International Journal of Plant & Soil Science

Volume 36, Issue 8, Page 28-35, 2024; Article no.IJPSS.114319
ISSN: 2320-7035

Evaluation of Different Botanicals Against *Sclerotium rolfsii* Causing Collar Rot Disease of Lentil

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: https://doi.org/10.9734/ijpss/2024/v36i84831

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/114319

Original Research Article

Received: 13/01/2024
Accepted: 15/03/2024
Published: 15/07/2024

ABSTRACT

Lentil (*Lens culinaris* Medik.) is an important pulse crop in semiarid regions of Iran, India, *Turkey* and Canada and originated in the fertile crescent of the Near East and dates back to the beginning of agriculture itself. Lentil suffer from attack of number seed borne diseases such as vascular wilt, collar rot, root rot, stem rot, rust, powdery mildew and downy mildew, which are caused by *Fusarium oxysporum* f.sp*. lentis, Sclerotium rolfsii*, *Rhizoctonia solani, Uromycis fabae*, *Erysiphe polygoni* and *Peronospora lentis*, respectively. Among the diseases, foot and root rot of lentil caused by *Sclerotium rolfsii* are common and the most severe disease. The fungi can attack the

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Cite as: *Chopra, Suman, Reeti Singh, Smriti Akodiya, Rajkumar Bajya, Ravi Regar, and Vedant Gautam. 2024. "Evaluation of Different Botanicals Against Sclerotium Rolfsii Causing Collar Rot Disease of Lentil". International Journal of Plant & Soil Science 36 (8):28-35. https://doi.org/10.9734/ijpss/2024/v36i84831.*

crop at any stage from seedling to flowering stage and are comparatively more destructive at the seedling stage. The effect of phyto extracts of nine plant species were tested *in vitro* by poisoned food technique to know their inhibitory effect on the growth of *Sclerotium rolfsii*. Significantly minimum mycelium growth was recorded in *Curcuma longa* (39.25 mm) while maximum mycelium growth was observed in *Ricinus communis* (90.00 mm).

Keywords: Lentil; pulse crop; borne diseases; fungal diseases; mustard seed; natural origin; plant pathology; Sclerotium rolfsii.

1. INTRODUCTION

"Lentil (*Lens culinaris* Medik.) is an important pulse crop. Lentil is utilized for human consumption as an edible protein resources.It is a good source of vitamin A, potassium, fiber, iron and vitamins B" [1].

"Among the fungal diseases, collar rot of lentil caused by *S. rolfsii* are generic and the most solemn disease*. Sclerotium rolfsii* can invade the crop during seedling to flowering stage. The pathogen *S. rolfsii* is more severe in the early stage. The fungus is soil-borne and produces survival structure sclerotia, which can survive very long time in the soil. *S. rolfsii* affects the lower stem and roots of lentil at or near the soil line. During infection whitish mycelia growth of the fungus can be seen at the junction of the branch with the stem close to the soil level, which is the most favorable point of attack. In advanced stage of disease, a white mycelial web dispersion over the soil and the basal plant part of the plant and sclerotia of mustard seed size are observed on the diseased area. In its advanced stage infection becomes prominent in the root system and subsequently the entire shoot withers and falls and the plant eventually die" [2].

"The use of plant extracts has been shown to be ecofriendly and effective against many plant pathogens" [3,4]. Botanicals are naturally occurring, environmentally benign and biodegradable. The current investigation was conducted with consideration for host target specificity, cost effectiveness, and nature safety. Based on the aforementioned information, Madhya Pradesh needs this kind of research. In order to evaluate the antifungal activity of additional plant extracts against the in vitro growth of *S. rolfsi*, the current investigation was conducted.

2. MATERIALS AND METHODS

The experiment was carried out at Department of Plant Pathology, College of Agriculture, Gwalior. The effect of botanicals of various plant species as listed in Table-1 were tested *in vitro* by poisoned food technique to know their inhibitory effect on the growth of *S. rolfsii*. The experiment was laid out in complete randomized design and replicated four times. Healthy fresh plant leaves were taken, washed thoroughly with fresh water and finally raised with sterilized water and air dried on blotter paper. Fifty gram of fresh plant parts were cut into small pieces and minced with help of grinder and 50 ml distilled water was added. The botanical extracts were filtered through double layered muslin cloth in 150 ml conical flasks and plugged with non absorbent cotton. The filtered extracts were autoclaved at 15 lb per square inch pressure for 20 minutes. Autoclaved extracts were individually added into molten sterilized potato dextrose agar @ 20 per cent mixed thoroughly at time of pouring in the previously sterilized Petri dishes. The Petri dishes were incubated aseptically after solidification by placing 5 mm diameter mycelia disc at the centre. Ten days old pure culture of *Sclerotium rolfsii* was used. The plate without phytoextract served as control. The Petri dishes were incubated at 25±2ºC. The observation was recorded at 120 and 168 hrs of inoculation. The per cent growth inhibition of the pathogen was worked out by using formula given by Vincent [5].

$$PGI = \frac{C-T}{C} \times 100$$

Where,

PGI = Per cent growth inhibition
C = Growth in control
T = Growth in botanicals

Table 1. List of botanicals

Serial Number	Botanicals	Plant part Used	%/100ml	Common name	Family	Constituents
1.	*Curcuma longa*	Powder	20%	Haldi	Zingiberaceae	Curcumenol, curdione, curcumin, isocurcumenol, curcumol, stigmasterol, zingiberene and curcumene
2.	*Michelia champaca*	Leaf	20%	Champaca	Magnoliaceae	Phenyl acetonitril, phenyl ethyl alcohol, alpha + beta ionone, methyl, methyl anthranilate, indole and methyl linoleate.
3.	*Ricinus communis*	Leaf	20%	Castor	Euphorbiaceae	Alkaloid, ricinoleic acid, stearic, linoleic, palmatic acid, sitosterol, squalenetocopherols and stearic acid
4.	*Nerium Oleander*	Leaf	20%	Kaner	Apocynaceae	Oleandrin, digitoxigenin and gitoxigenin
5.	*Ficus religiosa*	Leaf	20%	Pipal	Moraceae	Phenols, tannins, steroids, alkaloid and flavonoids, beta-sitosteryl-D-glucoside, vitamin K, n- octacosanol, methyl oleanolate, lanosterol, stigma sterol.
6.	*Pisidium guajava*	Leaf	20 %	Guava	Myrtaceae	Guajanic acid, beta-sitosterol, uvaol, oleanolic acid and ursolic acid.
7.	*Duranta erecta*	Leaf	20 %	Sky flower	Verbenaceae	Coumarinolignoids, (E)- cinnamic acid, (E)- p-methoxycinnamic acid, and lamiide, β- sitosterol, naringenin, acteoside, lamiide, sucrose, and raffinose
8.	*Saraca asoca*	Leaf	20 %	Ashok	Fabaceae	Quercetin, quercetin-3-O-α-Lrhamnoside, kaempferol 3-O- αL rhamnoside, amyrin, ceryl alcohol and β sitosterol
9.	*Tridax Procumbens*	Leaf	20 %	Tridax	Asteraceae	Alkyl esters, sterols, pentacyclic triterpenes, fatty acid and polysaccharides

3. RESULTS

3.1 Effect of Plant Extracts on Mycelial Growth of *Sclerotium rolfsii*

Fungicidal ability of nine botanicals *viz.,Curcurma longa* powder *,Michelia champaca* (champa), *Ricinus communis* (castor), *Nerium oleander* (kaner), *Ficus religiosa* (pipal), *Tridax procumbens, Psidium guajava*, *Duranta erecta* (hedge) and *Saraca asoca* (Ashoka) were tested against the growth of *S. rolfsii* at 20% concentration and was recorded at 3, 5 and 7 days after inoculation.

Significantly lowest mycelium growth (7.75mm) was recorded in case of *C. longa*, where inhibition was 74.17% at three days after inoculation. The next best botanical found was *N. oleander* with mean mycelium growth of 11.25 mm followed by *M. champaca* (19.00 mm), *S. asoca* (20.25 mm), *P. guajava* (21.50 mm) and *D. erecta* (24.00 mm). Mycelial growth was highest in case of untreated control (30.00mm); there was no inhibition of mycelia growth. Among the ten phytoextracts tested, *C. longa* extract gave higher level of inhibition of mycelial growth (70.52 %) at 20% concentration and recorded minimum mycelial growth (18.50 mm) which was statistically similar to *N. oleander* (21.50 mm) at five days after inoculation. The next best botanical found was *S. asoca* (41.50 mm) followed by *P. guajava* (46.00 mm), *D. erecta* (48.50 mm) and *M. champaca* (52.00 mm). Maximum mycelial growth (62.75 mm) was recorded in untreated control which was found at par with *R. communis* (60.75 mm) and *F. religiosa* (60.25 mm). Average radial mycelial growth of *S. rolfsii* in all the botanicals was varied from 30.00 mm to 90.00 mm at seven days after inoculation. However, significantly lowest mycelia growth was recorded in *C.longa* (39.25 mm).This was followed by *N. oleander* (43.75 mm), *S. asoca* (61.50 mm) and *P.guajava* (78.50 mm). Whereas, the botanicals named *R. communis* (90.00 mm) was found statistically similar with untreated control and recorded maximum mycelial growth followed by *F. religiosa* (86.00 mm), *D. erecta* (80.75 mm), *M. champaca* (80.50 mm) and *T*. *procumbens* (79.25 mm) (Fig. 1).

4. DISCUSSION

"The application of chemical fungicides, plant diseases can be managed but the hazardous impacts of such products in human health and environment are well known. Natural plant products have been found effective in plant disease managements and could be safely incorporated as suitable alternatives for chemical fungicides by Zaker [6]. Active principles from medicinal plants are being tried as replacements of synthetic fungicides in management of plant diseases inorganic farming system. Continous use of fungicides and chemicals causes pollution in environment and there is a requirement to minimize the amount of fungicides application to the plants. Thus, plant extracts and bio agents can be used for management of disease as an alternative source. Plants extracts are eco-friendly, protective, curative and antagonistic to many diseases" [7].

Fungicidal ability of nine botanicals was tested against the growth of *S.rolfsii* at 20% concentration. Significantly least mycelial growth was recorded with *C. longa* followed by *N. oleander*, *S. asoca* and *P. guajava*. Whereas, the botanicals named *R. communis* was found statistically similar with untreated control and recorded maximum mycelia growth. Similar experiment conducted by Bharathi and Benagi, [8] and reported that minimum mycelial growth was noticed in the combination of garlic bulb extract, rhizome extract of turmeric and black tulsi leaf extract (1:1:1). Srawani and Chandra [9] evaluated botanical against *S. rolfsii* by using food poison technique. Lemon grass leaf (*Cymbopogon citrates*) showed greater effect in reducing the pathogen growth followed by Datura leaf (*D. stramonium*) and Calotrophis leaf (*C.procera*). Farooq *et al.* [10] reported that maximum inhibition of *S. rolfsii* was recorded by *Azadirachta indica* (73.8%). Singh *et al.* [11] reported that foliar extract of neem followed by that of ashoka, caused maximum inhibition of mycelial growth, sclerotial production and viability of *S. rolfsii* causing collar rot of lentil.

Madhavi *et al*. [12] conducted experiment for *in vitro* evaluated eight different plant extracts against *S. rolfsii*. Neem leaf extract caused maximum inhibition of mycelial growth (80.74%). Sclerotial production was inhibited to an extent of 11% and the inhibition caused was maximum with neem extract, followed by *Polyalthia longifolia* [Ashoka]. Hanthe gowda and Adiver [13] tested different plant extract against *S. rolfsii.* Among different plant extracts 1:20 dilution of *Parthenium hysterophorus*, Ashoka and *A. indica* significantly inhibited the mycelia growth of *S. rolfsii.*

Harish *et al*. [14] conducted experiment for *in vitro* evaluated fifty plant extracts against *Bipolaris oryzae* (*Cochliobolus miyabeanus*), the causal agent of brown spot disease of rice and indicated that two leaf extracts, *Nerium oleander* and *Pithecolobium dulce* exerted the higher percent inhibition to mycelial growth and spore germination of *B. oryzae*. Gupta *et al.,* [15] medicinal plant extracts against collar rot of tomato *in vitro* and *in vivo* against *S. rolfsii*, causing collar rot of chickpea and found hundred % mycelia inhibition recorded in garlic followed by neem, ginger, marigold and lantana.

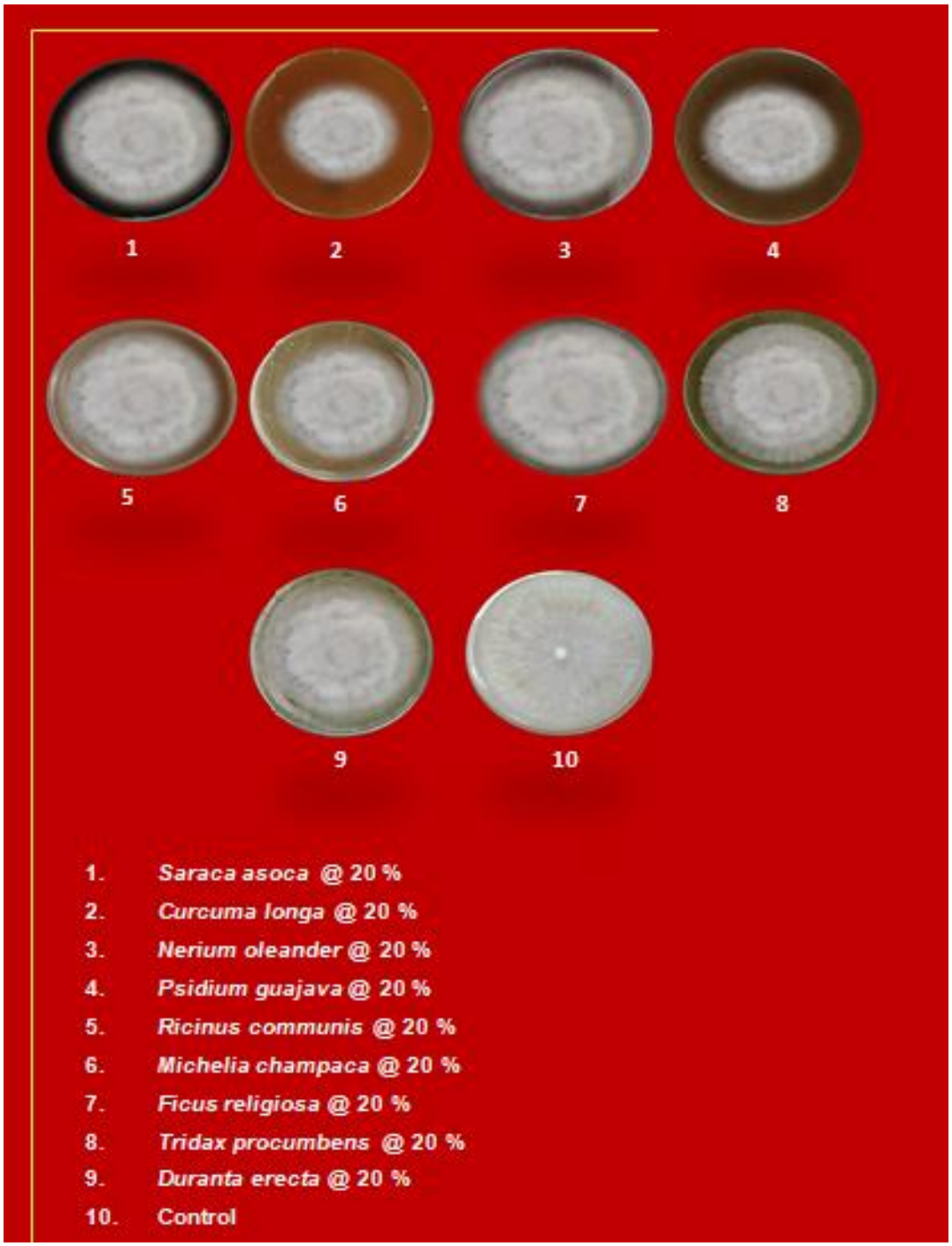

Fig. 1. *In-vitro* evaluation of botanicals against *Sclerotium rolfsii*

Table 2. *In-vitro* evaluation of botanicals against *Sclerotium rolfsii*

Botanicals	3DAI		5DAI		7DAI	
	Mycelium growth(mm)	% inhibition	Mycelium growth(mm)	% inhibition	Mycelium growth (mm)	% inhibition
Michelia champaca	19.00	36.67	52.00	17.13	80.50	10.56
Curcuma longa	7.75	74.17	18.50	70.52	39.25	56.39
Ficus religiosa	26.50	11.67	60.25	3.98	86.00	4.44
Nerium oleander	11.25	62.50	21.50	65.74	43.75	51.39
Tridax procumbens	26.50	11.67	53.00	15.54	79.25	11.94
Saraca asoca	20.25	32.50	41.50	33.86	61.50	31.67
Ricinus communis	27.50	8.33	60.75	3.19	90.00	0.00
Duranta erecta	24.00	20.00	48.50	22.71	80.75	10.28
Pisidium guajava	21.50	28.33	46.00	26.69	78.50	12.78
Control	30.00	0.00	62.75	0.00	90.00	0.00
SEm±	0.63	-	1.07	-	0.80	-
C.D. at 5 %	1.83	-	3.08	-	2.30	-

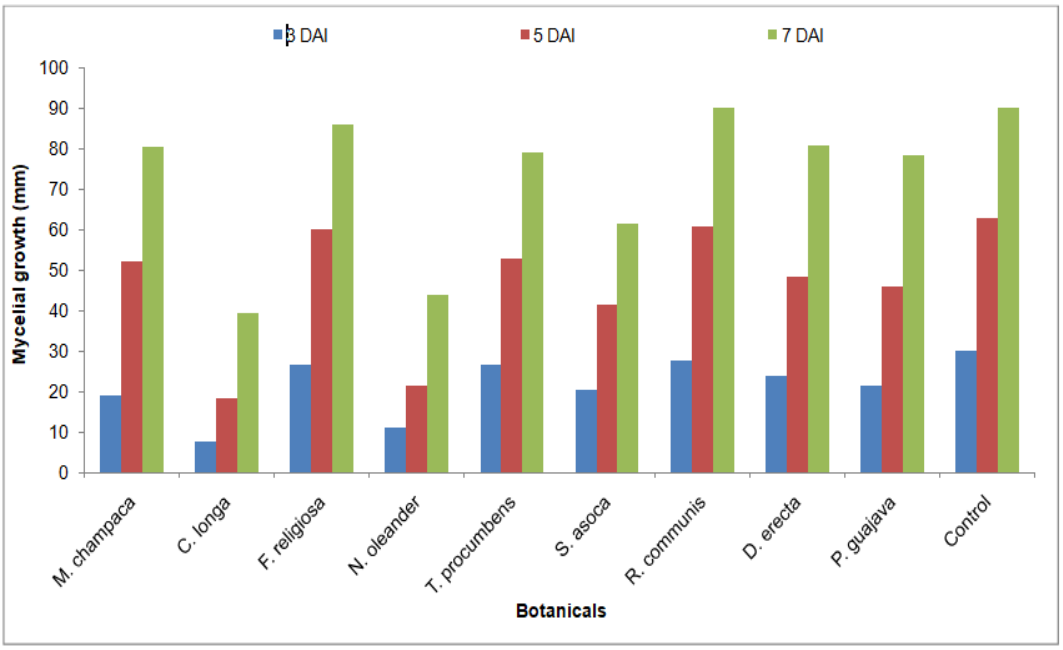

Fig. 2. *In-vitro* evaluation of botanicals against *Sclerotium rolfsii*

5. CONCLUSION

Collar rot disease caused by *Sclerotium rolfsii*, is a serious threat to lentil. Its control has acquired very limited success. Present investigation was carried out with management option by plant extract to minimize the mycelia growth of pathogen. A laboratory experiment was carried out for evaluation of different botanicals *viz. Curcurma longa* powder, *Michelia champaca* (champa), *Ricinus communis* (castor), *Nerium oleander* (kaner), *Ficus religiosa* (pipal), *Tridax procumbens, Psidium guajava*, *Duranta erecta* (hedge) and *Saraca asoca* (Ashoka) against *S. rolfsii*. Fungicidal ability of nine botanicals were tested against the growth of *S. rolfsii* at 20% concentration. Significantly least mycelial growth was recorded with *C. longa* followed by *N. oleander*, *S. asoca* and *P. guajava*. Whereas, the botanicals named *R. communis* was found statistically similar with untreated control and recorded maximum mycelial growth followed by *F. religiosa*, *D. erecta*, *M. champaca* and *T. procumbens.*

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.

ACKNOWLEDGEMENT

I would like to acknowledge to the Head of the department of Plant Pathology, College of Agriculture, Gwalior M.P. for providing me necessary facilities and encouragement for conducting the research work.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
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