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In vitro and field evaluation of novel fungicides against early blight disease of tomato (*Alternaria solani*)

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Early blight disease caused by *Alternaria solani* is one of the major diseases of tomato in Odisha. This disease has become a major constraint in getting potential yield from the tomato growing areas. Field experiments were carried out in the research field of Regional Research and Technology Transfer Station (RRTTS), Chiplima, Sambalpur, Odisha during two consecutive rabi seasons of 2022-23 and 2023-24. The study was carried out to find out the most effective novel fungicide against the disease. Three sprayings were given at 15 days interval, starting from the initiation of the disease. Under *in vitro* condition, the fungicides were tested following poisoned food technique. The result showed that, spraying of metiram 55% + pyraclostrobin 5% WG @ 3.0 g/lwas most effective to control early blight disease (77.8 % reduction over control) without any phytotoxic symptom and produced highest fruit yield (255.84 q/ha). Under in vitro condition also, (metiram + pyraclostrobin) gave 84.9% inhibition of radial growth even at 100 ppm and found to be the best of all the seven fungicide tested resulting 100% inhibition of radial growth at 250 and 500 ppm.

Keywords : Early blight, fungicide, management, tomato

INTRODUCTION

Tomato (*Solanum lycopersicum* L.), originated in western South America and Central America, is one of the most popular and widely consumed vegetable crops all over the world. Tomato is a good source of antioxidants (Borguini and Torres, 2009). It is the major dietary source of the antioxidant lycopene, which has been linked to many health benefits, including reduced risk of heart disease and cancer. A small (100-gram) ripe tomato fruit contains 1.9g protein, 3.6g carbohydrate, 45.8mg sodium, 20.0 mg calcium, 15.0 magnesium and 31.0 mg vitamin C (Singh, 2005).

In India, tomato is one of the major horticultural crops, grown on 8.5 lakh hectares producing 208.19 lakh tons whereas in Odisha 14.34 lakh tons from 1.07 lakh hectare area was produced during the year 2023-24 (www.agricoop.nic.in). Tomato is subjected to be attacked by various fungal, bacterial and viral diseases among which early blight caused by *Alternaria solani* (Ellis and Martin) Sorauer is one of the most destructive fungal diseases that may cause significant reduction in quantity and quality of fruit yield (Tewari and Vishunavat, 2012). The causal organism of this disease is air borne and soil inhabiting and it is considered as an important disease of tropical and subtropical areas. The disease appears on leaves, stems, petiole, twig and fruits under favourable conditions resulting in defoliation, drying off of twigs and premature fruit drop and causing serious damage in all stages of plant which ultimately reduce the yield (Abada *et al*. 2008). Yield losses due to this disease have been reported to be 15 to 100 % (Panthee and Chen, 2010; Kaur *et al*. 2016). In absence of resistant cultivars, fungicidal treatments can offer viable solution for disease management. Many new fungicides are available in the market against the disease. Therefore, evaluation of effective fungicides from time to time is essential for new recommendations for the farmers community to combat the disease. Hence, evaluation of effective fungicides must be

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a continuous process. The present study was carried out to know the efficacy of novel fungicides against early blight of tomato under both in vitro and field conditions.

MATERIALS AND METHODS

Field evaluation of novel fungicides against early blight of tomato

Field experiments were conducted during rabi season of 2022-23 and 2023-24 at Regional Research and Technology Transfer Station, Chiplima, Sambalpur, Odisha. A list of different fungicides used in the experiment along with their active ingredients, formulation and dose are presented in Table 1. The experiment was laid in RBD with 3 replications. Each plot measured 15 m^2 with a spacing of 75 × 60 cm with bunds all around the plots. Replications were separated with a gap of 1 meter for irrigation channels. The susceptible tomato variety Pusa rubi was planted with recommended package of practices except plant protection measures. Recommended fertilizer dose was applied in all the plots and standard agronomic practices were followed as and when necessary to raise the crop. Three foliar sprays of fungicides were given at 15 days interval. First spray was given just after initiation of disease symptoms in the experimental plots. Only water was sprayed for the untreated check. The data on disease severity was recorded 15 days after the last spray. From each treatment, 10 plants were selected randomly and the disease severity on foliage was scored based on 0-5 scales (0= no symptom on the leaf, 1=0-5% leaf area infected, 2=6-20% area infected, 3=21-40% area infected, 4=41-70% area infected, 5= >71% area infected) (Mayee and Datar, 1986). The Percent Disease Index was calculated by using the following formulae given byMckinny (1923)

$$PDI = \frac{\text{Sumof al the numerical rating}}{\text{Number of observation x Max rating}} x100$$

The fruit yield was recorded on plot basis and converted to kg/ha.

The data obtained were subjected to statistical analysis and were tested at 5% level of significance to interpret the treatment differences following Gomez and Gomez (1984).

In vitro evaluation of fungicides

Seven fungicide products were tested at different concentrations under *in vitro* condition against the early blight pathogen, *Alternaria solani* using poisoned food technique (Vincent, 1969). The culture of *Alternaria solani* was isolated from the leaf of diseased tomato plants and incubated at 28$\pm$2^{0}C. The culture was maintained on Potato Dextrose Agar (PDA) medium. Stock solution of each fungicide formulation was prepared by mixing required quantity of fungicides in sterilized distilled water. Required amount of stock solution was poured in sterilized melted PDA to get a series of concentration of 100, 250 and 500 ppm of fungicides in the medium. After thorough mixing of fungicides, the poisoned PDA medium of each flask was poured in three sterilized Petri plates of 90 mm diameter @ 20 ml/plate. The Petriplates poured with PDA having no fungicide served as control. After solidification, the Petriplates were inoculated aseptically with 5 mm mycelia disc taken from actively growing colonies of seven days old culture of *Alternaria solani* at the centre on agar surface with their mycelia side down and incubated at 28$\pm$2^{0}C. The experiment was laid in complete randomized design with three replications for each test concentration. After 72 h of incubation, when control plates were full with fungal growth, the radial colony growth was measured in each plate and growth inhibition of the test pathogen over control was calculated by using the standard formulae :

Percent Growth Inhibition (PGI) = $\frac{C- T}{C}$ X 100

Where C = Radial growth in check plate and T = Radial growth in treated plate.

RESULTS AND DISCUSSION

Field evaluation

All the treatments significantly reduced the disease as compared to the unsprayed control (Table2). Severity of early blight disease ranged from 12.67 to 60.67% in 2022-23 and 14.66 to 62.67% in 2023-24. It is evident that all the treatments significantly reduced disease severity

Table 1: Fungicides evaluated against early blight disease of tomato

Treatment	Active ingredients	Formulation type	Dose	
			Formulation	g ai/ha
T1	Metiram 55% + Pyraclostrobin 5%	WG	3.0 g L^{-1}	900-1050
T2	Thifluzamide 24%	SC	1.0 ml L^{-1}	120
T3	Captan70%+ Hexaconazole 5%	WP	2.0 g L^{-1}	375-750
T4	Fluxapyroxad 250g/l + Pyraclostrobin 250g/l	SC	0.4 ml L^{-1}	100 -125
T5	Azoxystrobin 11 % + Tebuconazole18.3%	WP	1.5 ml L^{-1}	82.5 + 137.25
T6	Tebuconazole 50% + Trifloxystrobin 25%	WG	0.4 g L^{-1}	100 + 50
T7	Mancozeb 75% WP	WP	2.5 g L^{-1}	1.125-1.5
T8	Control			

as compared to the untreated check in both the years of experiment (Table2). The reductions in disease severity ranged between 41.6 and 77.8%. However, treatment T1 (metiram + pyraclostrobin) proved to be the most effective one which provided 77.8% control of the disease with pooled PDI of 13.67 (12.67 and 14.66 in 2022-23 and 2023-24, respectively). The next best treatment was T4 (fluxapyroxad + pyraclostrobin) with PDI of 17.33. Other treatments also recorded significantly lower early blight disease severity compared to untreated control (Table2).

All the fungicidal treatments were significantly different from the check plot with respect to fruit yield (Table2).Highest fruit yield was obtained in T1 (metiram + pyraclostrobin) treatment plot in both the years recording 270.0 and 241.67 q/ha during 2022-23 and 2023-24 respectively. The pooled data on the effects of fungicides on tomato fruit yield revealed that T1 (metiram + pyraclostrobin) was the best among all the treatments. The treatment provided highest fruit yield of 255.84 q/ha and highest increase (53.4%) in fruit yield over control. Among different fungicidal treatments, T1 (metiram + pyraclostrobin) was most economical among all the fungicidal combinations. It was followed by T4 (fluxapyroxad + pyraclostrobin) recording the fruit yield of 250.56 q /ha (Table 2).

The results of present experiment are in conformity with the findings of many workers who also found that combination treatments are effective in disease control. Best early blight disease control with highest yield and fruit quality of tomato was reported where pyraclostrobin with metiram was used (Tofoli *et al*. 2003). Capriotti *et al*. (2005) reported the use of cabriotop-a combination product of pyraclostrobin + metiram for both early blight and late blight of tomato.

No phytotoxicity symptoms like necrosis, epinasty, hyponasty, chlorosis, leaf tip injury, stunting or wilting was recorded within seven days of each spray of the combination fungicides tested during both the years of experiment and

Table 2 : Effect of different fungicides on early blight disease severity and yield in tomato under field condition (rabi, 2022-23 and 2023-24)

Treatment	Disease incidence % (PDI)			Disease control (%)	Fruit yield (q ha^{-1})			% Yield increase
	2022-23	2023-24	Pooled		2022-23	2023-24	Pooled	
T1	12.67	14.66	13.67	77.8	270.0	241.67	255.84	53.4
	*(20.75)	*(22.44)	(21.61)					
T2	36.67	35.33	36.0	41.6	207.78	195.0	201.39	20.7
	(37.23)	(36.44)	(36.85)					
T3	24.0	26.67	25.33	58.9	245.56	223.33	234.45	40.5
	(29.31)	(31.06)	(30.20)					
T4	18.0	16.67	17.33	71.9	264.44	236.67	250.56	50.2
	(25.07)	(24.03)	(24.59)					
T5	30.0	32.67	31.33	49.2	231.11	210.83	220.98	32.5
	(33.19)	(34.83)	(34.02)					
T6	22.67	20.67	21.67	64.9	250.0	224.17	237.09	42.1
	(28.39)	(26.99)	(27.69)					
T7	28.67	30.67	29.67	48.6	224.44	201.67	213.06	27.7
	(32.34)	(33.60)	(32.98)					
T8 (Control)	60.67	62.67	61.67	51.9	174.45	159.17	166.81	-
	(51.16)	(52.35)	(51.73)					
CD (0.05)	3.6	4.01	2.50	-	35.06	28.49	26.40	-
SEm(±)	1.18	1.31	0.82	-	11.45	9.30	8.62	-

*Figures in the parenthesis are angular transformed values

hence the fungicides may be considered as safe for use in tomato crop.

In vitro evaluation of fungicides against *Alternaria solani*

All the seven fungicides exhibited varying level of efficacy against *Alternaria solani* for growth inhibition (Table3). The treatments T1 (metiram + pyraclostrobin) and T4 (tebuconazole + trifloxystrobin) showed high level of efficacy against *Alternaria solani*. The result indicates that T1 (metiram + pyraclostrobin) was highly inhibitory (100% inhibition) to *Alternaria solani* at 250 ppm concentration whereas in case of T4 (fluxapyroxad + pyraclostrobin) complete inhibition (100%) was recorded at 500 ppm. In case of other fungicidal combinations complete inhibition was not recorded at any of the concentrations but all of them controlled more than 80% mycelial growth of the early blight pathogen at 500 ppm (Table3).It is clear from the data that T1 (metiram + pyraclostrobin) and T4 (fluxapyroxad + pyraclostrobin) caused 100% inhibition of mycelial growth at 500 ppm and were most effective and statistically superior to others. Rao *et al*. (2015) found that combination product of metiram + pyraclostrobin showed 75% inhibition against *Alternaria porri* pathogen at 2500 ppm concentration *in vitro.* Treatments T6 (tebuconazole + trifloxystrobin) and T3 (captan + hexaconazole) were also found to be very effective against *Alternaria solani*and inhibiting the test pathogen at 500 ppm resulting 93.3 and 90.7% inhibition respectively. But T1 (metiram + pyraclostrobin) gave almost 84.9% inhibition of radial growth even at 100 ppm and proved to be the best amongst the seven fungicides tested.

Table 3 : Effect of fungicides on growth of *Alternaria solani* under in vitro condition

Treatment	Mean radial growth (mm)*			Growth Inhibition (%)		
	100 ppm	250 ppm	500 ppm	100 ppm	250 ppm	500 ppm
T1	6.8	0.0	0.0	84.9	100.0	100.0
				(67.1)**	(90.0)	(90.0)
T2	16.6	12.5	8.0	63.1	72.2	82.2
				(52.6)	(58.2)	(65.0)
T3	12.0	8.8	4.2	73.3	80.4	90.7
				(58.9)	(63.7)	(72.2)
T4	8.5	3.4	0.0	81.1	92.4	100.0
				(64.2)	(74.0)	(90.0)
T5	14.5	10.8	6.4	67.8	76.0	85.8
				(55.4)	(60.7)	(67.8)
T6	10.4	5.2	3.0	76.9	88.4	93.3
				(61.3)	(70.1)	(75.0)
T7	17.6	14.0	8.5	60.9	68.9	81.1
				(51.3)	(56.1)	(64.2)
T8 (Control)	45.0	45.0	45.0	0.0	0.0	0.0
SEm(±)	0.42	0.34	0.24	0.63	0.58	0.48
CD (0.05)	1.26	1.04	0.72	1.90	1.76	1.45

*Average of three replications.
**Figures in the parenthesis are angular transformed values.

So, considering the overall efficacy of the fungicides, it may be concluded that three foliar sprays of metiram 55% + pyraclostrobin 5% WG @ 3 g/l at 15 days interval starting from the initiation of the disease is highly effective for management of early blight disease of tomato and obtaining good yield of tomato crop.

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