Section A -Research paper



Efficacy of Botanicals, Organic amendments and Biocides against Groundnut Stem Rot pathogen *Sclerotium rolfsii* Sacc.

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ABSTRACT

Stem rot infection of peanut (Arachis hypogaea L.) encouraged by Sclerotium rolfsii Sacc. is acutely destructive disease in groundnut cultivated areas of the world. These examinations were executed to test the potency of aqueous extracts of 9 botanicals, 9 organic amendments and 9 antagonists against Sclerotium rolfsii Sacc inciting stem rot of groundnut in in-vitro during the period 2020-21. Among plant aqueous extracts of tested Garlic (Allium sativum) was recorded least fungal mycelial growth 0.20 mm and 0.10 mm at 15% and 20% concentrations respectively in contrast to control. The next greatest treatment against pathogen is C. longa (50.33 mm and 40.00 mm) followed L. camara (51.00 mm and 42.00 mm), E. globulus (72.66 mm and 60.33 mm), S. indicum (74.30 mm and 71.10 mm), M. pinnata (80.00 mm and 53.66 mm), Z. officianale (80.33 mm and 42.66 mm), A. cepa (87.33 mm and 61.00 mm) and A. indica (87.66 mm and 67.33 mm). In organic amendments fine decomposed Farm Yard Manure (FYM) and Groundnut oil cake were recorded maximum inhibited (100 percent) fungal mycelial growth for S. rolfsii followed by Mustard oil cake (88.88%), Sesamum (83.33%), Cotton (72.22%), Neem cake (70.36%), Karanj (60.74%) and Vermicompost (60.00%) which were importantly higher than control (0.00%). Bacillus subtilis which was most successful with least radial fungal mycelial growth (35.30 mm) with overhead mycelial zone of inhibition (60.70%) and least percent disease incidence (11.95) of the test organism and notably superior over all the biocides tested in *in-vitro* and *in- vivo* (pot assay) conditions respectively.

Keywords: Groundnut, Stem rot, botanicals, organic amendments, antagonists, S. rolfsii

INTRODUCTION

Peanut (*Arachis hypogaea* L.) excellent important economic oilseed crops in the world as well as cash crop of our country. It is also called as wonder nut and poor men's cashew nut. It is a low-priced commodity, but a valuable source of all the essential nutrients. It is also used in the manufacturing of lubricants, cakes, butter, soaps, shaving creams, cosmetics and lubricants. Narendra Kumar (1966) revealed that due to year-round cultivation, groundnut crop is subjected to various diseases produced by fungi, bacteria, virus, phytoplasma and phytonematodes etc. Out of various diseases of groundnut, soil borne disease stem rot incited by *Sclerotium rolfsii* (Sacc.) has been a major problem in growing regions; it causes economic yield and worth losses in different parts of India and throughout the world.

It is also called as southern-blight, southern-stem rot, sclerotium rot or white mold and is widely covered in India and USA. In the first half of 20th Century, peanut production sustained economic yield losses of 10 to 20 million dollars annually due to this disease. Soil borne nature of disease is especially very difficult to manage of convenient almost difficulty of scattering fungicides between the peanut awning to the soil profile. The chemical control method developed for disease management guidance too has its own restrictions like high resources interest, non-remunerative, poor availability, toxicity, selectivity, temporary effect, efficacy affected by morpho-physico-chemicals and biological factors, development of disease/pest tolerance, pollutant of food, feeds, health hazards, soil, water and air environmental pollution besides causing deleterious results on human health and biosphere.

In India, this pathogen causes appreciable losses in betel-vine, bean, beetroot, carrot, chilli, cucurbit, peanut, potato and spinach etc. Anamorph: *Sclerotium rolfsii* (Sacc) Teleomorph: *Athelia rolfsii* (Curzi) Tu and Kimbrough is a destructive fungal plant pathogen causes diseases in many monocotyledon and dicotyledon plants encompassing also 500 host species (Agrios (2005), Aycock (1966) and Punja (1985).

In spite of continual research over the earlier time century since its first report on guidance of the pathogen has remained a challenge. Guidance endeavors have often met with definite success, partially suitable to the wide host range, outline growth and capability of the fungal pathogenic organism to produce huge number of sclerotia bodies that may endure viable in soil for many years. Furthermore, management measures effective for a particular groundnut crop in an area may not be adaptable elsewhere owing to regulatory or economic constraints. Sprayings of chemical fungicides possess serious risk to beneficial microorganisms of eco-system. Since long, researchers are using aqueous extracts of botanicals, organic amendments and biocides are to manage plant disease.

Hence, the present experiment was undertaken to exploit the *in-vitro* efficacy of five species of *Trichoderma* for the keeping of stem rot infection of groundnut incited by *S. rolfsii* as test pathogen. *In-vitro* efficiency of phytoextracts, organic amendments and biocides are exploited within framework of integrated disease management.

MATERIALS AND METHODS

Isolation and mass multiplication of S. rolfsii Sacc.

Isolation and kept for the stem rot disease pathogen of ground nut *S. rolfsii* can be isolated from different plant growing parts viz., collar region of affected portion of the groundnut plants by tissue segment method. The disease at basal region of groundnut plants expressing stem rot symptoms from Prakasam district. The test pathogen was decontaminated by single hyphal tip procedure and identified as *Sclerotium rolfsii* Sacc (Rangaswami and Mahadevan (1999). The test organism pure culture was preserved on PDA for further investigations.

Evaluation of aqueous extracts of botanical parts for S. rolfsii Sacc

Nine locally available aqueous extracts of botanical plants were put out for the test organism in *invitro* to examine the inhibitory zone effect on mycelial growth (Table-1). Different parts of plants including leaves, rhizome, bulb, clove, inflorescence and others to be tested were first thoroughly washed, exterior disinfected with 2 percent sodium hypochlorite cleaned with disinfected distilled water and then air-dried. Fresh and chopped plant sample (100g) was lay hold of in a beaker containing water (100 ml) and boiled at 80°C to the 10 min in hot water bath. The material was homogenized to for 5 min, filtrated along muslin cloth and filtrate was centrifuged at 5000 rpm to the 15 min. The clear supernatant was collected. This was examined as 100 percent basic stock. To estimate of antifungal activities of the extracts, desired concentrations of 15 and 20 percents were examined for *S. rolfsii* with food poisoned

technique. Manmohan and Govindaiah (2012) and Nene and Thapliyal (1965). To avoid bacterial contamination, a pinch of streptomycin sulphate was added at the time of discharging of media in Petriplate. Three replications were kept-up for each aqueous botanical extract. The mycelial colony diameter was noticed and percent inhibition zone for *S. rolfsii* over check was evaluated in dual culture procedure at Department of Botany, College of Science and Technology, Andhra University, Visakapatnam, Andhra Pradesh. The effectiveness of botanical extracts was conveyed as percent inhibition data of radial mycelial growth over the controls which were deliberated by using the following formula (Vincent (1947).

Percent inhibition of fungal mycelial growth (I) inhibition for test organism was computed by the following method

Percent growth inhibition (I) =
$$\frac{C-T}{C} \times 100$$

Where,

I = Percent Inhibition/reduction in radial mycelial growth of the test pathogen;

C = The pathogen radial mycelial growth of (mm) in control after incubation;

T = The pathogen radial mycelial growth of (mm) in treatment after incubation;

S. No	Treatments	Botanical name	Plant part used	Concentratio	ns used (%)
1	Turmeric	Curcuma longa	Rhizome	15	20
2	Wild sage	Lantana camara	Inflorescence	15	20
3	Zinger	Zingiber officinale	Rhizome	15	20
4	Onion	Allium cepa	Bulb	15	20
5	Garlic	Allium sativum	Clove	15	20
6	Neem	Azadirachta indica	Leaves	15	20
7	Karanj	Millettia pinnata	Leaves	15	20
8	Eucalyptus	Eucalyptus globulus	Leaves	15	20
9	Sesamum	Sesamum indicum	Leaves	15	20
10	Control	-	-	-	-
	(Untreated)				

Table-1: Aqueous extracts of botanicals used for test pathogen S. rolfsii Sacc.

Further, matching square conversion values are created for facts and scrutiny statistically. The data gained *in-vitro* on percent growth inhibition zone of test organism was examined with completely randomized design (CRD) method.

In-vitro evaluation of organic amendments on mycelial growth (mm) of S. rolfsü Sacc.

In-vitro assessment of nine organic amendments with fine powder of fifty grams were captured into 250 ml conical flasks and 150 ml distilled water was added to each conical vessel and permitted to decomposed for 15 days then stained with muslin cloth to secure the extract and autoclaved at 1.2 Kg/cm² pressure to 20 min considered as 100 percent standard solution (Table-2). 50 ml quantity organic extracts of standard stock solution were embodied separately in melted sterilized PDA medium in Petriplates aseptically at the time of pouring the medium to gained desired 10 percent concentrations. After solidification, the Petriplates were injected in the centre by placing 5 mm mycelial disc at 6 days old actively developed test pathogen pure culture of S. rolfsii and keep up with 3 replications. The tests of organic amendments were demonstrated as percent of fungal mycelial radial growth over the check which was computed by applying the method as narrated earlier.

S. No	Treatments	Botanical name	Dosage (in Percent)	
1	Neem	Azadirachta indica	10	
2	Karanj	Millettia pinnata	10	
3	Ground nut	Arachis hypogaea	10	
4	Mustard	Brassica juncea	10	
5	Cotton	Gossypium hirsutum	10	
6	Sesamum	Sesamum indicum	10	
7	Castor	Ricinus communis	10	
8	FYM	-	10	
9	Vermicompost	-	10	
10	Control (Untreated)	-	-	

Table-2: Efficacy of organic amendments for S. rolfsii Sacc.

Antagonistic effect of bio-control agents/biocides for S. rolfsü Sacc.

In-vitro studies of biocides to the antagonistic activity of six *Trichoderma* spp., one *Aspergillus* spp. and two bacterial biocides for *S. rolfsii* using dual culture method as described by Coskuntuna and Ozer (2008) (Table-3). Potato dextrose agar (PDA) was use as medium and Petriplates were equally divided into two portions (Karunanithi and Usman, 1999). In the first half, actively growing culture of bio agent (5 mm) was incubated while in the opposite side (5mm) of each *S. rolfsii* isolates were placed. Three replicates were continued and experiments were repeated (twice) for one and all treatment along check was maintained without antagonist. All incubated Petriplates were kept at $25\pm2^{\circ}$ C until for *S. rolfsii* isolates in the control to reach to the edge of Petri dish at Department of Botany, College of Science and Technology, Andhra University, Visakapatnam, Andhra Pradesh. Data were recorded by using method.

The percentage inhibition of fungal mycelial radial growth was estimated

Percent growth inhibition (I) =
$$\frac{C-T}{C} \times 100$$

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S. No	Treatments	Type of biocide
1	Trichoderma asperellum	Fungal
2	T.harzianum	Fungal
3	T. hamatum	Fungal
4	T. longibrachiatum	Fungal
5	T. koningii	Fungal
6	T. atroviride	Fungal
7	Aspergillus niger	Fungal
8	Pseudomonas fluorescens	Bacterial
9	Bacillus subtilis	Bacterial
10	Control (No Bioagent)	_

Table-3: In-vitro test of biocides for fungal mycelial growth and inhibition zone of S.rolfsii Sacc.

Stem rot incidence percentage in different treatments in pot assay under greenhouse

To find out the potency of biocides against *S. rolfsii* in a pot assay was conducted under the greenhouse by applying randomized block design (RBD) with three replications, 10 treatments, $T_1,T_2,T_3,T_4,T_5,T_6,T_7,T_8,T_9$ are treated with biocides and T_{10} -untreated without biocide as a control (Table-4) in poly houses at Kothapatnam, Prakasam district. The disease infected symptoms were expressed on susceptible cultivar K-6 were used as standard check for comparison of yellowing, partial and complete vascular wilting of plant or branches. The culture of test fungi (*S. rolfsii* Sacc.) were multiplication with disinfected sorghum grains and sand; maize medium, respectively. Earthen pots were disinfestations with 5% solution of copper sulphate and loaded with autoclaved potting mixture of soil: sand: FYM (2:1:1). Those pots were served with pure culture of *S. rolfsii* watered slightly and injected at room temperature for a 2-3 week. The data on disease infected symptoms were recorded at 15 days intervals after 30 days of sowing. The seeds that as did not germinated were recorded as pre exigency seedling mortality and post exigency seedling mortality was calculated on the theory of germinate seedlings at 30, 45, 60, 75, 90 and 105 days after sowing and final plant stand was listed on the basis of total number of plants disease when seeds are sown. Serious infected plants showed drying of whole plant with damaged twigs and pods with discoloration and whitish mycelial growth on pod and seeds (Lukose *et al.*,2008).

Table-4. In-vivo test of bioclues for germination and minibition of b. roijsu bace	Table-4:	: In-vivo	test of bi	ocides for	germination	and inhibition	of S. rolfsii Saco
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S. No	Treatments	Dosage of biocide (in g/Kg of seed)
1	Trichoderma asperellum	10
2	T. harzianum	10
3	T. hamatum	10
4	T. longibrachiatum	10
5	T. koningii	10
6	T. atroviride	10
7	Aspergillus niger	10
8	Pseudomonas fluorescens	10

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9	Bacillus subtilis	10
10	Control (Treated with water)	

Statistical Analysis

The recorded data was scrutiny through analysis of variance (ANOVA) technique and presented at 5% level of significance (P = 0.05) by the procedure prescribed by Panse and Sukhatme (1967).

RESULTS AND DISCUSSION

In-vitro evaluation of aqueous extracts of botanicals parts for Sclerotium rolfsii Sacc.

Aqueous extracts of nine plant parts by poisoned food technique were evaluated *in-vitro* (each @ 15% and 20%) for *S. rolfsii* Sacc (Table-5) and results explained that the all 9 botanicals part extracts tested were fungistatic to *S. rolfsii* Sacc which significant decreased fungal radial mycelial growth and enhanced its inhibition zone over with the control (Fig-1).



Fig-1. Inhibitory effects of aqueous extracts of plants on mycelial growth of S. rolfsii Sacc.

Table-5: Inhibitory effects of aqueous extracts of plants for S. rolfsii Sacc.

			Phyto extracts concentrations (%)							
S. No			15%			20%				
	Treatments	Botanical name	Radial growth (mm)	% Inhibition	Growth reduction over control	Radial growth (mm)	% Inhibition	Growth reduction over control		
1	Turmeric	Curcuma longa	50.33	44.07	39.67	40.00	55.55	50.00		
2	Wildsage	Lantana camara	51.00	43.30	39.00	42.00	53.33	48.00		

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3	Zinger	Zingiber officinale	80.33	10.74	9.67	42.66	63.71	47.34
4	Onion	Allium cepa	87.33	2.96	2.67	61.00	32.22	29.00
5	Garlic	Allium sativum	0.20	100.00	89.80	0.10	100.00	89.90
6	Neem	Azadirachta indica	87.66	2.60	2.34	67.33	25.18	22.67
7	Karanj	Millettia pinnata	80.00	11.11	10.00	53.66	40.37	36.34
8	Eucalyptus	Eucalyptus globulus	72.66	19.26	17.34	60.33	32.96	29.67
9	Sesamum	Sesamum indicum	74.30	17.40	15.70	71.10	21.00	18.90
10	Control	-	90.00	0.00	-	90.0	0.00	-
	CD (5%)		2.73	1.36		1.63	2.32	
	SE(m) <u>+</u>		0.80	0.40		0.47	0.68	
	CV %		2.37	3.19		2.42	3.21	

The results at 15 percent concentration were revealed that radial fungal mycelial growth of the test organism was ranged from 87.66 mm (*A. indica*), to 50.33 mm, (*C. longa*) as against in control (90.00 mm) Table-5. However, signified minimum fungal radial mycelial out growth was noticed with *A. sativum* extract (0.20 mm) which was found significances superior to inhibit the test fungal pathogen mycelial growth over all other plant/botanical extracts tested. It is followed by viz., *C. longa* (50.33 mm) followed by viz., *L. camara* (51.00 mm), *E. globulus* (72.66 mm), *S. indicum* (74.30), *M. pinnata* (80.00 mm), *Z. officianale* (80.33 mm), *A. cepa* (87.33 mm) and *A. indica* (87.66 mm) other botanicals. The botanicals *A. indica* (87.66 mm) and *A. cepa* (87.33 mm) were found lowest useful for inhibition zone of the test fungal pathogen with maximum fungal radial mycelial out growth of the test pathogen at 15 percent concentration.

The botanicals examined at 20 percent concentrations (Table-5) exhibits nearly foremost trend of fungal radial mycelial out growth during the time at 15 percent concentrations. Significant minimum fungal radial mycelial out growth was noticed with Α. sativum extract (0.10 mm), which was found significances superior to inhibit the test fungal pathogen mycelial growth over all the other botanicals tested. It is ranged from 40.00 mm (C. longa) to 71.00 mm (S. indicum) as against in control (90.00 mm) and C. longa (40.00 mm), followed by L. camara (42.00 mm), Z. officinale (42.66 mm), M. pinnata (53.66 mm) and E. globulus (60.33 mm) other botanicals. S. indicum (71.10 mm) and A. indica (67.33 mm) were found lowest effective for zone of inhibition with maximum of the test organism at 20 percent concentration.

Results of inhibitory work of aqueous extracts of plants on fungal mycelial growth on *S. rolfsii* Sacc is in consonance with such earlier inscribed by several researchers Madhavi and Bhattiprolu (2011), Kumar *et al.*, (2011), Sumi and Tiameren (2015), Rabeya *et al.*, (2016), Kuldhar and Suryawanshi (2017), Murthy *et al.*, (2018) and Sarita, Shankar Soyal and Ratnoo (2018). Botanicals/plant extracts Garlic (*A. sativum*), Onion (*A. cepa*), Nirgudi (*Vitex* spp.), Turmeric (*C. longa*), Tulsi (*Ocimum sanctum*), Ginger (*Z. officinale*), Neem (*A. indica*), Shatavari (*Asparagus racemosus*) were reported earlier as antifungal/fungistatic for *S.rolfsii* (Sacc.) causing disease on groundnut and many other major cultivable crops by several workers. Suryawanshi, (2015), Sneha *et al.*, (2016) and Suranjit *et al.*, (2018).

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In-vitro evaluation of organic amendments on mycelial growth (mm) to S. rolfsii Sacc.

Organic amendments/oil extracts i.e., cakes were tested *in-vitro* for *S. rolfsii* at 10 percent concentrations each.

S. No	Treatments	Botanical name	Dosage (%)	Radial growth (mm)	% Inhibition	Reduced the growth over control
1	Neem	Azadirachta indica	10	26.67	70.36	63.33
2	Karanj	Millettia pinnata	10	35.33	60.74	54.67
3	Ground nut	Arachis hypogaea	10	0.10	100.00	89.90
4	Mustard	Brassica juncea	10	10.00	88.88	80.00
5	Cotton	Gossypium hirsutum	10	25.00	72.22	65.00
6	Sesamum	Sesamum indicum	10	15.00	83.33	75.00
7	Castor	Ricinus communis	10	40.00	55.55	50.00
8	FYM	-	10	0.10	100.00	90.00
9	Vermicompost	-	10	36.00	60.00	54.00
10	Control (Untreated)		-	90.00	0.00	-
	CD (5%)		-	2.24	4.80	-
	SE(m) <u>+</u>	-	0.65	1.40	-	
	CV %	-	4.73	4.07	-	

Table-6: Inhibitory effects of organic cakes for S. rolfsii Sacc.

Results showed that well decomposed FYM and Groundnut oil cake inhibited fungal mycelial outgrowth of S. rolfsii by 100 percent followed by Mustard oil cake (88.88%), Sesamum (83.33%), Cotton (72.22%), Neem cake (70.36%), Karanj (60.74%), Vermicompost (60.00%) and Castor (55.55%) which considerable higher control (Table-6) were than and (Fig-2). Predominant findings have been reported by Senjaliya, (2015) that among 9 different organic extracts (FYM, groundnut, mustard, castor, cotton cakes, neem, karanj, vermicompost and castor) tested all plant extracts significantly and results shows that the reserved for the S. rolfsii in-vitro except castor. However, present study divested that 100 percent inhibition zone by well decomposed FYM. Governance of stem rot infection in groundnut crop has also have been exhibited with different doses of amendments such as organic cakes and FYM by Jonson et al., (2003).

100.00 90.00 80.00 70.00 60.00 50.00 40.00 30.00 Radial growth (mm) 20.00 % Inhibition 10.00 0.00 Gosspurnhrsuturn Azadirachta indica Sesonum indicum Ricinus communis Willettia pinnota Arachis Mpogoed Prosice ince Vermicompost Control

Fig-2. Inhibitory effects of organic cakes (10%) on mycelial growth of pathogen S. rolfsii Sacc.

Antagonistic effect of bio-control agents/biocides for S. rolfsii Sacc.

Results of all the 9 biocides are examined to exhibit fungistatic/antifungal, antibacterial activity for *S. rolfsii* (Sacc.) and significant inhibited zone its fungal mycelial growth over control (Table-7) and (Fig-3).



Fig-3. Inhibitory effects of biocides on mycelial growth of S. rolfsii Sacc. (In-Vitro)

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S. No	Treatments	Colony Dia.(mm)* of pathogen/radial growth (mm)	Inhibition (%)	Reduced the growth over control
1	Trichoderma asperellum	41.45	53.92	48.55
2	T. harzianum	45.02	50.00	44.98
3	T. hamatum	66.38	25.91	23.62
4	T. longibrachiatum	59.43	33.97	30.57
5	T. koningii	86.42	04.00	3.58
6	T. atroviride	70.20	22.00	19.80
7	Aspergillus niger	49.36	45.16	40.64
8	Pseudomonas fluorescens	57.05	36.67	32.95
9	Bacillus subtilis	35.30	60.70	54.70
10	Control (No Bioagent)	90.00	00.00	-
	CD (5%)	2.44	2.40	-
	SE(m) <u>+</u>	0.82	0.80	-
	CV %	2.37	4.21	-

Table-7: Inhibitory effects of biocides for S. rolfsii Sacc.

However, maximum inhibition zone of fungal mycelial out growth was obtained with B. subtilis which was found most successful with lowest radial mycelial growth (35.30 mm) with highest fungal mycelial inhibition zone (60.70 percent) of the fungal pathogen and significantly best over all the biocides towards the test pathogen. (Pawar et al., (2022). The second and third most best antagonists found were T. asperellum (zone of inhibition-53.92 percent and recorded colony mycelial growth diameter 41.45 mm) were followed by T. harzianum (zone of inhibition of 50.00 percent and mycelial colony diameter: 45.02 mm), A. niger (zone of inhibition: 45.16 percent and colony mycelial growth diameter: 49.36 mm), P. fluorescens (zone of inhibition: 36.67 percent and colony mycelial growth diameter: 57.05 mm), T. longibrachiatum (zone of inhibition: 33.97 percent and colony mycelial growth diameter: 59.43 mm), T. hamatum (zone of inhibition: 25.91 percent and colony mycelial growth diameter: 66.38 mm), T. atroviride (zone of inhibition: 22.00 percent and colony mycelial growth 70.20 diameter: mm) and T. koningii (zone of inhibition: 4.00 percent and colony mycelial growth diameter: 86.42 mm).

T. longibrachiatum and *P. fluorescens* were found equal to each other treatments. *T. koningii* was established least effective compared the untreated (control).

In vivo efficacy biocides against groundnut stem rot disease percentage in pot assay

There was significant variation in stem rot disease frequency of groundnut due to antifungal and antibacterial reactions to *S. rolfsii* Sacc (Table-8). *B. subtilis* was found antagonistic potential against *S. rolfsii* and *T. asperellum* attack directly and lyses the fungal mycelium and sclerotial bodies of

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S. rolfsii (Sacc.) by hyphal coiling, entry through haustoria like structure and direct penetration in hyphae and sclerotia gave efficient control over stem rot disease.

G N	Treatments	Stem rot incidence after DAS (%)					Mean	Mean reduction	
S. No		30	45	60	75	90	105	PDI (%)	(%) over control
1	Trichoderma asperellum	25.37	19.20	14.30	10.43	6.40	1.62	12.89	67.25
2	T. harzianum	41.47	35.63	28.87	21.10	14.53	10.62	25.37	35.56
3	T. hamatum	49.53	42.30	38.27	31.57	24.40	15.00	33.51	14.88
4	T. longibrachiatum	22.00	39.67	34.47	28.90	21.10	15.00	26.86	31.77
5	T. koningii	45.33	26.23	21.10	16.60	10.00	7.50	21.13	46.32
6	T. atroviride	32.00	31.17	28.90	27.78	26.86	25.37	19.80	49.70
7	Aspergillus niger	31.17	22.40	18.87	14.43	7.77	5.62	16.71	57.55
8	Pseudomonas fluorescens	26.43	30.27	15.53	11.10	5.57	3.75	15.44	60.78
9	Bacillus subtilis	23.63	18.30	12.27	10.00	4.40	3.12	11.95	69.64
10	Control (Treated with water)	26.62	31.13	36.60	41.24	46.20	54.43	39.37	-
	CD (5%)	2.99	2.72	2.17	1.85	2.85	1.32		
	SE(m) <u>+</u>	1.01	0.92	0.73	0.62	0.95	0.44		
	CV %	5.39	5.35	5.07	5.05	9.95	5.49		

Table-8: Inhibitory effects of biocides for S. rolfsii Sacc in pot assay

Lowest data on percent disease incidence (PDI) was obtained with *B. subtilis* for *S. rolfsii* (Sacc.)(11.95) than control (39.37) Fig-4. It is also recorded with highest disease recovery (69.64 percent) than control of the fungal test pathogen and significantly outstanding over all the biocides treatments (Pawar *et al.*, (2022). The second and third most best antagonists were found with *T. asperellum* (PDI is 12.89) and *P. fluorescens* (PDI is 15.44) which recorded disease recovery (57.55 percent), *T. koningii* PDI is 21.13 with disease recovery (46.32 percent), *T. harzianum* PDI is 25.37 with disease recovery (35.56 percent), *T. longibrachiatum* PDI is 26.86 with disease recovery (31.77 percent) were found at par with each other and *T. hamatum* was found PDI is 33.51 with lowest disease recovery (14.88 percent).

Fig-4. Inhibitory effects of biocides on mycelial growth of S. rolfsii Sacc. Pot assay (In-Vivo)

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Karthikeyan *et al.*, (2006) recorded that one among the five isolates of *Trichoderma* one isolate each of *T. harzianum* and *P. fluorescence* were inhibitory zone to the fungal mycelial radial growth of *S. rolfsii* (Sacc.) the induced for stem rot infection in groundnut that are also supported to our study.

CONCLUSION

In these studies, out of all 9 aqueous botanical extracts A. *sativum* was recorded maximum inhibition zone was found (100 percent) at 15% and 20% concentrates on the part of radial fungal mycelial growth with 0.20 mm and 0.10 mm (*A. sativum*) respectively. In organic amendments out of 9 tested well decomposed FYM and Groundnut oil cake were recorded maximum inhibited zone fungal mycelial out growth (100 percent) of *S. rolfisii*. *B. subtilis* which was most induced with least radial fungal mycelial growth (35.30 mm) with greatest fungal mycelial inhibition zone (60.70%) of the test fungal pathogen and significant lowest percent disease incidence (PDI- 11.95%) compared to overall treatments are evaluated in *in-vitro*, *In-vivo* conditions. The outcome of these study can be further used for formulating Bio Intensive Disease Management schedule of stem rot infection of groundnut.

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