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Evaluation of Different Fungicides and Bio-agents to Minimize the Collar Rot Disease of Groundnut Caused by *Aspergillus niger* Van Tiegham

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Abstract: Collar rot disease of groundnut caused by soil inhibiting fungus, *Aspergillus niger* Van Tiegham is a most prevalent and destructive nature of groundnut disease under stress conditions. Present investigation was carried out to minimize collar rot disease incidence through application of different bio-agents and fungicides to reduced crop yield losses. *Trichoderma viridae* and *T. harzianum* were most effective bio-control agents to inhibit the mycelia growth of *A. niger* under *invitro* conditions. Out of eight fungicides four fungicides *viz*., pyraclostrobin +epoxiconaxole, tebuconazole, carbendazim 12%+mancozeb 63% and azoxystrobin11.5% + mancozeb30 %, completely inhibit the mycelia growth of *A. niger* under *invitro* condition. Moreover, fungicides *viz.,* trifloxystrobin 25% + tebuconazole 50% (91.67%), thiram (82.19%) and azoxystrobin (68.09 %) also found effective to inhibited mycelia growth of *A.niger*. The experiment was conducted during *Kharif* -2018 to manage collar rot disease of groundnut due to *A. niger*. The present investigation revealed that minimum plant mortality (9.25%) was observed in seeds treated with azoxystrobin 23SC @ 1.5 ml/ kg followed by seeds treated with *T.harzianum* @10 g /kg seeds. Similarly maximum disease control (69.13%) was recorded in seeds treated with azoxystrobin 23SC followed by seeds treated with *T.harzianum*. While highest plant mortality (29.97%) was recorded in control treatment. In case of pods/ plant, maximum (25.13) was recorded in seeds treated with azoxystrobin 23SC followed by seeds treated with *T.harzianum* (24.27). Highest pod yield was recorded in seeds treated with azoxystrobin 23SC (1581 kg/ha.) at par with seeds treated with *T.harzianum* with 1542 kg/ha. Lowest pod yield (925 kg/ha) was recorded in control treatment. Highest haulm yield was recorded in seeds treated with *T.harzianum* which was at par with seeds treated with azoxystrobin 23SC. The present study indicated that groundnut seeds treatment with with azoxystrobin 23SC @ 1.5 ml/ kg seeds and seed treatment with *T.harzianum* @10 g /kg reduced the plant mortality due to collar rot disease and gave higher pod and haulm yield per hectare.

Keywords: Groundnut, collar rot, *Aspergillus niger,* bioagents, fungicides

1. INTRODUCTION

Groundnut (*Arachis hypogaea* L.) is an important oilseed and supplementary food crop of the world. India is the largest groundnut growing country in the world with an area of around 4.83 million ha., produce 9.95 million tones with 2063 kg/ha. productivity (Anon., 2021). India is the second largest producer accounting 14.50 percent of the world production while fifth position in productivity. Gujarat is the largest groundnut producer share 46.68 percent in India production. Gujarat, produce 4.65 million tones production from 1.69 million ha. with 2751 kg/ ha productivity (Anon., 2021). The collar rot of groundnut also known as "Kalijad" (in Hindi) caused by *Aspergillus niger* Van Tiegham is one of important seed and soil borne disease. This disease was first reported by Jochem (1926). However, Jain and Nema (1952) first reported the *Aspergillus* blight of groundnut caused

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by *A. niger* in India. It is an important disease in the major groundnut growing areas of the country. Therefore, in view of destructive nature of groundnut disease thus, present work was carried out to minimized collar rot disease incidence caused by *Aspergillus niger* through application of different bio-agents and fungicides and reduced crop yield losses.

2. MATERIAL AND METHODS

2.1. Evaluation of bio-agents against the *A.niger* pathogen under in vitro:

For this study, the pathogen was isolated from collar rot diseased groundnut plants from Regional Rice Research Station N.A.U., Vyara. *Aspergillus niger* pathogen was maintained on potato dextose agar Petri-plates at 27±1^{0}C and its used for the studies. Bio-agents *i.e., Trichoderma harzianum, T. viridae, Psuedomonas fluorescens* and *Bacillus subtilis* were brought from Department of Plant Pathology, N.M. College of Agriculture, NAU, Navsari and evaluated, antagonistic activity of these bio-agent against *A. niger* was determined by "dual culture technique" (Dennis and Webster, 1971). Five millimeter disk of pathogen was taken from the actively growing colonies of the test pathogen and antagonist. Only pathogen in Petri plate served as control. Each treatment was repeated four times with CRD design and incubated at 27±1^{0}C. Growth of antagonists and pathogen were recorded recorded after 5 days of incubation and per cent inhibition was worked out using the Vincent's (1927) formula.

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

Where,

I = Percent inhibition

C = Radial growth in control

T = Radial growth in treatment

2.2. Evaluation of fungicides against the *A.niger* pathogen under *in vitro*

Eight fungicides *viz.*, pyraclostrobin 133 g/ I+epoxiconaxole 50 g/I SE, thiophanate methyl 450 g/I+pyraclostrobin 509 g/I FS, azoxystrobin 23 SC, tebuconazole 2 DS, carbendazim 12%+mancozeb 63% 75WP, azoxystrobin11.5% + mancozeb 30%, trifloxystrobin 25% + tebuconazole 50% WG and thiram 75% WS were evaluated against *A. niger* under *invitro* conditions. All fungicides were evaluated at 500 ppm concentrations to screen out the best fungicides depending upon their inhibitory effect on the growth of the fungus. The different fungicides were screened for their efficacy against the pathogen by "Food Poison Techniques" described by *Schmitz* (1930) in which required quantity of each fungicide was thoroughly mixed with 100 ml well sterilized potato dextrose agar medium contained in 250 ml flasks. Each treatment was repeated three times under CRD design. One set of control was also kept in which the medium was not mixed with fungicides. 5 mm of active fungal growth, cut by the cork borer was inoculated in each petri-plate at the center and incubated at 27±1^{0}C for 5 days. Redial mycelial growth of fungus was recorded in each Petri-plates at 5 DAI. Per cent growth inhibition of pathogen was calculated by using the Vincent's (1927) formula.

2.3. Evaluation of different bio-agents and fungicides to manage collar rot disease of groundnut

The experiment was conducted during *kharif*-2018 to evaluated different bio-agents and fungicides against collar rot disease of groundnut under field conditions. Most effective bio-control agents and fungicides found under *invitro* condition were selected to test under field conditions. Treatment details as; T_1: Seeds treated with *T.harzianum* @10 g /kg, T_2: Seeds treated with *T.viridae*@10 g /kg, T_3: Soil application of *T.harzianum* @2.5 kg /ha, T_4: Soil application of *T.viride* @ 2.5kg/ha, T_5: Seeds treated with tebuconazole 2SD @3g/ kg , T_6: Seeds treated with carbendazim + mancozeb @ 3g/ kg , T_7: Seeds treated with azoxystrobin 23SC @ 1.5 ml/ kg T_8: Seeds treated with thiophanate methyl + pyroclostrobin @ 1.5ml/ kg seeds and T_9: Control with no treatments. Bio-control agents applied in soil mixed in 100 kg FYM 4 days before application. Groundnut variety GJG-9 used in present study All treatments were layout in RBD deign with three replications. Gross plot size was

5.0 x 2.25 m and net plot size was: 4.8 x 1.35 m, NPK: 12.5: 25: 0.0 fertilizer was applied as basal dose. Pathogen multiplied on sorghum grains and 10 days old culture added in soil before the sowing. Percent plant mortality, pods/ plant, pod yield and Haulum yield were recorded in treated as well untreated plots. Plant mortality was calculate by following formula

$$Plant\ mortility(\%) = \frac{Initial\ plant\ population - Final\ Plant\ population}{Initial\ plant\ population} \times 100$$

2.4. Statistical Analysis

All statistical analysis was performed by OPSTAT software developed by Sheoran (1998). All the data were statistical analysis by one way analysis of variance (ANOVA) if probability significant level ($P<0.05$) and the mean comparison of treatments was carried out by Duncan's Multiple Range Test using OPSTAT software.

3. RESULTS AND DISCUSSION

3.1. Evaluation of different bio-control agents against A.niger under in vitro conditions

The result on antagonistic activity of bio-control agents from table1 revealed that a significant variation among the treatments were recorded. Minimum mycelia growth was recorded in *T. viridae* with 27.20 mm it was at par with *T. harzianum* with 28.50 mm while maximum mecelial growth (75.00 mm) recorded in control treatment at 5 days after incubation. In case of mycelia growth inhibition, among the treatments, *T. viridae* showed maximum inhibition over the control by 63.72 per cent it was at par with *T. harzianum* (61.95 %). Whereas, least mycelial growth inhibition was recorded by *Bacillus subtilis* (24.96%).

The present results are more or less agreement with earlier workers who reported mycelia growth of *A.niger* inhibited by various bio-control agents (Kumari and Singh, 2017, Nathawat and Mahendra, 2014, Latha, 2013, Gajera *et al*., 2011, Devi and Prasad, 2009, Rao and Sitaramaih, 2001, Kishore *et al*,. 2001 and Dasgupta and Raj, 1998). The results found that fungal bio-control agents are fast colonizing in nature as compared to pathogen and also over grew on the pathogen. while, *A.niger* growth covered the *Bacillus subtilis* growth.

3.2. Evaluation of different fungicide against *A.niger* under *in vitro* conditions

The results about *in vitro* evaluation of eight different fungicides were recorded at 5 DAI and found that no fungus growth was observed in pyraclostrobin +epoxiconaxole, tebuconazole, carbendazim 12%+mancozeb 63% and azoxystrobin11.5% + mancozeb30 %. Very less mycelium redial growth was recorded in trifloxystrobin 25% + tebuconazole 50% thiram and azoxystrobin with 9.00, 12.83 and 23 mm, respectively. While, maximum (72.17 mm) mycelia redial growth was recorded in control treatment. In case of per cent inhibitory effect of fungicides on the growth of *A. niger* were found significantly mycelium growth inhibition over the control. The result presented in table 2 showed that among all the fungicides, pyraclostrobin +epoxiconaxole, tebuconazole, carbendazim 12%+mancozeb 63% and azoxystrobin 11.5% + mancozeb 30% were found most effective to inhibited cent per cent mycelial growth of *A.niger* under *in vitro* condition at 500 ppm concentration. While, trifloxystrobin 25% + tebuconazole 50% thiram and azoxystrobin were found effective with 91.67, 82.19 and 68.09 per cent mycelial growth inhibition, respectively. Thiophanate methyl +Pyraclostrobin fungicide was found least effective with 53.57 per cent mycelia growth inhibition over the control treatment. The present results is in agreements with earlier findings in efficacy among the fungicides reported by Vineela *et al.* (2018) found that thiophanate methyl (@ 0.15%) was inhibited cent per cent mycelial growth of fungus followed by tebuconazole, difenconazole, azoxystrobin with 60.60, 50.80 and 49.69 per cent, respectively. Rani *et al.* (2017) recorded cent per cent mycelial growth inhibition of radial growth of *A. niger* was obtained with tebuconazole followed by mancozeb+ carbandazim, thiram with 92.94, 83.92 per cent inhibition, respectively. Andge *et al.* (2017) *invitro* tested different fungicides carboxin+ thiram, carbendazim+thiram, carbendazim, thiram and carbendazim+ mancozeb against *A. niger* and found that cent

per cent radial growth inhibited at 250, 500, 1000 ppm concentration at 7 days after incubation. Kumari *et al*. (2016) observed mycelial inhibition was highest in carbandazim+ mancozeb (71.83%) followed by carbendazim (69.16%), carboxin+ thiram (65.80%) at 50, 150, and 250 ppm concentrations against *A. niger in vitro*. Nathawat and Partap (2014) observed cent percent mycelial growth inhibition of *A. niger* by tebuconazole and propiconazole at 100 ppm followed by carbendazim and difenconazole at 750 and 1000 ppm concentration, respectively. Nandeesha *et al.* (2013) observed efficacy of systemic fungicides *viz*., carbendazim, propiconazole, tebuconazole and hexaconazole and non-systemic fungicides *viz*., mancozeb and captan against *A. niger* at 250, 500, 1000 and 1500 ppm concentrations. All systemic fungicides were found highly effective and completely inhibited the mycelial growth of the *A. niger invitro* condition. Sharma *et al.* (2012) observed carbendazim and mancozeb completely inhibited the mycelium growth of *A. niger* at 500 and 1000 ppm and partially inhibited the growth at 100 and 250 ppm *in vitro*, respectively. Raju and Naik (2006) reported that pre-harvest spray of carbendazim (0.1%) reduced the 73.53 per cent disease incidence of black mold of onion caused by *A. niger* followed by carbendazim + mancozebwith 67.82 per cent disease reduction.

3.3. Evaluation of different bio-agents and fungicides to manage collar rot disease of groundnut

Experiment was conducted during *Kharif* -2018 and results presented in table 3 revealed that minimum plant mortality (9.25%) was observed in T_7 which was at par with T_1, T_5, and T_8 with 11.65, 15.14 and 15.19 per cent, respectively. While highest plant mortality (29.97%) was recorded in T_9 treatment. Maximum disease control (69.13%) was recorded in T_7 followed by T_1 treatment (61.13%). In case of pods/ plant maximum (25.13) was recorded in T_7 which was at par with T_1, T_6 and T_3 by 24.27, 23.40 and 22.73 pods per plant, respectively. Highest pod yield was recorded in treatment T_7 (1581 kg/ha.) at par with T_1 treatment with 1542 kg/ha. Similarly highest per cent pod yield was increase observed in T_7 treatment (69.09%) and T_1 treatment (64.92%) over the control. Lowest pod yield (925 kg/ha) was recorded in T_9 treatment. But highest haulum yield (3400 kg/ha.) was recorded in T_1 treatment which was at par with T_7 treatment (3394 kg/ha.). Highest haulum yield increase was observed in treatment T_1 (122.37%) followed by T_7 treatment (121.98%) over the control. Moreover T_7 treatment (seeds treated with azoxystrobin 23SC @1.5 ml/ kg) and T_1 treatment (seeds treated with *T.harzianum* @10 g /kg) was

Table 1: Evaluation of different bio-control agents against *Aspergillus niger* under *invitro* conditions

Treatment No.	*Name of bio-control agents*	*Mycelial growth (mm)*	*Mycelial growth inhibition (%)*
T_1	Trichoderma harzianum	28.50	61.95[a] (52.22)
T_2	Trichoderma viridae	27.20	63.72[a] (53.27)
T_3	Psuedomonasfluorescenas	42.25	43.51[b] (41.53)
T_4	Bacillus subtilis	56.25	24.96[c] (30.29)
T_5	Control	**75.00**	-
S.Em.±		1.22	1.07
C.D. at 5 %		3.71	3.25
C.V. %		5.36	6.08

() mean of four repetitions of Arcsine transformed value (X+0.5). X= Original value.
In each column, values with different letters show significant difference ($P \leq 0.05$) as determined by Duncan's Multiple Range Test.

Table 2: Evaluation of different fungicides agents against *Aspergillus niger* under *invitro* conditions

Treatment No.	*Name of fungicides*	*Mycelial growth (mm)*	*Mycelial growth inhibition (%)*
T_1	Pyraclostrobin 133 g/I+Epoxiconaxole 50 g/I	0.00	100.00[a] (90.00)
T_2	Thiophanate methyl 450 g/ I+Pyraclostrobin 509 g	33.50	53.57[e] (47.03)
T_3	Azoxystrobin 23 SC	23.00	68.09[d] (55.60)
T_4	Tebuconazole 2 DS	0.00	100.00[a] (90.00)
T_5	Carbendazim 12%+Mancozeb 63% 75WP	0.00	100.00[a] (90.00)
T_6	Azoxystrobin11.5% + Mancozeb30 %	0.00	100.00[a] (90.00)
T_7	Trifloxystrobin 25% + Tebuconazole 50% WG	6.00	91.67[b] (73.24)
T_8	Thiram 75% WS	12.83	82.19[c] (65.03)
T_9	Control	72.17	-
S.Em.±			0.48
C.D. at 5 %			1.43
C.V. %			1.24

() mean of four repetitions of Arcsine transformed value (X+0.5). X= Original value.
In each column, values with different letters show significant difference ($P \leq 0.05$) as determined by Duncan's Multiple Range Test.

Table 3: Effect of seed treatment on collar rot of groundnut on plant mortality and yield during *Kharif*- 2018

Treatments	*Mortality (%)*	*Plant mortality reduced over control*	*Pods/ plant*	*Pod yield Kg/ha.*	*Pod yield Increased over check*	*Haulum yield Kg/ha.*	*Haulum yield Increased over check*
T_1	11.65	61.12	24.27	1542	64.92	3400	122.37
T_2	17.11	42.92	21.73	1240	32.62	2507	63.96
T_3	22.97	23.35	22.73	1010	8.02	2009	31.39
T_4	26.52	11.52	21.00	1028	9.95	1914	25.18
T_5	15.14	49.49	20.53	1074	14.87	1689	10.46
T_6	16.17	46.06	23.40	1227	31.23	2335	52.71
T_7	9.25	69.13	25.13	1581	69.09	3394	121.98
T_8	15.19	49.31	21.07	1176	25.78	2505	63.83
T_9	29.97	-	18.20	935	-	1529	-
S.Em.±	2.29		0.95	140.00		310.00	
C.D. at 5 %	6.85		2.85	419.73		929.44	
C.V. %	21.73		7.46	20.19		14.53	

found best over the rest treatment to manage the collar rot disease of groundnut. These results were similar to the findings earlier reported by Kumari and Singh, 2017. He observed that *T. harzianum* + *P. fluorescens* was most effective to control collar rot disease (52.21%) in groundnut followed by *T. harzianum* (45.73%). *Trichoderma* spp. used as seed treatment + soil treatment shown effective reduction in groundnut collar rot disease incidence in pot conditions (Rao and Sitaramaih, 2001). Devi and Prasad (2009) reported that *T. viridae* was most effective under in pot condition. They also observed Combined effect of seed treatment with *T. viridae* and captan significantly reduced the collar rot disease of groundnut in field condition. Minimum disease incidence (4.77%) and maximum pod yield (1632 kg/ha) was recorded in seed treated

with carboxin + thiram (4 g/kg seed) followed by ipconazole and thiram against collar rot of groundnut in field condition (Rakholiya *et al*., 2012). Mohapatra and Beher (2012) recorded minimum collar rot incidence (4.66%) at 30 days after sowing in seeds treated with bavistin followed by saaf (5.33%), thiram and benlate (6.33%).

The present study revealed that seed treatment with with Azoxystrobin 23SC @ 1.5 ml/ kg seeds and seed treatment with *T.harzianum* @10 g /kg reduced the plant mortality due to collar rot disease and gave higher pod and haulum yield per ha.

4. SUMMERY AND CONCLUSION

From the present studies found that *T. viridae* and *T. harzianum* were most effective bio-control agents to inhibit the mycelia growth of *A. niger* under *invitro* conditions. Out of eight fungicides, pyraclostrobin +epoxiconaxole, tebuconazole, carbendazim 12%+mancozeb 63% and azoxystrobin 11.5% + mancozeb30 %, trifloxystrobin 25% + tebuconazole 50% thiram and azoxystrobin were found most effective fungicides to inhibit the mycelia growth of *A. niger* under *invitro* condition. Groundnut seeds treated with with Azoxystrobin 23SC @ 1.5 ml/ kg seeds and seeds treated with *T.harzianum* @10 g /kg reduced the plant mortality due to collar rot disease and gave higher pod and haulum yield per ha.

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