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Efficacy of different fungicides and weedicides against *Macrophomina phaseolina* (Tassi) Goid causing groundnut root rot

Badana Manoj Kumar and Kelaiya DSDOI: <https://doi.org/10.22271/chemi.2020.v8.i2ak.9119>**Abstract**

In this study, different systemic, non-systemic, combination fungicides and weedicides at four different concentration were evaluated *in vitro* against groundnut root rot pathogen *Macrophomina phaseolina*. Among the non-systemic fungicides, Mancozeb 75% WP and Propineb 70% WP were found most effective and showed (99.98 and 99.96 per cent) mycelia growth inhibition at all concentrations. Whereas in systemic fungicides, Carbendazim 50% WP were found most effective with (94.24 per cent) mean mycelial growth inhibition. Among different combination fungicides tested, Carboxin 37.5% + Thiram 37.5% WP were found most effective with mean mycelial growth inhibition of (97.16 per cent).. In case of weedicides tested, Quizalofop-p-ethyl 5% EC found as most effective inhibiting the mycelial growth of *M. phaseolina* at all concentrations with (90.92 per cent) mean mycelial growth inhibition. Sclerotial formation was also absent in all this treatments.

Keywords: Fungicides, weedicide, *Macrophomina phaseolina*, root rot, groundnut**Introduction**

Groundnut (*Arachis hypogaea* L.) is a cultivated annually belonging to the plant family *Leguminosae* and sub family *Papilionaceae*. It is believed to be originated from Brazil in South America and introduced into India in 16th century. It is cultivated in tropical and sub tropical countries of the world. It contains 48-50% oil and 26-28% protein. It provides 12% recommended nutrients and has dietary fibre that reduce the risk of some kinds of cancer and helps control blood sugar. Among 13 essential vitamins, 7 are found in groundnut and among 20 minerals necessary for growth, 7 are present in groundnut. The major groundnut producer countries of the world are India, China, USS, Senegal, Sudan, Nigeria and Burma. Cultivation of groundnut has also considerably increased in Australia, Japan and South America. Among the groundnut producing nations, India shows the highest area under this crop and is second largest producer next to China. Groundnut is affected by many diseases like as collar rot, cercospora leaf spot, alternaria leaf spot etc. Among all diseases, root rot caused by *Macrophomina phaseolina* is a major problem in India and it has now become a limiting factor in stepping up the groundnut yield. *Macrophomina phaseolina* is a soil borne fungus causing the root rot disease on groundnut and more than 500 plant species from more than 100 families distributed worldwide and is one of the cosmopolitan fungi (Wyllie, 1988 and Mihail, 1992) [18]. The fungus invades the host both inter & intracellular, it grows rather fast covering large areas of the host tissue & eventually killing them in short time. It produces numerous sclerotial bodies on host tissue, which measure about 110-130-µm in diameter. The first symptom of disease is yellowing of the leaves which droop in next 2 or 3 days and withers off. The plant may wilt within a week after the appearance of first symptom. When stem is examined closely, dark lesion may be seen on the bark at the ground level. If the plants are pulled from soil the basal stem & main root may show dry rot symptoms the tissues are weakened and break off easily in advanced cases sclerotial bodies may be seen scattered on the affected tissues. *Macrophomina phaseolina* (Tassi) Goid has been noticed to cause 33.33% seed rotting and 23.80% post emergence mortality (Gupta and Kolte, 1982) [8]. The main objective of this study was to evaluate the different fungicides at different concentrations for testing their efficacy against mycelia growth inhibition and sclerotial formation of *Macrophomina phaseolina* under *in vitro* condition.

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Materials and Methods

Collection of samples

The plants showing typical symptoms of root rot, caused by *M. phaseolina* (Tassi) Goid were collected from Main Oilseeds Research Station farm, JAU, Junagadh

Isolation

Isolation from infected roots of groundnut plant showing typical root rot symptoms were done by using tissue isolation method.

Purification

The tissue were surface sterilized with 0.1 per cent mercuric chloride (HgCl₂) solution for 30 seconds followed by 3 times washing with sterilized distilled water and transferred aseptically in sterilized Petri plates. These Petri plates were incubated at room temperature (27 ± 2 °C) for 6 to 7 days. The fungal hypha developed from the infected tissue was sub cultured aseptically on PDA slants for the maintenance of pure culture.

In vitro evaluation of fungicides and weedicides

Different concentrations of systemic, non-systemic, combination fungicides and weedicides were tested for the growth inhibition and sclerotial formation of *M. phaseolina* by using poisoned food technique given by Bagchi and Das (1968). The quantity of each fungicides and weedicides 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 were 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 were transferred under aseptic conditions over the solidified PDA medium. The mycelial disc were 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 28 ± 2 °C for 7 days and observations were recorded on radial growth of mycelium in treated and control plates. Inoculated Petri dishes containing PDA medium without fungicides and weedicides were served as control. The radial growth 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) ^[17].

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

Where,

I = Percent inhibition of mycelial growth

C = Radial growth of fungus in control (mm)

T = Radial growth of fungus in treatment (mm)

Sclerotial formations were counted in fungal culture suspension under microscope at low power (10x). The fungal culture suspension was prepared by vigorously shaking the 4 mm mycelial disc of the fungus in 10 ml sterilized distilled water after 15 days of incubation. The relative degree of sclerotial formation was recorded as below Table 1.

Table 1: Relative degree of sclerotial formation

No. of sclerotia per microscopic field (10x)	Grade	Sign
0	Absent	-
1-4	Scanty	+
5-8	Moderate	++
9-15	Good	+++
>15	Abundant	++++

Results and Discussion

The growth inhibition and sclerotial formation of *M. phaseolina* causing root rot in groundnut has been tested at various concentration of systemic, non-systemic, combination fungicides and weedicides *in vitro* recorded in Table 2, 3, 4 and 5.

Non- systemic fungicides

The perusal of results showed that (Table 2) among the seven different non-systemic fungicides tested under *in vitro* condition, Mancozeb 75% WP and Propineb 70% WP were proved most effective fungicides, which inhibited (99.98 and 99.96 per cent) mycelia growth at all concentration (500, 1000, 1500 and 2000 ppm) respectively of *M. phaseolina*. The effectiveness of Mancozeb in inhibition the growth of *M. phaseolina* has been reported by Dubey (2003) ^[7]. Propineb and Mancozeb shows maximum mean mycelial growth inhibition of *M. phaseolina* in present study, in agreement with results obtained by Parmar *et al.* (2017) ^[14]. Followed by Thiram 75% WP with (81.89, 83.80 and 86.25 per cent) growth inhibition at (1000, 1500 and 2000 ppm) respectively. The effectiveness of Thiram against *M. phaseolina* has been recorded by (Ammajamma and Hegde 2009, Lambhate *et al.* 2002) ^[1, 12]. Copper hydroxide 50% WP were found least effective non-systemic fungicide with lowest (1.07, 2.33, 4.21 and 5.76 percent) mycelia growth inhibition at (500, 1000, 1500 and 2000 ppm) concentrations tested. The effect of different concentrations of non-systemic fungicides on sclerotial formation was found negatively correlated with the inhibition of growth. No sclerotial formation was found in Mancozeb 75% WP and Propineb 75% WP. While abundant sclerotial formation was found in Copper hydroxide 50% WP and Copper oxychloride 50% WP at all concentration tested.

Table 2: Effect of different non-systemic fungicides on mycelial growth inhibition and sclerotial formation of *M. phaseolina* under *in vitro* condition after seven days incubation at 28 ± 1 °C

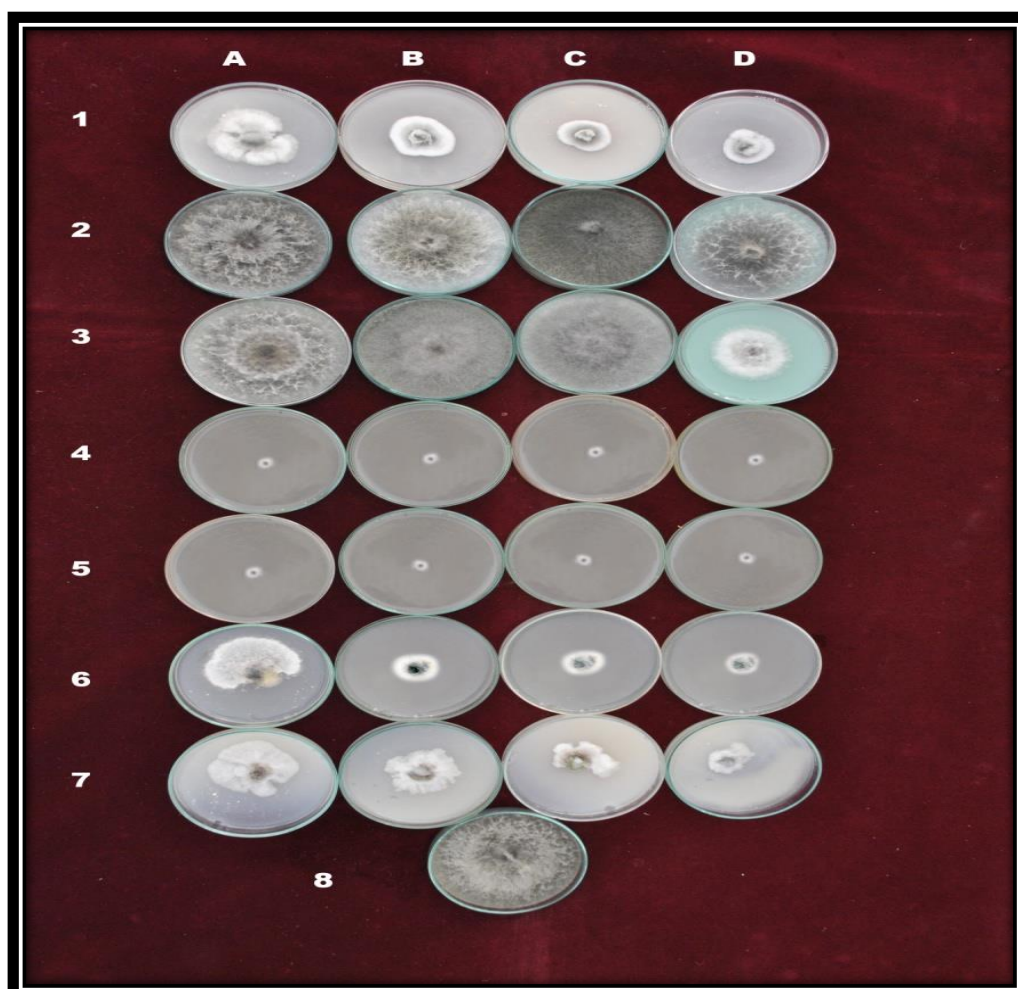
Fungicide	Concentration (ppm) and Sclerotial formation				Mean fungicide inhibition (%)
	500	1000	1500	2000	
Chlorothalonil 75% WP	45.07 (50.14)	47.13 (53.72)	48.42 (55.92)	53.29 (64.27)	48.45 (56.01)
	+++	+++	++	++	
Copper hydroxide 50% WP	5.80 (1.07)	8.72 (2.33)	11.79 (4.21)	13.59 (5.76)	10.53 (3.34)
	++++	++++	++++	++++	
Copper oxychloride	7.44	9.56	11.16	38.70	20.14

50% WP	(1.72)	(2.77)	(3.78)	(39.15)	(11.85)
	++++	++++	++++	++++	
Mancozeb 75% WP	89.19 (99.98)	89.19 (99.98)	89.19 (99.98)	89.19 (99.98)	89.19 (99.98)
	-	-	-	-	
Propineb 75% WP	88.85 (99.96)	88.85 (99.96)	88.85 (99.96)	88.85 (99.96)	88.85 (99.96)
	-	-	-	-	
Thiram 75% WP	54.58 (66.23)	64.79 (81.80)	66.29 (83.80)	68.29 (86.25)	63.09 (79.52)
	++	-	-	-	
Zineb 75% WP	53.04 (63.84)	56.32 (69.21)	56.81 (70.03)	58.62 (72.88)	56.16 (68.99)
	++	+	+	+	
Concentration Mean (%)	47.70 (54.70)	49.91 (58.53)	50.57 (59.66)	54.87 (66.89)	
	Fungicide (F)	Conc. (C)	F x C		
S.Em. ±	0.44	0.33	0.89		
C. D. at 5%	1.26	0.95	2.53		
C.V.%	2.99				

Sclerotial formation

++++ = Abundant; +++ = good; ++ = moderate; + = scanty; - = no sclerotial formation.

Values in parentheses are re-transformed values while outside were transformed with arcsine transformation before analysis.



Non-systemic fungicides

1. Chlorothalonil 75% WP
2. Copper hydroxide 77% WP
3. Copper oxychloride 50% WP
4. Mancozeb 75% WP
5. Propineb 70% WP
6. Thiram 75% WP
7. Zineb 75% WP

Concentrations (ppm)

- 500
- 1000
- 1500
- 2000

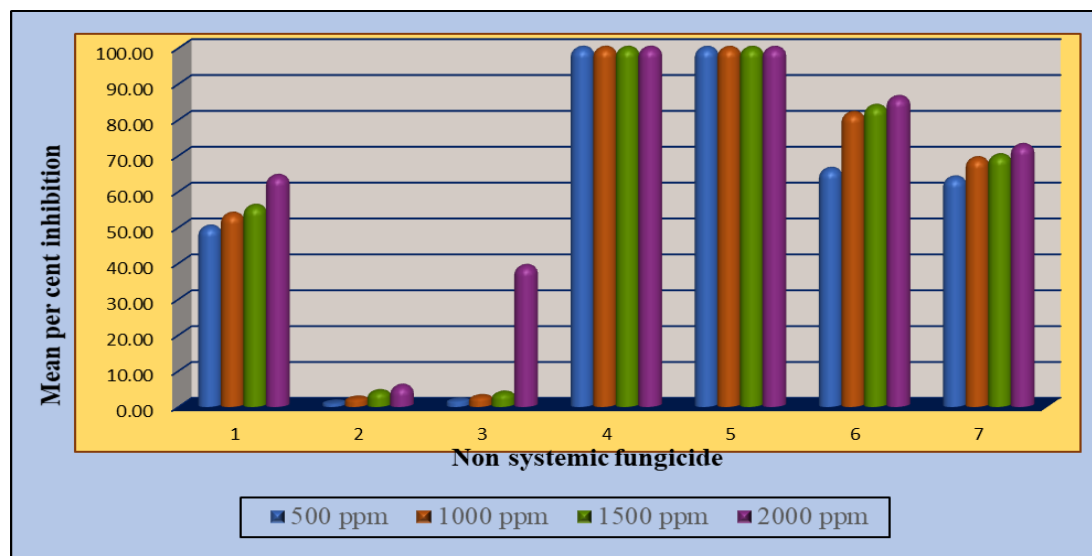


Plate 1: Effect of different non-systemic fungicides on mycelial growth inhibition of *M. phaseolina* under *in vitro*

Systemic fungicides

Among the seven different systemic fungicides (Table 3) tested under *in vitro* condition, Carbendazim 50% WP were proved most effective among all treatments, which inhibited (85.98, 91.05, 99.97 and 99.97 per cent), mycelial growth of the fungus at (50, 100, 250 and 500 ppm) concentrations respectively. Similarly the effectiveness of Carbendazim against *M. phaseolina* which inhibit maximum mycelia growth and sclerotial formation has been reported by Bhatia *et al.* (1997) [4], Chaudhari and Sharma (1998) [5], lambhate *et al.* (2002) [12], Dubey (2003) [7], jaiman and jain (2010) [10], khalikar *et al.* (2011) [11] and Rekha *et al.* (2012) while working with various crops. Difenonazole 25% EC were found second best fungicides with (41.17, 66.16, 74.13 and 80.32 per cent) mycelial growth inhibition followed by Hexaconazole 5% EC with (52.58, 61.31, 64.76 and 66.80 per cent) mycelial growth inhibition at (50, 100, 250 and 500

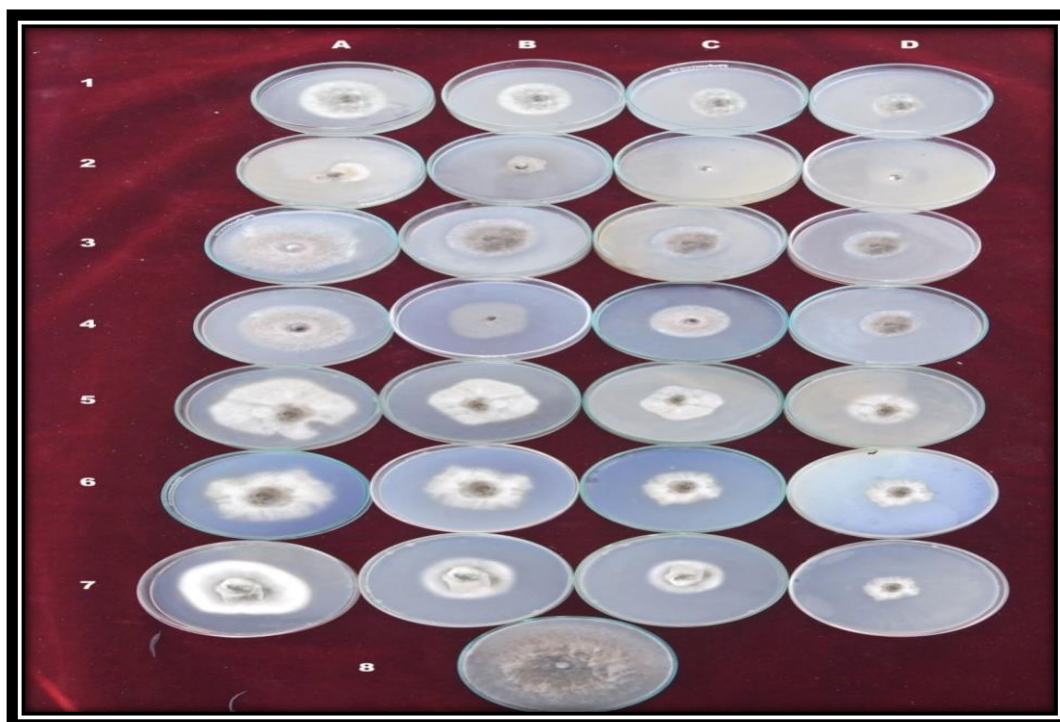
ppm) concentrations respectively. Moradia (2011) [13] reported that Difenonazole 25% EC was found most effective and capable of inhibiting the growth of *M. phaseolina* at all the concentration tried. And the effectiveness of Hexaconazole have already been reported to be the best control of *M. phaseolina* by earlier workers while working with this fungus in different crops (Ammajamma and Hegde, 2009; Hegde and chavhan, 2009 and Khalikar *et al.* 2011) [1, 9, 11]. Among all systemic fungicides tested, Pyroclostrobin 20% WP was found least effective in mycelial growth inhibition with (45.23 per cent) mean inhibition. The effect of different concentrations of systemic fungicides on sclerotial formation was found negatively correlated with the inhibition of growth. No sclerotial formation was found at any concentration of Carbendazim 50% WP. While abundant sclerotial formation was observed in all concentrations of Pyroclostrobin 20% WP.

Table 3: Effect of different systemic fungicides on mycelial growth inhibition and sclerotial formation of *M. phaseolina* under *in vitro* condition after seven days incubation at $28 \pm 1^\circ\text{C}$.

Fungicide	Concentration (ppm) and Sclerotial formation				Mean fungicide inhibition (%)
	50	100	250	500	
Difenonazole 25% EC	39.89 (41.17) +++	54.43 (66.16) +	59.43 (74.13) -	63.68 (80.32) -	53.99 (65.44)
Carbendazim 50% WP	68.04 (85.98) -	72.83 (91.05) -	89.01 (99.97) -	89.01 (99.97) -	76.11 (94.24)
Fosetyl-Al 80% WP	31.72 (27.69) +++	47.80 (54.89) ++	54.85 (66.86) +	57.72 (71.49) -	48.00 (55.23)
Hexaconazole 5% EC	46.49 (52.58) ++	51.54 (61.31) +	53.59 (64.76) +	54.82 (66.80) +	51.57 (61.36)
Pyroclostrobin 20% WP	18.49 (10.19) ++++	40.05 (41.41) +++	52.37 (62.73) ++	54.68 (66.59) +	42.26 (45.23)
Propiconazole 25% EC	36.47 (35.37) +++	41.92 (44.65) ++	53.57 (64.72) +	58.44 (72.61) +	47.48 (54.33)
Thiophanate methyl 70% WP	34.35 (31.77) +++	51.49 (61.23) ++	53.98 (65.42) +	62.71 (78.96) -	50.38 (59.34)
Concentration Mean (%)	39.62 (40.67)	50.86 (60.15)	57.56 (71.23)	61.12 (76.67)	
	Fungicide (F)	Conc. (C)	F x C		

S.E.m. \pm	0.47	0.35	0.94
C. D. at 5%	1.33	1.00	2.66
C.V.%	3.06		

Values in parentheses are re-transformed values while outside were transformed with arcsine transformation before analysis.



Systemic fungicides Concentrations (ppm)

1. Difenconazole 25% EC	50
2. Carbendazim 50% WP	100
3. Fosetyl-Al 80% WP	250
4. Hexaconazole 5% EC	500
5. Pyraclostrobin 20% WP	
6. Propiconazole 25% EC	
7. Thiophanate methyl 70% WP	

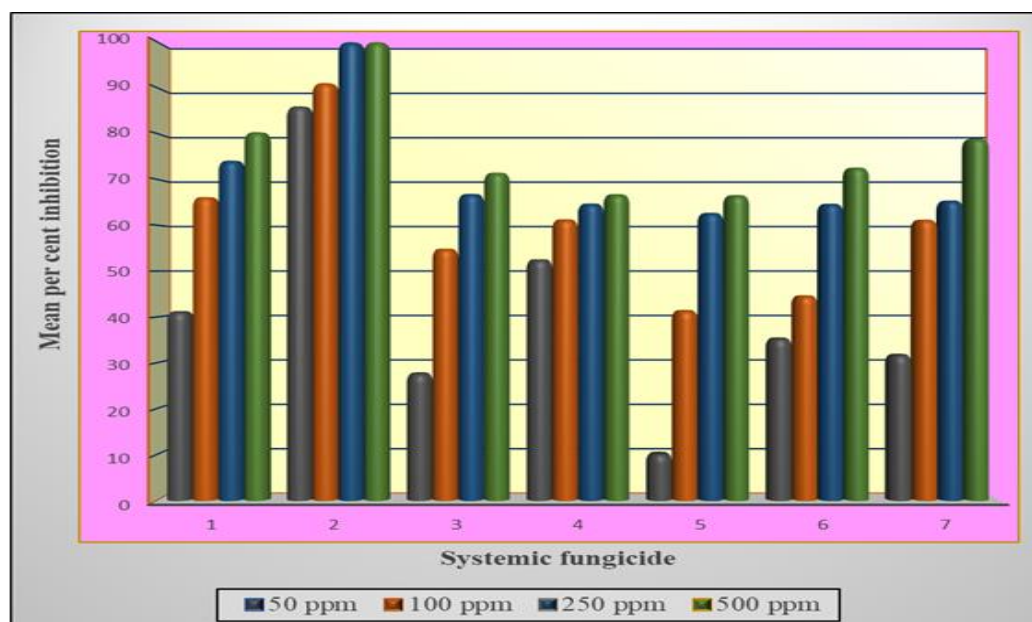


Plate 2: Effect of different systemic fungicides on mycelial growth inhibition of *M. phaseolina* under *in vitro*

Combination fungicides

All combination fungicides (Table 4) were found effective for mycelia growth inhibition of *M. phaseolina*. Among the seven different combination fungicides tested under *in vitro* condition, Carboxin 37.5% + Thiram 37.5% WP which inhibited (93.39, 97.54, 98.87 and 98.87 per cent) mycelial

growth at (100, 250, 500 and 1000 ppm) concentrations respectively were found the most effective combination fungicide. Hegde and Chavhan (2009) [9] reported that drenching with carboxin + thiram @ 0.1% have managed the root rot of *Jatropha* effectively. Followed by Carbendazim 12% + Mancozeb 63% WP with (86.30, 88.27, 95.96 and

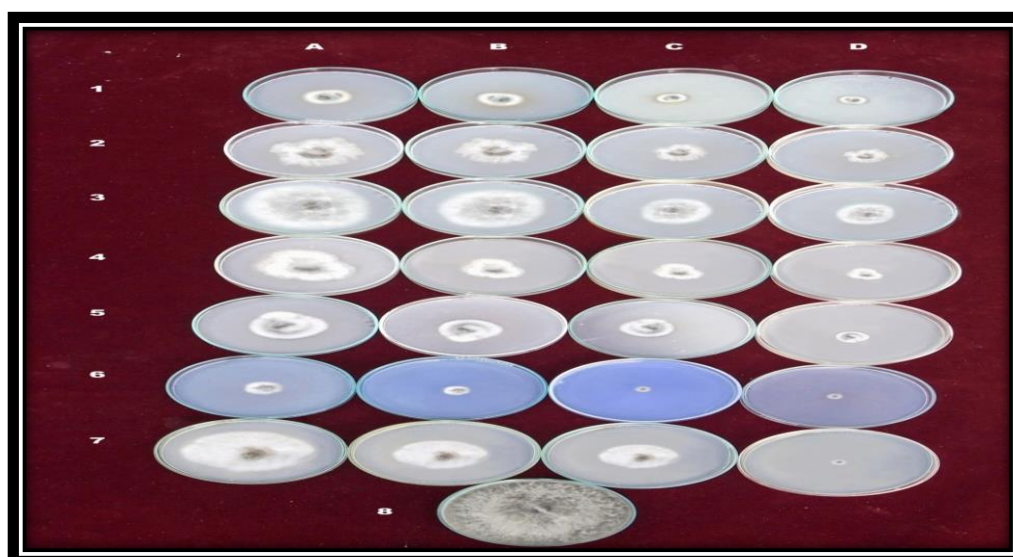
95.96 per cent) mycelial growth inhibition at (100, 250, 500 and 1000 ppm) concentrations, respectively. Tandel *et al.* (2010) [16] studied Carbendazim + mancozeb was found significantly superior over the rest as it resulted minimum (8.13%) disease intensity of *M. phaseolina* in mungbean. Azoxystrobin 11% + Tebuconazole 18.30% WP were found least effective combination in mycelial growth inhibition with

41.17 per cent mean inhibition. No sclerotial formation was found in all concentration of Carboxin 37.5 + Thiram 37.5% WP, Cymoxanil 8% + Mancozeb 64% WP and Carbendazim 12% + Mancozeb 63% WP. While, abundant sclerotial formation was found in all concentrations of Azoxystrobin 11% + Tebuconazole 18.30% WP.

Table 4: Effect of different combination fungicides on mycelial growth inhibition and sclerotial formation of *M. phaseolina* under *in vitro* condition after seven days incubation at 28 ± 1 °C

Fungicide	Concentration (ppm) and Sclerotial formation				Mean fungicide inhibition (%)
	100	250	500	1000	
Carbendazim 12% + Mancozeb 63% WP	68.29 (86.30)	70.01 (88.27)	78.76 (95.96)	78.76 (95.96)	73.17 (91.26)
	-	-	-	-	
Cymoxanil 8% + Mancozeb 64% WP	46.57 (52.74)	47.99 (55.18)	66.18 (83.68)	68.23 (86.24)	56.45 (69.46)
	-	-	-	-	
Azoxystrobin 11% + Tebuconazole 18.30% WP	30.03 (25.38)	33.85 (31.27)	44.66 (49.41)	51.23 (60.78)	40.23 (41.17)
	+++	+++	++	+	
Tricyclazole 18% + Mancozeb 62% WP	43.57 (47.51)	55.55 (67.99)	59.21 (73.77)	66.57 (84.16)	55.77 (68.35)
	+	-	-	-	
Hexaconazole 4% + Zineb 68% WP	46.52 (52.64)	53.52 (64.66)	56.40 (69.38)	66.22 (83.74)	55.30 (67.60)
	+	-	-	-	
Carboxin 37.5% + Thiram 37.5% WP	75.12 (93.39)	81.09 (97.54)	83.91 (98.87)	83.91 (98.87)	80.30 (97.16)
	-	-	-	-	
Tebuconazole 50% + Trifloxystrobin 25% WG	37.78 (37.53)	45.57 (51.26)	52.94 (63.69)	81.89 (98.01)	52.27 (62.55)
	++	+	-	-	
Concentration Mean (%)	48.73 (56.49)	53.81 (65.13)	60.93 (76.39)	68.71 (86.82)	
	Fungicide (F)	Conc. (C)	F x C		
S.Em. ±	0.66	0.50	1.33		
C. D. at 5%	1.89	1.43	3.79		
C.V.%	3.88				

Values in parentheses are re-transformed values while outside were transformed with arcsine transformation before analysis.



Combination fungicides

1. Carbendazim 12% + Mancozeb 63% WP
2. Cymoxanil 8% + Mancozeb 64% WP
3. Azoxystrobin 11% + Tebuconazole 18.3% SL
4. Tricyclazole 4% + Mancozeb 62% WP
5. Hexaconazole 4% + Zineb 68% WP
6. Carboxin 37.5% + Thiram 37.5% WP
7. Tebuconazole 50% + Trifloxystrobin 25% WG

Concentrations (ppm)

- 100
- 250
- 500
- 1000

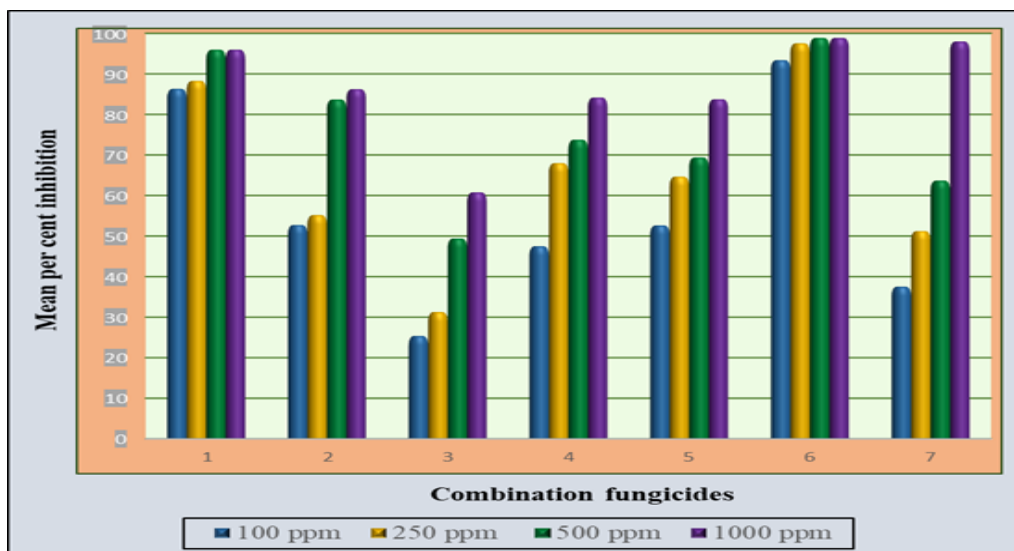


Plate 3: Effect of different combination fungicides on mycelial growth inhibition of *M. phaseolina* under *in vitro*

Weedicides

Among the seven different weedicides (Table 5) tested in *in vitro* condition, Quizalofop-p-ethyl 5% EC inhibited (83.47, 86.79, 95.06 and 98.39 per cent) growth of the fungus. Maximum inhibitory effect of Quizalofop-p-ethyl against *R. bataticola* has also been reported by De *et al.* (2007) [6]. Followed by Pendimethalin 30% EC with (72.73, 81.21, 93.54 and 95.50 per cent) inhibition of growth of the fungus at (100, 150, 250 and 500 ppm) concentrations respectively. Which was proved as most effective treatment for mycelium

growth inhibition of *M. phaseolina* under *in vitro* condition. Bauske and Kirby (1992) [3] noted that Pendimethalin herbicide did not cause an increase in disease caused by *R. bataticola* on field location and soybean cultivar. Metribuzin 70% WP was found least effective in inhibiting the mycelial growth of *M. phaseolina* at all concentration tested with 46.69 per cent mean inhibition and abundant sclerotial formation was observed in all concentration of Quizalofop-p-ethyl 5% EC.

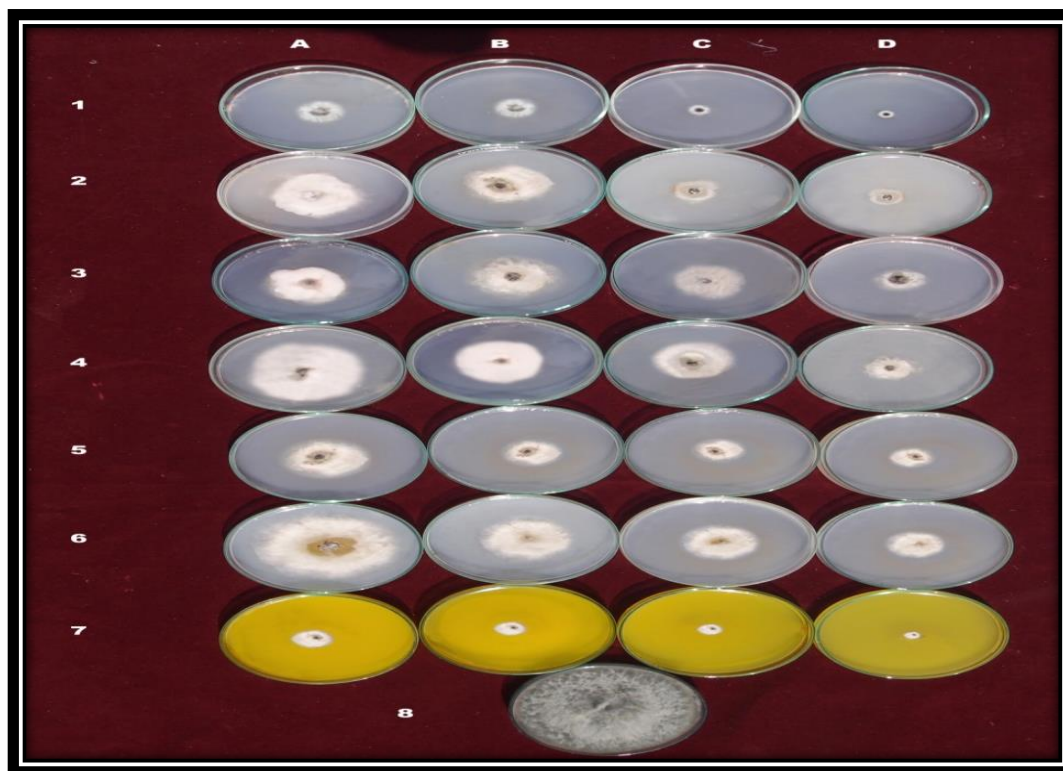
Table 5: Effect of different weedicides on mycelial growth inhibition and sclerotial formation of *M. phaseolina* under *in vitro* condition after seven days incubation at 28 ± 1 °C.

Weedicides	Concentration (ppm) and Sclerotial formation				Mean weedicide inhibition (%)
	100	150	250	500	
Quizalofop-p-ethyle 5%EC	66.02 (83.47)	68.75 (86.79)	77.43 (95.06)	82.64 (98.36)	72.46 (90.92)
	-	-	-	-	
Atrazine 50% WP	40.07 (41.44)	45.64 (51.11)	54.23 (65.83)	55.32 (67.62)	48.75 (56.52)
	++	++	+	+	
Imazethapyr 10% SL	42.65 (45.91)	45.71 (51.24)	52.44 (62.84)	64.32 (80.77)	50.88 (60.19)
	++	++	+	-	
Metribuzin 70% WP	34.34 (31.84)	37.87 (37.70)	44.13 (48.49)	56.02 (68.76)	43.10 (46.69)
	+++	+++	++	+	
Propaquizafop 10% EC	49.58 (57.97)	55.86 (68.52)	57.52 (71.09)	59.24 (73.84)	55.46 (67.85)
	++	-	-	-	
Oxyfluorfen 23.5% EC	30.94 (26.45)	43.62 (47.60)	51.46 (61.09)	57.11 (70.45)	45.80 (51.39)
	++++	++	+	+	
Pendimethalin 30% EC	58.53 (72.73)	64.34 (81.21)	75.34 (93.54)	77.87 (95.50)	67.81 (85.74)
	+	-	-	-	
Concentration Mean (%)	45.80 (51.40)	51.11 (60.59)	57.50 (71.13)	62.95 (79.32)	
	Weedicide (W)	Conc. (C)	W x C		
S.Em. ±	0.46	0.34	0.92		
C. D. at 5%	1.34	0.98	2.60		
C.V.%	2.87				

Sclerotial formation

++++ = Abundant; +++ = good; ++ = moderate; + = scanty; - = no sclerotial formation.

Values in parentheses are re-transformed values while outside were transformed with arcsine transformation before analysis.

**Weedicides****Concentrations (ppm)**

- | | |
|-------------------------------|-----|
| 1. Quizalofop-p- ethyle 5% EC | 100 |
| 2. Atrazine 50% WP | 150 |
| 3. Imazethapyr 10% SL | 250 |
| 4. Metribuzin 70% WP | 500 |
| 5. Propaquizafop 10% EC | |
| 6. Oxyfluorfen 23.5% EC | |
| 7. Pendimethalin 30% EC | |

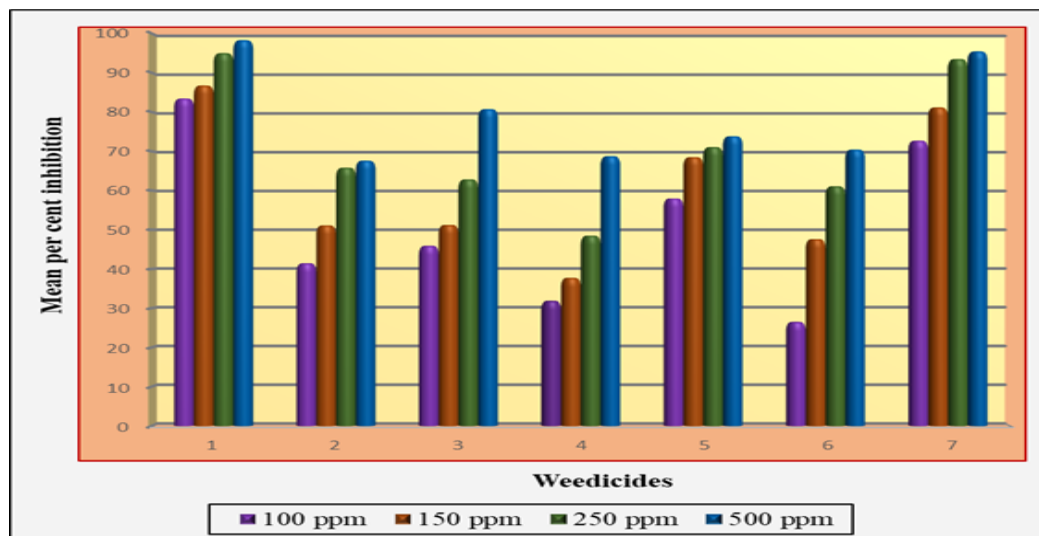


Plate 4: Effect of different weedicides on mycelial growth inhibition of *M. phaseolina* under *in vitro*

Conclusion

Based on present investigation, it can be concluded that groundnut (*Arachis hypogaea* L.) is susceptible to *Macrophomina phaseolina* (Tassi) Goid. And it causes enormous yield loss in groundnut so various chemicals are used to control. Among all chemicals, non-systemic (mancozeb 75% WP and propineb 75% WP); systemic (carbendazim 50% WP); combination fungicide (carboxin 37.5% + thiram 37.5% WP and carbendazim 12% + mancozeb 63% WP) and weedicide (quizalofop-p-ethyle 5% EC) emerged out effective against *Macrophomina phaseolina* *in vitro* condition.

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