Agriways
(A Multidisciplinary Peer –Reviewed Refereed Journal)
www.agriwaysjournal.com
ISSN (Print) 2321-8614 &ISSN (Online) 2454-2318
Vol 10 Issue 02 July-Dec 2022

Evaluation of different fungicides against stem rot on tomato caused by *S.rolfsii* under *invitro* condition

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Abstract:
Tomato (*Solanum lycopersicum* L.) is one of the most popular and widely grown vegetable in the India belongs to family *Solanaceae*. Tomato is considered "protective food" and year-round production throughout the world. The stem rot of tomato is the most severe threat for the tomato industry and also for foreign exchange earnings. The disease caused by soil borne fungi, *Sclerotium rolfsii* that cause stem rot or collar rot in tomato, has become more serious among plant pathogenic fungi. In laboratory screening of different fungicides, non-systemic fungicides @ 1000, 1500 and 2000 ppm (mancozeb 75 % WP, thiram 75 % WS, chlorothalonil 75 % WP and propineb 70 % WP), systemic fungicides @ 100, 250 and 500 ppm (tebuconazole 25.9 % EC, hexaconazole 5% EC, propiconazole 25 % EC, difenoconazole 25 % EC and pyraclostrobin 20 % WG) and ready mix fungicides @ 250, 500 and 1000 ppm (azoxystrobin 11 % + tebuconazole 18.30 % SC, carboxin 37.5 % + thiram 37.5 % WS, tebuconazole 50 % + trifloxystrobin 25 % WG, carbendazim 12 % + mancozeb 63 % WP and fluxapyroxad 250 g/l + pyraclostrobin 250 g/l SC) were found cent per cent inhibition of mycelial growth of *S. rolfsii*under in vitro condition.

Key words: Tomato, stem rot, *Sclerotium rolfsii*, fungicides

Date of Submission: 017-11-2022 **Date of Acceptance: 01-12-2022**

Introduction:

Tomato (*Solanum lycopersicum*L.) is one of the most popular and widely grown vegetable in the India. The common tomato belongs to an extremely diverse and large family the *Solanaceae*. The species originated in Western South America and Central America. Tomato is considered "protective food" because of its nutritional benefits and year-round production throughout the world (Prasad *et al.*, 2017). The crop area of tomato is continuously increasing and the consumption quantity also enhanced by 3 % annually average rate (Abedin *et al.*, 2018). The tomatoes contain a large amount of water, vitamins and minerals, a low amount of protein, fat and some carbohydrate. Tomato can help combat the formation of free radicals known to cause cancer.

Tomato crop is mostly susceptible to biotic (fungi, bacteria, viruses and nematodes) and abiotic (temperature, sunlight and malnutrition *etc*.) stresses (Balanchard, 1992). The *S. rolfsii*Sacc. is

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www.agriwaysjournal.com Vol 10 Issue 02 July-Dec 2022

widely spread across the tropics, subtropics and warmer regions of the temperate zone of the world (Mahato*et al.*, 2018). The disease caused by *S. rolfsii*, soil-borne fungi that cause stem rot, foot rot or collar rot in tomatoes, has become more serious among plant pathogenic fungi. Among these, stem rot has caused severe infections in tomato growing areas of India. This disease causes approximately 1-60 % crop loss in field condition (Kator*et al.*, 2015).

It causes collar rot disease in several plants, including tomatoes it affects the pre and post-emergence plants in nursery beds and pots (Prasad *et al.*, 2017). Seedlings are very susceptible and die quickly once they become infected. Wilted plants often decline and die rapidly as a result of an extensive lower stem rot. White mat was often spread out onto the nearby soil surface. Soft water-soaked, sunken and slightly yellowish lesions develop. The skin of the fruit often cracks open and fine white mycelium and developing sclerotia spreads over the surface and quickly fills lesion cavities (Kator*et al.*, 2015).Close examination of the diseased plants showed deep cracks near collar region. On all infected plant tissues and even on nearby soil, *S. rolfsii*produced numerous, small sclerotia. Sclerotia contain viable hyphae and serve as the primary inoculum source in the disease cycle. Oxalic acid plays an important role in the virulence of *S. rolfsii* (Punja, 1985).Since it is a very serious problem, a study on certain basic aspects of this disease is very much essential for its management.

Material and methods:

Isolation and Identification

In vitro assessment of different non-systemic, systemic and ready-mix fungicides were tested against *S. rolfsii* at Department of Plant Pathology, Junagadh Agricultural University in 2020-2021. The pathogen was isolated from naturally infected stem rot on tomato plant.The freshly collected plants of tomato which showed symptoms of infection which were usually yellowing and wilting of leaves and stem rot at the collar region and pulled out very easily. At that time directly picked white mycelial growth adhered to collar region along with dark brown colour mustard seed like sclerotia from infected stem. 3-4 mm bits were cut from infected collar region with blade and pieces were dipped in 1 % sodium hypochlorite solution for 1 minute and finally washed well with three changes of sterilized distilled water eliminate excess sodium hypochlorite. After that excess water was removed with sterilized blotting paper then the pieces were transferred on to PDA medium in Petri plates and incubated at 28 ± 1° C and observed periodically for growth of the fungus.

Poisoned food technique

The required quantities of each test fungicides were added in a conical flask containing 100 ml PDA medium so as to get required concentration. The flask containing poisoned medium was well shake to facilitate uniform mixture of fungicides and 20 ml of medium was poured in sterilized Petri plates. On solidification of the medium, the plates were inoculated in the centre by placing 5 mm diameter culture disc cut aseptically with the help of cork borer from 7-10 days old pure culture of *S. rolfsii.* Three repetitions were kept for each concentration of respective fungicide. The inoculated plates were incubated at 28 ± 1° C for *S. rolfsii*. The growth of test fungus on non-poisoned PDA was served as a control (Grover and Moore, 1962).

Toxicity index for each fungicide were calculated by total of growth inhibition per cent of all concentrations for respective fungicide. The per cent mycelial growth inhibition of fungus in each treatment was calculated as per following formula.

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

Where, I = Per cent inhibition of mycelia growth

C = Radial growth of fungi in control (mm)

T = Radial growth of fungi in treatment (mm)

The experiment was laid out with twenty-four treatments with three repetitions with different concentration according to its group. Completely Randomized Block Design with factorial concept was used for analysing the data.

Result and discussion

Efficacy of eight commonly used non-systemic fungicides were evaluated against *S. rolfsii* at different concentrations *viz*., 1000, 1500 and 2000 ppm using poisoned food technique. The perusal of data presented in Table 1 revealed that cent per cent mean growth inhibition of *S. rolfsii* was recorded in mancozeb, thiram, chlorothalonil and propineb which was followed by wettablesulphur(43.50 %). While, eight commonly used systemic fungicides were evaluated against *S. rolfsii* at different concentrations *viz*., 100, 250 and 500 ppm using poisoned food technique. The perusal of data presented in Table 2 revealed that cent per cent mean growth inhibition of *S. rolfsii* was recorded in tebuconazole, hexaconazole, propiconazole, difenoconazole and pyraclostrobin which was followed by azoxystrobin (56.31 %), carbendazim (4.03 %) which was at par with thiophanate methyl (2.95 %). Maximum toxicity index was found in tebuconazole, hexaconazole, propiconazole, difenoconazole and pyraclostrobin (300) which was followed by azoxystrobin (168.94) and eight commonly used ready mix fungicides were evaluated against *S. rolfsii* at different concentrations *viz*., 250, 500 and 1000 ppm using poisoned food technique. The perusal of data presented in Table 3 revealed that cent per cent mean growth inhibition of *S. rolfsii* was recorded in five ready mix fungicides such as, azoxystrobin 11 % + tebuconazole 18.30 % SC, carboxin 37.5 % + thiram 37.5 % WS, tebuconazole 50 % + trifloxystrobin 25 % WG, carbendazim 12 % + mancozeb 63 % WP and fluxapyroxad 250 g/l + pyraclostrobin 250 g/l SC which was followed by metiram 55 % + pyraclostrobin 5 % WG (98.32 %). Tebuconazole 10 % + sulphur 65 % WG and mancozeb 40 % + azoxystrobin 7 % WG showed 96.39 and 88.15 per cent mycelia inhibition growth, respectively.

Consonant denouement with Torray*et al*. (2007), who recorded cent per cent growth inhibition of *S. rolfsii* by captan, thiram, mancozeb and propineb at 1000, 1500, 2000 ppm. The effectiveness of mancozeb and thiram against *S. rolfsii*has been recorded by Rangarani*et al*. (2017) and Archana *et al*. (2018). While Mahato*et al*. (2014)**,** Rakholiya (2015) and Archana *et al*. (2018) also observed that copper oxychloride has negligible effect on reduction of mycelial growth of *S. rolfsii*. Similar observations were also obtained by Vineela*et al*. (2017) revealed that triazole group fungicides such as tebuconazole 25.9 % SL, hexaconazole 5 % EC, difenoconazole 10 % WP, propiconazole 25 % EC, showed 100 % inhibition against *S.rolfsii*. The effectiveness of hexaconazole and propiconazole against *S. rolfsii* has beenreported by Rakholiya (2015) and Gopikaand Jagadeeshwar (2017). Rakholiya (2015), Rangarani*et al*. (2017) and Archana *et al*. (2018) found carbendazim and thiophanate methyl were least effective on growth inhibition of *S. rolfsii* at 500 ppm.

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(A Multidisciplinary Peer –Reviewed Refereed Journal)
www.agriwaysjournal.com Vol 10 Issue 02 July-Dec 2022

The present results congruent with those obtained by Prasad *et al.* (2017), who tested tebuconazole + trifloxystrobin and metiram + pyraclostrobin were recorded cent per cent growth inhibition under *in vitro* condition. Madhavi and Bhattiprolu (2011), Kumar *et al*. (2014), Rakholiya (2015) and Archana *et al.* (2018)recorded maximum inhibition of *S. rolfsii* with carbendazim 12 % + mancozeb 63 % WP. Maximum inhibition of mycelial growth of *S. rolfsii* with carboxin 37.5 % + thiram 37.5 % WP was also observed byAkgul*et al*. (2011), Das *et al.* (2014) and Mahato*et al*. (2014).

Conclusion:

The different fungicides were tested against *S. rolfsiiviz.*, non-systemic, systemic and ready-mix fungicides. Non-systemic fungicides tested @ 1000, 1500 and 2000 ppm, systemic fungicides @ 100, 250 and 500 ppm, while ready mix fungicides tested @ 250, 500 and 1000 ppm. Among them, non-systemic fungicides (mancozeb 75 % WP, thiram 75 % WS, chlorothalonil 75 % WP and propineb 70 %WP) systemic fungicides (tebuconazole 25.9 % EC, hexaconazole 5 % EC, propiconazole 25 % EC, difenoconazole 25 % EC and pyraclostrobin 20 % WG) and ready mix fungicides (azoxystrobin 11 % + tebuconazole 18.30 % SC, carboxin 37.5 % + thiram 37.5 % WS, tebuconazole 50 % + trifloxystobin 25 % WG, carbendazim 12 % + mancozeb 63 % WP and fluxapyroxad 250 g/l + pyraclostrobin 250 g/l SC) were found the bestwith cent per cent mean inhibition of mycelial growth of *S. rolfsii* under *in vitro* condition.

Acknowledgement:

I am thankful to Department of Plant pathology, College of Agriculture, JAU, Junagadh and authority of Junagadh Agricultural University for funding and providing facility to conduct the research work.

Table 1Effect of non-systemic fungicides on mycelial growth inhibition of*S. rolfsii*under *invitro* condition

Sr. No.	Treatments	Growth inhibition (%)			Mean inhibition (%)	Toxicity index
		1000 ppm	1500 ppm	2000 Ppm		
1.	Copper oxychloride 50 % WP	0.00 (0.00) *	24.09 (16.66)	30.35 (25.53)	18.15 (14.06)	42.19
2.	Mancozeb 75 % WP	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
3.	Thiram 75 % WS	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
4.	Captan 75 % WP	21.85 (13.85)	31.81 (27.78)	41.81 (44.44)	31.82 (28.69)	86.07
5.	Chlorothalonil 75 % WP	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
6.	Propineb 70 % WP	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
7.	Copper hydroxide 53.8	13.50	24.06	90.00	42.52	122.08

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Vol 10 Issue 02 July-Dec 2022

	% WP	(5.45)	(16.63)	(100)	(40.69)	
8.	Wettable sulphur 80 % WP	35.26 (33.32)	40.18 (41.63)	48.19 (55.56)	41.21 (43.50)	130.50
	Mean	53.83 (56.58)	60.02 (62.84)	71.29 (78.19)	61.71 (60.78)	-
		Fungicide (F)		**Concentration (C)**		**F x C**
	S.Em. ±	0.27		0.16		0.46
	C.D. at 5%	0.76		0.47		1.32
	C.V.%	1.30				

*Data outside the parentheses are arcsine transformed, whereas inside are re-transformed values

Table 2Effect of different systemic fungicides on mycelial growth inhibition of *S.rolfsii*under*in vitro* condition

Sr. No.	Treatments	Growth inhibition (%)			Mean inhibition (%)	Toxicity index
		100 ppm	250 ppm	500 ppm		
1.	Carbendazim 50 % WP	0.00 (0.00)*	5.91 (1.06)	19.40 (11.04)	8.44 (4.03)	12.10
2.	Tebuconazole 25.9 % EC	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
3.	Hexaconazole 5 % EC	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
4.	Azoxystrobin 23 % SC	45.32 (50.56)	45.64 (51.12)	55.10 (67.27)	48.69 (56.31)	168.94
5.	Thiophanate methyl 70 % WP	0.00 (0.00)	0.00 (0.00)	17.30 (8.84)	6.79 (2.95)	8.84
6.	Propiconazole 25 % EC	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
7.	Difenconazole 25 % EC	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
8.	Pyraclostrobin 20 % WG	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
	Mean	61.91 (68.82)	62.69 (69.02)	67.73 (73.39)	64.11 (70.41)	-
		Fungicide (F)		**Concentration (C)**		**F x C**
	S.Em. ±	0.35		0.21		0.61
	C.D. at 5%	1.00		0.61		1.73
	C.V.%	1.64				

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(A Multidisciplinary Peer –Reviewed Refereed Journal)
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Vol 10 Issue 02 July-Dec 2022

Table 3 Effect of different ready-mix fungicides on mycelial growth inhibition of *S. rolfsii* under *in vitro* condition

Sr. No.	Treatments	Growth inhibition (%)			Mean inhibition (%)	Toxicity index
		250 ppm	500 ppm	1000 ppm		
1.	Azoxystrobin 11 % + Tebuconazole 18.30 % SC	90.00 (100)*	90.00 (100)	90.00 (100)	90.00 (100)	300.00
2.	Tebuconazole 10 % + Sulphur 65 % WG	70.78 (89.16)	90.00 (100)	90.00 (100)	83.59 (96.39)	289.16
3.	Carboxin 37.5 % + Thiram 37.5 % WS	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
4.	Metiram 55 % + Pyraclostrobin 5 % WG	77.02 (94.95)	90.00 (100)	90.00 (100)	85.67 (98.32)	294.95
5.	Tebuconazole 50 % + Trifloxystrobin 25 % WG	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
6.	Carbendazim 12 % + Mancozeb 63 % WP	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
7.	Fluxapyroxad 250 g/l + Pyraclostrobin 250 g/l SC	90.00 (100)	90.00 (100)	90.00 (100)	90.00 (100)	300.00
8.	Mancozeb 40 % + Azoxystrobin 7 % WG	53.40 (64.46)	90.00 (100)	90.00 (100)	77.80 (88.15)	264.46
	Mean	81.40 (93.35)	90.00 (100)	90.00 (100)	87.13 (97.85)	-
		Fungicide (F)		**Concentration (C)**		**F x C**
	S.Em. ±	0.30		0.18		0.52
	C.D. at 5%	0.85		0.52		1.48
	C.V.%	1.03				

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