

International Journal of Statistics and Applied Mathematics



ISSN: 2456-1452

Maths 2023; SP-8(5): 880-883

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<https://www.mathsjournal.com>

Received: 13-05-2023

Accepted: 15-06-2023

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Management of *Sclerotium rolfsii* by the use of botanical

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Abstract

Tomato (*Solanum lycopersicon* L.) is a globally significant vegetable crop facing substantial challenges due to collar rot disease caused by *Sclerotium rolfsii*. The study aimed to assess the antifungal potential of thirteen botanical extracts against *S. rolfsii* mycelial growth *in vitro*. *Allium sativum* (garlic) displayed the highest fungicidal efficacy, completely inhibiting mycelial growth at all tested concentrations (25%, 50%, 75%, and 100%). *Jasminum angustifolium* also exhibited notable inhibition, 57.14% to 70.95% across concentrations. Other botanical extracts showed varying degrees of inhibition. The study provides an eco-friendly alternative to chemical fungicides for controlling *S. rolfsii*, mitigating environmental risks and reducing the emergence of resistant pathogens. This research contributes to the growing emphasis on organic agriculture and sustainable disease management strategies further research should explore field applications and investigate the broader implications of these botanical extracts in integrated disease management programs.

Keywords: *Sclerotium rolfsii*, management, *Allium sativum*, plant extract, botanical, minimal concentrations

Introduction

The Tomato (*Solanum lycopersicon* L.), a plant of the solanaceae family, is the most widely grown and valuable vegetable crop in the world due to its flavour, colour, high nutritional content, and variety of uses. It is produced in practically every nation in the world and is frequently consumed either fresh or processed. While miller classified Tomatoes in a new genus, *lycopersicon*, and gave it the name *lycopersicon esculentum* mill, Linnaeus put the Tomato in the genus *Solanum* in 1753 (Diez & Nuez, 2008) [3].

In most warm temperate and subtropical regions of the world, Tomato (*Solanum Lycopersicon* L.) Production is severely hampered by the collar rot disease, which is brought on by *Sclerotium rolfsii*. According to reports, the disease affects practically all of India's tomato-growing regions, including, and significantly reduces yields. The pathogen prefers soft tissues and produces collar zone rot, which results in death by interfering with the movement of food from the top to the root zone (begum *et al.*, 2011) [2].

The significance of organic agriculture is on the rise, and the global market for organic products has experienced substantial growth (Suja, 2013) [8]. In the field of plant pathology, Rangaswami and Mahadevan (1999) [7] provide valuable insights into crop diseases in India. J. Tuite (1996) [10] presents comprehensive plant pathological methods that encompass fungi and bacteria. For further understanding, H Barnett and BB Hunter (1972) [11] present a detailed guide to imperfect fungi genera. These sources contribute to the broader knowledge in the field.

In recent years, there has been a heightened global emphasis on plant research, leading to a substantial accumulation of evidence showcasing the considerable botanical plants potential, widely utilized in diverse traditional medical systems. Plant extracts are recognized for their distinct antimicrobial attributes. Numerous researchers have highlighted that plant extracts and their secondary metabolites, encompassing phenolic acids, glycosides, alkaloids, and terpenoids, exhibit a range of medicinal qualities, including antimicrobial effects, impacting biological processes even at minimal concentrations (Singh SK *et al.*, 1999) [9].

Currently, diseases are primarily controlled through the application of chemicals like fungicides. Nevertheless, the unrestricted application of these chemicals poses risks not only to living organisms but also disrupts the natural ecological equilibrium by eliminating beneficial and antagonistic microorganisms. Conversely, plant growth promoting fungi (PGPF), botanical extracts, and soil amendments offer a sustainable and cost-effective substitute to chemicals. These alternatives not only subdue pathogens and safeguard plants but also ensure environmental safety (Mukhopadhyay, 1994)^[5]. This approach mitigates the emergence of resistant pathogen strains.

Collection, isolation and identification of pathogen

Tomato plants exhibiting characteristic Sclerotium rot symptoms were identified and transported to the laboratory. The infected stem segments were collected for isolation purposes. The fungus was extracted from the stems of the afflicted Tomato plants utilizing the tissue segment method as described by Rangaswami G and Mahadevan A in 1999^[7]. This isolation was performed on a potato dextrose agar medium. The process of obtaining pure cultures of the fungal isolates was accomplished using the hyphal tip technique, following the approach outlined by Tuite J. in 1996^[10]. The identification of the fungus was carried out based on its distinctive mycelial and sclerotial characteristics, in accordance with the criteria established by Barnett H.H. and Hunter BB in 1972^[1]. To attain a pure culture of the identified *Sclerotium rolfsii*, a series of repeated cultures were conducted from the tips of individual hyphae. The resulting pure culture was then stored within PDA slants for future utilization.

Materials and Methods

Evaluation of botanical extracts on mycelial inhibition of *Sclerotium rolfsii* (in vitro)

The fungitoxicity of thirteen plant extracts viz. *Leaves Parthenium hysterophorus*, *Ricinus communis*, *Zingiber officinale*, *Jasminum angustifolium*, *Allium sativum*, *Salvia officinalis*, *Nicotiana tabacum*, *Allium cepa*, *Datura stramonium*, *Calotropis gigantea*, *Senna occidentalis* *Ocimum tenuiflorum* and *Azadirachta indica* against *Sclerotium rolfsii* was determined in a laboratory experiment. To extract their properties, hot water extraction was performed using a weight/volume ratio of 100 g plant material per 100 ml water. After filtering by muslin cloth then for 30 minutes centrifuged at 5000 rpm, the extract was sterilized. The resulting concentrate was stored in a refrigerator.

To evaluate the antifungal characteristics of these extracts, a poisoned food technique was employed. The concentrate was added to sterilized potato dextrose agar (PDA) medium during pouring, creating concentrations of 25%, 50%, 75%, and 100%. Each Petri plate received 20ml of the medium, with 5mm diameter mycelial discs taken from 7-day-old PDA cultures of the pathogen and placed at the centre of the plates. Twenty millilitres of the medium were poured into each Petri plate, and mycelial discs of 5 mm in diameter were excised from 7-day-old PDA cultures of the pathogen, utilizing a sterile cork borer. These discs were then aseptically transferred to the centre of the Petri plates containing the modified PDA medium. Control plates with PDA lacking plant extracts were maintained. Incubation occurred at 25±2 °C in an incubator. The experiment was replicated three times with the mycelial

discs of 5 mm in the center of petri plates. Radial growth of the pathogen's mycelium was observed at 24-hour intervals until control colonies reached the plate rims following the approach described by Islam *et al.*, in 2001^[4]. The percentage of inhibition over control was determined in accordance with the equation provided by Vincent in 1947^[11].

The measurement of mycelium radial growth on each plate was conducted by calculating the average of two diameters taken at right angles for each colony. The linear expansion of the causal pathogen's mycelium was observed and recorded at 24-hour intervals until the colony growth extended to the rim of the control plates, in accordance with the procedures outlined by Islam *et al.*, in 2001^[4] and Nene and Thapliyal in 1979^[16].

To determine the inhibition percentage of radial growth, the colony diameter at 4 days after inoculation (DAI) on the control plate was used as a reference. The calculation was performed using the following formula:

$$I = C-T/C \times 100$$

Where

I = per cent inhibition of mycelial growth

C= growth of mycelium in control.

T = growth of mycelium in treatment

Results and Discussion

Evaluation of botanical extracts on mycelial inhibition of *Sclerotium rolfsii* (in vitro)

The effectiveness of thirteen extracts from botanical plants was examined at four distinct concentrations each, using the poisoned food technique. The outcomes for the mycelial growth of *Sclerotium rolfsii* and the percentage of growth inhibition have been displayed in a table. The findings indicated that notable distinctions were observed in all instances between the untreated control group and the various treatments.

Among the thirteen evaluated botanicals, *Allium sativum* revealed highest fungicidal efficacy against *Sclerotium rolfsii* at all tested concentration. It showed 100% of mycelial growth inhibition at 25%, 50%, 75% and 100% of botanical concentrations respectively. Followed by *Jasminum angustifolium*. It showed 57.14%, 61.90%, 65.71% and 70.95% of mycelial growth inhibition at 25%, 50%, 75% and 100% of botanical concentrations respectively.

The latest research revealed that *Allium sativum* displayed the highest effectiveness against *Sclerotium rolfsii* at various concentrations compared to all other treatments. The current findings align with prior researcher's observations. The well-established fungicidal properties of *Allium sativum* are particularly evident in contrast with human pathogens and also extend to plant pathogens. This plant contains diverse antimicrobial components like allicin, iso-E- 10devinylojoene, E-and Z-ajoene, and similar compounds that demonstrate efficacy in contrast to bacteria, yeasts, and plant-damaging fungi (Prithviraj *et al.*, 1998; Yoshida *et al.*, 1999)^[12, 13]. Garlic harbors specific sulfur-containing compounds such as ajoene, dithin, allicin, diallylsulphide, and sallylcysteine (Cavallito *et al.*, 1945 and Ross *et al.*, 2000)^[14, 15]. These constituents exhibited superior antifungal properties compared to both streptomycin and ampicillin antibiotics (ILIC *et al.*, 2012)^[16].

Table 1: Effect of selected botanical extracts on mycelial inhibition of *Sclerotium rolfsii* in vitro

Treatment No.	Botanical name	25%	50%	75%	100%
T ₁	<i>Parthenium hysterophorus</i>	0(0)	0(0)	0(0)	0(0)
T ₂	<i>Ricinus communis</i>	0(0)	0(0)	0(0)	0(0)
T ₃	<i>Zingiber officinale</i>	40.47(39.50)	49.52(44.72)	56.67(48.8)	70.47(57.08)
T ₄	<i>Jasminum angustifolium</i>	57.14(49.10)	61.90(51.88)	65.71(54.16)	70.95(57.38)
T ₅	<i>Allium sativum</i>	100(90.00)	100(90.00)	100(90.00)	100(90.00)
T ₆	<i>Salvia officinalis</i>	8.571(16.98)	10.47(18.87)	17.14(24.44)	31.42(34.09)
T ₇	<i>Nicotiana tabacum</i>	20.95(27.23)	42.85(40.89)	60(50.77)	66.67(54.73)
T ₈	<i>Allium cepa</i>	0(0)	24.76(28.10)	15.71(23.34)	40.47(39.50)
T ₉	<i>Datura stramonium</i>	21.42(27.57)	20.95(27.23)	21.42(27.57)	21.42(27.57)
T ₁₀	<i>Calotropis gigantea</i>	0(0)	3.333(10.47)	4.285(11.83)	9.523(17.96)
T ₁₁	<i>Senna occidentalis</i>	18.57(25.51)	31.42(34.09)	44.28(41.71)	57.61(49.38)
T ₁₂	<i>Ocimum tenuiflorum</i>	5.714(13.75)	7.142(15.45)	15.71(23.34)	18.57(25.51)
T ₁₃	<i>Azadirachta indica</i>	0(0)	4.285(11.83)	9.523(17.96)	14.28(22.19)
T ₁₄	Control	0(0)	0(0)	0(0)	0(0)
	Mean	19.49	25.47	49.47	44.92
	SEd	0.804	5.628	0.604	0.577
	CD 1%	2.224	15.55	1.669	1.595
	CV	5.058	27.05	1.495	1.573

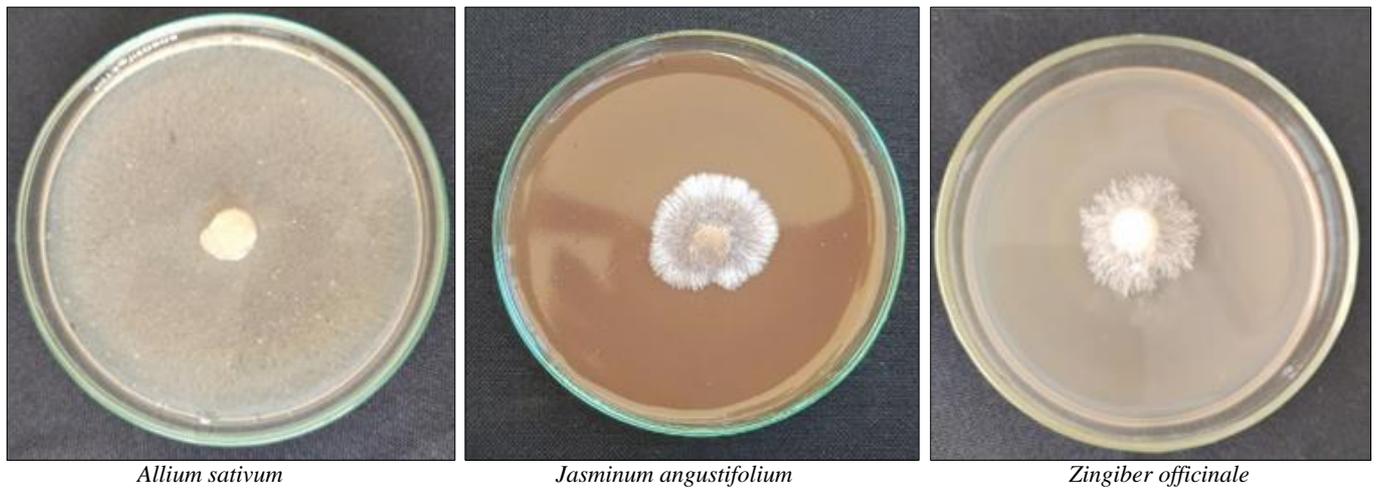


Plate 1: Effect of botanicals in mycelial inhibition of *Sclerotium rolfsii* (in vitro) at 100% concentration

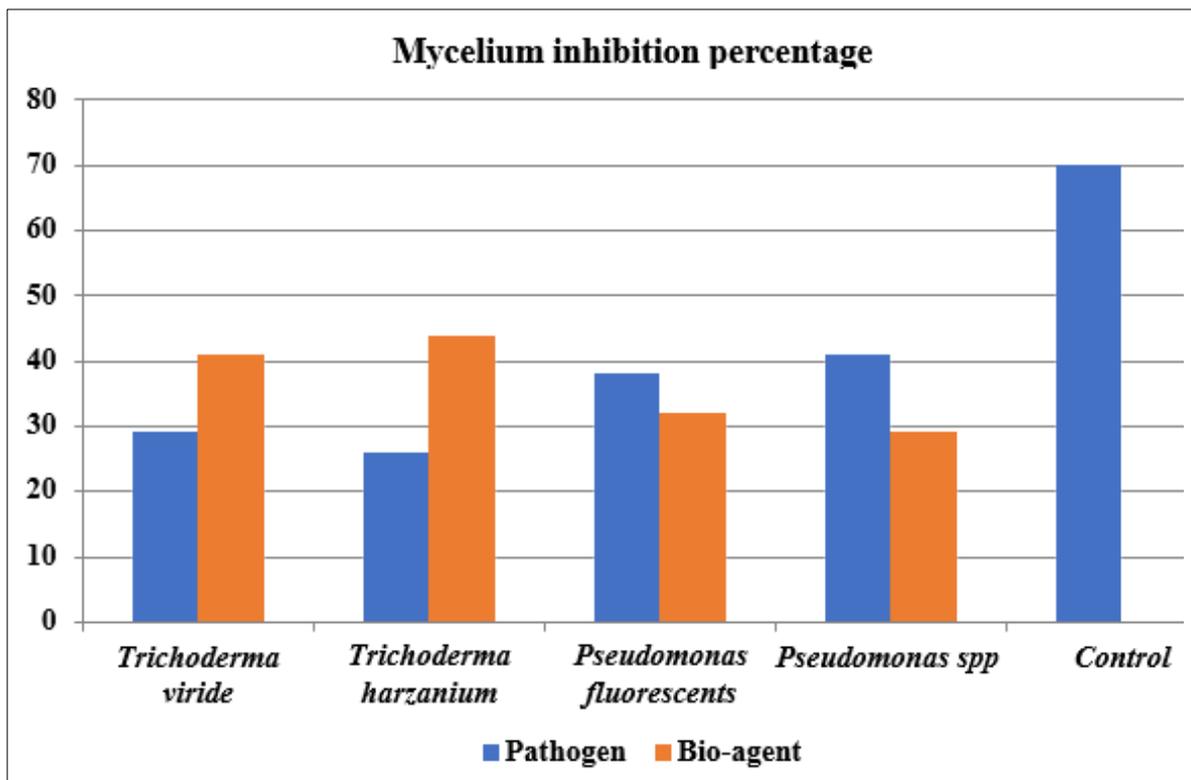


Fig 1: Effect of selected botanical extracts on mycelial inhibition of pathogen Reference

Conclusion

In this present investigation, an attempt has been made to manage the disease using various botanical under *in vitro* condition. Further research and exploration of these plant extracts can contribute to the development of eco-friendly and sustainable strategies for disease control in Tomato cultivation. These results highlight the potential of *Allium sativum* and *Jasminum angustifolium* as natural alternatives for managing collar rot resulting from *Sclerotium rolfsii* in Tomato plants. The antifungal properties of garlic extract have been attributed to its bioactive compounds, such as allicin.

Acknowledgement

We extend our heartfelt gratitude to guide as well as dean and all individuals who generously contributed to this research endeavor and provided invaluable guidance throughout the process. Their support and expertise were instrumental in shaping the trajectory of this study. Their dedication to sharing knowledge and insights has been pivotal in making this work possible. We are deeply indebted to their assistance and mentorship.

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