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Efficacy of botanicals and bio-agents against web blight caused by *Rhizoctonia solani* (Kuhn) of Mungbean *in vitro*

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Abstract
Web blight caused by *Rhizoctonia solani* (Kuhn) is one of the most important fungal diseases which appear every year in varying intensity and causes heavy reduction in yield. Though the web blight could be managed by the use of fungicide but due to the emergence of several problems like environmental pollution. Residual effect in grains, killing non targeted organisms its use should be discouraged. Hence, for minimizing the losses caused by web blight need in-expensive and environmentally safe management practices. Several botanicals and Bio-agents have been found effective against web blight of mungbean caused by *Rhizoctonia solani.* The botanicals were tested at different doses like 5.0, 7.5 and 10.0 per cent against web blight of Mungbean. Where 10.0 per cent concentration was found most effective which inhibited maximum per cent growth of fungus *i.e.* Garlic (82.53%) and followed by Ginger, Neem, Onion, Tulsi, Clerodendron and Sadabahar. However Per cent Inhibition in all treatments differed significantly to each other. The maximum per cent inhibition was obtained in 5.0, 7.5 and 10.0 per cent concentration in Garlic. Followed by Ginger after 48 hours of incubation. It was very clear from the studies that effectively of extracts increased with the increase in concentration and time of incubation. The per cent inhibition was also higher in *T. viride* (59.13, 68.47 and 71.78 per cent) and *T. harzianum* (49.35, 57.75 and 56.66 per cent) significantly inhibition the growth of *R. solani* as compared to control at 24, 36 and 48 hours of incubation. *Trichoderma. viride* was better in inhibiting radial growth of *R. solani* as compared to *T. harzianum in vitro.*

Keywords: Botanicals, bio-agents, *Rhizoctonia solani*, Mungbean

Introduction
Pulses constitute an important ingredient of the predominantly vegetarian diet in Indian. Production of pulses in the country is far below the requirement to meet even the minimum level of per capita consumption. According to Indian Council of Medical Research (ICMR) optimum requirement of the pulse for a person to maintain his normal health is 104 g per day. However, not even half of this quality is available to people, which is causing malnutrition among the growing people. Therefore, it is necessary that agricultural scientists should evolve the strategy of increasing the production of pulses to meet the protein requirement of increasing population of the country.

In 1924, web blight was reported for the first time on mungbean from Philippines (Nacien, 1924) [10]. While in India, Dwivedi and Saksena (1974) [4] first reported it on mungbean from Kanpur, Uttar Pradesh. Further, it has also been reported from Assam (Saikia, 1976), Punjab (Bains *et al*., 1988) [2], Madhya Pradesh (Tiwari and Khare, 1998) [12], Bihar, Rajasthan, Haryana, Himachal Pradesh and Jammu & Kashmir (Anonymous, 2004) [1]. The pathogen causes huge losses in yield of mungbean and urdbean in India (Dubey 2003) [3].

Web blight caused by *Rhizoctonia solani* (Kuhn) is one of the most important fungal diseases which appear every year in varying intensity and causes heavy reduction in yield. Though the web blight could be managed by the use of fungicide but due to the emergence of several problems like environmental pollution. Residual effect in grains, killing non targeted organisms its use should be discouraged. Hence, for minimizing the losses caused by web blight need in-expensive and environmentally safe management practices. Several botanicals and Bio-agents have been found effective for management of different crops including web blight of mungbean caused by *Rhizoctonia solani.*

Materials and Methods

Efficacy of plant extracts against *R. solani*

In order of find out the efficacy of various plant extracts against *R. solani*, sixteen plants extract *viz*., leaves of Neem, Tulsi, Sadabahar, Clerodendron, bulbs of Garlic, onion and rhizome of Ginger were used. Fresh leaves, bulb and rhizome were collected and washed thoroughly in clean water. Hundred gram of each washed plant material was grinded in Pestle and Mortar by adding equal amounts (100 ml) of sterilized water (1:I w/v) and boiled at 80 °C for 10 minute in hot water bath. The material was filtered through double layered muslin cloth followed by filtering through sterilized Whatman No.1 filter paper and treated as standard plant extract (100 per cent). The 5.0, 7.5 and 10.0 percent concentration were made by adding in requisite amount of sterilized PDA medium.

All the plant extracts were tested at 5.0,7.5 and 10.00 percent concentration under *in vitro* condition by using poison food technique to study the inhibitory effect of these botanicals on mycelial growth of *R. solani*, 7.5 and 10.0 ml plant extract of each stock solution were added to the 95.0.92.5 and 90.0 ml of sterilized cooled PDA medium. The flasks were thoroughly shaken to get uniform mix of the extract under aseptic condition before pouring it into the Petri dishes. Twenty ml medium was poured into each Petri dishes. Sixteen treatments having four replications were maintained. Control treatment was maintained by pouring PDA medium without plant extracts. Five mm discs of 3 days old culture of *R. solani* were cut with sterilized cork borer and placed in the centre of plant extracts amended Petri dishes. The Petri dishes having PDA alone were inoculated in the same manner. These Petri dishes were incubated at 26±1℃. The observations were recorded on radial growth at 24, 36 and 48 hours of incubation in plant extracts amended Petri dishes as well as in control.

Per cent growth inhibition was calculated by using formula (Vincent, 1947).

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

Where,

I = Per cent inhibition of fungal growth

C = Radial growth of control

T = Radial growth in treated Petri dish

Results and Discussion

At 24 hours of incubation

In 5.0 per cent concentration the maximum per cent inhibition was recorded in Garlic (59.08%) followed by Ginger (56.92%), Neem (50.27%), Onion (48.66%), Tulsi (46.91%), Clerodendron (34.00%), Sadabahar (28.12%). The per cent inhibition in Garlic and Ginger, Tulsi and Onion and Neem, were at par to each other, while Clerodendron, Sadabahar differed significantly to each other (Table 4). The similar pattern were obtained in 7.5 per cent concentration and per cent inhibition ranged from 66.0% to 36.00%. Among 10.0 per cent concentration the maximum per cent inhibition was recorded in Garlic (79.52%) followed by Ginger, Neem, Onion, Tulsi, Clerodendron, Sadabahar, however Onion and Neem were at par to each other while Per cent inhibition in all the treatments differed significantly to each other (Table 4). Thus, it is very clear that Garlic was most effective in suppressing the growth of *Rhizoctonia solani in vitro* at different concentration.

At 36 hours of incubation

In 5.0 per cent concentration minimum radial growth was obtained in Garlic (26.51 mm) followed by Ginger (28.00 mm), Neem (29.51 mm). Onion (31.01 mm), Tulsi (33.05 mm), Clerodendron (37.00 mm), Sadabhar (41.01mm), as compared to control (45.26 mm), however Garlic and Ginger, Ginger and Neem, Neem and onion were at par to each other, while Tulsi, Clerodendron and Sadabahar were significantly different to each other (Table 2). The similar results were obtained in 7.5 per cent concentration as 5.0 per cent concentration and the radial growth ranged from 20.50 mm to 72.00 mm. However Garlic, Ginger, Neem, Clerodendron and Sadabahar were statistically differed to each other, while Tulsi and Onion were at par to each other (Table 2). At 10.0 per cent concentration lowest radial growth was obtained in Garlic (14.01 mm) followed by Ginger, Neem, Onion, Tulsi, Clerodendron and Sadabahar. However Garlic, Ginger, Tulsi, Clerodendron and Sadabahar were significantly different to each other, while Neem and Onion were at par to each other (Table 2).

At 48 hours of incubation

In 5.0 per cent concentration the minimum radial growth was obtained in Garlic (29.50 mm) followed by Ginger (32.0 mm), Neem (34.00 mm), Onion (36.10 mm), Tulsi (38.00 mm), Clerodendron (42.50mm) and Sadabahar (50.25 mm). Which were significantly different from each other (Table 3). The similar pattern was obtained in 7.5 per cent concentration and radial growth ranged from 25.00 mm to 89.00 mm. Every treatment statistically differed to each other (Table 3). Among 10.0 per cent concentration minimum radial growth was observed in Garlic (15.5 mm), followed by Ginger, Neem, Onion, Tulsi, Clerodendron and Sadabahar which were significantly superior to each other (Table 3). Minimum radial growth was obtained in 5.0, 7.5 and 10.0 per cent concentration in Garlic. The results clearly indicated that plants extracts reduced the radial growth of *R. solani* at 5.0, 7.5 and 10.0 per cent concentration after 24, 36 and 48 hours of incubation and effectiveness of extracts increased with the increase of their concentration.

Efficacy of plant extract against *R. solani* on per cent inhibition.

At 24 hours of incubation

At 5.0 per cent concentration the maximum per cent inhibition was recorded in Garlic (59.08%) followed by Ginger (56.92%), Neem (50.27%), Onion (48.66%), Tulsi (46.91%), Clerodendron (34.00%), Sadabahar (28.12%). The per cent inhibition in Garlic and Ginger, Tulsi and Onion Neem, were at par to each other (Table 4). The similar pattern were obtained in 7.5 per cent concentration and per cent inhibition ranged from 66.0% to 36.00% Garlic and Ginger, Onion and Neem, were at par to each other (Table 4). Among 10.0 per cent concentration the maximum per cent inhibition was recorded in Garlic (79.52%) followed by Ginger (75.66%), Neem (70.18%), Onion (66.84%), Tulsi (62.42%), Clerodendron (49.16%), Sadabahar (40.30%), however Onion and Neem were at par to each other while Per cent inhibition in all the other treatments differed significantly to each other (Table 4). The maximum per cent inhibition was obtained in 5.0, 7.5 and 10.0 per cent concentration in Garlic after 24 hours in incubation.

At 36 hours of incubation

At five per cent concentration the maximum per cent inhibition in mycelium growth of R. solani was recorded in Garlic (41.44%), followed by Ginger (38.11%), Neem (34.81%), Onion (31.44%), Tulsi (27.00%), Clerodendron (18.23%) and Sadabahar (11.60%). The per cent inhibition in all the treatments differed significantly to each other (Table 5). Similar pattern were obtained in 7.5 per cent concentration and per cent inhibition ranged from 72.00% to 44.00%. However Tulsi and Onion were at par to each other; all other treatments significantly differed to each other (Table 5). Among 10.0 per cent concentration maximum per cent inhibition was recorded in Garlic (80.32%) and followed by Ginger, Neem, Onion, Tulsi, Clerodendron and Sadabahar. However Per cent inhibition in Onion and Neem were at par to each other (Table 5). The maximum per cent inhibition was obtained in 5.0, 7.5 and 10.0 per cent concentration in Garlic followed by Ginger, Neem, Onion, Tulsi, Clerodendron and Sadabahar after 36 hours of incubation.

At 48 hours of incubation

In 5.0 per cent concentration the maximum per cent inhibition in mycelium growth of *R. solani* was recorded in Garlic (66.77%), followed by Ginger (63.94%), Neem (61.69%), Onion (59.10%), Tulsi (57.19%), Clerodendron (52.11%) and Sadabahar (44.79%) The per cent inhibition in all the treatments differed significantly to each other (Table 6). Similar pattern were obtained in 7.5 per cent concentration and per cent inhibition ranged from 72.00% to 47.00%. However all treatments significantly differed to each other (Table 6). Among 10.0 per cent concentration maximum per cent inhibition was recorded in Garlic (82.53%) and followed by Ginger, Neem, Onion, Tulsi, Clerodendron and Sadabahar. However Per cent inhibition in all treatments differed significantly to each other (Table 6). The maximum per cent inhibition was obtained in 5.0, 7.5 and 10.0 per cent concentration in Garlic followed by Ginger after 48 hours of incubation. It was very clear from the studies that effectively of extracts increased with the increase in concentration and time of incubation.

Though, the effect of different plant extracts are lacking specifically in mungbean caused by *R. solani*. However, different reports are available in literature against *R. solani* causing different diseases in different crops.

Meena *et al*. (2002) [7] were also found that extract of Garlic at 5.0 per cent concentration (w/v) completely inhibited the mycelial growth of *R. solani* causing sheath blight of rice and similarly Shinde and Patel (2004) [11] reported that bulb extract of Garlic gave hundred per cent inhibition of mycelia growth of R solani causing black scurf of potato followed by Ginger, Tulsi, Eucalyptus and Neem. Mittal and Goswami (2004) [9] also found that in lab condition, Garlic extract completely inhibited the mycelia growth of *R. solani* causing black scurf of potato, followed by Eucalyptus, Tulsi, bulb extract of onion, rhizome extract. Yadav (2007) [13] reported extract of Garlic gave maximum per cent inhibition in mycelial growth of *R. solani* causing web blight of French bean, followed by Ginger, Neem. Onion, Datura, Tulsi, Eucalyptus and Congress grass. This is in agreement with present findings. In contrary to present findings Mishra *et al.* (2005) [8] found Ginger was most effective in inhibiting radial growth of *R. solani* as compared to *Calotropis gigantia, Vinca rosea, Ocimum sanctum, Azadirachta indica, Pongamia pinnata, Lantana camara, Eucalyptus citriodora, Allitun cepa in vitro.*

Table 1: Effect of different Concentration of plant extracts against *R. solani* on mycelia growth *in vitro* at 24 hrs.

Plant extract	Mycelia growth (mm) Concentration (%)		
	5.00	**7.50**	**10.00**
Neem	22.51	18.51	13.51
Garlic	18.50	15.51	9.26
Tulsi	24.01	22.01	17.01
Onion	23.25	20.01	15.01
Ginger	19.75	16.34	11.00
Sadabahar	32.51	29.01	27.01
Clerodendron	29.51	25.51	23.01
Control	45.26	45.26	45.26
CD at 5%	1.83	1.783	1.728

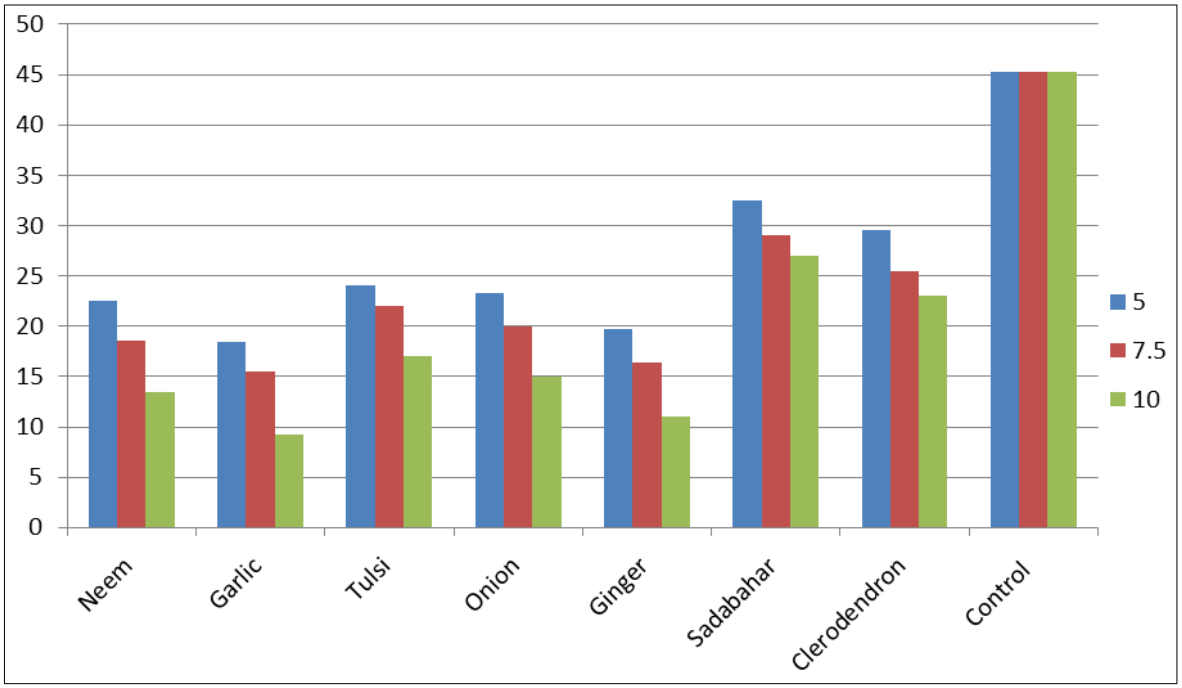

Fig 1: Effect of different Concentration of plant extracts against *R. solani* on mycelia growth *in vitro* at 24 hrs.

Table 2: Effect of different Concentration of plant extracts against *R. solani* on mycelia growth *in vitro* at 36 hrs.

Plant extract	Mycelia growth (mm)		
	Concentration (%)		
	5.00	**7.50**	**10.00**
Neem	29.51	25.02	18.51
Garlic	26.51	20.58	14.01
Tulsi	33.05	29.88	22.00
Onion	31.01	28.13	20.00
Ginger	28.00	22.88	16.00
Sadabahar	41.01	39.63	32.00
Clerodendron	37.00	34.88	27.0
Control	45.26	71.76	71.26
CD at 5%	1.723	1.752	1.732

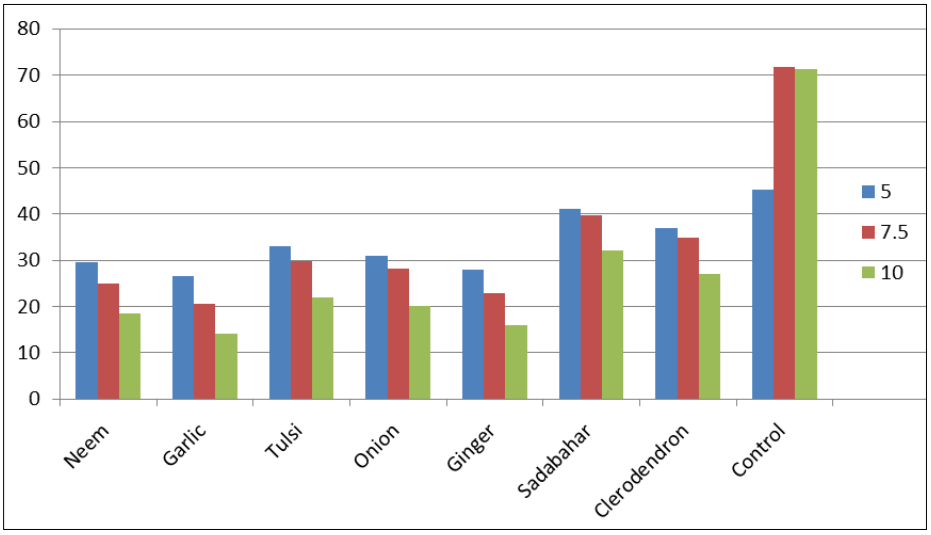

Fig 2: Effect of different Concentration of plant extracts against *R. solani* on mycelia growth *in vitro* at 36 hrs.

Table 3: Effect of different Concentration of plant extracts against *R. solani* on mycelia growth *in vitro* at 48 hrs.

Plant extract	Mycelia growth (mm)		
	Concentration (%)		
	5.00	**7.50**	**10.00**
Neem	34.0	31.5	20.1
Garlic	29.5	25.13	15.5
Tulsi	38.00	36.3	24.6
Onion	36.1	34.1	22.2
Ginger	32.0	27.6	17.8
Sadabahar	49.0	47.1	33.6
Clerodendron	42.5	40.8	29.3
Control	88.76	88.76	88.76
CD at 5%	1.730	1.731	1.735

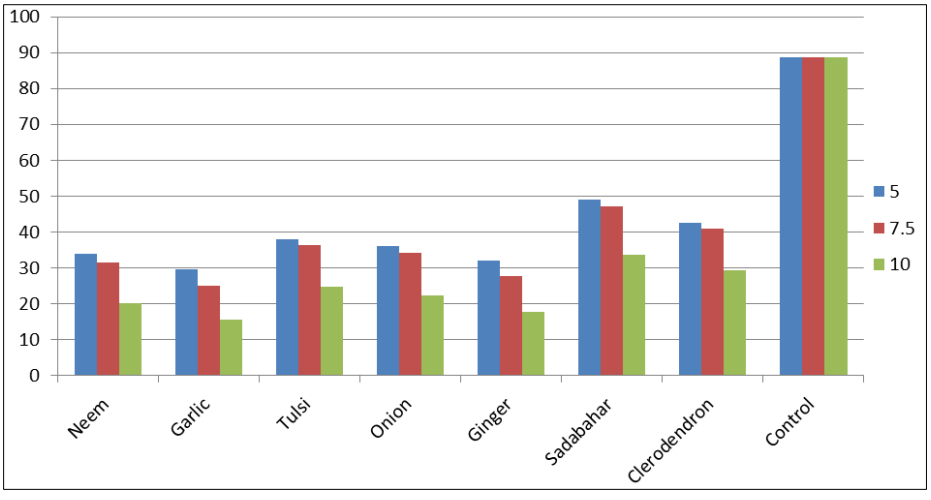

Fig 3: Effect of different Concentration of plant extracts against *R. solani* on mycelia growth *in vitro* at 48 hrs.

Table 4: Effect of different Concentration of plant extracts against *R. solani* on per cent inhibition *in vitro* at 24 hrs.

Plant extract	Percent inhibition		
	Concentration (%)		
	5.00	**7.50**	**10.00**
Neem	50.27(45.153)	59.11(50.250)	70.18(56.904)
Garlic	59.08(50.233)	65.71(54.164)	79.52(63.116)
Tulsi	46.91(43.226)	51.27(45.727)	62.42(52.197)
Onion	48.66(44.232)	55.75(48.307)	66.84(54.851)
Ginger	56.92(48.983)	63.86(53.058)	75.66(60.458)
Sadabahar	28.18(32.060)	35.91(36.813)	40.30(39.404)
Clerodendron	34.80(36.152)	43.59(41.315)	49.16(44.512)
Control	0.00(1.281)	0.00(1.281)	0.00(1.281)
CD at 5%	1.996	2.452	2.627

Figure given in parenthesis are transformed value.

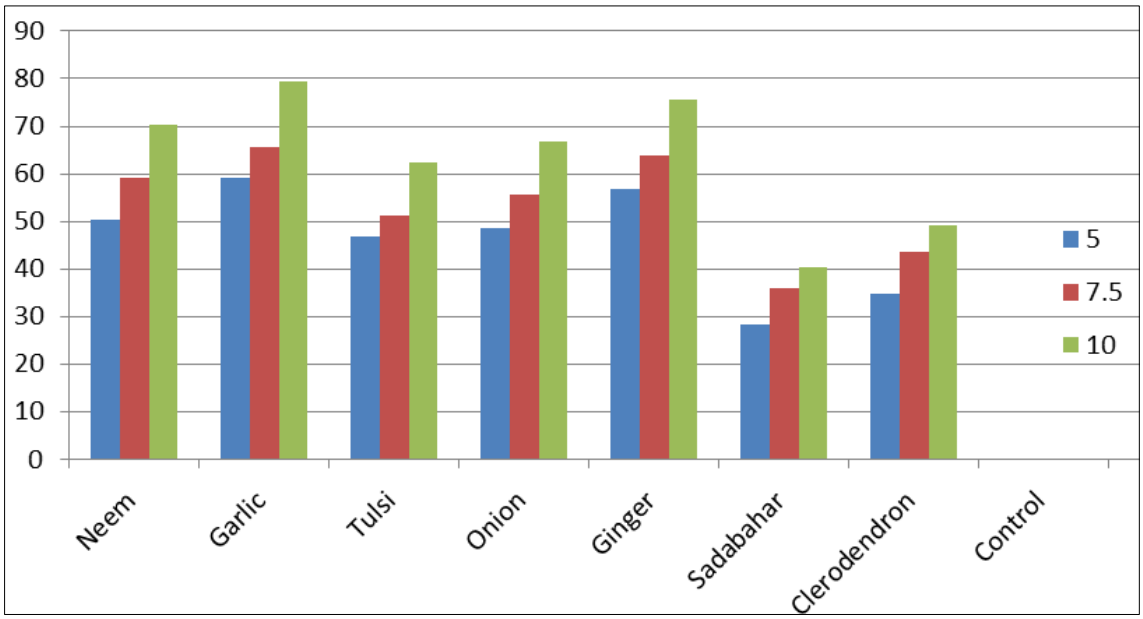

Fig 4: Effect of different Concentration of plant extracts against *R. solani* on per cent inhibition *in vitro* at 24 hrs.

Table 5: Effect of different Concentration of plant extracts against *R. solani* on per cent inhibition *in vitro* at 36 hrs.

Plant extract	Percent inhibition		
	Concentration (%)		
	5.00	**7.50**	**10.00**
Neem	34.81(36.156)	65.14(53.814)	74.03(59.364)
Garlic	41.44(40.073)	71.35(57.645)	80.32(63.683)
Tulsi	27.00(31.310)	58.38(49.827)	69.12(56.24)
Onion	31.44(34.088)	56.79(48.907)	71.92(58.009)
Ginger	38.11(38.120)	68.11(55.618)	77.53(61.716)
Sadabahar	11.60(19.909)	44.75(41.986)	55.08(47.919)
Clerodendron	18.23(25.251)	51.41(45.808)	62.09(52.000)
Control	0.00(1.281)	0.00(1.281)	0.00(1.281)
CD at 5%	1.746	1.498	1.724

Figure given in parenthesis are transformed value

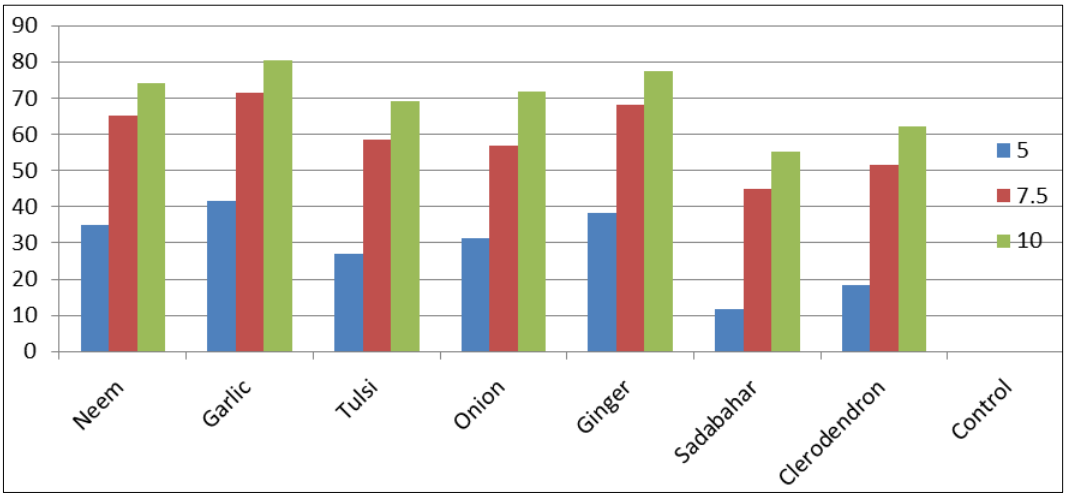

Fig 5: Effect of different Concentration of plant extracts against *R. solani* on per cent inhibition *in vitro* at 36 hrs.

Table 6: Effect of different Concentration of plant extracts against *R. solani* on per cent inhibition *in vitro* at 48 hrs.

Plant extract	Percent inhibition		
	Concentration (%)		
	5.00	7.50	10.00
Neem	61.69(51.726)	64.50(53.433)	77.36(53.433)
Garlic	66.77(54.799)	71.72(57.433)	82.53(57.878)
Tulsi	57.19(49.132)	59.10(50.246)	72.26(50.246)
Onion	59.33(50.376)	61.57(51.694)	74.98(51.694)
Ginger	63.94(53.095)	68.90(56.106)	79.94(56.106)
Sadabahar	44.79(42.007)	46.94(43.246)	62.15(43.246)
Clerodendron	52.11(46.208)	54.03(47.314)	67.01(47.314)
Control	0.00(1.281)	0.00(1.281)	0.00(1.281)
CD at 5%	1.174	1.093	1.093

Figure given in parenthesis are transformed value.

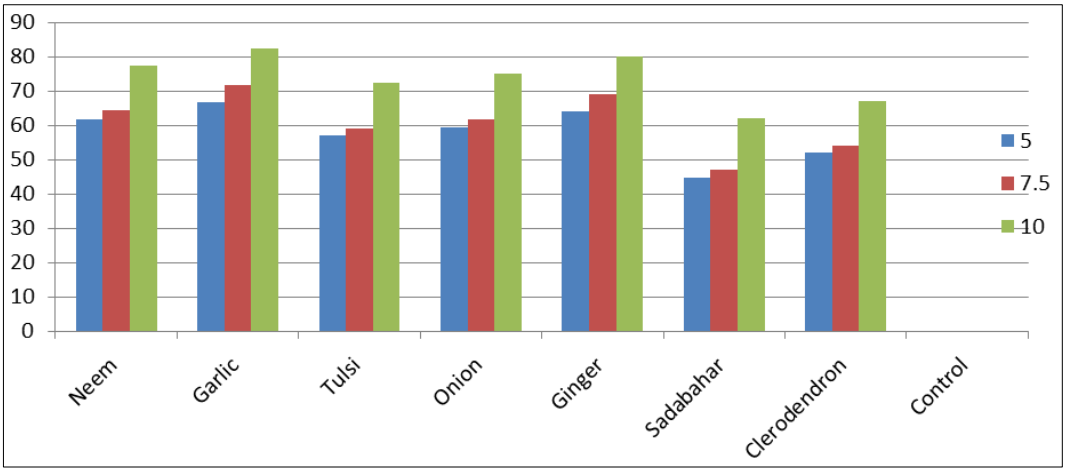

Fig 6: Effect of different Concentration of plant extracts against *R. solani* on per cent inhibition *in vitro* at 48 hrs.

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