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Evaluation of chemicals for the management of lentil wilt, caused by *Fusarium oxysporum* f.sp. *lentis*

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Abstract
The present investigation is on the evaluation of chemicals to see their effect on the fungal pathogen *Fusarium oxysporum* f.sp. *lentis*, causing wilt disease of lentil crop. Fungal pathogen was evaluated in the laboratory by employing the Poisson Food Technology along with seed treatment to the lentil seeds was given for evaluation of the chemicals against the pathogen under *in vivo* conditions. Percent inhibition in the radial growth of mycelia of pathogen in the petri dish was used for evaluating the potential of chemicals in laboratory to manage pathogen growth and reduction in the disease incidence was recorded to see the effect of chemicals under field conditions. The eight chemicals namely Azoxystrobin, Propiconazole, Provax, Captaf, Carbendazim, Thiram, Raxil and Vitavax Power, were used against the pathogen. Among all the chemicals evaluated, Raxil was found most effective giving 100 percent mycelium inhibition in laboratory at 500ppm followed by 88.75 percent, Captaf (75%), Vitavax power (71%), Azoxystrobin (69.5%), Carbendazim (56.66%), Thiram (53.75%), Provax (37.5%) and control where no inhibiton of mycelium was observed. The seed treatment with the chemicals showed minimum disease incidence in case of Raxil treatment i.e.0.97 percent in the year 2016 and 0.87 percent in the year 2017, and the maximum disease incidence was recorded in case of seed control (9.23%). The present study concluded that there are several new chemicals those have the potential to manage the disease even in a lower concentrations.

Keywords: *Fusarium oxysporum* f.sp. *lentis*, chemicals, disease incidence, bioassay

Introduction
Lentil is one of the most important pulse crops after chickpea. It is rich in digestive proteins and in some other amino acids. This pulse crop is affected by wilt disease causing pathogen *Fusarium oxysporum* f.sp. *lentis***.** This disease is prevalent in many lentil growing areas of world and in India it occurs in UP, MP and in some other lentil growing states. The prominent symptom of the disease is yellowing, drying and wilting of the plant, thereby reducing the yield of the crop upto 75 percent in many areas. The fungus is soil borne in nature and commonly found in all crop growing areas in world. The fungus belongs to ascomycetes and cause wilt disease of several important crops. Fungus produces microconidia, macroconidia and chlamydospores which persist in soil for several years. It was also reported by Erskine *et al.*, (1990) [3] that the pathogen is seed borne in nature too and transmit from one place to another through seed material. Management of the pathogen is done through several biological, cultural and chemical practices. Several systemic and nonsystemic fungicides have been tested against the pathogen which showed variability in the results. The systemic fungicides were found superior for the management of pathogen by the several scientists (Rafique *et al*., 2016) [9]. Use of synthetic chemicals in the management of the disease plays an important role. Chemicals can protect the plant from the infection of microbes and can eradicate the pathogen from filed. Gullino *et al.*, (2002) [4] showed the higher efficacy of azoxystrobin against the Fusarium wilt of carnetion, cyclamen and paris daisy. Singh and Jha (2003) [10] used Bavistin (carbendazim), Blitox [copper oxychloride], Indofil M-45 [mancozeb+thiophanate-methyl], thiram, Captaf [captan], Kitazin [iprobenfos], and Ridomil MZ [mancozeb+metalaxyl] against chickpea wilt and found all effective against the pathogen. Somu *et al.*, (2014) [11] observed the total mycelia growth inhibition of *Fusarium oxysporum* f.sp. *cubense* at 500, 1000 and 2000 ppm with carbendazim, carboxin, propiconazole and benomyl. Mohamed *et al.*, (2016) [8] managed chickpea wilt through the use of botanicals and chemicals and found thiram as the most effective chemical for the management of pathogen. Kumar and Mane (2017) [7] in their study on *Fusarium oxysporum* f.sp. *ciceri* found Fosetyl AL @0.2per cent, Carbendazim @ 0.1 per cent, Thiram+Carbendazim @0.3 per cent and Fosetyl AL+Carbendazim @0.3 per cent most effective with maximum 100 per cent inhibition

Of mycelial growth. Management of cumin wilt pathogen was done by Khalequzzaman *et al.,* (2016) [6] along with an increased yield with the treatment of bavistin followed by provax. Jat *et al.,* (2017) [5] found Bavistin gave hundred percent inhibition of mycelia growth of *Fusarium oxysporum* f.sp. *corianderii* followed by Companion, Topsin–M and Vitavax. The present investigation reveals the best chemical to be used for the management of wilt disease through seed treatment.

Material and Method

Isolation of pathogen

Lentil wilt pathogen *Fusarium oxysporum* f.sp. *lentis*, was isolated from the infected plant parts taken from the field. The roots of the lentil plant are most susceptible area of the plant which is preferably affected by the wilt pathogen. The small pieces of the infected plant part was cut with the help of knife and then surface sterilized with Sodium Hypochloride @1 percent for 60seconds, subsequently washed with distilled water for three times to eliminate extra chemical retained on the surface of the infected plant tissue. The sterilized plant tissue was then dried up by placing it on the sterilized blotter sheet for 1-2 minutes. The dried infected plant tissue was then transferred to the potato dextrose agar medium poured petriplate and placed in BOD incubator at 25±2 °C temperature for 8 days for growth of the fungus.

Food poison Technique

Eight chemicals namely Azoxystrobin, Propiconazole, Provax, Captaf, Carbendazim, Thiram, Raxil and Vitavax Power were tested against the pathogen by employing Food poison technique. The chemicals were incorporate in the 15ml medium before pouring it into petriplate separately, and a disc of 5mm of seven days old culture of fungus was transferred to the each petriplate and kept in BOD incubator for 8 days for the full growth of fungus as observed in case of control where no chemicals were applied in medium. The inhibition in the radial growth of pathogen mycelium was recorded and percent inhibition in the mycelium of pathogen was recorded by comparing the mycelail growth of fungus in the treated plates with control. Per cent growth inhibition was calculated by using the formula given by Vincent (1947).

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

Where,
PI = Inhibition percentage
C = Colony diameter in check plate (mm)
T = Colony diameter in treatments (mm)

The different concentrations of the chemicals taken for the laboratory experiment were 1ppm, 10ppm, 25ppm, 50ppm, 100ppm, 200ppm, 350ppm, 500ppm and 800ppm.

Field Experiment

Field treatment was done for 2 years in 2016-17 and 2017-18 at NEBCRC, GBPUAT, Pantnagar, US Nagar, Uttarakhand. Eight chemicals viz; Azoxystrobin (1ml/litre), Propiconazole (1ml/litre), Provax (1ml/litre), Captaf (2g/kg seed), Carbendazim (1g/kg seed), Thiram (2g/kg seed), Raxil (1g/kg seed), and Vitavax Power (1g/kg seed), was used for the seed treatment. Seeds were treated with chemicals before sowing and dried porperly. Line sowing of the seeds was done 3 lines per plot and plot size was 1.5mX4m for the experiment. The seeds germination was counted 25Days After Sowing along with infected plants with the wilt disease. The disease incidence was calculated using following formula.

$$\text{Disease Incidence} = \frac{\text{No. of infected plants}}{\text{Total no. of plants assessed}} \times 100$$

Some growth parameters were also recorded to test the effect of chemicals on the crop height, root length, no. of branches and finally the yield of crop.

Results and Discussion

Present study on the management of lentil wilt caused by *Fusarium oxysporum* f.sp. *lentis* revealed that the systemic fungicides have great impact on the fungal plant pathogens they can manage the pathogen very effectively as comparison to non systemic fungicides. In laboratory experiments as well as in field experiment systemic fungicides perform better. In this study presented in table1 showed that aomg the chemicals evaluated Raxil gave the maximum radial growth inhibition of fungal pathogen i.e 53.12 percent at 1ppm concentration, 70.62 percent at 10ppm concentration, 72.5 percent at 50ppm concentration, 73.75 percent at 100ppm concentration, 80.41 percent at 200ppm, 93.33 per cent inhibition at 350ppm concentration and 100 per cent inhibition at 500ppm concentration under *in vitro* conditions. Propiconazole also performed better at 500ppm and gave 88.75 percent mycelia growth inhibition followed by captaf (75%), Vitavax power (71%), Azoxystrobin (69.5%), Carbendazim (56.66%), Thiram (53.75%), Provax (37.5%) and control where no inhibition was recorded. At 800 ppm concentration of chemicals maximum radial growth inhibition of mycelium was recorded in the treatment with Raxil i.e.100 percent, followed by Carbendazim (99.66%), Propiconazol (99.16%), Provax (98.75%), Captaf (89.75%), Vitavax Power (83.25%), Thiram (79.95%) and control where no inhibition of mycelium growth was given.

The field studies shown in table 2 revealed that the seed treatment with chemicals have a significant effect on the reduction of disease incidence of lentil wilt and the minimum disease incidence i.e.0.97 and 0.87 percent for the years 2016 and 2017 respectively, was observed in case of seed treatment with the Raxil (1g/kg seed), followed by propiconazole 1.56 percent and 0.96 percent, vitavax power 1.69 percent and 1.17 percent, thiram 1.71 percent and 2.11percent, provax 2.39 percent and 2.39 percent, azoxystobin 2.65 percent and 2.77 percent, carbendazim 5.03 percent and 3.16 percent, captaf 6.55 percent and 4.79 percent and control 9.23 percent and 19.54 percent for the years 2016 and 2017, respectively. Chemicals showed significant positive impact on the management of the disease for both the years and found effective to control the disease with achieving good yield. Although the plant height, root length, number of the branches and the total plant vigour of the plant has been increased as compare to control but there was no significant effect was observed between the treatments upon these parameters of lentil plant. Difference in the yield of the crop per plot was observed among the treatment due to more crop stand as a result of less disease incidence. The maximum yield was observed in the seed treatment with Raxil i.e 933.33 kg/ha and 646.66 kg/ha followed by provax 834.8kg/ha and 633.33 kg/ ha, propiconazole 784.44 kg/ha and 577.77kg/ha, Carbendazim 760 kg/ ha and 231.11 kg/ ha, Vitavax Power 722.22 kg/ ha and 576.28 kg/ ha, Captaf 669.62 kg/ ha and 657.77 kg/ ha, Thiram 577.77 kg/ ha and 354.066 kg/ ha and

control 366.66 kg/ ha and 222.22 kg/ ha for the year 2016 and 2017, respectively. The significant impact of the treatments on the crop yield was observed in the present investigation.

Table 1: Percent fungal mycelium radial growth inhibition at different concentrations of chemicals used for evaluation

Treatments	Percent inhibition in mycelia growth of fungus								
	1ppm	10 ppm	25ppm	50PPM	100ppm	200ppm	350ppm	500ppm	800PPM
CARBENDAZIM	12.16	20.33	26.66	30.41	32.91	37.08	52.91	56.66	99.66
CAPTAF	0.41	33.33	42.5	44.58	60	63.75	72.5	75	89.75
RAXIL	53.12	70.62	72.25	72.5	73.75	80.41	93.33	100	100
AZOXYSTROBIN	2.08	6.25	17.7	48.75	56.25	61	65	69.5	62.58
VITAVAX POWER	12.5	27.5	37.5	48.75	58.75	60	65	71	83.25
THIRAM	25	27.91	31.66	32.08	42.5	53.12	53.75	53.75	79.95
PROPICONAOLE	45	65	66.04	66.04	67.5	77.5	78.75	88.75	99.166
PROVAX	0.41	21.87	30	32.5	35	36.25	37.5	37.5	98.75
CONTROL	0	0	0	0	0	0	0	0	0
CD at 5%	LEVEL A=0.799, LEVEL B 0.789, AXB 2.39								
CV	3.07								

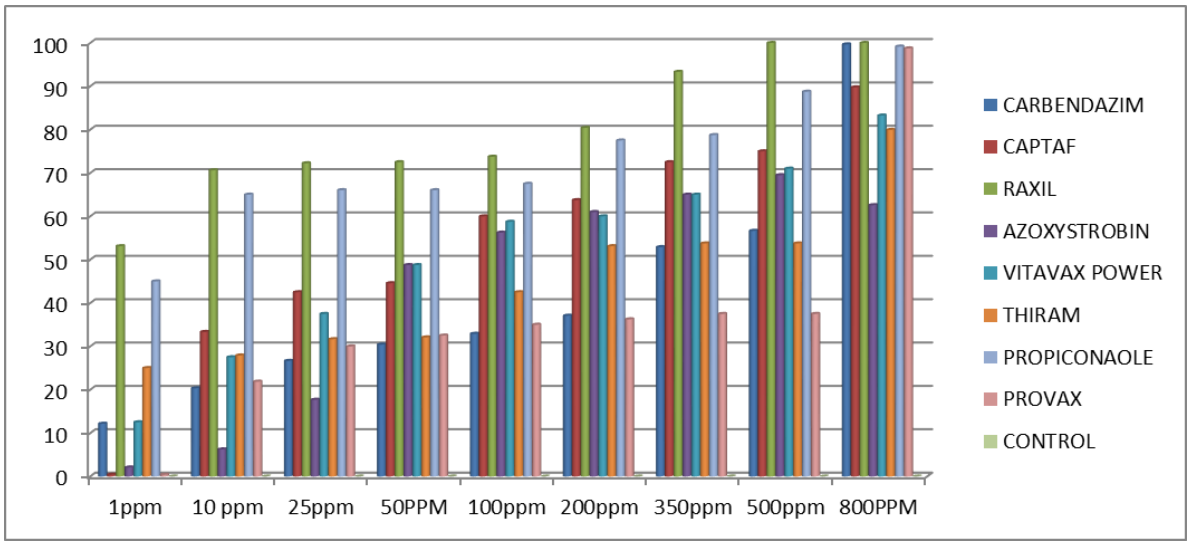

Fig 1: Percent fungal mycelium radial growth inhibition at different concentrations of chemicals used for evaluation

Table 2: Effect of chemicals on the growth parameters, yield and disease incidence of pathogen on lentil crop

Treatments	Crop stand		Mortality		Disease incidence		Pl. Ht.		Root Lt.		Branches		Yield (kg/ha)	
	2016	2017	2016	2017	2016	2017	2016	2017	2016	2017	2016	2017	2016	2017
AZOXYSTROBIN	378	498	10	13	2.65	2.77	32.83	22.66	9.166	9.16	8.5	3.83	395.556	297.02
PROPICONAZOLE	821.66	483.66	8	21	1.56	0.96	28.83	22.33	7.33	6.5	10	2.5	784.44	577.77
PROVAX	709	369.66	17	4	2.39	2.39	26.33	17.58	9.33	8.66	13.16	3.83	834.8	633.33
CAPTAF	776.66	375.66	83	6	6.55	4.79	32.66	18.83	8	10.16	13.66	2.83	669.62	657.77
CARBENDAZIM	592	515	29	16	5.03	3.16	33	24.41	7.83	7.66	7.33	2.83	760	231.11
THIRAM	600.33	490.67	10	10	1.71	2.11	28.91	24.08	8.16	8.66	13	3.16	577.77	354.06
RAXIL	748.67	44.33	12	4	0.97	0.87	27.33	22.83	7.5	8	10.33	2.5	933.33	646.66
VITAVAX POWER	453	356	7	3	1.69	0.86	29.75	19.75	8.33	9	17.83	2.33	722.22	576.28
CONTROL	341	126	8	25	9.23	19.54	20.33	13.66	6	4.76	8.33	1.33	366.67	222.22
CD at 5%	90.54	63.55	3	2.68	0.43	1.42	3.25	2.38	1.88	1.46	6.07	1.22	16.42	33.66
CV	8.68	9.007	8.73	13.98	8.06	290.95	6.77	6.95	14.24	10.93	32.19	26.37	10.88	23.15

Conclusion

The present study on the lentil wilt concluded that the management of the lentil Wilt disease causing pathogen *Fusarium oxysporum* f. sp. *lentis* can be achieved with the seed treatment. The results are in accordance with the Dastogeer, 2013 [2] who found seed treatment an important management tool for the seed and soil borne pathogens. Triazoles (Raxil and Propiconzole) fungicides are found superior over other as they have potential to manage the soil borne pathogen as they are demethylation inhibitors (DMIs) and inhibit the C-14 α-demethylation of 24-methylenedihydrolanosterol, a precursor of ergosterol in fungi (Brent, 1995) [1]. Triazole are used widely for the management of soil borne plant pathogens (Spolti *et al*., 2012) [12]. This study concludes that among the seed treatments with chemicals Raxil has more potential to control the fungal pathogen in laboratory as well as in field and could be a better option for the application in field for the management of the diseases.

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