitrogen fixers and phosphate solubilizers, resulting in poor performance of applied microbial inoculants, is a subject of great concern; several studies have conclusively shown that, some of these chemicals are incompatible with *Rhizobium* (Welty *et al.*, 1988) [19]. Hence keeping view, aspects of compatibility of agrochemicals and bioinoculants, present study on "Compatibility of *Rhizobium* spp. with Agrochemicals used to control Chickpea Wilt incited by *Fusarium oxysporum* f. sp. *ciceri* (Padwick) Snyder and Hansen" was conducted during 2021-22 at Department of Plant Pathology.

2. Materials and Methods
2.1 Isolation and Identification of *Fusarium oxysporum* f. sp. *ciceri* from infected plants
Chickpea plant showing typical wilt symptoms were collected from the chickpea growing fields of farmer and isolation of wilt fungus was done by following Standard Tissue Isolation method under aseptic condition. Based on morphological and cultural characteristics, the

pathogen involved in chickpea wilt were identified (Booth, 1971 and Aneja, 2007) [3, 1]. Pathogenicity was proved by Sick soil method in Pot culture using chickpea susceptible cultivar (JG 62).

2.2 Isolation and Identification of *Rhizobium* from root nodules of chickpea

Healthy root nodules of young and healthy chickpea plant were collected from different location of Osmanabad, Latur and Beed districts of Marathwada region of Maharashtra. Root nodules were washed with running tap water to remove adhering soil and plant debris. Nodules were dipped in 0.1% of Mercuric chloride ($HgCl^2$) solution for 30 seconds and later washed successively four times with sterilized distilled water to remove the traces of toxic $HgCl^2$. Surface sterilized nodules were transferred and crushed with a sterile glass rod in a test tube containing 5 ml distilled water. One loopful of nodule suspension were streaked on Petri plates containing Congo-Red Yeast Extract Mannitol Agar media and incubated at 28 ℃ for 2-3 days. The purity of *Rhizobium* was checked by microscopic examination and further confirmation of identity were performed by Gram's staining, Congo-red test, Catalase oxidation test, Potassium hydroxide (KOH) test.

2.3 *In vitro* efficacy of *Rhizobium* spp. against *Fusarium oxysporum* f. sp. *ciceri*

All isolates of *Rhizobium* spp. were tested *in vitro* against *Fusarium oxysporum* f. sp. *ciceri.* A culture disc (5mm) for the *Fusarium* was placed along with the periphery of the YEMA plate and exactly opposite to it, pure culture suspension of the different *Rhizobium* spp. was streaked with wire / inoculation needle loop. The YEMA plate inoculated (in the centre) only with pure culture disc of the *Fusarium* was maintained as untreated control. After 96 hours radial mycelium growth was recorded.

2.4 *In vitro* compatibility of *Rhizobium* spp. with various agrochemicals

Compatibility of *Rhizobium* with fungicides (systemic, contact and combi-product), insecticides and herbicides (pre/post-emergence) were tested *in vitro.* Systemic fungicides were evaluated at 1000 ppm, whereas 2500 ppm concentration was used for contact and combi-product fungicides and recommended dose of insecticides and herbicides used to access their compatibility with *Rhizobium* spp., by employing paper disc method/inhibition zone technique using YEMA as basal culture medium. Three Petri plates per treatment were maintained.

Table 1: *In vitro* compatibility *Rhizobium* spp. with systemic fungicides Treatment Details

Tr. No.	Treatments	Tr. No.	Treatments
T_1	Carbendazim 50% WP	T_5	Difenoconazole 25% EC
T_2	Azoxystrobin 23% SC	T_6	Tebuconazole 25.9% EC
T_3	Propiconazole 25% EC	T_7	Hexaconazole 5% EC
T_4	Thiophanate methyl 70% WP	T_8	Control (untreated)

Treatment Details

Table 2: *In vitro* compatibility *Rhizobium* spp. with contact and combi product fungicides

Tr. No.	Treatments	Tr. No.	Treatments
T_1	Copper oxychloride 50% WP	T_6	Tebuconazole 50% + Trifloxystrobin 25% WDG
T_2	Mancozeb 75% WP	T_7	Thiophanate methyl 45% + Pyraclostrobin 5% FS
T_3	Chlorothalonil 75% WP	T_8	Captan 70% + Hexaconazole 5% WP
T_4	Captan 50% WP	T_9	Control (untreated)
T_5	Carbendazim 12% + Mancozeb 63% WP		

Treatment Details

Table 3: *In vitro* compatibility *Rhizobium* spp. with Insecticide

Tr. No.	Treatments	Tr. No.	Treatments
T_1	Imidacloprid 17.8% WSL	T_5	Spinosad 45% SC
T_2	Quinalphos 25% EC	T_6	Emamectin Benzoate 5% WG
T_3	Dimethoate 30% EC	T_7	Chlorantraniliprole 18.5% SC
T_4	Profenofos 50% EC	T_8	Control (untreated)

Treatment Details

Table 4: *In vitro* compatibility with Herbicides

Tr. No.	Treatments	Conc. in ppm.	Tr. No.	Treatments	Conc. in ppm.
T_1	Pendimethalin 30% EC	500	T_6	Imazethapyr 10% EC	1500
T_2	Pendimethalin 30% EC	1000	T_7	Quizalofop ethyl 5% EC	500
T_3	Metribuzin 70% WP	500	T_8	Quizalofop ethyl 5% EC	1000
T_4	Metribuzin 70% WP	1000	T_9	Control (untreated)	
T_5	Imazethapyr 10% EC	750			

Compatibility of *Rhizobium* spp. with best compatible fungicide, insecticide and herbicide studied *in vitro* were used for seed treatment with JG-62 (Susceptible) cultivar of chickpea in pot culture by using Sick pot culture technique.

Treatment details

Table 5: Evaluation of compatible isolate of *Rhizobium* with agrochemicals in control of wilt of chickpea in pot culture

Tr. No.	Treatments	Tr. No.	Treatments
T_1	ST with potential *Rhizobium* (Rh)	T_6	ST with compatible fungicide + compatible insecticide + Rh
T_2	ST with compatible fungicide	T_7	Spraying of pre-emergence herbicide + Rh
T_3	ST with compatible insecticide	T_8	Spraying with post-emergence herbicide + Rh
T_4	ST with compatible fungicide + Rh	T_9	Control (untreated)
T_5	ST with compatible insecticide + Rh		

ST = Seed Treatment

3. Results and Discussions

3.1 Isolation and identification of *Fusarium oxysporum*

The test pathogen (*Fusarium oxysporum*) was isolated successfully on Potato Dextrose Agar (PDA) from the roots of infected chickpea plants showing typical symptoms of wilt by Standard Tissue Isolation method. The pure culture of fungus was identified on the basis of cultural and morphological character. On PDA fungus produced hyaline, septate, profusely branched whitish pink mycelium 2-3 septate, fusoid (curved) macroconidia and variable shape micro conidia.

Nikam *et al*. (2011) [16] who also isolated *F. oxysporum* f. sp. *ciceri* from wilted plant and purify by hyphal tip method. Minakshi *et al*. (2017) [14] also isolated *Fusarium oxysporum* f. sp. *ciceri* from wilted plant and purify by Tissue isolation method on Potato Dextrose Agar.

3.2 Pathogenicity test

Isolated pathogen was proved pathogenic by Sick soil method. Symptoms of Drooping, yellowing and withering of leaves with finally death of susceptible cultivar JG-62 were observed after artificial inoculation of pathogen. On re-isolation of pathogen on PDA the cultural and morphological characters of the pathogen showed similarity with the original pathogen isolated from naturally diseased chickpea plant in the field and thus Koch's postulates were proved. Nikam *et al*. (2011) [16] confirmed pathogenicity of the *Fusarium oxysporum* f. sp. *ciceri* by Sick Soil Inoculation.

3.3 Isolation of *Rhizobium* spp.

Root nodules was collected from the young and healthy chickpea plant from farmer's field from different location of Osmanabad, Latur and Beed districts of Marathwada region of Maharashtra and isolated on Congo-Red Yeast Extract Mannitol Agar (CRYEMA) media by streek plate method. Isolated colonies were designated as (Table 6).

Table 6: Isolates of *Rhizobium* from chickpea growing areas of Marathwada region

Sr. No.	District	Tehsils	Name of Village	Isolate Code
1.	Osmanabad	Kalamb	Kalamb	Rh1
2.	Latur	Latur	Latur	Rh2
3.	Beed	Kaij	Chincholi Mali	Rh3

3.4 Biochemical characterization

Different biochemical tests *viz*., Gram staining, Catalase test, Potassium hydroxide (KOH) solubility test and Starch hydrolysis test were attempted for biochemical characterization of *Rhizobium* spp. The results revealed that, all the *Rhizobium* isolates were small, rod shaped and negative in gram reaction. All these isolates were found positive for catalase test, KOH test and starch hydrolysis test.

3.8 *In vitro* efficacy of *Rhizobium* spp. against *Fusarium oxysporum* f. sp. *ciceri*

All three isolates of *Rhizobium* spp. *viz.*, Rh1, Rh2 and Rh3 were tested against *F. oxysporum* f. sp. *ciceri in vitro.* All isolates inhibited mycelial growth of test pathogen (90 mm), but none of *Rhizobium* spp. isolates showed complete inhibition of pathogen. Among three isolates tested (Table 7) Rh2 was found most effective in inhibition of mycelial growth (46.66%) where least mycelial growth (48 mm) was observed. The Rh1 and Rh3 isolates recorded mycelium growth of pathogen up to 55 mm with per cent inhibition of 38.88% and 57.5 mm with per cent inhibition 36.11%, respectively. Buonassisi *et al.* (1986) [4] also showed antagonistic properties of *Rhizobium* against wilt pathogen.

Table 7: *In vitro* efficacy of *Rhizobium* spp. against *Fusarium oxysporum* f. sp. *ciceri*

Tr. No.	Treatments	*Colony diameter of test pathogen (mm)	Percent inhibition
T_1	Rh1	55.00	38.88
T_2	Rh2	48.00	46.66
T_3	Rh3	57.50	36.11
T_4	Control	90.00	00.00
SE±		0.412	
CD at 1%		1.225	

*Colony diameter of test pathogen = Average of six replications.

3.9 *In vitro* compatibility of *Rhizobium* spp. (Rh2) with various agrochemicals

3.9.1 *In vitro* compatibility *Rhizobium* spp. (Rh2) with systemic fungicides

The results (Table 8) revealed that all of the seven systemic fungicides *viz*., Carbendazim 50% WP, Azoxystrobin 23% SC, Propiconazole 25% EC, Thiophanate methyl 70% WP, Difenoconazole 25% EC, Tebuconazole 25.9% EC and Hexaconazole 5% EC tested at various concentrations, exhibited significant differences in the amount of inhibition zone (mm) recorded at 48, 72 and 96 hrs of incubation. Further, the zone of inhibition was found to be increased steadily with increase in concentrations of the test fungicides (Table). Carbendazim 50% WP was found most compatible as didn't inhibit the growth of *Rhizobium*. Hexaconazole 5% EC and Azoxystrobin 23% EC were found at par with each other in producing inhibition zone after 96 hrs of observation.

Table 8: *In vitro* compatibility of *Rhizobium* spp. (Rh2) with systemic fungicides at 48, 72 and 96 hrs

Tr. No.	Treatments	Inhibition zone* (mm) at 48 hrs	Inhibition zone* (mm) at 72 hrs	Inhibition zone* (mm) at 96 hrs
T_1	Carbendazim 50% WP	0.00	0.00	0.00
T_2	Azoxystrobin 23% SC	7.50	8.00	8.50
T_3	Propiconazole 25% EC	13.50	15.00	16.50
T_4	Thiophanate Methyl 70% WP	9.00	10.50	11.50
T_5	Difenoconazole 25% EC	10.50	12.50	13.00
T_6	Tebuconazole 25.9% EC	11.50	13.00	14.00
T_7	Hexaconazole 5% EC	6.00	7.50	8.00
T_8	Control (untreated)	0.00	0.00	0.00
S.E ±		0.500	0.612	0.468
C.D. (P=0.01)		1.512	1.852	1.414

* = Mean of three replications

Rest of the fungicides *viz*., Propiconazole 25% EC, Thiophanate methyl 70% WP, Difenoconazole 25% EC and Tebuconazole 25.9% EC tested at 1000 ppm concentration were found least compatible with the test bacterium, as they expressed greater inhibition zones, at both 48, 72 and 96 hrs of incubation.

Dinkwar *et al.* (2020) [5] who also reported that Carbendazim 50% WP at all four dosages were found compatible with *B. japonicum*, as they didn't show any zone of inhibition, at 72 hrs of incubation. Mishra *et al*. (2013) [15] evaluated *in vitro* the compatibility of *B. japonicum* and PGPR with five fungicides *viz*., Carboxin 75% WP, Carboxin 37.5% + Thiram 37.5% DS, Thiram 70% WS, Carbendazim 50% WP and reported that all of the test chemicals at their lower dosages were compatible with both, whereas, at higher dosages (>0.4%) were non-compatible.

3.9.2 *In vitro* compatibility of *Rhizobium* spp. (Rh2) with contact and combi product fungicides

The results (Table 9) revealed that all of the eight contact and combi product fungicides tested at 2500 ppm concentration also exhibited significant differences in the amount of inhibition zone (mm) recorded at 48, 72 and 96 hrs of incubation. Further, the zone of inhibition was found to be increased steadily with increase in concentrations of the test fungicides.

The fungicides, Chlorothalonil 75% WP and Thiophanate methyl 45% + Pyraclostrobin 5% FS at 2500 dose were found highly compatible with *Rhizobium* spp. (Rh2), as they didn't show any zone of inhibition, at 48, 72 and 96 hrs of incubation. Whereas, rest of the six contact and combi-product fungicides *viz*., Copper oxychloride 50% WP, Mancozeb 75% WP, Captan 50% WP, Carbendazim 12% + Mancozeb 63% WP, Tebuconazole 50% + Tryfloxystrobin 25% WDG and Captan 70% + Hexaconazole 5% WP tested at 2500 ppm concentration were found least compatible with the test bacterium, as they expressed significant inhibition zones, at 48, 72 and 96 hrs of incubation as compared to Chlorothalonil 75% WP and Thiophanate methyl 45% + Pyraclostrobin 5% FS.

Table 9: *In vitro* compatibility of *Rhizobium* spp. (Rh2) with contact and combi fungicides at 48, 72 and 96 hrs at 2500 ppm

Tr. No.	Treatments	Inhibition zone* (mm) at 48 hrs	Inhibition zone* (mm) at 72 hrs	Inhibition zone* (mm) at 96 hrs
T_1	Copper oxychloride 50% WP	18.00	20.50	21.50
T_2	Mancozeb 75% WP	6.00	8.50	10.00
T_3	Chlorothalonil 75% WP	0.00	0.00	0.00
T_4	Captan 50% WP	8.50	10.50	11.50
T_5	Carbendazim 12% + Mancozeb 63% WP	6.50	7.50	8.00
T_6	Tebuconazole 50% + Tryfloxystrobin 25% WDG	7.00	7.50	9.50
T_7	Thiophanate methyl 45% + Pyraclostrobin 5% FS	0.00	0.00	0.00
T_8	Captan 70% + Hexaconazole 5% WP	10.50	13.50	14.50
T_9	Control (untreated)	0.00	0.00	0.00
S.E ±		0.667	0.577	0.471
C.D. (P=0.01)		1.996	1.723	1.411

* = Mean of three replications.

Curley and Burton (1975) [20] also observed less toxic effect of Captan on rhizobia than was PCNB. Thiram had no adverse effect on viable rhizobia or taproot nodulation Panwar *et al*. (2015) [17] studied *in vitro* compatibility of *Rhizobium* with fungicides *viz*., Thiram 75% WS, Carbendazim 50% WP, Captan 50% WP and Metalaxyl 35% WS (@ 50, 100, 200, 300 and 500 ppm), by Poison Food Technique. They reported that Metalaxyl and Thiram even at lower concentration as non-compatible and rest of the fungicides as compatible with the bacterium, even at higher concentration.

3.9.3 *In vitro* compatibility of *Rhizobium* spp. (Rh2) with insecticides

The results (Table 10) revealed that all of the seven insecticides tested at recommended dose, exhibited significant differences in the amount of inhibition zone recorded at 48, 72 and 96 hrs of incubation. Further, the zone of inhibition was found to be increased steadily with increase in concentrations of the test insecticides.

Table 10: *In vitro* compatibility of *Rhizobium* spp. (Rh2) with insecticides at 48, 72 and 96 hrs at recommended dose

Tr. No.	Treatments	Inhibition zone* (mm) at 48 hrs	Inhibition zone* (mm) at 72 hrs	Inhibition zone* (mm) at 96 hrs
T_1	Imidacloprid 17.8% WSL	6.50	8.50	10.00
T_2	Quinalphos 25% EC	9.50	10.50	12.00
T_3	Dimethoate 30% EC	0.00	0.00	0.00
T_4	Profenofos 50% EC	10.00	12.00	13.00
T_5	Spinosad 45% SC	11.00	12.50	14.00
T_6	Emamectin Benzoate 5% WG	0.00	0.00	0.00
T_7	Chlorantraniliprole 18.5% SC	0.00	0.00	0.00
T_8	Control (untreated)	0.00	0.00	0.00
S.E ±		0.408	0.645	0.540
C.D. (P=0.01)		1.234	1.952	1.633

* = Mean of three replications

The insecticides, Emamectin Benzoate 5% WG and Chlorantraniliprole 18.5% SC at the recommended dose were found highly compatible with *Rhizobium* spp. (Rh2), as they didn't show any zone of inhibition, at 48, 72 and 96 hrs of incubation. Whereas rest of the Five insecticides *viz*., Imidacloprid 17.8% WSL, Quinolphos 25% EC, Dimethoate 30% EC, Profenofos 50% EC and Spinosad 45% SC at the recommended dose were found least-compatible with the test bacterium, as they expressed significant inhibition zones, at both 48, 72 and 96 hrs of incubation.

Ghosh *et al*. (2003) who also observed the compatibility of moong rhizobia (M-1006) in terms of nodule occupancy, nodule number, nitrogen fixation and grain yield in green gram with different doses of Forate, Furadan, Monocrotophos and Chloropyriphos. Cheema *et al*. (2009) [21] conducted field trial and evaluated those insecticides Endosulfan @ 15 ml/kg and Chlorpyrifos @ 10 ml/kg seed had no adverse effect on germination, nodulation, yield-attributing characters and grain yield when applied alone or in combination with the recommended fungicide Captan and *Rhizobium* inoculant, thus, indicating the compatibility amongst all the 3 components *viz*., insecticide, fungicide and *Rhizobium* inoculant for seed treatment in chickpea.

3.9.4 *In vitro* compatibility of *Rhizobium* spp. (Rh2) with herbicides

The results (Table 11) revealed that all of the herbicides tested each at two concentrations (50% RD and 100% RD), exhibited significant differences in the amount of inhibition zone recorded a 48, 72 and 96 hrs of incubation. Further, the zone of inhibition was found to be increased steadily with increase in concentrations of the test herbicides.

The herbicide, Imazethapyr 10% EC were found highly compatible with *Rhizobium* spp. (Rh2), as it didn't show any zone of inhibition, at 48, 72 and 96 hrs of incubation. Whereas, rest of the three herbicides *viz*., Pendimethalin 30% EC, Metribuzin 70% WP and Quizolofop ethyl 5% EC tested at two concentrations were found least compatible with the test bacterium, as they expressed significant inhibition zones, at 48, 72 and 96 hrs of incubation.

Table 11: *In vitro* compatibility of *Rhizobium* spp. (Rh2) with herbicides at 48, 72 and 96 hrs

Tr. No.	Treatments	Conc. (ppm)	Inhibition zone* (mm) at 48 hrs	Inhibition zone* (mm) at 72 hrs	Inhibition zone* (mm) at 96 hrs
T_1	Pendimethalin 30% EC	500	6.00	6.50	8.50
T_2	Pendimethalin 30% EC	1000	11.00	12.50	13.00
T_3	Metribuzin 70% WP	500	6.50	7.50	8.00
T_4	Metribuzin 70% WP	1000	10.50	11.50	12.50
T_5	Imazethapyr 10% EC	750	0.00	0.00	0.00
T_6	Imazethapyr 10% EC	1500	6.50	7.50	9.00
T_7	Quizalofop ethyl 5% EC	500	9.00	12.50	13.50
T_8	Quizalofop ethyl 5% EC	1000	16.50	18.00	18.50
T_9	Control (untreated)		0.00	0.00	0.00
S.E ±			0.481	0.509	0.451
C.D. (P=0.01)			1.441	1.525	1.351

* = Mean of three replications

Jeenie *et al*. (2011) [12] who also evaluated *in vitro* sensitivity of *Rhizobium* and PSB to herbicides *viz.*, Fluchloralin 75% WG and Pendimethalin30% EC and reported both the herbicides as compatible with both *Rhizobium* and PSB. Khanna *et al*. (2012) [13] evaluated *in vitro* compatibility of herbicides *viz*., Pendimethalin 30% EC (@ 1980 and 396 ppm), Imazethapyr 10% SL (@ 200 and 400 ppm) and Paraquat 24% SL (@ 1272 and 255 ppm) with *Rhizobium*, by Inhibition zone method and observed that Pendimethalin at both doses as compatible with the bacterium, whereas Imazethapyr and Paraquat were non-compatible. Indradevi *et al.* (2017) [10] studied the effect of pesticides *viz.*, Carbendazim 50% WP, Thiram 75% WS, Imazethapyr 10% SL and Monocrotophos 36% on *Rhizobium* spp. in *Vigna mungo*. They reported that at lower dosages, the test chemicals significantly increased the number of nodules, shoot height, root height and total plant height.

3.10.5 Evaluation of compatible isolate of *Rhizobium* spp. (Rh2) with agrochemicals in control of chickpea wilt in pot culture

The pot culture experiment conducted during *Rabi*, 2021-22 to evaluate efficacy of compatible isolate of *Rhizobium* spp. with different agrochemicals in control of wilt of chickpea in pot culture using susceptible variety JG-62 against *F. oxysporum* f. sp. *ciceri.* Result (Table 12) revealed that, all the

treatments were found effective against the test pathogen and significantly enhance the seed germination, reduced the percent pre- and post-emergence mortality, and reduced the per cent disease incidence in chickpea over untreated control. All treatments increased germination percentage in the ranged 73.33 to 93.33 per cent compared to control (66.66%). Among all treatments the Carbendazim 50% WP + Rh2 and Carbendazim 50% WP + Emamectin Benzoate 5% WG + Rh2 gave 93.33 per cent of seed germination. This was followed by Rh2 (86.66%) Carbendazim 50% WP + Rh2 (86.66%), Imazethapyr 10% EC + Rh2 (86.66%), Carbendazim 50% WP (80.00%) and Pendimethalin 30% EC + Rh2 (80.00%). The least germination percentage (73.33%) was observed with seed treatment of Emamectin Benzoate 5% WG.

3.10.5.2 Pre emergence seedling mortality

Compatible isolate of *Rhizobium* spp. with fungicide, insecticide and herbicide significantly minimized the seedling mortality both at pre-emergence and post emergence stage and mortality of seedling recorded in the range of 6.66 to 26.66 per cent at pre-emergence stage as compared to control (33.33%). Least pre-emergence seedling mortality of 6.66 per cent was observed with Carbendazim 50% WP + Rh2 and Carbendazim 50% WP + Emamectin Benzoate 5% WG + Rh2. This was followed by the treatment *viz*., Rh2 (13.33%), Imazethapyr 10% EC + Rh2 (13.33%), Imazethapyr 10% EC + Rh2 (13.33%), Carbendazim 50% WP (20.00%) and Pendimethalin 30% EC + Rh2 (20.00%). Seed treatment with Emamectin Benzoate 5% WG was found least effective with highest pre-emergence seedling mortality (26.66%) of chickpea cultivar JG-62.

3.10.5.3 Post emergence seedling mortality

All treatments of compatible *Rhizobium* spp. with agrochemicals minimized post emergence seedling mortality, which observed from 7.69 to 16.66 per cent, whereas mortality was up to 20.00 per cent in control. The least post emergence seedling mortality was found in Rh2 (7.69%). This was followed treatments *viz*., Carbendazim 50% WP (8.33%), Emamectin Benzoate 5% WG (9.09%), Carbendazim 50% WP + Rh2 (14.28%), Carbendazim 50% WP + Emamectin Benzoate 5% WG + Rh2 (14.28%), Emamectin Benzoate 5% WG + Rh2 (15.38%) and Imazethapyr 10% EC + Rh2 (15.38%). The seed treated with Pendimethalin 30% EC + Rh2 was found least effective with highest percentage of post emergence seedling mortality (16.16%).

3.10.5.4 Per cent wilt incidence

All treatments significantly influenced the disease incidence and per cent disease control which was recorded against untreated control. The lowest wilt incidence and highest disease control was recorded with treatment combination of Carbendazim 50% WP + Emamectin Benzoate 5% WG + Rh2 followed by Imazethapyr 10% EC + Rh2 Seed treatment of. Carbendazim 50% WP + Rh2, Rh2, Emamectin Benzoate 5% WG, Pendimethalin 30% EC and Carbendazim 50% WP recorded 16.16%, 18.18%, 18.18%, 22.22% and 27.27% wilt incidence, respectively. The highest wilt incidence observed in Emamectin Benzoate 5% WG + Rh2 (30.00%) and which was found least effective in controlling disease.

Table 12: Evaluation of compatible isolate of *Rhizobium* spp. (Rh2) with agrochemicals in control of wilt of chickpea in pot culture

Tr. No.	Treatment Details	*Per cent Germination	*Pre-emergence mortality	*Post-emergence mortality (8 days)	*Per cent Wilt (45 days)	Disease inhibition per cent over control
T_1	ST with potential *Rhizobium* (Rh2)	86.66 (68.57)	13.33 (21.41)	7.69 (16.09)	18.18 (25.23)	63.64
T_2	ST with compatible fungicide	80.00 (63.43)	20.00 (26.56)	8.33 (16.77)	27.27 (31.48)	45.46
T_3	ST with compatible insecticide	73.33 (58.90)	26.66 (31.08)	9.09 (17.54)	18.18 (25.23)	63.64
T_4	ST with compatible fungicide + Rh2	93.33 (75.03)	6.66 (14.95)	14.28 (22.20)	16.16 (23.70)	67.68
T_5	ST with compatible insecticide + Rh2	86.66 (68.57)	13.33 (21.41)	15.38 (23.08)	30.00 (33.21)	40
T_6	ST with compatible fungicide + compatible insecticide + Rh2	93.33 (75.03)	6.66 (14.95)	14.28 (22.20)	8.33 (16.77)	83.34
T_7	Spraying of pre-emergence herbicide + Rh2	80.00 (63.43)	20 (26.56)	16.66 (24.09)	22.22 (28.12)	55.56
T_8	Spraying with post-emergence herbicide + Rh2	86.66 (68.57)	13.33 (21.41)	15.38 (23.08)	15.38 (23.08)	69.24
T_9	Control (untreated)	66.66 (54.73)	33.33 (35.26)	20.00 (26.56)	50.00 (45)	-
S.E ±		0.667	0.624	0.645	0.521	
C.D. (P=0.01)		1.996	1.867	1.993	1.561	

*Observation = Average of three replications

Figures in parenthesis are arcsine transformation value Indradevi *et al.* (2017) [10] who studied the effect of pesticides *viz.*, Carbendazim 50% WP, Thiram 75% WS, Imazethapyr 10% SL and Monocrotophos 36% SL (each @ 100, 200 and 300 ppm/kg) on *Rhizobium* spp. in *Vigna mungo.* They reported that at lower dosages (100 and 200 ppm), the test chemicals significantly increased the number of nodules, shoot height, root height and total plant height. Dinkwar *et al.* (2020) [5] evaluated the compatibility of *B. japonicum* with systemic, non-systemic/contact, combi-fungicides at 50%, 75%, 100% and 125% of the recommended doses by inhibition zone technique and resulted that the fungicides Carbendazim 50% WP and Mancozeb 75% WP at all four dosages were found compatible with *B. japonicum*, as they didn't show any zone of inhibition, at 72 hrs of incubation.

4. References

1. Aneja KR. Experiments in Microbiology, Plant

Pathology and Biotechnology. (IV Ed.) New Age International (P) Limited, Publishers. New Delhi; c2007. p. 145-156.
2. Anonymous. Statistical year book by Ministry of Statistics and Programme Implementation Government of India; c2020.
3. Booth C. The genus *Fusarium* commonwealth mycological Institute. Kew, Surrey, England; c1971. p. 237.
4. Buonassisi AJ, Copeman RJ, Pepin HS, Eaton GW. Effect of *Rhizobium* spp. on *Fusarium solani* f. sp. *phaseoli.* Canadian Journal of Plant Pathology. 1986;8:140-146.
5. Dinkwar GT, Thakur KD, Bramhankar SB, Pillai TS, Isokar SS, Ravali T, *et al*. *In vitro* compatibility of *B. japonicum* with fungicides. International Journal of Chemical Studies. 2020;8(2):692-694.
6. Ghosh R, Sharma M, Telangre R, Pande S. Occurrence & Distribution of Chickpea Diseases in Central & Southern Parts of India. American Journal of Plant Science. 2013;4:940-944.
7. Gupta SK, Upadhyay JP, Ojha KH. Effect of fungicidal seed treatment on the incidence of chickpea wilt complex. Annals of Plant Protection Science. 1997;5:184-187.
8. Halila MH, Strange RN. Identification of the causal agent of wilt of chickpea in Tunisia as *Fusarium oxysporum* f. sp. *ciceri* race 0. Phytopathologia Mediterranea. 1996;35:67-74.
9. Hashem FM, Saleh SA, Berkum VP, Voll M. Survival of *Bradyrhizobium* sp. on fungicide treated peanut in relationship to plant growth and yield. World Journal of Microbiology and Biotechnology. 1997;13:335-340.
10. Indradevi M, Madanraj P, Panneerselvam A. Effect of pesticides on the suppression on the growth and development of *Vigna mungo* treated with *Rhizobium* sp. International Journal of Current Microbiology and Applied Science. 2017;6(2):446-452.
11. Jalali BL, Chand H. Chickpea wilt. In: Plant Diseases of International Importance. 1. Diseases of cereals & pulses. (Singh US, Mukhopadhayay AN, Kumar J & Chaube HS eds.) Prentice Hall, Englewood Cliffs, NJ; c1992. p. 429-444.
12. Jeenie P, Sharma P, Khanna V. *In vitro* sensitivity of *Rhizobium* and phosphate solubilising bacteria to herbicides. Indian Journal of Microbiology. 2011;51(2):230-233.
13. Khanna V, Singh G, Sharma P, Kaur H. Influence of herbicides on *Rhizobium* growth & its symbiosis with pigeonpea. Trends in Bioscience Journal. 2012;5(2):133-135.
14. Minakshi P, Gupta O, Rathod PK. Morphological, cultural & pathogenic variation in races & variant of *Fusarium oxysporum* f. sp. *ciceri* from seven locations of central zone of India. International Journal of Applied and Pure Science and Agriculture; c2017. p. 66-74.
15. Mishra G, Kumar N, Giri K, Pandey S. *In vitro* interaction between fungicides & beneficial plant growth promoting rhizobacteria. Academic Journal. 2013;8(45):5630-5633.
16. Nikam PS, Jagtap GP, Sontakke PL. (b). Survey, surveillance & cultural characteristics of chickpea wilt caused by *Fusarium oxysporum* f. sp. *ciceri.* African Journal of Agricultural Research. 2011;6(7):1913-1917.
17. Panwar A, Sharma YK, Meena RS, Aishwath OP, Choudhary S, Jethra G. *In-vitro* interaction between fungicides and rhizobacteria isolated from fenugreek root nodules. Annals of Plant and Soil Research. 2015;17(4):336-341.
18. Patil VB, Gawade DB, Suryawanshi AP, Zagde SN. Biological & fungicidal management of chickpea wilt caused by *Fusarium oxysporum* f. sp. *ciceri.* International Quarterly Journal of Life Science. 2015;10(2):685-690.
19. Welty LE, Prestbye LS, Hall JA, Mathre DE, Ditterline RL. Effect of fungicide seed treatment and rhizobia inoculation on chickpea production. International Journal of Applied Agricultural Research. 1988;3:17-20.
20. Curley RL, Burton JC. Compatibility of Rhizobium japonicum with chemical seed protectants 1. Agronomy Journal. 1975 Nov;67(6):807-8.
21. Archin NM, Espeseth A, Parker D, Cheema M, Hazuda D, Margolis DM. Expression of latent HIV induced by the potent HDAC inhibitor suberoylanilide hydroxamic acid. AIDS research and human retroviruses. 2009 Feb 1;25(2):207-212.