International Journal of Bio-resource and Stress Management https://ojs.pphouse.org/index.php/IJBSM

Crossref

IJBSM August 2024, 15(8): 01-08 **Article** AR5474

Research Article *Natural Resource Management*

DOI: HTTPS://DOI.ORG/10.23910/1.2024.5474

Evaluation of Homoeopathic Drugs against *Sclerotium rolfsii* Sacc. in Groundnut (*Arachis hypogea* L.)

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ABSTRACT

The study was conducted from June to October, 2022 at the Department of Plant Pathology, College of Agriculture, Professor Jayashankar Telangana State Agricultural University, Rajendranagar, Hyderabad, India to screen the thirty homeopathic drugs under in-vitro condition against the groundnut stem rot pathogen *Sclerotium rolfsii*. Bio-efficacy of homeopathic drugs was tested by agar well diffusion method to identify the potential drug against stem rot pathogen. In this method, notably, *Chelidonium* (88.88%) and *Colchicum* (82.21%) emerged as particularly potent, significantly inhibiting *Sclerotium rolfsii* mycelial growth and reducing sclerotial formation compared to the control, while *Acidium phosphoricum* showed a very low mycelial inhibition percentage (13.33%). *Chelidonium* exhibited the lowest sclerotia production (52), followed by *Colchicum* (62), compared to the control (400). Sclerotial initiation delayed in *Arnica montana* (18 days) and *Thuja occidentalis* (14 days), whereas in the control group, it occurred within six days. Early ooze formation from sclerotia was observed in treatments with *Cina* and *Nux vomica* (8 days), while it was delayed in *Arnica montana* (18 days). The cultural characteristics, such as mycelial pattern, sclerotial arrangement, and color of sclerotia, also changed among the treatments. Significant inhibition of sclerotial germination was observed, particularly with *Chelidonium* and *Colchicum* treatments at 15 and 20-minute durations. *Chelidonium* and *Colchicum*, shown significant antifungal properties against the groundnut stem rot pathogen. These findings suggest that, homeopathic drugs could potentially be used to manage stem rot in groundnut crop, reducing the reliance on chemical fungicides.

KEYWORDS: Groundnut stem rot, homoeopathic drugs, *Sclerotium rolfsii*

***Citation* (VANCOUVER):** Sagar et al., Evaluation of Homoeopathic Drugs against *Sclerotium rolfsii* Sacc. in Groundnut (*Arachis hypogea* L.). *International Journal of Bio-resource and Stress Management,* 2024; 15(8), 01-08. HTTPS://DOI.ORG/10.23910/1.2024.5474.

Data Availability Statement: Legal restrictions are imposed on the public sharing of raw data. However, authors have full right to transfer or share the data in raw form upon request subject to either meeting the conditions of the original consents and the original research study. Further, access of data needs to meet whether the user complies with the ethical and legal obligations as data controllers to allow for secondary use of the data outside of the original study.

Conflict of interests: The authors have declared that no conflict of interest exists.

RECEIVED on 23rd May 2024 RECEIVED in revised form on 15th July 2024 ACCEPTED in final form on 30th July 2024 PUBLISHED on 01st August 2024

1. INTRODUCTION

Groundnut (*Arachis hypogea* L.) is an important edible oilseed crop, has a wide range of adaptability to varying Agro-climatic conditions and soils, which made its cultivation possible in most of the tropical and subtropical countries in the world. In India, the major groundnut-growing states are Gujarat, Andhra Pradesh, Telangana, Tamil Nadu, Karnataka and Maharashtra (Meena and Meena, 2023). In Telangana, the cultivated area is 0.74 lakh ha with a production of 0.10 lakh tonnes (Anonymous, 2024). Diseases including seedling rot, leaf spots, rust, stem rot, pod rot, and bud necrosis hamper groundnut production (Akash et al., 2022; Joshi et al., 2020; Hawaladar et al., 2022). Among the soil-borne fungal diseases, stem rot caused by *Sclerotium rolfsii* is a potential threat to successful groundnut cultivation (Priya et al., 2013). The yield losses caused by *Sclerotium rolfsii* generally is 25%, but sometimes it reaches 80 to 90% (Aydogdu, 2023; Goud et al., 2013; Adiver, 2003).

The pathogen is characterized by white fluffy, branched, septate mycelium, and spherical or irregular shaped brown sclerotia, which range from 0.5 to 2.0 mm in diameter and at maturity, resemble mustard seed (Ayyandurai et al., 2022; Kumar et al., 2021). The symptomatology of stem rot disease includes the presence of mycelium enveloping the plant stem close to the soil surface (Motlagh et al., 2022; Wilson, 1953). The disease causes damage on root and stem of groundnut plant (Motlagh et al., 2022; Akash et al., 2022). The pathogenhas an ability to produce a large number of sclerotia which persist in soil for several years (Hawaladar et al., 2022; Akash et al., 2022; Burlakoti, 2012; Goud et al., 2013).

Different practices are recommended for the management of groundnut stem rot disease *viz*., deep summer ploughing, destruction of plant debris, crop rotation with jowar and bajra, seed treatment with contact and trizole group fungicides, soil drenching with hexaconazole, application of ammonium sulphate or calcium ammonium nitrate instead of urea, application of gypsum at flowering stage and biological control (Rani et al., 2023; Sunkad et al., 2016; Kumar et al., 2013; Rudresh et al., 2005). However, the indiscriminate use of chemical fungicides may cause development of resistance among the pathogens. In addition to this, synthetic fungicides also exhibit harmful impacts on the environment. Using homeopathic alternatives to manage plant diseases reduces public worries about the harmful effects of pesticides on crops. Homeopathy in agriculture is transforming traditional farming into agro-ecological practices. Most homeopathic medicines are made from natural herbs. They are generally safe, eco-friendly, and target-specific with minimal side effects, helping to prevent or cure human and plant diseases. (Kumar et al., 2023). Homeopathic medicine helps reduce negative effects on vital energy and restore balance by boosting the plant's defence mechanism (Biswas et al., 2002). To eliminate white mold in bean plants, growers used homeopathic remedies containing phosphorus and *Calcarea carbonica* (Rissato et al., 2016). Hanif and Dawar (2016) reported that *A. montana* significantly inhibited the mycelial growth of *F. oxysporum, Rhizoctonia solani*, and *Macrophomina phaseolina.T. occidentalis*, an evergreen coniferous tree, is widely used in homeopathic medicine for its antiviral, antifungal, antidiarrheal, and antioxidant properties (Alam and Mistry, 2022; Caruntu et al., 2020).

However, there is lack of information on influence of homoeopathic drugs on *Sclerotium rolfsii* development (cultural and morphological characters). Hence, the present study is planned to identify the potential homoeopathic drug against *Sclerotium rolfsii* causing stem rot disease in groundnut.

2. MATERIALS AND METHODS

The study was conducted from June to October, 2022 at the Department of Plant Pathology, College of Agriculture, Professor Jayashankar Telangana State Agricultural University, Rajendranagar, Hyderabad, India.

2.1. Isolation of sclerotium rolfsii

Groundnut (*Arachis hypogea* L.) plants typically showing stem rot symptoms were collected from Seed Research and Technology Centre, PJTSAU, Rajendranagar, Hyderabad, Telangana during June to October, 2022. The pathogen was identified based on mycelial and sclerotial characters mentioned in standard mycological keys (Barnett and Hunter, 1972). Infected stem tissues were surface sterilized aseptically with 1% NaClO (Sodium hypochlorite) for 1 minute followed by three subsequent washing with sterilized distilled water in aseptic condition (Bashyal et al., 2021). Sterilized petri plates (90 mm diameter) were taken and poured with 20 ml of PDA medium at lukewarm condition and the media allowed to solidify. After solidifying the medium, the sterilized stem pieces were transferred aseptically under laminar air flow on sterilized petri plates containing 20 ml of solidified potato dextrose agar medium. The petri plates were incubated in B.O.D at 25±2°C for optimum growth. The fungal hyphae developing from the infected tissues were sub cultured aseptically on petri plates containing PDA media. The pure culture was obtained by hyphal tip method and stored in PDA slants at 4°C for future use (Bashyal et al., 2021).

2.2. In vitro screening of homoeopathic drugs against sclerotium rolfsii

Nearly thirty homoeopathic drugs were evaluated to test

their bio-efficacy against *Sclerotium rolfsii* causing groundnut stem rot by agar well diffusion method (Jimenez-Esquilin et al., 2005). The sterilized PDA medium was poured into pre-sterilised petri plates (90 mm in diameter) (20 ml plate^{-1}) and allowed to solidify. Four wells were cut at four corners of the petri plate with a sterilised cork borer (5mm) and 100 µl of homoeopathic drugs potency 200 C and 300 C were poured in each well. One week old culture of *S. rolfsii* was placed in the middle of the petri plate. PDA plate without homoeopathic drugs serve as control. Each treatment replicated three times. Plates were incubated in BOD at 25±2°C for optimum growth. Mycelial growth of *S. rolfsii* taken daily (mm day^{-1}) until the end of the assay. Mycelial inhibition was measured when the mycelium covers whole plate in the control. The per cent mycelial inhibition was calculated by using the following formula (Vincent, 1927).

PCI=(C-T)/C×100

Where, I=Per cent inhibition of mycelium

C=Mycelial growth in control (un treated)

T=Mycelial growth in treated.

2.3. Homoeopathic drugs impact on sclerotial characters

Influence on the development of sclerotial characters viz.,initiation of sclerotia, ooze formation, maturity of sclerotia, number of sclerotia upon treatment of homeopathic drugs were noted down.The studies were conducted under *in vitro* conditions using PDA media. Each treatment replicated three times (Ayed et al., 2018)

2.4. Cultural and morphological characteristics of the pathogen

The cultural and morphological characteristics of *Sclerotium rolfsii* viz., mycelial growth, sclerotial colour, sclerotial shape, sclerotial arrangement were studied on PDA media under *in vitro*.

2.5. Effect of homoeopathic drugs on sclerotial germination

The efficacy of homoeopathic drugs on the sclerotial germination of *S. rolfsii* was studied by dipping the sclerotia in homoeopathic drugs at different time intervals i.e., 5, 10, 15 and 20 minutes. Five sclerotia were placed on 90mm Petri plates containing PDA media for each treatment, and the plates were then incubated in a BOD at 25±2°C. After 72 hours of incubation, the germination of sclerotia was assessed by counting the number showing outgrowing hyphae. Each treatment was replicated three times, with sclerotia treated with sterile water serving as the control. The percentage of sclerotial germination was calculated based on the number of germinated sclerotia in each treatment (Ayed et al., 2018).

2.6. Statistical analysis

Data were analysed as (ANOVA) analysis of variance by using OPSTAT software one and two factor analysis in Complete Randomised Design. All the data were considered as significant at the level of $p \leq 0.05$, results were presented±standard error.

3. RESULTS AND DISCUSSION

3.1. Mycelial inhibition of sclerotium rolfsii

The effect of homoeopathic drugs on mycelial growth of *Sclerotium rolfsii* was studied by using agar well diffusion method. The homoeopathic drugs was found more effective in inhibiting the mycelial growth of *Sclerotium rolfsii* under *in vitro* (Table 1). Maximum inhibition was recorded by *Chelidonium* (88.88%) followed by *Colchicum* (82.21%) and *Natrum muriaticumn* (81.47%), *Thuja occidentalis* (80.66%), *Arnica montana* (80.66%)and *Kali iodatum* (80.25%), *Chelidonium*, *Iodium* was also effective compare to control while in *Acidium phosphoricum* mycelia inhibition percentage was very less (13.33%). The significant inhibition of mycelial growth of *S. rolfsii* treated with homoeopathic drugs (*Chelidonium colchicum, Natrum muriaticum, Thuja occidentalis, Arnica montana, Kali iodatum* and *Chelidonium* was identified as potential drugs against mycelial growth of *S. rolfsii*.

The study presents compelling evidence on the efficacy of homoeopathic drugs in inhibiting the mycelial growth and sclerotial development of *Sclerotium rolfsii*. Notably, *Chelidonium* emerged as the most effective treatment, demonstrating an 88.88% inhibition of mycelial growth. This is consistent with previous findings, such as those of Khanna and Chandra (1987, 1989, 1992), who reported significant suppression of spore germination and mycelial inhibition across various pathogens when treated with homoeopathic drugs. Hanif and Dawar (2015) used homeopathic drugs as substitute technique in reducing the incidence of root rot fungi like *Rhizoctonia solani, Fusarium spp* and *Macrophomina phaseolina*. Homeopathic drugs such as *Arnica montana* and *Thuja occidentalis* with 100, 75 and 50% v/v concentrations was used to investigate growth parameters and for the control of root rot fungi. The inhibition of sclerotial germination by homoeopathic drugs, especially *Chelidonium* and *Colchicum*, at various durations and potencies, highlights another critical aspect of their antifungal activity. This finding is supported by similar studies on other fungal pathogens, such as those by Chaudhari et al. (2011), who reported substantial inhibition of mycelial growth of *Alternaria brassicae* with homoeopathic treatments.

3.2. Top of form

3.2. Cultural, morphological characters of mycelium and sclerotia of sclerotium rolfsii

The cultural characteristics viz., mycelial pattern, sclerotial arrangement and colour of sclerotia, was observed among

Table 1: Efficacy of homoeopathic drugson mycelial inhibition of *Sclerotium rolfsii* under *in vitro*

Homoeopathic drug	Mycelial inhibition (%)	
	Potency (C)	
	200 C	300 C
Acidium phosphoricum	2.96±0.74	13.33±1.28
Arsenic album	16.29±1.96	49.62*±5.92
Arnica montana	30.29±8.34	80.21±0.34
Bryonia alba	18.51±7.40	26.66*±2.22
Calcaria carbonica	22.22±0	25.18±1.95
Chelidonium	37.03*±1.48	88.88*±2.56
Chimaphila	26.66±3.39	40.73*±3.70
Cina	12.59±1.48	67.40*±5.33
Ferrum sulphoricum	22.22±0	40.73*±7.40
Iodium	38.51*±5.18	77.03*±7.73
Kali iodatum	33.33±12.12	80.25*±6.41
Lachesis	20.73±2.67	71.10*±2.22
Magnesium carbonicum	23.70±1.48	28.14*±3.22
Natrum muriaticum	11.11±0	81.47*±2.67
Rhus toxicodendron	24.44±2.22	25.92*±7.40
Sambucus nigra	30.36*±8.34	65.18*±4.12
Sanguinaria canadensis	19.25±1.96	66.66*±11.11
Selenium	6.66±0	34.07*±1.95
Sepia	18.51±3.70	20.73±2.67
Silicea terria	17.77±6.66	75.55*±2.56
Spongia tosta	38.51*±1.96	69.25*±1.96
Staphysagria	25.18±11.85	45.18*±0.74
Tarentula hispana	73.33*±11.40	74.81*±9.71
Thuja occidentalis	11.11±0	80.66*±12.63
Tuburculinum	30.36*±4.12	40.73*±3.70
Colchicum	20.73±8.54	82.21*±4.62
Borax	29.52*±7.40	47.40*±5.18
Phosphorus	29.65*±1.33	39.99*±1.14
Belladonna	48.14*±3.22	54.07*±6.58
Colchicum autumnale	44.44*±1.69	78.51*±1.96
Aswagandha	10± 0	33.33*±1.69

Data were considered as significant at the level of $p \leq 0.05$, results were presented±standard error

the treatments (Table 2). Pluffy growth of mycelium was observed in the treatment with *Chelidonium* while it was thread-like or cottony growth in control. Variations in the arrangement of sclerotia and sclerotial colour was also observed in comparison with control. The sclerotia was arranged at different patterns i.e., centre, and scattered upon treatment with homoeopathic drugs while sclerotia arranged in peripheral region in control plates. Sclerotia was brown to dark brown in colour upon treatment with homoeopathic drugs while it was brown in control. The shape of sclerotia also varied from treatment to treatment when compared to control. The sclerotial shape was irregular in homoeopathic drugs treated plates while it was round in control.

The cultural and morphological characteristics of *S. rolfsii* was markedly influenced by homoeopathic treatments, with notable differences in mycelial patterns, sclerotial arrangement, and color. Such changes can affect the pathogen's ability to colonize and infect host plants, thus providing a multifaceted approach to disease management. Previous studies, such as those by Meinerz et al. (2010), Bekriwala et al. (2016) have also documented the impact of homoeopathic treatments on the morphological characteristics of fungal pathogens, reinforcing the potential of these drugs in altering pathogen development.

3.3. Slerotial development and germination of sclerotium rolfsii

The impact of homeopathic drugs on sclerotial development, including initiation, ooze formation, maturity, and quantity of sclerotia formed on media, exhibited significant variability as detailed in Table 3. Sclerotial initiation varied across treatments, spanning from 6 to 16 days, while ooze formation on sclerotia showed differences ranging from 8 to 20 days. Maturity of sclerotia also exhibited variations among treatments, ranging from 13 to 28 days, except for the control group, which matured within a week. A similar trend was observed in the number of sclerotia formed on media across different treatments (Table 3), with counts ranging from 52 to 400. Notably, *Chelidonium* exhibited the lowest sclerotia production (52), followed by *Colchicum* (62), compared to the control (400). Sclerotial initiation was notably delayed in *Arnica montana* (18 days) and *Thuja occidentalis* (14 days), whereas in the control group, it occurred within six (6) days. Early ooze formation from sclerotia was observed in treatments with *Cina* and *Nux vomica* (8 days), while it was delayed in *Arnica montana* (18 days).

The impact of homoeopathic drugs, administered at potencies of 200 and 300 C, on the germination of *Sclerotium rolfsii* sclerotial bodies was investigated over durations of 5, 10, 15, and 20 minutes on PDA medium. The results, outlined in Table 4, demonstrate significant inhibition of sclerotial germination. Particularly, the highest inhibition was observed with *Chelidonium*, and *Colchicum* treatments at 15 and 20-minute durations, along with *Thuja occidentalis*, and *Spongia* treatments showing maximum inhibition at the 20-minute mark, as compared to the control group.

The observed variations in sclerotial development, initiation,

Table 2: Effect of homoeopathic drugs on cultural and morphological characters of *Sclerotium rolfsii* of groundnut stem rot pathogen

Homoeopathic drug	Colony character	Sclerotial arrangement	Sclerotial colour
Control	Light cottony thread like structure growth	Peripheral	Light brown
Acidium phosphoricum	Light cottony growth, dense at margins	Peripheral	Brown
Arsenic album	Thick cottony, pluffy at edges and upright growth habit, white to pale olive colour	Peripheral	Brown
Bryonia alba	Thick pluffy growth	peripheral	Brown
Calcaria carbonica	Thick pluffy growth	Centralised	Dark brown
Chelidonium	Pluffy growth upto middle, cottony growthy from middle to edges	Peripheral	Light brown
Chimaphila	Thick pluffy growth	Peripheral	Brown
Cina	Thick pluffy growth	Scattered	Light brown
Ferrum sulphoricum	Thick pluffy growth	peripheral	Brown
Iodium	Light pluffy growth	Peripheral	Brown
Kali iodatum	Thick Cottony growth	Centralised	Thick brown
Lachesis	Thick pluffy growth, Cottony in middle	Centralised	Dark brown
Magnesium carbonicum	Cottony growth upto middle, pluffy from middle to edges	Peripheral	Brown
Natrum muriaticum	Cottony pluffy growth upto middle, dense at margins	Centralised	Dark brown
Rhus tox	Light pluffy growth	peripheral	Brown
Sambucus nigra	Cottony upto middle, pluffy from middle to edges	Peripheral	Brown
Sanguinaria canadensis	Cottony growth, pluffy in middle	Peripheral	Brown
Selenium	Light cottony growth	Scattered	Brown
Sepia	Pluffy at middle, dense at margins	Scattered	Dark brown
Silicea terria	Thick cottony growth	Scattered	Brown
Spongia tosta	Cottony upto middle and Pluffy from middle to edges	Centralised	Brown
Staphysagria	Pluffy growth	Peripheral	Brown
Tarentula hispana	Light pluffy growth	peripheral	Dark brown
Thuja occidentalis	Cottony, dense at centre	Centralised	Brown
Tuburculinum	Cottony growth	Scattered	Dark brown
Arnica montana	Pluffy Cottony growth at centre, dense at margins	Starting portion	Light brown
Arsenic album	Pluffy white cottony, dense at margins	Centralised	Brown
Iodium	Pluffy in middle and cottony upto edges	Scattered	Brown
Sulphur	Cottony thread like at starting and Pluffy at centre and edges	Centralised	Brown
Colchicum	White thick cottony, pluffy growth upto edges	Centralised	Brown
Belladonna	Pluffy at middle, dense at margins	Peripheral	Brown
Colchicum	Pluffy cottony growth	Scattered	Dark brown
Borax	Thick pluffy growth	Scattered	Brown
Phosphorus	Dense upto middle and pluffy in middle and dense at margins	Margins	Dark brown
Ashwagandha	Cottony upto middle, pluffy from middle to edges	Centralised	Dark brown

Table 2: Continue...

Homoeopathic drug	Colony character	Sclerotial arrangement	Sclerotial colour
Lycopodium	Thick pluffy upto middle dense at margins	Scattered	Dark brown
Nux vomica	Thick cottony pluffy growth	Scattered	Dark brown

Table 3: Impact of homoeopathic drugs on sclerotial development of Sclerotium rolfsii under *in vitro*

Homoeopathic drug	SI	OF	SM	NS
Control	6	8	13	450
Acidium phosphoricum	6	8	15	228
Arsenic album	8	11	23	199
Arnica montana	16	18	24	126
Bryonia alba	8	12	24	215
Calcaria carbonica	11	14	24	98*
Chelidonium	9	12	28	52*
Chimaphila	7	9	21	121
Cina	10	13	21	170
Ferrum sulphoricum	9	12	23	117
Iodium	11	13	23	148
Kali iodatum	14	17	23	142
Lachesis	9	12	24	157
Magnesium carbonicum	9	11	23	135
Natrum muriaticum	7	10	16	161
Rhus toxicodendron	9	12	20	139
Sambucus nigra	9	13	24	146
Sanguinaria canadensis	9	12	21	149
Selenium	8	12	24	212
Sepia	10	14	22	137
Silicea terria	6	11	23	183
Spongia tosta	8	11	24	158
Staphysagria	10	12	23	184
Tarentula hispana	7	9	21	150
Thuja occidentalis	14	16	22	165
Tuburculinum	7	9	23	176
Colchicum	9	12	19	62*
Borax	8	11	18	125
Phosphorus	11	15	23	189
Belladonna	11	14	21	159
Aswagandha	12	15	22	179

SI: Sclerotia initiation (day); OF: Ooze formation (day); SM: Sclerotial maturity (day); NS: No. of Sclerotia; Values were mean of three replications.*Lowest number of sclerotia production

Table 4: Inhibition of sclerotial germination treated with homoeopathic drugs at 72 hrs of incubation under *in vitro* on PDA

Homoeopathic drug	Scleorotial germination (%)			
	Time (min)			
	5	10	15	20
Control	100	100	100	100
Colchicum @ 1000C	100	100	100	100
Natrum muriaticum @ 200C	100	100	100	40*
Arnica montana @ 30C	100	100	0*	0*
Staphysagria @ 30C	100	80*	0*	0*
Chelidonium @ 30C	100	100	100	0*
Spongia @ 30C	100	80*	60*	0*
Iodium @ 30C	100	100	80*	60*
Tarentula @ 30C	100	100	60*	40*
Thuja occidentalis @ 200C	100	100	60*	0*
Colchicum @ 6C	100	100	100	40

and maturity among different homoeopathic treatments further underscore the potential of these drugs in managing *S. rolfsii*. *Chelidonium* and *Colchicum* not only delayed the initiation and maturity of sclerotia but also significantly reduced their quantity, which is crucial for managing the persistence and spread of this pathogen. This aligns with the findings of Hanif and Dawar (2015, 2016, 2017), who reported that homoeopathic drugs, particularly *Arnica montana* and *Thuja occidentalis*, effectively reduced the colonization of various root rot fungi and enhanced plant growth parameters. Additionally, the broad spectrum of efficacy demonstrated by various homoeopathic drugs against different fungal pathogens, as reported in studies by Alam and Mistry (2022) suggests that these treatments can be effectively integrated into comprehensive disease management strategies. The consistent results across different studies emphasize the reliability and potential of homoeopathic drugs as viable alternatives to conventional fungicides.

The overall study showed the significant inhibition of *S. rolfsii* mycelial growth, sclerotial development, and germination by homoeopathic drugs presents a promising avenue for sustainable and eco-friendly plant disease management. Further research should explore the mechanisms underlying these effects and the potential for integrating homoeopathic

treatments with other disease management practices to enhance their efficacy and reliability.

5. CONCLUSION

The experiment revealed that, homeopathic drugs, specifically *Arnica montana*, *Chelidonium*, and *Thuja occidentalis*, effectively inhibited the radial growth of mycelium and sclerotial germination, confirming their fungicidal properties against *Sclerotium rolfsii*. These remedies were cost-effective, readily available, biodegradable, non-phytotoxic, and non-pollutant.

6. ACKNOWLEDGEMENT

The authors are thankful to CCRH (Central Council for Research in Homoeopathy), Ministry of AAYUSH, Government of India, New Delhi for providing the financial support to conducting this Research.

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