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Evaluation of Different Fungicides against Leaf Blight (*Alternaria triticina*) of Wheat under *in vitro* Condition

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A B S T R A C T

Considering the importance of leaf blight of wheat caused by *Alternaria triticina*, a *in vitro* study was conducted using different fungicides using poisoned food technique. In laboratory screening, non-systemic fungicides *viz.*, mancozeb 75 % WP and propineb 70 % WP were found most effective with 89.41 and 88.70 per cent fungal growth inhibition, respectively. While systemic fungicides *viz.*, propiconazole 25 % EC and hexaconazole 5 % EC proved most effective with 96.00 and 90.33 per cent fungal growth inhibition, respectively. But in combination fungicides *viz.*, azoxystrobin 11 % + tebuconazole 18.30 % WP, hexaconazole 4 % + zineb 68 % WP and carboxin 37.5 % + thiram 37.5 % SD were found most effective with cent per cent growth inhibition of fungus.

Introduction

Wheat (*Triticum aestivum* L.) is the world most widely cultivated food crop. In India wheat is second important staple food crop. Globally wheat has 224.98 million hectares area, 735.50 million metric tonnes production and 3.27 metric tonnes per hectare productivity and in India wheat has 30.78 million hectares area, 98.51 million metric tonnes production and 3.20 metric tonnes per hectare productivity during 2016-17 (Anon., 2018). In Gujarat wheat has 0.99 million hectares area, 2.73 million metric tonnes production and 2.75 metric tonnes per hectare productivity during 2016-17 (Anon., 2017). Soil with a clay loam or loam texture, good structure and moderate water holding capacity are ideal for wheat cultivation. The major wheat growing countries are United States, China, Morocco, Algeria, Italy, Syria, USSR, Iran, Argentina and India. Alternaria leaf spot of wheat caused by *Alternaria triticina* has attained importance in northeastern and north-western plain zone during recent years due to significant losses The loss of yield due to this

disease vary upto 29.4- 43.2 per cent beside reduction of 15.2- 30.5 percent in 1000 grain weight. The disease first appears as small, oval, discoloured lesion which are irregularly scattered on the leaves. These spots become irregular in shape with increase in size and appear brown to gray in colour. A bright yellow halo surrounds the spots. Several lesions coalesce to cover large areas and cause death of the entire leaf. In severe cases the leaf starts drying from the tip. Host range of the species is confined to wheat varieties only. Thus, the present investigation entitled ‘‘evaluation of different fungicides against leaf blight (*Alternaria triticina*) of wheat under *in vitro* condition’’ was proposed with the objective to evaluate the efficacy of fungicides on disease intensity.

Materials and Methods

Mycelial growth inhibition activities of different concentration of fungicides were tested against *Alternaria triticina* under *in vitro* condition by employing poisoned food technique of Bagchi and Das (1968) using Potato Dextrose Agar (PDA) as a germinating medium. The quantity of each fungicides required were incorporated into autoclaved measured PDA medium before solidification with micropipette and then medium were poured into sterilized Petri dishes (90 mm dia.) in equal quantity (20 ml per Petri dish) to form a uniform layer. The experiment was arranged in FCRD with three repetitions.

These plates were then allowed to solidify. After solidification the plates were inoculated with an actively growing fungal mycelial bit of 4 mm diameter which was transferred under aseptic conditions over the solidified PDA medium. The mycelial disc was placed in the center of plates in an inverted position to make a direct contact with the poisoned medium. Then Petri dishes were incubated at 25 $\pm$ 1°C for 10 days and observations were recorded on radial growth of mycelium in treated and control plates. Inoculated Petri dishes containing PDA medium without fungicides were served as control. The radial growths of the fungal colonies were measured from two different angles in millimeter (mm) and the average values were calculated. The per cent growth inhibition of the fungus in each treatment was calculated by using following formula (Vincent, 1947)

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

Where,

PGI = Per cent growth inhibition

C = Average growth diameter of test pathogen in control (mm^2)

T = Average growth diameter of test pathogen in treatments (mm^2)

Experiment was laid out with seven treatments with three repetitions. Completely Randomized Block Design with factorial concept was used for analyzing the data.

Results and Discussion

The results were discussed in Tab.1, Tab.2, Tab.3. Among different non-systemic fungicides mancozeb 75 % WP and propineb 70 % WP gave mycelial growth inhibition of 87.74, 88.89, 90.04, 90.96 and 86.15, 87.56, 90.96 90.15 per cent at concentrations 500, 1000, 1500 and 2000 ppm, respectively which were found best treatments for inhibiting the growth of mycelium of *A. triticina* under *in vitro* condition. Metiram 55 % WG was found next best effective fungicide gave 75.78, 86.85, 90.00, 92.37 per cent mycelial growth inhibition at 500, 1000, 1500 and 2000 ppm concentrations, respectively. Thiram 75 %

WP gave 80.74, 84.07, 88.78 and 91.26 per cent and zineb 75 % WP gave 64.22, 65.44, 70.44 and 84.33 per cent mycelial growth inhibition of the fungus at 500, 1000, 1500, 2000 ppm concentration, respectively. Copper oxychloride 50 % WP showed mycelial growth inhibition 53.70, 65.96, 75.52 and 76.41 per cent and sulphur 80 % WP inhibited the mycelial growth 33.74, 42.07, 60.85 and 75.00 per cent at 500, 1000, 1500 and 2000 ppm concentrations, respectively these two were found as a least effective non-systemic fungicides. The cumulative mycelial growth inhibition was increased with increased concentration of fungicides. The interaction effect studied between fungicide and its concentrations showed metiram 55 % WG (92.37 %), thiram 75 % WP (91.26 %), propineb 70 % WP (90.89 %) and mancozeb 75 % WP (90.96 %) at 2000 ppm were on par with each other whereas mancozeb 75 % WP at 1000 ppm (88.89 %) and 1500 ppm (90.04 %), propineb 70 % WP at 1500 ppm (90.21 %) and 2000 ppm (90.89 %), metiram 55 % WG at 1500 ppm (90.00 %) were on par with each other in inhibiting radial growth of mycelium. The effectiveness of mancozeb in inhibiting the growth of *A. triticina* has been reported by Sankhla *et al.,* (1972). Mancozeb showed maximum mean mycelial growth inhibition of *Alternaria triticina* in present study, agreement with results obtained by Bhowmick (1974) and Sahu *et al.,* (2013).

Among different systemic fungicides tested propiconazole 25 % EC found the best among all treatments and gave 84.00, 100.00, 100.00 and 100.00 per cent inhibition of growth of the fungus at 50, 100, 250 and 500 ppm concentrations, respectively under *in vitro* condition. Hexaconazole 5% EC was found as a second best fungicide with 85.07, 85.93, 90.30 and 100.00 per cent mycelial growth at 50, 100, 250 and 500 ppm concentrations, respectively. Next to it azoxystrobin 23 % SC was found effective with mycelia growth inhibition per cent of 65.96, 67.81, 77.85 and 81.63 at 50, 100, 250 and 500 ppm concentrations respectively. Carbendazim 50 % WP showed mycelia growth inhibition (48.93, 52.15, 66.30 and 70.81), thiophanate methyl 70 % WP showed (54.44, 57.15, 60.74 and 63.44), fosetyl -Al showed (43.11, 51.48, 63.30 and 73.52) and picoxystrobin 25 % EC showed 38.93, 54.44, 60.48 and 73.30 mycelial growth inhibition at 50, 100, 250 and 500 ppm, respectively. The cumulative mycelial growth inhibition was increased with increased concentration of fungicides. The interaction effect studied between fungicide and its concentrations showed hexaconazole 5 % EC at 500 ppm (100 %) and propiconazole 25 % EC at 100,250 and 500 ppm (100 %) were on par with each other whereas hexaconazole 5 % EC at 50 ppm (85.07 %), 100 ppm (85.93 %) and propiconazole 25 % EC at 50 ppm (84.0 %) were on par with each other. This findings are in conformity with earlier studies of Patel (2008) and Ginoya and Gohel (2015) who reported cent per cent inhibition of radial growth of fungus *A. alternata* with propiconazole.

Among different combination fungicides tested azoxystrobin 11 % + tebuconazole 18.30 % WP, hexaconazole 4 % + zineb 68 % WP and carboxin 37.5 % + thiram 37.5 % SD were significantly superior over rest of the treatments gave mean mycelial growth inhibition cent per cent. Next to it, cymoxanil 8 % + mancozeb 64 % WP was found effective with mycelial growth inhibition per cent of 75.81, 80.89, 86.15 and 90.89 at 100, 250, 500 and 1000 ppm, respectively. Carbendazim 12 % + mancozeb 63 % WP showed mycelial growth inhibition of 64.96, 77.33, 80.52 and 88.52, tricyclazole 18 % + mancozeb 62 % WP showed 55.93, 66.15, 84.07 and 87.19 and tebuconazole 50 % + trifloxystrobin 25 % WG showed 54.56, 58.07, 87.89 and 90.70 at 100, 250, 500 and 1000 ppm, respectively (Table 1–3).

Table.1 Effect of different non-systemic fungicides on mycelial growth inhibition of *A. triticina* under *in vitro* condition

Sr. No.	Fungicides	*Growth inhibition (%)				Mean inhibition (%)
		500 ppm	1000 ppm	1500 ppm	2000 ppm	
1	Mancozeb (75%WP)	87.74 (69.54)♯	88.89 (70.56)	90.04 (71.64)	90.96 (72.54)	89.41 (71.07)
2	Propineb (70 %WP)	86.15 (68.18)	87.56 (69.38)	90.21 (71.80)	90.89 (72.47)	88.70 (70.46)
3	Copper Oxy chloride (50% WP)	53.70 (47.15)	65.96 (54.34)	75.52 (60.37)	76.41 (60.97)	67.90 (55.71)
4	Metiram (55 %WG)	75.78 (60.55)	86.85 (68.77)	90.00 (71.60)	92.37 (74.00)	86.25 (68.73)
5	Sulphur (80 %WP)	33.74 (35.53)	42.07 (40.46)	60.85 (51.29)	75.00 (60.03)	52.92 (46.83)
6	Thiram (75 %WP)	80.74 (64.00)	84.07 (66.51)	88.78 (70.46)	91.26 (72.84)	86.21 (68.45)
7	Zineb (75 %WP)	64.22 (53.29)	65.44 (54.02)	70.44 (57.10)	84.33 (66.72)	71.11 (57.78)
Mean		68.87 (56.89)	74.41 (60.58)	80.83 (64.07)	85.89 (67.97)	-
		Between Fungicide(F)		**Within fungicide Concentration(C)**		**F × C**
S.Em. ±		0.34		0.26		0.69
C.D at 5%		0.98		0.76		1.96
CV%		1.90				

* Mean of three replications

♯ Data in parentheses are arcsine transformed and outside are retransformed values.

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

Sr. No.	Fungicides	*Growth inhibition (%)				Mean inhibition (%)
		50 ppm	100 ppm	250 Ppm	500 ppm	
1	Azoxystrobin 23 % SC	65.96 (54.34)♯	67.81 (55.46)	77.85 (61.96)	81.63 (64.65)	73.31 (59.10)
2	Carbendazim 50 % WP	48.93 (44.41)	52.15 (46.25)	66.30 (54.54)	70.81 (57.33)	59.54 (50.63)
3	Fosetyl-Al 80 % WP	43.11 (41.06)	51.48 (45.87)	63.30 (52.74)	73.52 (59.06)	57.85 (49.68)
4	Hexaconazole 5 % EC	85.07 (67.31)	85.93 (68.00)	90.30 (71.89)	100.00 (90.05)	90.33 (74.31)
5	Picoxystrobin 25 % EC	38.93 (38.62)	55.44 (48.15)	60.48 (51.08)	73.30 (58.91)	57.03 (49.19)
6	Propiconazole 25 % EC	84.00 (66.46)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	96.00 (84.15)
7	Thiophanate methyl 70 % WP	54.44 (47.57)	57.15 (49.13)	60.74 (51.23)	63.44 (52.83)	58.94 (50.19)
Mean		60.06 (51.40)	67.14 (57.56)	74.14 (61.93)	80.38 (67.55)	-
		Between Fungicide (F)		**Within fungicide Concentration (C)**		**F x C**
S.Em. ±		0.37		0.28		0.74
C. D. at 5%		1.05		0.81		2.11
C.V. %		2.16				

* Mean of three replications

♯ Data in parentheses are arcsine transformed and outside are retransformed values.

Table.3 Effect of different combination fungicides on mycelial growth inhibition of *A. triticina* under *in vitro* condition

Sr. No.	Fungicide	*Growth inhibition (%)				Mean inhibition (%)
		100 ppm	250 ppm	500 Ppm	1000 ppm	
1	Carbendazim 12 % + Mancozeb 63 % WP	64.96 (53.73)♯	77.33 (61.60)	80.52 (63.84)	88.52 (70.23)	77.83 (62.35)
2	Cymoxanil 8 % + Mancozeb 64 % WP	75.81 (60.57)	80.89 (64.11)	86.15 (68.18)	90.89 (72.47)	83.44 (66.33)
3	Azoxystrobin 11 % + Tebuconazole 18.30 % WP	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)
4	Tricyclazole 18 % + Mancozeb 62 % WP	55.93 (48.43)	66.15 (54.45)	84.07 (66.51)	87.19 (69.06)	73.34 (59.61)
5	Hexaconazole 4 % + Zineb 68 % WP	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)
6	Carboxin 37.5 % + Thiram 37.5 % SD	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)
7	Tebuconazole 50 % + Trifloxystrobin 25 % WG	54.56 (47.64)	58.07 (49.67)	87.89 (69.67)	90.70 (72.28)	72.81 (59.82)
Mean		78.75 (68.65)	83.21 (71.43)	91.23 (76.91)	93.90 (79.17)	-
		Between Fungicide (F)		**Within fungicide Concentration (C)**		**F x C**
S.Em. ±		0.23		0.17		0.45
C. D. at 5%		0.65		0.50		1.29
C.V. %		1.06				

* Mean of three replications

♯ Data in parentheses are arcsine transformed and outside are retransformed values

The cumulative mycelial growth inhibition was increased with increased concentration of fungicides. The interaction effect studied between fungicide and its concentrations showed cymoxanil 8 % + mancozeb 64 % WP at 1000 ppm (90.89 %) and tebuconazole 50 % + trifloxystrobin 25 % WG at 1000 ppm (90.70 %) on par with each other whereas tebuconazole 50 % + trifloxystrobin 25 % WG at 500 ppm (87.89 %), tricyclazole 18 % + mancozeb 62 % WP at 1000 ppm (87.19 %) and carbendazim 12 % + mancozeb 63 % WP at 1000 ppm (88.52 %) were on par with each other. Next to it, cymoxanil 8 % + mancozeb 64 % WP at 500 ppm (86.15 %) and tricyclazole 18 % + mancozeb 62 % WP at 1000 ppm (87.19 %) on par with each other in inhibiting radial growth of mycelium. In present investigation, hexaconazole 4 % + zineb 68 % WP, azoxystrobin 11 % + tebuconazole 18.30 % WP and carboxin 37.5 % + thiram 37.5 % SD were found as a most effective fungicide in inhibition of radial growth of *A. triticina* has reported by Patel (2008) and Ginoya and Gohel (2015).

Among the non-systemic fungicides tested under *in vitro* condition, mancozeb 75 % WP and propineb 70 % WP were found the most effective with 89.41 and 88.70 per cent fungal growth inhibition respectively. While copper oxychloride 50 % WP was least effective even at higher concentration. In case of systemic fungicides propiconazole 25 % EC and hexaconazole 5 % SC proved the most effective with 96.00 and 90.33 per cent mean fungal growth inhibition. During laboratory screening of combination fungicides, azoxystrobin 11 % + tebuconazole 18.30 % WP, hexaconazole 4 % + zineb 68 % WP and carboxin 37.5 % + thiram 37.5 % SD were found most effective with cent per cent growth inhibition of fungus. In all the fungicides bioassay studies, with increase in the concentration there was corresponding decrease in the growth of fungus.

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