

## Evaluation of Botanical Extracts for their efficacy against In-vitro growth of *Sclerotium rolfsii*

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### Abstract

*Sclerotium rolfsii* is a necrotrophic, soil-borne plant pathogenic fungus associated with economically significant diseases causing mainly collar rot, southern blight, seedling blight, and stem rot on more than 500 plant species including almost all agronomical and horticultural crops. In contrast to chemical fungicides, plant extracts do not cause pollution and are easily biodegradable, therefore the current study focuses on the affordable, environmentally friendly, safe, and sustainable control of diseases through botanical extracts. An experiment was conducted at the Institute of Agriculture and Animal Science, Lamjung Campus, Plant Pathology Laboratory to ascertain the effectiveness of botanical extracts against this pathogen. Under the in-vitro study, five botanical extracts viz. *Allium sativum*, *Zingiber officinale*, *Datura stramonium*, *Azadirachta indica*, and *Allium cepa* were evaluated at three different concentrations (5%, 10%, and 15%) using the poisoned food technique in a Completely Randomized Design and each concentration was replicated three times. The purpose of the study was to assess their efficacy against the mycelial growth of *S. rolfsii*. The data were analyzed using R-software in R-studio and means were compared at a 5% significance level using Duncan's Multiple Range Test (DMRT). All tested chemicals reduced the pathogen's mycelial development compared to the control. Data on pathogen mycelial growth were collected after 24, 48, and 72 hours of incubation. After 72 hours, among all the tested botanicals, the pathogen's growth was significantly inhibited by 15% *Allium sativum* (growth inhibition of 80.9%) followed by 15% *Azadirachta indica* (74.8%), and by 15% *Zingiber officinale* (70.1%). *Datura stramonium* at 5% showed the least growth inhibition (12.1%) which was statistically at par with the effect of *Allium cepa* at 5% concentration (14.9%). Further research and field trials should be conducted to select appropriate concentrations of botanical extracts to control the disease pathogen economically and sustainably.

**Keywords:** Botanical extracts, Completely Randomized Design, Growth inhibition, Poisoned food technique

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## 1. Introduction

*Sclerotium rolfsii* is a destructive soil-borne fungal pathogen with a necrotrophic lifestyle that, under conducive conditions, develops profuse white mycelium on infected plants and in culture (Mullen, 2000). It has a wide host range that includes more than 500 plant species of horticulture and agronomic crops (Okabe & Matsumoto, 2000). This pathogen is globally distributed, particularly in tropical, subtropical, and other warm-temperate regions (Aycock, 1966). Numerous symptoms, including seed rot, seedling blight, collar rot, damping-off, stem rot, wilt, crown and root rot, stem canker, and sclerotium blight, are brought on by the fungus in diverse host plants (Mullen, 2001; Arunasri, 2011). This disease causes significant economic losses due to its wide host range, prolific proliferation, and ability to create chronic sclerotia (Kokub et al., 2007). Mayee and Datar (1988) stated that these diseases usually result in yield losses of over 25%, and in extreme cases, this can be as high as 80%.

*S. rolfsii* is primarily characterized as a polyphagous, non-targeting, moisture-loving, ubiquitous facultative parasitic basidiomycetes fungus that produces oxalic acid, polygalacturonase, and cellulase as pathogenic components (Chen et al., 2020). White fungal mycelia can be visible on the soil around the plant at the base; a characteristic of *S. rolfsii* growth in infected plants. The pathogen's broad host range, rapid development, and propensity to produce persistent sclerotia all contribute to the pathogen's massive economic losses. In adequate organic matter, the pathogen persists successfully in the soil as sclerotia, even under unfavorable climate conditions (Kumar et al., 2018). The occurrence and severity of *S. rolfsii* have been found to increase when beans are grown in rotation with legumes, cucurbits, and other vegetable crops. The pathogen poses a significant management challenge since it can infect crops at different stages of growth.

From an economic standpoint, this pathogen is recognized as the most devastating and destructive to plants, with a wide range of hosts including cereal crops, and different vegetables such as tomatoes, chili, onions, and so on (Abang, 2003). *S. rolfsii* is thought to have caused losses of 10-20 million dollars worldwide, with field yields depleted by 1- 60% (Laiamnhee et al., 2015). Over the past five years, *Sclerotium rolfsii* Sacc. has become more severe in a variety of crops, including rice, onions, and chilies in Nepal. It typically affects legumeous and solanaceous crops (Adhikari et al., 2022). The following areas were infected by *S. rolfsii*: lentil, rajma, chickpea, and mustard in Lumbini and Sudur Paschim province since 2015 (NGLRP, 2015); rice in the Sunsari, Jhapa, Morang, and Udaypur districts during 2016/2017 (Annual Report, 2073); and onion in Dhading district during 2018/2019 (Annual Report, Kathmandu, 2018) and chili in Chitwan since 2015. In Nepal, this pathogen has been regarded as one of the serious in agricultural production, posing a serious threat to all producers.

The diseases caused by *S. rolfsii* can be managed using physical and cultural approaches, resistant cultivars, and chemical and biological control strategies (Vargas Gil et al., 2008). Effective strategies for managing the pathogen include various soil management techniques. Deep plowing reduces the viability of sclerotia or destroys the

pathogen's hyphae in the fields by burying infected tissues under 6–20 cm. Rotation with non-host crops increases the nutritional condition of the soil and may harm pathogen inoculum density. Furthermore, soil solarization is also one of the effective management strategies. Moreover, different fungicides are predominantly used for the control of the disease viz tebuconazole, carbendazim, pentachloronitrobenzene, and mancozeb. Mancozeb suppressed *S. rolf sii* in vitro by approximately 55%, 73%, and 83% at 500, 750, and 1000 ppm, respectively (Manu & Nagaraja, 2012). At 4 DAI, *S. rolf sii* growth inhibition was 100.00, 78.15, 75.64, 59.10, and 44.53% for Provox-200, Score 250 EC, Tilt 250 EC, Pencozeb 80 WP, and Rovral 50 WP, respectively (Rahman et al., 2020). These five fungicides were noted as highly effective. Besides chemical fungicides, biological controls have effectively managed multiple plant diseases based on their antagonistic nature (Sivan, 1987). Fungal biocontrol agents and *Trichoderma* species are well agent for controlling plant diseases caused by soilborne fungi (Prasad & Naik, 2008). *S. rolf sii* was successfully controlled by *Trichoderma* species in both in-vitro and in-vivo conditions (Singh et al., 1997). In the same way, it has been demonstrated that several organic plant products such as Alfalfa (*Medicago sativa*), Basil (*Ocimum basilicum*), Chamomile (*Matricaria chamomilla*), Palmarosa (*Cymbopogon martinii*), and Thyme (*Thymus vulgaris*) (Sekhar et al., 2020) don't exhibit phytotoxicity and have systemic action, reduce pathogen activity, and monitor the spread of disease. Plant extracts can be employed as environmentally friendly substitutes, cost-effective, and crucial elements in integrated pathogen management (Perello et al., 2013). The reasonable integration of all these methods can provide the economic and sustainable management of *S. rolf sii*.

The antifungal properties of botanical extracts have been demonstrated against a variety of plant pathogenic fungi. They are biodegradable and free from environmental contamination and health hazards (Rahman, 2020). Different botanicals' antifungal potency varies due to variations in the active chemical makeup of the extracts (Shrestha & Tiwari, 2009). The medicinal and aromatic herbs contain antifungal and antioxidant properties that not only slow the spread of disease but also result in goods that are free of hazardous residue (Dalbeer, 2015; Pun et al., 2020). Numerous studies have highlighted the antifungal potential of botanical extracts against various fungal pathogens, including *S. rolf sii*. These botanical extracts were selected due to their diverse therapeutic value and potential anti-microbial properties. Garlic has a strong flavor and therapeutic value due to its high starch and aromatic component content. Diallyl sulphide, diallyl disulphide, diallyl trisulphide, allyl methyl trisulphide, dithiins, and ajoene are some of the antibacterial compounds which are obtained from the breakdown of allicin, a phytochemical found in garlic (Khan & Katiyar, 2000). In addition to allicin, Ajoene is a significant antifungal component in garlic that can harm fungi's cell walls (Yoshida et al., 1987) and inhibits the activity of certain enzymes necessary for fungal growth (Kutawa et al., 2018). The secondary metabolites of the Dhatura plant have a potent anti-microbial and antifungal effect (öz arık, 2017). Alkaloids, triterpenoids, steroids, flavonoids, triterpenes, phenolic chemicals, and tannins are said to be responsible for the antibacterial characteristics of the Dhatura plant. *Datura*

*sp.* produces pharmacologically important alkaloids like hyoscyamine and scopolamine. The quantity of triterpenoids, peaks, and pure compounds (isomeldenin and nimonol) in Neem gives it antifungal properties. Neem organic extracts contain active components such as Quercetin, sitosterol, 6-deacetyl nimbin, azadiradione, Nimbin, salannin, and epoxy-azadiradione, which have antifungal and antibacterial qualities (Govindachari et al., 1998). The onion's phytochemical components and total phenolic contents (quercetin and kaempferol) helped to effectively prevent fungal growth (Singh, 2017). The high content of antioxidants in onions and onion by-products makes them powerful scavengers of free radicals. As a result, it has anti-inflammatory and anti-mutagenic properties. Ginger possesses antibacterial, anti-inflammatory, and antioxidant properties. Over 400 distinct chemicals, including zingerone, shogaols, gingerols, sesquiterpenoid, and a minor monoterpene portion, are thought to be responsible for ginger's antibacterial activities (Pun et al., 2020). Additionally, ginger contains non-volatile pungent components and 1% to 3% volatile oils, oleoresin (Zick et al., 2008).

The heavy use of agrochemicals for disease control causes toxicity to non-target organisms, neurological effects, reproductive health effects, population development of resistant pathogens, and pollution in the environment (Ganie et al., 2013). This has prompted research into safe and economical substances that don't have any harmful effects on the general health of consumers or the environment (Nxumalo et al., 2021). Utilizing plant materials as innovative chemotherapeutic agents in plant protection has recently received interest in many parts of the world. Finding new plant protectants that can reduce fungal pathogenicity has become more important because of the emergence of resistance to conventional fungicides and tighter regulations on the use of hazardous compounds in the environment. Plant extracts are now utilized against numerous plant pathogenic fungi due to their antifungal properties, which have received a lot of attention (Swami et al., 2013).

## **Methodology**

### **Experimental Site**

The present study was conducted at the Central Plant Pathology Laboratory of the Institute of Agriculture and Animal Science (IAAS), Sundarbazar, Lamjung, in 2022, under strictly controlled conditions to prevent contamination. A pure culture of *Sclerotium rolfsii* was sourced from the Division of Plant Pathology, NARC, Khumaltar, Lalitpur, Nepal, and preserved at 4°C in a refrigerator. The experiment followed a Completely Randomized Design (CRD) with six distinct treatments, as detailed in Table 1. Three concentrations (5%, 10%, and 15%) of five different plant extracts, excluding the control, were evaluated. Each concentration was tested in three replications. The details of the various treatments are as follows:

**Table 1: Treatment details used in the experiment**

Treatments Symbol	Botanical Extracts (Common Name)	Scientific Name	Plant parts used
T1	Datura	<i>Datura stramonium</i>	Leaves
T2	Garlic	<i>Allium sativum</i>	Bulb
T3	Ginger	<i>Zingiber officinale</i>	Rhizome
T4	Neem	<i>Azadirachta indica</i>	Leaf
T5	Onion	<i>Allium cepa</i>	Bulb
T6	Control (Distilled Water)		

### General Laboratory Procedure

#### Equipment, Apparatus, and their sterilization

Test tubes, beakers, conical flasks, glass funnels, and petri plates were cleaned with a detergent solution under running water, rinsed with distilled water, and allowed to air dry. Following that, the glass wares were wrapped in aluminum foil and autoclaved for 20 minutes at 15 psi and 121°C to sterilize it. The inoculation needle, inoculation loop, forceps, blades, and cork borer were all sterilized by heating them to a red-hot over the flame. The procedure was carried out two or three times. The laminar flow chamber was sterilized by activating UV light for fifteen minutes, and the base and wall of the chamber were cleaned using 70% ethanol.

#### Media Preparation

Potato Dextrose Agar (PDA), which contains 2% agar was used as the growth medium for *S. rolf sii*. PDA powder was mixed with 1000 ml of distilled water in a reagent bottle at a rate of 39 gm per 1000 ml, as directed, to provide the necessary number of media. The media was then autoclaved at 15 pressure, 121 °C for 20 minutes to sterilize it, after which it was allowed to cool. When the media's temperature reached about 40 °C, it was placed into sterile Petri plates in a laminar flow environment and allowed to solidify. After solidification, the media was infused with botanical extracts and used to inoculate the pathogen.

#### Preparation of Mother Culture

The mother culture was utilized for further multiplication and treatment purposes, prepared using pure culture, which was refrigerated. A small amount of mycelial thread was excised from the pure culture plate using a sterile

inoculation loop and transferred to a sterile Petri plate containing about 20 ml of sterile PDA media. Those mycelial inoculated Petri plates were called mother cultures which were incubated in an incubator at  $27\pm 2^{\circ}\text{C}$  for 3 days.

### Preparation and In vitro evaluation of selected botanical extracts

The plant extract was prepared by following the guidelines used by Ul-Haq et al. (2014). The essential plant parts such as matured fresh leaves, bulbs, and rhizomes were collected, thoroughly cleansed with tap water, sterilized with distilled water, and then allowed to dry in the shade. Separately, the materials were pulverized in a mortar and pestle with sterilized distilled water (1:1 w/v). The mixture was filtered using a four-layered muslin cloth and boiled at  $80^{\circ}\text{C}$  for ten minutes in the hot water bath. After that, the extract was centrifuged for 5 minutes at 4000 rpm. Whatman's filter paper No. 1 was used to filter the supernatant. Thus, the resulting filtrate was regarded to have a concentration of 100 % and was taken as a basic stock solution. Streptomycin (0.25 g/l) was added in sterilized and cooled PDA ( $40^{\circ}\text{C}$ ) to check bacterial growth.

The pathogen was examined at three concentration levels in a comparative study: 5%, 10%, and 15% for the botanicals listed below. Three replications of each concentration were used in the completely randomized design (CRD) experiment, which was carried out using the poisoned food approach.

### Food poisoning technique

The Poisoned Food Technique was used to evaluate botanical extracts in vitro against *Sclerotium rolfsii*. To make the necessary concentrations for food poisoning the determined amount of stock solution of botanical extracts was mixed with sterilized PDA (Potato Dextrose Agar) medium. Each sterilized Petri plate received 20 ml of adjusted or amended PDA, which was allowed to solidify. The control treatment was maintained where no amended medium was used. In the center of the solidified PDA plates, identical circular discs of mycelial growth from a 3-day-old culture of *Sclerotium rolfsii* were transferred aseptically using a sterilized cork borer. Three Petri plates were used to replicate each treatment. The Petri plates were then incubated for 3 days at  $27\pm 2^{\circ}\text{C}$ .

### Growth inhibition test

The measurements of mycelial growth were performed using a Vernier caliper in the 24, 48, and 72 hours of incubation for each treatment. Two readings at a perpendicular angle from each other, forming a "+" sign, were used to compute the average diameter of mycelial growth. Both control and amended plates were used to take readings. The percentage inhibition of mycelial growth compared to the control was computed using the formula of Vincent (1947):

$$PGI = (C - T)/C * 100$$

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Where PGI= Percent growth inhibition, C = Average Growth of hyphae in control (mm), and T = Average Growth of hyphae in Treatment (mm).

### Statistical analysis

All the data were entered in Ms. Excel (2019) and analysis of variance was done using R-Stat software version (4.2.2). The mean comparison was done using the Duncan Multiple Range Test (DMRT) and Least Significant Difference (LSD) at a 0.05 level of significance.

### Results and Discussion

Five different botanical extracts were evaluated against *Sclerotium rolfsii* at different concentrations viz., 5%, 10%, and 15% in laboratory conditions by poisoned food technique. Our result (Table 2) shows that all the tested botanicals inhibited pathogen growth over unamended media. Different plant extracts showed different levels of fungicidal properties against the pathogen. Plant extracts are now utilized against numerous plant pathogenic fungi due to their antifungal properties, which have received a lot of attention (Swami et al., 2013).

The inhibitory effect of the various extracts was found to differ significantly ( $P \leq 0.001$ ). Regardless of the concentration used, the range of growth inhibition ranged from 12.06% to 80.91% after 72 hours of inoculation.

**Table 2: Invitro evaluation of botanical extracts against *Sclerotium rolfsii* at 27±2 °C**

S.N.	Botanical Extracts	Concentrations (%)	Growth Inhibition Percentage (%)		
			24 hours	48 hours	72 hours
1.	Garlic ( <i>Allium sativum</i> )	5	32.36 <sup>f</sup>	34.92 <sup>g</sup>	37.23 <sup>g</sup>
		10	61.08 <sup>c</sup>	63.84 <sup>d</sup>	65.03 <sup>d</sup>
		15	69.54 <sup>a</sup>	77.77 <sup>a</sup>	80.91 <sup>a</sup>
2.	Neem ( <i>Azadirachta indica</i> )	5	28.88 <sup>g</sup>	31.76 <sup>h</sup>	33.67 <sup>gh</sup>
		10	58.61 <sup>c</sup>	61.95 <sup>d</sup>	64.82 <sup>d</sup>
		15	67.45 <sup>a</sup>	72.07 <sup>b</sup>	74.80 <sup>b</sup>
3.	Ginger ( <i>Zingiber officinale</i> )	5	21.15 <sup>i</sup>	23.36 <sup>j</sup>	26.60 <sup>hi</sup>
		10	51.92 <sup>d</sup>	54.80 <sup>e</sup>	57.11 <sup>e</sup>
		15	64.18 <sup>b</sup>	66.80 <sup>c</sup>	70.13 <sup>c</sup>
4.s	Onion ( <i>Allium cepa</i> )	5	10.67 <sup>j</sup>	12.38 <sup>k</sup>	14.88 <sup>j</sup>
		10	27.81 <sup>g</sup>	30.65 <sup>h</sup>	32.01 <sup>h</sup>
		15	50.41 <sup>d</sup>	53.61 <sup>e</sup>	55.76 <sup>e</sup>

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5.	Dhatura ( <i>Datura stramonium</i> )	5	9.22 <sup>j</sup>	10.88 <sup>k</sup>	12.06 <sup>j</sup>
		10	24.76 <sup>h</sup>	26.53 <sup>i</sup>	29.96 <sup>hi</sup>
		15	42.39 <sup>e</sup>	45.12 <sup>f</sup>	48.48 <sup>f</sup>
Grand Mean			41.36	44.42	46.89
SEM (±)			1.74	1.50	2.73
CV (%)			4.22	3.39	5.82
LSD at 5%			2.91	2.51	4.55

LSD: Least Significant Difference      SEM: Standard Error of Mean      CV: Coefficient of Variation

The mycelial growth of *Sclerotium rolfsii* was observed to decrease as the concentration of the plant extracts increased. After 24 hours of inoculation, maximum growth inhibition was recorded in 15% Garlic i.e. inhibition percentage of 69.54% which was statistically at par with 15% Neem (67.45%) with each other. It was followed by 15% Ginger (64.18%), 10% Garlic (61.08%) which was statistically at par with 10% Neem (58.61%), 10% Ginger (51.92%) which was statistically at par with 15% onion (50.41%), and by 15% Dhatura (42.39%). Minimum inhibition was obtained in 5% Dhatura (9.22%) which was statistically at par with 5% Onion (10.67%).

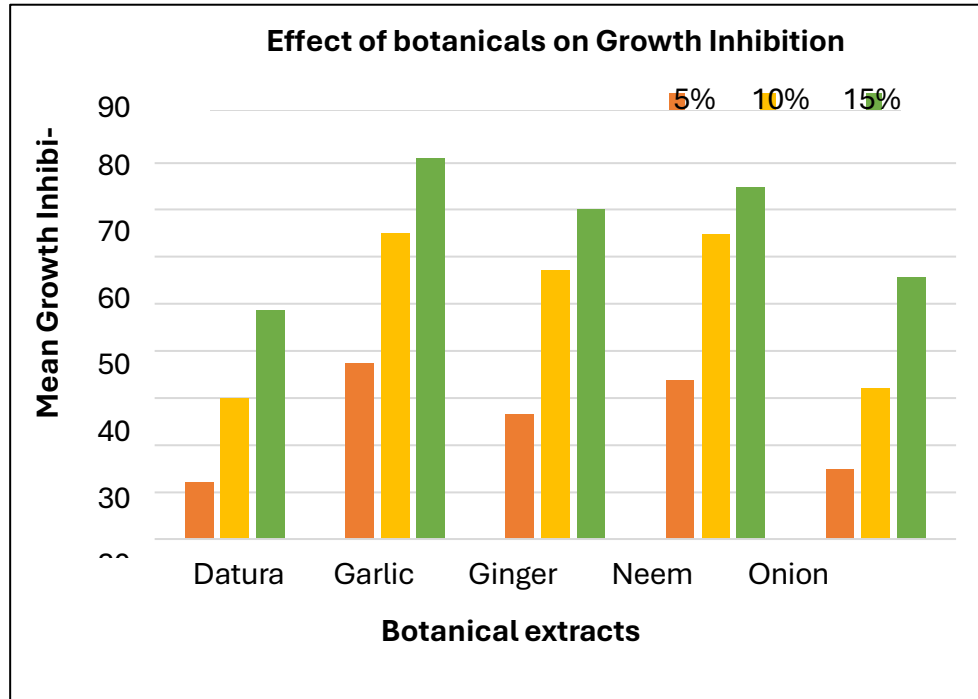


Figure 1: Mycelial Inhibition (%) after 72 hours of inoculation at different treatments



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After 48 hours of inoculation, the inhibition percentage varied from 10.88% to 77.77%. Garlic was proved to be the most effective botanical extract at 15% concentration showing 77.77% inhibition followed by 15% Neem (72.07%) and 15% Ginger (66.80%). 10% Garlic with an inhibition percentage of 63.84% which was statistically at par with 10% Neem (61.95%). 5% Dhatura (10.88%) was least effective in restricting the mycelial growth of pathogen which was statistically at par with 5% Onion (12.38%).

Similarly, after 72 hours of inoculation, maximum growth inhibition was recorded by 15% Garlic (80.91%) followed by 15% Neem (74.80%) and 15% Ginger (70.13%). 10% garlic (65.03%) was statistically at par with 10% Neem (64.82%). Minimum growth inhibition was recorded by 5% Dhatura (12.06%) followed by 5% Onion (14.88%) which were statistically at par with each other.

Figure 2: Mycelial growth of *S. rolfsii* in PDA- unamended media as a control

The highest inhibitory effect was observed in Garlic extract at 15% concentration as compared to control against the tested pathogen in the present investigation. The result obtained agreed with (Mahato et al., 2018), where *Allium sativum* recorded maximum growth inhibition of *S. rolfsii*, significantly superior to all other botanical



extracts. (Rahman et al., 2020) also evaluated the antifungal property of phytoextracts against *S. rolfsii* and obtained the highest growth inhibition with garlic extract than other botanicals at all concentrations tested. Bio-efficacy of ten botanical extracts on growth of *S. rolfsii* and *A. niger* were tested invitro at two concentrations viz., 10 and 20%. *Allium sativum* was proved to be an effective botanical and recorded a maximum reduction of growth of both *A. niger* and *S. rolfsii* by 100% which was significantly superior to all the plant extracts at 20% concentration. Chrysanthemum at both concentrations was least effective in reducing fungal growth (Vineela et al., 2020).

The antifungal metabolites present in plants may be responsible for the fungitoxicity of plant extracts. Different botanicals' antifungal potency varies due to variations in the active chemical makeup of the extracts (Shrestha

& Tiwari, 2009). The primary component of garlic, allicin, which is produced by the phosphopyridoxal enzyme allinase, is believed to be responsible for the antibacterial properties of garlic (Arunachalam, 1980). Garlic contains other bioactive compounds such as garlicin, ajoene, and allylsulfides. Ajoene derived from garlic causes morphological changes such as the disappearance of surface ornaments, thickening of a cell wall, and destruction of cell organelle by acting on the cell wall of a fungus (Yoshida et al., 1987). The efficacy of garlic clove extracts is attributed to the volatile oil, which contains diallyl disulphide, diallyl trisulphide, and sulphodoxides, which are all obtained from allicin (Chethana et al., 2012). The allicin or ajoene restricts the performance of some enzymes that are important to fungal growth (Kutawa et al., 2018). Along with sulfur compounds, garlic contains arginine, 17 amino acids, and their glycosides, as well as other amino acids and other amino acids. minerals like selenium, as well as enzymes like myrosinase, allinase, peroxides, and others. Numerous studies have noted that garlic has a similar impact.

The second-best inhibitory effect was shown by 15% of Neem i.e. (74.80%). The presence of triterpenes or limonoids such as azadirachtin, nimonol, quercetin, nimbin,  $\beta$ -sitosterol, and other butter substances viz., alkaloids, glycosides, and gums contribute to the antimicrobial properties of Neem (Mahmoud et al., 2011). Neem is known to inhibit protease activity and alter the hydrophobicity of fungal cells, leading to anti-adhesion effects (Polaquini et al., 2006). According to Begum et al. (2014), Neem (*Azadirachta indica*) had the highest average inhibition (74.81%) of the investigated botanicals at 5 and 10% concentrations, followed by Tulsi (*Ocimum sanctum*) (67.10%) and Nirgudi (*Vitex negundo*) (65.81%) against *S. rolfsii*.

The third-best inhibitory effect was observed in Ginger. Citral-containing chemicals in ginger essential oil are lipophilic, making the cell wall and cytoplasmic membrane more permeable and causing membrane integrity loss in fungus. The phenolic compounds in ginger are mainly gingerol and shagelol as antifungal compounds that inhibit the mycelial growth of pathogens (Jain et al., 2011). In the in-vivo condition, Mahato et al. (2018) tested botanicals such as ginger, neem, garlic, kalmegh, turmeric, periwinkle, tulsi, and onion at 5, 10, and 20% against *Sclerotium rolfsii*. *Azadirachta indica* (Neem) showed the best effectiveness against collar rot disease by reducing 64.9% incidence which was followed by *Allium sativum* (60.34%), and *Zingiber officinale* (58.83%) and discovered that the concentrations of 5%, 10%, and 20%, resulted in the greatest reduction of mycelial growth (35.31%, 68.50%, and 84.89% respectively) in in-vitro condition.

Dhatura i.e. inhibition percentage of 12.06% was found to be the least effective which is by (Hosen et al., 2016) who also showed the least inhibition of Dhatura and maximum inhibition of Garlic against *S. rolfsii* at all tested concentrations (5, 10 and 15%).

According to an evaluation of the effectiveness of botanicals by Bharathi et al. (2018), the combination of black tulsi rust extract, turmeric rhizome extract, and garlic bulb extract at a concentration of 15% produced the

highest level of mycelial growth inhibition (85.00%). The five botanical essential oils—Palmarosa, Karanja, Thyme, Menthol, and Lemongrass oils—were examined by (Sekhar et al., 2020) at varying concentrations (0.5, 1, 1.5, and 2%). Palmarosa (*Cymbopogon martini*) and Thyme (*Thymus vulgaris*) demonstrated the most significant efficacy, delivering 100% growth inhibition at 1.5 and 2.0%, regardless of the various concentration levels of essential oils evaluated against the mycelial growth of *S. rolfsii*. The effectiveness of several botanicals, including the leaves of *Bauhinia purpurea*, *Caesalpinia gilliesii*, *Cassia fistula*, *Cassia senna*, *Chrysanthemum frutescens*, *Euonymus japonicus*, and *Thespesia populnea var. acutiloba*, was assessed separately and found that the leaf extracts considerably inhibited the radial growth of *S. rolfsii* when compared to the control. The leaf extracts of *T. populnea var. acutiloba* were the most effective against the pathogen, with an inhibition percentage of 82.8% followed by *Chrysanthemum frutescens*, *Caesalpinia gilliesii*, *Euonymus japonicus*, *Cassia senna*, *Bauhinia purpurea*, and *Cassia fistula* with the inhibition percentage of 79.5, 78.3, 78.0, 77.2, 75.0, and 74.0 %, respectively at 200 ppm concentration (Derbalah et al., 2012). In conclusion, botanical extracts have been shown to have potent inhibitory activity against a wide variety of phytopathogenic fungi, making them a viable alternative to chemical fungicides.

## Conclusion

*Sclerotium rolfsii* is one of the devastating pathogens that has been regarded as one of the serious in agricultural production, posing a serious threat to all farmers. To combat obvious environmental pollution issues and prevent the hazardous effects of synthetic chemicals on non-target organisms, research on the use of pesticides with plant origin is becoming more and more significant in the field of plant pathology. Plant products must be periodically examined before they can be included as a component in integrated disease management. Likewise, the study showed that all the botanical extracts examined had a significant inhibitory effect compared to the control. In this experiment, garlic was found to be an effective botanical extract for radial growth inhibition of *Sclerotium rolfsii*. The use of botanicals could be a cheap, safe, and environmentally sound approach to disease management. The present research finding is limited to laboratory conditions so, more in-vitro and field trials are required to validate this finding for further recommendation in field conditions.

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