



Antifungal Activity of Plant Extracts in Different Solvents against *Rhizoctonia solani* Causing Sheath Blight disease in Rice

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

Article Information

DOI: <https://doi.org/10.9734/jsrr/2024/v30i92384>

Open Peer Review History:

This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: <https://www.sdiarticle5.com/review-history/122290>

Original Research Article

Received: 28/06/2024

Accepted: 31/08/2024

Published: 05/09/2024

ABSTRACT

Sheath blight of rice caused by *Rhizoctonia solani* (*R. solani*) leads to significant yield losses under severe disease conditions. This study investigates the efficacy of various aqueous and ethanol plant extracts against the mycelial growth and sclerotial production of *R. solani* under *In vitro* condition and sheath blight disease in rice under polyhouse condition. Among the aqueous extracts, *Datura stramonium* showed the highest efficacy, reducing mycelial growth from 13.17 mm at 20% concentration to 3.34 mm at 50%, with 85.37% to 96.29% inhibition. *Cannabis sp.* and *Calotropis gigantean* also exhibited strong antifungal properties, while *Aegle marmelos*, *Aloe barbadensis* and *Azadirachta indica* showed moderate effectiveness. *Nerium oleander* was the least effective. Ethanol extracts of *Aegle marmelos*, *Calotropis gigantean*, and *Datura stramonium* completely

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inhibited mycelial growth at all concentrations, with *Azadirachta indica*, *Cannabis sp.* and *Aloe barbadensis* also demonstrating high efficacy. *Datura stramonium* aqueous extract was the most effective in reducing sclerotium production, while ethanol extracts of *Aegle marmelos*, *Calotropis gigantean*, *Datura stramonium*, and *Azadirachta indica* completely inhibited it. In polyhouse conditions, aqueous and ethanol extracts of *Datura stramonium* and *Cannabis sp.* were the most effective in reducing rice sheath blight severity, with *Datura stramonium* reducing severity to 10.96% and 7.67%, respectively and *Cannabis sp.* to 11.78% and 8.25%. *Calotropis gigantean* and *Aegle marmelos* also showed notable effectiveness. These findings highlight the potential of *Datura stramonium* and *Cannabis sp.* as eco-friendly agents for managing *R. solani* and rice sheath blight disease.

Keywords: Sheath blight of rice; *Rhizoctonia solani*; plant extracts.

1. INTRODUCTION

Rice (*Oryza sativa* L.) is a crucial and extensively cultivated food crop worldwide. Annually, global rice production reaches 503 million metric tons [1]. In India, during the 2021-22 period, rice was cultivated on 46.38 million hectares, yielding 130.29 million tons with an average productivity of 2.809 tons per hectare [2]. Despite its significance, rice production is severely threatened by various biotic and abiotic stresses, with diseases and pests causing substantial yield losses [3,4]. Sheath blight, caused by *Rhizoctonia solani* [*Thanatephorus cucumeris* (teleomorph)], is the second most critical fungal disease after rice blast [5,6], can lead to yield reductions ranging from 20 to 50%, depending on disease severity [7,8]. The fungus *R. solani* infects over 188 genera across 32 plant families and is challenging to control due to its soil-borne nature and survival as sclerotia [9]. In rice, sheath blight disease starts as lesions on lower leaf sheaths that expand into water-soaked spots, and under high humidity (>95%) and temperatures (29-32°C), it spreads to upper plant parts, causing 'banded blight' with tan lesions and brown margins [10,6].

Traditionally, the control of sheath blight disease has relied heavily on the application of synthetic fungicides. However, this approach poses several problems, including the development of resistance, the presence of pesticide residues, and increased production costs [11,12]. In light of these issues, there is a growing interest in exploring alternative, sustainable methods for disease management. Recent studies have shown that plant-based products with phytochemicals like steroids, tannins, flavonoids, alkaloids, and saponins offer effective alternatives to chemical fungicides due to their antimicrobial properties. Numerous studies have documented the antifungal properties of various plant extracts against plant pathogenic fungi [6,13,14]. For

instance, Khoa et al. [15] demonstrated that seed soaking and foliar spraying with extracts from either fresh or dried leaves of *Chromolaena odorata* could reduce sheath blight disease by 68% under controlled and semi-field conditions. Similarly, other researchers have reported the inhibitory effects and disease control mechanisms of different plant extracts against *R. solani* [16,17,18].

For example, botanical extracts from *Meliaceae* species have been tested against brown spot of rice [19], while aqueous extracts of leaves from *Azadirachta indica*, *Embllica officinalis*, *Pongamia glabra*, and *Acacia nilotica* have shown effectiveness against both rice blast and brown spot [13]. Moreover, extracts of *Tagetes patula*, *Canna gigantea*, *Chamaedorea curassavica*, *Allium fistulosum*, and *Aegle marmelos* exhibited inhibition of *Magnaporthe oryzae*, the pathogen responsible for rice blast [14]. Kumar and Simon [20] also evaluated various plant extracts for their effectiveness against brown spot in rice, and extracts from plants such as *Azadirachta indica*, *Nerium oleander*, *Curcuma longa*, *S. indicum*, and *Cymbopogon citratus* have been found suitable for managing brown spot in rice [21]. Given the increasing recognition of the role of botanicals in sustainable agriculture, the present study aimed to evaluate the antifungal properties of aqueous and ethanol extracts from various botanicals (*Aegle marmelos*, *Aloe barbadensis*, *Calotropis gigantea*, *Cannabis sp.*, *Datura stramonium*, *Nerium oleander*, *Azadirachta indica*) against *R. solani* causing sheath blight disease in rice.

2. MATERIALS AND METHODS

The study was conducted during the period of January to May 2020 at the Department of Plant Pathology, Dr. Rajendra Prasad Central Agricultural University, Pusa, Samastipur, Bihar, India.

2.1 Isolation of *R. solani*

The plant samples with severe sheath blight symptoms were collected from the Rice fields of Dr. Rajendra Prasad Central Agricultural University, Pusa (Samastipur), Bihar, India. The samples were transferred to the Plant Pathology laboratory for isolation of the pathogen. The rice tiller with disease symptom including a piece of healthy tissue were cut into small pieces (0.5–1.0 cm) and surface sterilized with 1% sodium hypochlorite for two minutes. The samples were then washed three times with sterile distilled water and placed onto PDA plates for incubation at 28 ± 2 °C for 3 days. The identification was performed by observing the morphology and mycelial characteristics of the pathogen. After identification, a mycelial disc (5 mm) from the actively growing zone of the 3 days old culture was placed onto PDA plates to obtain pure culture [6].

2.2 Collection and Preparation of Plant Extracts

Fresh and healthy parts of eleven plants (*Aegle marmelos*, *Aloe barbadensis*, *Calotropis gigantea*, *Cannabis sp.*, *Datura stramonium*, *Nerium oleander*, *Azadirachta indica*) were collected from surrounding areas of Dr. Rajendra Prasad Central Agricultural University, Pusa (Samastipur), Bihar, India. The samples were washed with running tap water and then rinsed two times with sterile distilled water and air dried for 2–3 h. The samples were then cut into small pieces (1–2 cm). For preparation of aqueous extracts One hundred gram of sample was ground with 100 ml of sterile distilled water (1:1 W/V) using mortar and pestle and filtered through a double layer white muslin cloth. The filtrate was collected and stored in a conical flask at 25–28 °C for further study. This served as 100% stock solution of the plant extract. Distilled water was replaced with ethanol for ethanol plant extract [6].

2.3 Evaluation of Plant Extracts Against *R. solani* under *In vitro*

Aqueous and ethanol extracts of botanicals were tested at 20%, 30%, 40% and 50% of concentrations. To get desired concentrations, 20 ml, 30 ml and 40 ml of standard stock solutions were poured in 80 ml, 70 ml and 60 ml of sterilized molten PDA respectively. After that, they were poured into Petri plates under aseptic condition. Later 9 mm of pathogen (3 to 4 days old culture) containing PDA disc was transferred

on to solidified treated media. Culture transferred media which was not treated with aqueous/ethanol extracts of botanicals was used as control. Each treatment has been done with three replications. Inoculated plates were kept in BOD at 28 ± 1 °C. The diameter of fungal growth was recorded after 72 hours of incubation.

By using following formula, per cent growth inhibition of *Rhizoctonia solani* was calculated.

$$\text{Inhibition percentage} = \frac{(C-T) \times 100}{C}$$

Where

C = Diameter of fungal growth (mm) in control plate.

T = Diameter of fungal growth (mm) in treated plate.

After 120 hours of incubation in different treatments the number of sclerotia formation was recorded in three replications.

2.4 Statistical Analysis

The experiments were carried out using a completely randomized design (CRD), and the data were analyzed through a one-way analysis of variance (ANOVA) at a 5% significance level ($P \leq 0.05$) in OPSTAT. Each experiment included three replications.

3. RESULTS

3.1 Efficacy of Plant Extracts Against *R. solani* under *In vitro*

The study revealed that among the various aqueous plant extracts tested (Table 1), *Datura stramonium* demonstrated the highest efficacy against *R. solani*, significantly reducing mycelial growth from 13.17 mm at 20% concentration to 3.34 mm at 50%, and achieving percent growth inhibition ranging from 85.37% to 96.29%. *Cannabis sp.* also showed strong antifungal properties, with mycelial growth decreasing from 26.17 mm at 20% concentration to 14.37 mm at 50%, and percent inhibition ranging from 70.92% to 84.03%. *Calotropis gigantean* exhibited notable effectiveness, with mycelial growth reducing from 46.83 mm at 20% concentration to 18.64 mm at 50%, and percent growth inhibition between 47.97% and 79.29%. Other extracts, such as *Aegle marmelos*, *Aloe barbadensis* and *Azadirachta indica*, also displayed varying degrees of effectiveness. *Nerium oleander* exhibited the least effectiveness among the tested extracts, with mycelial growth reducing

from 58.50 mm at 20% concentration to 41.51 mm at 50%, and percent inhibition ranging from 35.00% to 53.88%.

The antifungal activity of ethanol extracts of botanicals against *R. solani* (Table 2), revealing that, the extracts from *Aegle marmelos*, *Calotropis gigantean*, and *Datura stramonium* demonstrated the highest efficacy, achieving complete inhibition of mycelial growth (100%) at all tested concentrations (20%, 30%, 40%, and 50%). *Azadirachta indica* was also highly effective, showing complete inhibition at concentrations of 30% and above, with 85.00% inhibition at 20%. *Cannabis sp.* displayed significant inhibition, reducing mycelial growth to 0.00 mm at 50% concentration, with percent inhibition ranging from 72.97% to 100%. *Aloe barbadensis* exhibited moderate to strong antifungal activity, with mycelial growth decreasing from 53.50 mm at 20% concentration to 10.54 mm at 50%, achieving up to 88.29% inhibition. *Nerium oleander* showed moderate effectiveness, with mycelial growth reducing from 14.67 mm at 20% concentration to 0.00 mm at 50%, with percent inhibition ranging from 83.70% to 100%.

Similar to the study conducted by Persaud et al. [6] against *Rhizoctonia solani*, which revealed that extracts of lemon grass, thick leaf thyme, and clove recorded significantly lowest mycelial growth (5.00 mm each) and highest percent inhibition (94.44% each) at 15% concentration *in vitro*, our study demonstrated the high efficacy of various plant extracts in reducing mycelial growth. Specifically, our results showed that ethanol extracts of *Aegle marmelos*, *Calotropis gigantean*, and *Datura stramonium* achieved complete inhibition of mycelial growth (100%) at all tested concentrations (20, 30, 40 and 50%). Additionally, *Azadirachta indica* showed complete inhibition at concentrations of 30% and above, while *Cannabis sp.* significantly inhibited mycelial growth, achieving 100% inhibition at 50% concentration. For aqueous extracts, *Datura stramonium*, *Calotropis gigantean*, and *Aegle marmelos* also exhibited high efficacy, achieving complete inhibition of mycelial growth (100%) at all concentrations tested. These findings corroborate the antifungal potential of both ethanol and aqueous plant extracts against *R. solani*, underscoring their promise as eco-friendly alternatives for disease management. Furthermore, in addition to *R. solani*, Persaud et al. [14] demonstrated the effect of plant extracts on the mycelial growth of *Magnaporthe oryzae*. Extracts of *Tagetes patula* (5%), *Calotropis*

gigantea (5%), *C. curassavica* (10%), *A. fistulosum* (10%), and *A. marmelos* (15%) showed greater than 81% inhibition to *M. oryzae in vitro*, further supporting the broad-spectrum antifungal efficacy of these plant extracts.

3.2 Effect of Plant Extracts on Sclerotia Production of *R. solani*

The investigation on effectiveness of aqueous and ethanol extracts of botanicals in reducing sclerotium production of *R. solani* after 120 hours of incubation revealed that, among the aqueous extracts, *Datura stramonium* was the most effective, reducing sclerotium production from 21.00 to 5.33 across 20% to 50% concentrations. *Cannabis sp.* and *Aegle marmelos* also demonstrated significant reductions, with *Cannabis sp.* reducing production from 42.33 to 23.67 and *Aegle marmelos* from 50.33 to 29.00. *Aloe barbadensis* and *Azadirachta indica* showed moderate effectiveness, while *Nerium oleander* was less effective, reducing sclerotium production from 91.00 to 36.67. Conversely, ethanol extracts from *Aegle marmelos*, *Calotropis gigantean*, *Datura stramonium*, and *Azadirachta indica* were highly effective, completely inhibiting sclerotium production at all concentrations. *Cannabis sp.* and *Nerium oleander* also demonstrated strong efficacy, with significant reductions observed. *Aloe barbadensis* showed moderate effectiveness, with reductions from 76.33 to 7.00. In contrast, the control group showed no reduction, maintaining consistent sclerotium production at 146.00 across all concentrations.

These findings are consistent with those of Sriraj et al. [22], who reported that the leaf extracts of *Azadirachta indica*, *Lumnitzera littorea*, and the seed extract of *Melia longifolia* completely inhibited sclerotial formation at all tested concentrations (10%, 15%, and 20%). Moreover, Singh et al. [23] found that the extracts of *Tegetes erecta* and *Azadirachta indica* caused maximum inhibition of sclerotial production and its size in *Sclerotium rolfsii*. Our results align with these findings, as the ethanol extract of *Azadirachta indica* showed complete inhibition of sclerotial production at concentrations of 30% and above, with significant reduction at 20%. Overall, both aqueous and ethanol extracts of *Datura stramonium* and ethanol extracts of *Aegle marmelos*, *Calotropis gigantean*, and *Azadirachta indica* demonstrated high efficacy in reducing sclerotial formation of *Rhizoctonia solani*. These extracts have potential as eco-friendly agents for managing rice sheath blight.

Table 1. Efficacy of aqueous plant extracts against *R. solani* under *In vitro*

Aqueous plant extract	Mycelial growth of <i>R. solani</i> (mm)				Per cent growth inhibition			
	20%	30%	40%	50%	20%	30%	40%	50%
<i>Aegle marmelos</i>	34.17 ^c ±0.85*	31.83 ^c ±0.81	28.17 ^c ±0.18	25.12 ^d ±0.08	62.03 ^e ±1.07	64.63 ^f ±1.31	68.70 ^d ±0.36	72.09 ^e ±1.24
<i>Aloe barbadensis</i>	58.33 ^f ±0.64	49.83 ^f ±0.93	44.33 ^e ±0.71	36.75 ^f ±0.92	35.19 ^b ±0.711	44.63 ^c ±0.98	50.75 ^b ±1.11	59.17 ^c ±0.12
<i>Azadirachta indica</i>	55.33 ^e ±0.58	46.50 ^e ±0.34	37.83 ^d ±0.28	29.38 ^e ±0.03	38.52 ^c ±0.40	48.33 ^d ±0.95	57.97 ^c ±0.42	67.36 ^d ±0.84
<i>Calotropis gigantean</i>	46.83 ^d ±1.00	36.50 ^d ±0.15	27.00 ^c ±0.24	18.64 ^c ±0.44	47.97 ^d ±0.73	59.44 ^e ±1.24	70.00 ^d ±1.79	79.29 ^f ±1.73
<i>Cannabis sp.</i>	26.17 ^b ±0.13	22.50 ^b ±0.48	18.83 ^b ±0.43	14.37 ^b ±0.29	70.92 ^f ±1.00	75.00 ^g ±0.74	79.08 ^e ±0.66	84.03 ^g ±0.87
<i>Datura stramonium</i>	13.17 ^a ±0.24	9.50 ^a ±0.20	5.17 ^a ±0.06	3.34 ^a ±0.06	85.37 ^g ±2.04	89.44 ^h ±2.00	94.26 ^f ±1.18	96.29 ^h ±1.10
<i>Nerium oleander</i>	58.50 ^f ±0.43	53.67 ^g ±1.31	46.33 ^f ±1.18	41.51 ^g ±0.73	35.00 ^b ±0.33	40.36 ^b ±0.44	48.52 ^b ±0.96	53.88 ^b ±0.76
Control	90.00 ^g ±0.00	90.00 ^h ±0.00	90.00 ^g ±0.00	90.00 ^h ±0.00	0.00a	0.00a ±	0.00a ±	0.00a ±
C.D. (p<0.05)	1.763	2.045	1.606	1.378	2.948	3.358	2.921	2.988
SEm±	0.583	0.676	0.531	0.456	0.975	1.111	0.966	0.988

*Mean standard error, means followed by different letters are significantly different from each other according to Duncan's Multiple Range Test at the 0.05 significance level.

Table 2. Efficacy of ethanol plant extracts against *R. solani* under *In vitro*

Ethanol plant extract	Mycelial growth of <i>R. solani</i> (mm)				Per cent growth inhibition			
	20%	30%	40%	50%	20%	30%	40%	50%
<i>Aegle marmelos</i>	0.00 ^a ±0.00*	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	100.00 ^f ±0.00	100.00 ^e ±0.00	100.00 ^d ±0.00	100.00 ^c ±0.00
<i>Aloe barbadensis</i>	53.50 ^e ±0.72	41.83 ^d ±0.90	24.67 ^c ±0.36	10.54 ^b ±0.27	40.56 ^b ±0.80	53.52 ^b ±0.99	72.59 ^b ±0.40	88.29 ^b ±0.30
<i>Azadirachta indica</i>	13.50 ^b ±0.13	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	85.00 ^d ±0.14	100.00 ^e ±0.00	100.00 ^d ±0.00	100.00 ^c ±0.00
<i>Calotropis gigantean</i>	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	100.00 ^f ±0.00	100.00 ^e ±0.00	100.00 ^d ±0.00	100.00 ^c ±0.00
<i>Cannabis sp.</i>	24.33 ^d ±0.54	13.50 ^c ±0.28	4.33 ^b ±0.09	0.00 ^a ±0.00	72.97 ^c ±0.61	85.00 ^c ±0.31	95.19 ^c ±0.10	100.00 ^c ±0.00
<i>Datura stramonium</i>	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	100.00 ^f ±0.00	100.00 ^e ±0.00	100.00 ^d ±0.00	100.00 ^c ±0.00
<i>Nerium oleander</i>	14.67 ^c ±0.21	10.83 ^b ±0.13	0.00 ^a ±0.00	0.00 ^a ±0.00	83.70 ^d ±0.24	87.97 ^d ±0.14	100.00 ^d ±0.00	100.00 ^c ±0.00
Control	90.00 ^f ±0.00	90.00 ^e ±0.00	90.00 ^d ±0.00	90.00 ^c ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00
C.D. (p<0.05)	1.003	1.013	0.396	0.289	1.115	1.121	0.442	0.318
SEm±	0.332	0.335	0.131	0.096	0.369	0.371	0.146	0.105

*Mean standard error, means followed by different letters are significantly different from each other according to Duncan's Multiple Range Test at the 0.05 significance level.

Table 3. Effect of aqueous and ethanol plant extracts on number of sclerotia production by *R. solani* under *In vitro*

Plant extract	Aqueous plant extract				Ethanol plant extract			
	20%	30%	40%	50%	20%	30%	40%	50%
<i>Aegle marmelos</i>	50.33 ^c ±3.38*	44.00 ^c ±1.73	35.67 ^b ±1.76	29.00 ^b ±2.31	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00
<i>Aloe barbadensis</i>	92.00 ^f ±3.22	77.67 ^f ±1.86	64.00 ^e ±2.31	49.67 ^e ±2.60	76.33 ^d ±2.01	57.67 ^d ±1.76	30.33 ^c ±2.33	7.00 ^b ±1.16
<i>Azadirachta indica</i>	84.00 ^e ±2.08	67.33 ^e ±2.13	55.67 ^d ±2.60	40.67 ^d ±2.60	22.00 ^b ±1.53	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00
<i>Calotropis gigantean</i>	76.00 ^d ±1.53	59.33 ^d ±2.40	43.67 ^c ±2.33	30.33 ^{bc} ±2.03	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00
<i>Cannabis sp.</i>	42.33 ^b ±1.45	36.67 ^b ±3.18	30.33 ^b ±2.03	23.67 ^b ±2.13	29.67 ^c ±2.00	12.00 ^b ±1.73	3.00 ^b ±1.53	0.00 ^a ±0.00
<i>Datura stramonium</i>	21.00 ^a ±1.53	15.00 ^a ±2.89	8.67 ^a ±0.88	5.33 ^a ±1.20	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00
<i>Nerium oleander</i>	91.00 ^{ef} ±2.89	78.00 ^f ±1.53	53.33 ^d ±2.03	36.67 ^{cd} ±2.04	23.67 ^b ±1.35	17.67 ^c ±1.45	0.00 ^a ±0.00	0.00 ^a ±0.00
Control	146.00 ^g ±2.65	146.00 ^g ±2.65	146.00 ^f ±2.65	146.00 ^f ±2.65	146.00 ^e ±2.65	146.00 ^e ±2.65	146.00 ^d ±2.65	146.00 ^c ±2.65
C.D. (p<0.05)	7.424	7.207	6.474	6.733	4.741	4.171	4.11	3.086
SEm±	2.455	2.383	2.141	2.227	1.568	1.379	1.359	1.021

*Mean standard error, means followed by different letters are significantly different from each other according to Duncan's Multiple Range Test at the 0.05 significance level.

Table 4. Effect of aqueous and ethanol plant extracts against sheath blight (*R. solani*) disease of rice in polyhouse condition

Plant extract	Solvent used for the extraction	Initial (7DAI) ^A	14DAI ^B	21DAI	28DAI	35DAI	42DAI
<i>Aegle marmelos</i> @50%	Water	2.32 ^a ±0.01*	5.96 ^{abc} ±0.46	7.60 ^{abc} ±1.31	10.17 ^{bc} ±1.21	11.36 ^{bc} ±1.01	12.05 ^{bcd} ±1.13
<i>Aloe barbadensis</i> @50%	Water	2.31 ^a ±0.02	8.52 ^{cd} ±0.73	11.33 ^{de} ±1.83	15.71 ^d ±1.30	17.74 ^e ±1.16	18.92 ^f ±1.21
<i>Azadirachta indica</i> @50%	Water	2.33 ^a ±0.03	8.14 ^{cd} ±0.67	10.80 ^d ±0.72	14.96 ^d ±1.41	15.89 ^{de} ±0.98	17.01 ^{ef} ±1.35
<i>Calotropis gigantean</i> @50%	Water	2.32 ^a ±0.03	7.47 ^{bcd} ±0.66	9.72 ^{cd} ±0.55	12.23 ^{cd} ±1.14	13.87 ^{cd} ±1.02	14.82 ^{cdf} ±1.01
<i>Cannabis sp.</i> @50%	Water	2.33 ^a ±0.01	5.12 ^{ab} ±1.18	7.37 ^{abc} ±0.61	9.34 ^{abc} ±0.93	10.25 ^{ab} ±1.29	11.78 ^{bc} ±1.23
<i>Datura stramonium</i> @50%	Water	2.21 ^a ±0.04	4.61 ^{ab} ±1.25	6.25 ^{ab} ±0.12	8.24 ^{ab} ±1.13	9.69 ^{ab} ±1.28	10.96 ^{ab} ±1.00
<i>Nerium oleander</i> @50%	Water	2.32 ^a ±0.01	8.55 ^{cd} ±1.19	11.36 ^{de} ±1.20	15.75 ^d ±1.20	17.80 ^e ±0.98	18.98 ^f ±0.93
<i>Aegle marmelos</i> @50%	Ethanol	2.34 ^a ±0.04	4.71 ^{ab} ±1.23	5.89 ^{ab} ±0.15	7.16 ^{ab} ±1.13	8.29 ^{ab} ±1.12	9.36 ^{ab} ±1.33
<i>Aloe barbadensis</i> @50%	Ethanol	2.31 ^a ±0.01	6.01 ^{abc} ±0.57	8.59 ^{bcd} ±1.20	12.61 ^{cd} ±1.25	14.47 ^{cde} ±1.57	15.56 ^{def} ±1.13
<i>Azadirachta indica</i> @50%	Ethanol	2.23 ^a ±0.05	4.76 ^{ab} ±0.56	6.41 ^{ab} ±0.58	7.42 ^{ab} ±1.03	8.89 ^{ab} ±0.54	10.17 ^{ab} ±1.24
<i>Calotropis gigantean</i> @50%	Ethanol	2.32 ^a ±0.06	3.72 ^a ±0.63	5.89 ^{ab} ±0.68	7.62 ^{ab} ±1.24	8.81 ^{ab} ±1.16	9.93 ^{ab} ±1.10
<i>Cannabis sp.</i> @50%	Ethanol	2.33 ^a ±0.04	3.91 ^a ±0.57	6.08 ^{ab} ±1.47	6.91 ^{ab} ±1.22	7.76 ^{ab} ±0.44	8.25 ^{ab} ±0.24
<i>Datura stramonium</i> @50%	Ethanol	2.35 ^a ±0.05	3.45 ^a ±1.18	4.51 ^a ±0.56	5.61 ^a ±1.02	6.65 ^a ±1.00	7.67 ^a ±1.09
<i>Nerium oleander</i> @50%	Ethanol	2.23 ^a ±0.02	5.88 ^{abc} ±1.24	8.59 ^{bcd} ±1.30	9.69 ^{bc} ±0.94	10.20 ^{ab} ±1.12	10.50 ^{ab} ±1.22
Pathogen inoculated control	-	2.34 ^a ±0.02	9.58 ^d ±1.01	13.91 ^e ±1.15	20.67 ^e ±1.12	23.81 ^f ±1.56	25.63 ^g ±2.12
C.D. (p<0.05)		0.094	2.678	2.743	3.354	3.171	3.366
SEm±		0.032	0.923	0.945	1.156	1.093	1.16

^AInitial observation and treatment applied, ^BSecond treatment applied; DAI- Days after inoculation, *Mean standard error, means followed by different letters are significantly different from each other according to Duncan's Multiple Range Test at the 0.05 significance level.

3.3 Efficacy of Plant Extracts Against Sheath Blight of Rice in Polyhouse Condition

The aqueous and ethanol plant extracts showed significant reductions in rice sheath blight disease severity compared to the pathogen-inoculated control. At 42 days after inoculation (DAI), the aqueous extract of *Datura stramonium* was the most effective, reducing disease severity to 10.96%, while the pathogen-inoculated control exhibited the highest disease severity with 25.63%. Similarly, *Cannabis sp.* aqueous extract reduced severity to 11.78%, demonstrating strong antifungal activity. *Calotropis gigantean* and *Aegle marmelos* aqueous extracts also showed notable effectiveness, reducing disease severity to 14.82% and 12.05%, respectively. Among the ethanol extracts, *Datura stramonium* again showed the highest efficacy, with disease severity reduced to 7.67%. Other effective ethanol extracts included *Cannabis sp.*, *Calotropis gigantean*, *Aegle marmelos*, and *Azadirachta indica*, with reductions ranging from 8.25% to 10.17%. *Nerium oleander* aqueous extract was less effective, with disease severity at 18.98%, while its ethanol extract was more effective, reducing severity to 10.50%. Overall, both aqueous and ethanol extracts of *Datura stramonium* and *Cannabis sp.* were the most effective in reducing disease severity, highlighting their potential as eco-friendly agents for managing rice sheath blight.

4. DISCUSSION

The study revealed that among the various aqueous plant extracts tested, *Datura stramonium* demonstrated the highest efficacy against *R. solani*, significantly reducing mycelial growth and achieving high growth inhibition percentages. *Cannabis sp.* and *Calotropis gigantean* also showed strong antifungal properties, with notable reductions in mycelial growth and high inhibition rates. Other extracts, such as *Aegle marmelos*, *Aloe barbadensis*, and *Azadirachta indica*, displayed varying degrees of effectiveness, while *Nerium oleander* was the least effective. The ethanol extracts of *Aegle marmelos*, *Calotropis gigantean*, and *Datura stramonium* were the most potent, achieving complete inhibition of mycelial growth at all tested concentrations. *Azadirachta indica* also showed high efficacy, particularly at concentrations of

30% and above. These results are consistent with previous studies, such as those conducted by Persaud et al. [6], which demonstrated the effectiveness of plant extracts like lemon grass, thick leaf thyme, and clove in significantly reducing mycelial growth of *R. solani*. Our findings further corroborate the antifungal potential of both ethanol and aqueous plant extracts against *R. solani*, highlighting their promise as eco-friendly alternatives for disease management. Additionally, Persaud et al. [14] showed the broad-spectrum antifungal efficacy of plant extracts against *Magnaporthe oryzae*, supporting the potential of these botanicals in managing a range of fungal pathogens.

The investigation into the effectiveness of aqueous and ethanol extracts of botanicals in reducing sclerotium production of *R. solani* revealed that *Datura stramonium* was the most effective among the aqueous extracts, significantly reducing sclerotium production from 21.00 to 5.33 across 20% to 50% concentrations. *Cannabis sp.* and *Aegle marmelos* also demonstrated notable reductions, while *Aloe barbadensis* and *Azadirachta indica* showed moderate effectiveness. Conversely, *Nerium oleander* was less effective. Among the ethanol extracts, *Aegle marmelos*, *Calotropis gigantean*, *Datura stramonium*, and *Azadirachta indica* completely inhibited sclerotium production at all concentrations. *Cannabis sp.* and *Nerium oleander* also showed strong efficacy, while *Aloe barbadensis* exhibited moderate effectiveness. These findings align with previous studies, such as those by Sriraj et al. [22] and Singh et al. [23], which reported similar inhibitory effects of botanical extracts on sclerotial formation. Overall, both aqueous and ethanol extracts of *Datura stramonium*, and ethanol extracts of *Aegle marmelos*, *Calotropis gigantean*, and *Azadirachta indica* demonstrated high efficacy in reducing sclerotial formation of *R. solani*, highlighting their potential as eco-friendly agents for managing rice sheath blight.

The aqueous and ethanol plant extracts significantly reduced rice sheath blight severity. At 42 days after inoculation, *Datura stramonium* aqueous extract was the most effective, reducing severity to 10.96%, while the control had 25.63%. *Cannabis sp.* aqueous extract reduced severity to 11.78%, and *Calotropis gigantean* and

Aegle marmelos extracts reduced it to 14.82% and 12.05%, respectively. Among ethanol extracts, *Datura stramonium* was most effective, reducing severity to 7.67%, followed by *Cannabis* sp., *Calotropis gigantean*, *Aegle marmelos*, and *Azadirachta indica*, with reductions from 8.25% to 10.17%. Overall, *Datura stramonium* and *Cannabis* sp. were the most effective, demonstrating potential as eco-friendly agents for managing rice sheath blight. These findings align with the study conducted by Persaud et al. [6], who reported that the extracts of lemon grass (7.21%; 8.04%; 4.85%) and thick leaf thyme (6.71%; 7.28%; 4.71%) at 15% recorded low disease severity of rice sheath blight in greenhouse, field trial I and II compared to untreated control (26.25%; 31.16%; 20.43%). Similarly, Persaud et al. [14] demonstrated that in trials against rice blast disease, the application of *Aegle marmelos* (16.60 mm; 46.67%), *C. curassavica* (18.87 mm; 48.15%), and *C. gigantea* (18.87 mm; 48.89%) significantly reduced lesion length and disease severity in Trial I. In Trial II, *C. curassavica* (19.00 mm; 56.29%), *A. marmelos* (18.07 mm; 57.78%), and *C. gigantea* (20.84 mm; 70.37%) again significantly reduced lesion length and disease severity compared to the untreated control.

5. CONCLUSION

This study demonstrated the significant potential of plant extracts, particularly *Datura stramonium* and *Cannabis* sp., in managing sheath blight of rice caused by *Rhizoctonia solani*. Both aqueous and ethanol extracts of these plants were effective in inhibiting mycelial growth and sclerotial production of *R. solani* under *In vitro* conditions, with *Datura stramonium* showing the highest efficacy. The polyhouse experiments further confirmed the ability of these extracts to reduce sheath blight severity in rice, suggesting that they could serve as eco-friendly alternatives to chemical fungicides. The strong antifungal properties of *Datura stramonium*, *Cannabis* sp., *Calotropis gigantean*, and *Aegle marmelos* highlight their potential in integrated disease management strategies for sustainable rice cultivation. Further research is warranted to optimize their application methods and evaluate their effectiveness under field conditions.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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