

Screening of Medicinal Plants against *Sclerotium rolfsii* Isolated from Root Rot of Chilli

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ABSTRACT: The enormous progress made in recent decades in vegetable cultivation, fungal diseases are still an unsolved problem, mainly due to the fact that available fungicides are of hazard out effect on soil and environment. Antifungal activities of leaves extract of *Argemone mexicana* Linn, *Eucalyptus globulus* Labill and *Murraya koenigii* (curry leaf) was tested against chilli rot causing pathogenic fungi *Sclerotium rolfsii* by poisoned food technique. The medicinal plant leaves extract of 10%, 25%, 50%, 75% and 100% (without treatment served as control) of aqueous medium was studied. The inhibition values were recorded as percentage of inhibition recorded as range from 0% percentage to 31.03 percent.

The maximum percent of inhibition 31.03 recorded in *Eucalyptus globulus* Labill (Neelgrii) about 40mm mycelia growth after 48 hours at 75 per cent of aqueous leaves extract followed by 30.95 percentage of inhibition on with 14.50mm inhibited at 50 percent of aqueous leaves extract after 24 hours of incubation periods.

After *Eucalyptus globulus* Labill, *Argemone mexicana* Linn gave 20.93 & 18.54 percent of inhibition with 68mm & 50.5mm mycelial growth after 72 hours and 48 hours respectively.

The minimum percentage of inhibition found in case of *Murraya koenigii* (curry leaf) 17.5 and 14.28 with 49.5 mm & 18mm inhibited after 48 hours and 24 hours of incubation period respectively.

These effects of medicinal plant leaves extract were categorized as least in *Murraya koenigii*(17.5%), moderate in *Argemone mexicana* Linn (20.93) and strong in *Eucalyptus globulus* Labill.

KEYWORDS: Antifungal activity, *Argemona mexicana* Linn, *Eucalyptus globulus* Labill, *Murraya koenigii*, Plant extract and *Sclerotium rolfsii*.

I. INTRODUCTION

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II. RELATED WORK (REVIEW OF LITERATURE)

The effect of plant extracts on pathogenic fungi has been studied by a very large number of researchers in different in part of the world. The herbal products today symbolized safely in contrast to the synthetics that are regarded as unsafe to human and environmentally. Therefore plant extract may be use as alternative source for controlling soil born diseases since they comprises a rich source of bioactive substance.

The development of resistant strains of pathogens against various chemical fungicides and their toxic properties make the used of these chemicals limited (Lin, 1981) [1]. In recent times, the plant extract and isolate have been subjected to rigorous biochemical, pharmacological, toxicology investigations and many new therapeutic applications have been emerged out (Pinheiro, 1987) [2], and (Takahashi 2001) [3]. The plant fungal diseases are traditional been control by chemical fungicides. However their (chemical fungicides) adverse effects affect on the human health and environment,

International Journal of Innovative Research in Science, Engineering and Technology

(An ISO 3297: 2007 Certified Organization)

Vol. 5, Issue 5, May 2016

promote man to produce natural pesticides (Hayes et al., 1991) [4]. Consequently, biological controls, including the use of microorganisms or their antibiotics, offer an attractive alternative or supplement to pesticides for the management of plant diseases without negative impact of chemical control (Wang et al., 1999) [5]. The uses of plant-derived product as diseases controlled agents have been studied they tend to have less environmental effects and wide public acceptance (Lee et al., 2007) [6].

Among the various diseases of *Sclerotium rolfsii* causing root rot of chilli as treated and consider as the major constraints of vegetables.

In this study aim to investigate the characteristics of bioactive compound produced by against *Sclerotium rolfsii* and the influence of additive supplements on the antagonistic activity of medicinal antifungal plant extracts. The objectives of the research work was to study the antifungal activity of *Argemone mexicana* Linn, *Eucalyptus globulus* Labill and *Murraya koenigii* (curry leaf) medicinal plants. The present investigations evaluate the antifungal effects of crude extracted of medicinal plants.

III. MATERIALS AND METHODS

Isolation of *Sclerotium rolfsii*.

Infected root rot disease of chilli plant caused by *Sclerotium rolfsii* was collected from the field of Aurangabad District. The cut of infected portion into small pieces of about 3-5mm thick and sterilized with 0.1% (HgCl₂) mercuric chloride solution for few seconds and rinsed thrice in sterilized distilled water, and then placed on filter paper at room temperature. The tissue sections were then placed on potato dextrose agar and incubated at room temperature for seven days. Ultimately the pure culture of the pathogen was isolated subsequently maintained on the potato dextrose agar medium. Potato dextrose agar medium was prepared and after room temperature is poured in petriplates of the medium, mycelial disk 4mm diameter were cut from 4-5 day-old actively growing culture of *Sclerotium rolfsii* and each was placed in the center of petriplates containing potato dextrose agar. The effect of plant extracts on inhibition of mycelial on the growth of *Sclerotium rolfsii* was studied using poisoning food technique, (Dhingra and Sinclair, 1985) [7].

Plant species with families selected.

Plant species with families were selected for in-vitro evaluation. These plant species used for the study were presented in the Table 1 below.

Table 1

Sr.No.	Botanical name of medicinal Plants	Families
1	<i>Argemone mexicana</i> Linn.	Papaveraceae
2	<i>Eucalyptus globulus</i> Labill	Myrtaceae
3	<i>Murraya koenigii</i> (curry leaf)	Rutaceae

Preparations' of plant extract:

Fresh leaves of *Argemone mexicana* Linn, *Eucalyptus globulus* Labill and *Murraya koenigii* (curry leaf) were used medicinal plants leaves extract preparation. These leaves were collected from phulambari roadside field in Aurangabad District, Maharashtra Education Planning & Administration Institute campus and local vegetable market respectively during January, February 2015. These plants leaves, were rinsed in sterile distilled water in two to three times and dried in shade at room temperature, after which they were milled motor and pestle and electric blender to make powder. The powders were packed in to bottles and in air tight plastic pouches.

Preparation plant leaves extract medium for different concentration:

The plant leaves extract were made with at the rate of one ml/one gm or one gm / one ml of sterilized distilled water, autoclaved, cooled and then strained through muslin cloth. This formed a standard plant extract were made in aqueous medium of 10%, 25%, 50%, 75% and control (a without plant leaves extracts) concentrations.

Studies effect of plant leaves extracts of different concentrations:

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The effect of plant leaves extracts of *Argemone mexicana* Linn, *Eucalyptus globulus* Labill and *Murraya koenigii* (curry leaf) by inhibition the *Sclerotium rolfisii* was studied using poisoning food technique, (Dhingra and Sinclair, 1985) [7]. From standard stock solutions of plants leaves 10, 25, 50, 75 percentage concentrations was prepared separately by adding the required quantity of plants extract to the molten potato dextrose agar medium. One set is made without plant extract was kept as controlled. All these poured in to sterilized Petri plates. A mycelial disc cut from the periphery to 3-4 days old colony of *Sclerotium rolfisii* grown on potato dextrose agar medium were centrally placed in each of the Petri plates containing the potato dextrose agar medium having different three above medicinal plant leaves of *Argemone mexicana* Linn, *Eucalyptus globulus* Labill and *Murraya koenigii* (curry leaf) at different 10%, 25%, 50%, 75% concentrations and control under aseptic conditions. The Petri plates contains the potato dextrose agar medium inoculated with the pathogen alone served as control. All these Petri plates were incubated at room temperature. There are three replication were maintained for each treatment. The diameter of the colony was measured in two directions and average was recorded. The inhibition the growth of *Sclerotium rolfisii* was calculated by using the formula given below.

$$\text{Percentage of inhibition} = \frac{[\text{percentage of colony control}] - [\text{Diameter in colony treatment}]}{\text{Diameter of colony of control}}$$

Table 1- Medicinal plants in aqueous medium of following concentrations impress radial growth of *Sclerotium rolfisii* measured in mm after 24 hours interval.

Serial. No	Medicinal plants	Concentrations (%)	Growth <i>S.rolfisii</i> given		
			extracts 24 hr	measured 48 hr	mm 72 hr
1	<i>Argemone mexicana</i> Linn.	10 %	20	62	86
		25 %	20.5	57.5	84.50
		50 %	20	55.5	82
		75 %	20.5	50.5	68
		Control			
2	<i>Eucalyptus globules</i> labill.	10 %	21	55.50	79.50
		25 %	19.50	52	82
		50 %	14.50	45.75	71.25
		75 %	15.50	40	65
		Control			
3	<i>Murraya keonigii</i>	10 %	20.50	59	86
		25 %	20.50	56	85
		50 %	18.50	49.5	80
		75 %	18	53	76.50
		Control			

- * Values are average of triplicate.
- * Values measured after deducting or reducing 4mm mycelium disk.
- * (I) is denoted for inhibition.
- * Note 1- 10%, 25%, 50%, 75% and Control (100%) are aqueous medium leaves extract.

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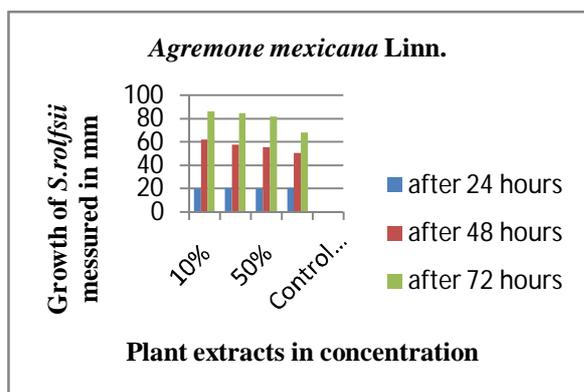
Table 2- The percentage of Inhibition (% of I) of *Sclerotium rolfsii* at given plant extract concentration in aqueous medium.

Serial. No	Medicinal plants	Concentrations (%)	Percentage Of Inhibition (I) Of <i>S. Rolfsii</i> in following					
			At Given plant Concentration		Extract		in following	
			24 hr	% of I	48 hr	% of I	72 hr	% of I
1	<i>Argemone mexicana</i> Linn.	10 %	20	13.04	62	0	86	0
		25 %	20.5	10.86	57.5	7.258	84.50	1.744
		50 %	20	13.04	55.5	11.29	82	4.651
		75 %	20.5	10.86	50.5	18.54	68	20.93
		Control						
2	<i>Eucalyptus globules</i> labill.	10 %	21	0	55.50	4.310	79.50	4.286
		25 %	19.50	7.142	52	10.34	82	1.204
		50 %	14.50	30.95	45.75	21.12	71.25	14.15
		75 %	15.50	26.19	40	31.03	65	21.68
		Control						
3	<i>Murraya keonigii</i>	10 %	20.50	2.380	59	1.666	86	0
		25 %	20.50	2.380	56	6.666	85	1.162
		50 %	18.50	11.90	49.5	17.5	80	6.976
		75 %	18	14.28	53	11.66	76.50	11.04
		Control						

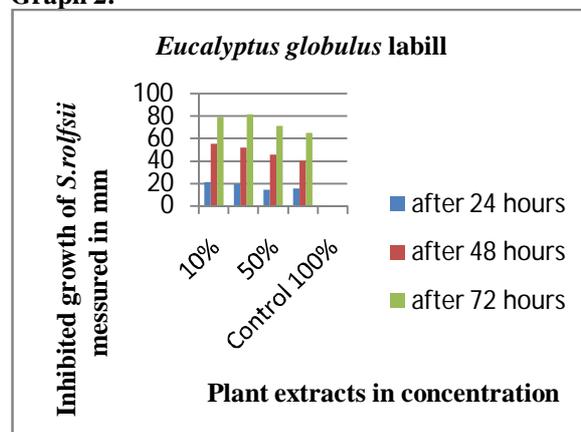
- * Values are average of triplicate.
- * Values measured after deducting or reducing 4mm mycelium disc.
- * (I) is denoted for inhibition.
- * Note 2- 10%, 25%, 50%, 75% and Control (100%) are aqueous medium leaves extract.

Graph 1 & 2 - Inhibited growth of *Sclerotium rolfsii* in *Argemone mexicana* Linn. and *Eucalyptus globulus* Labill leaves extract.

Graph 1:



Graph 2:



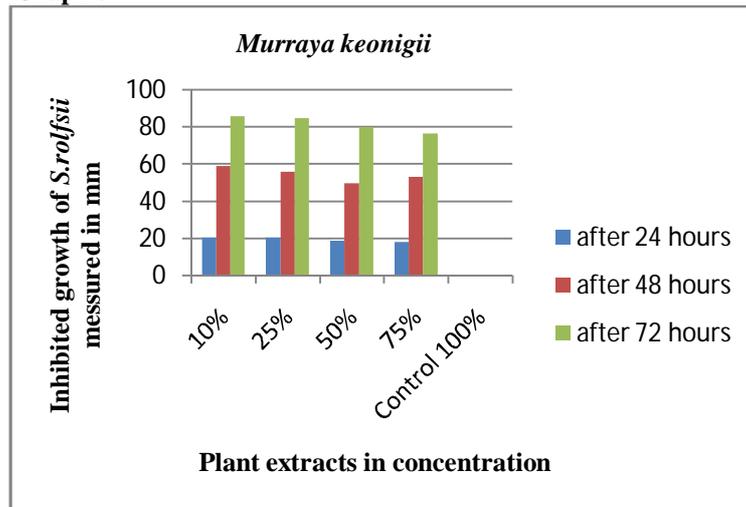
International Journal of Innovative Research in Science, Engineering and Technology

(An ISO 3297: 2007 Certified Organization)

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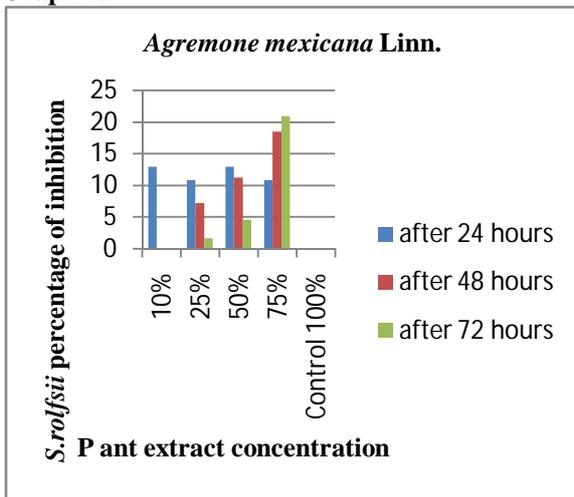
Graph 3- Inhibited growth of *Sclerotium rolfisii* in *Murraya keonigii* leaves extract.

Graph 3:

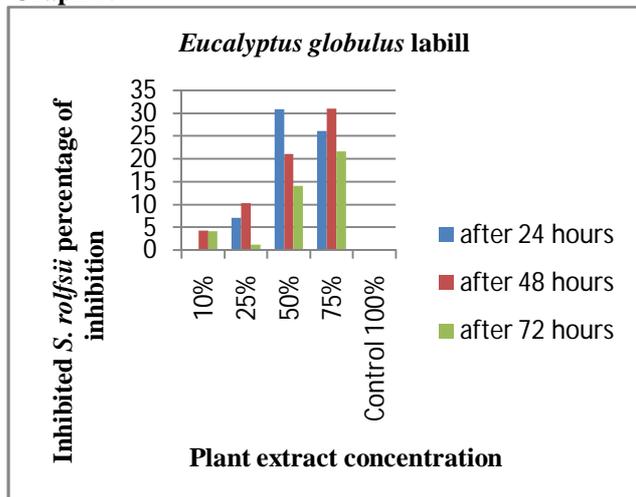


Graph 4 & 5- The percentage of inhibition of *Sclerotium rolfisii* of *Argemone mexicana* Linn. and *Eucalyptus globulus* Labill.

Graph 4:



Graph 5:



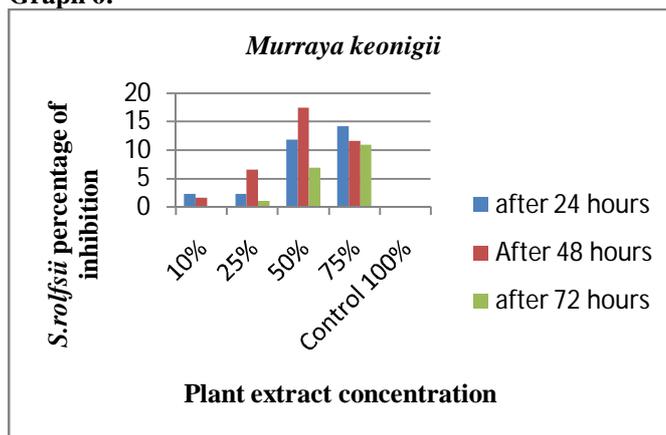
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(An ISO 3297: 2007 Certified Organization)

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Graph 6- The percentage of inhibition of *Sclerotium rolfsii* of *Murraya keonigii*.

Graph 6:



IV. RESULT AND DISCUSSION

The result of given *Argemone mexicana* Linn., *Eucalyptus globulus* Labill and *Murraya keonigii* plant extract showed in graph 1-6 and table 1,2,3. The strong percentage of inhibition on *Sclerotium rolfsii* recorded at 25% concentration as 24.51 percent of inhibition with 61.9mm mycelial inhibited. The *Eucalyptus globules* Labill plant leaves extract observed moderate percentage if inhibitions 1.70 to 21.90 with 64mm to 80.6mm growth range. This moderate activity of *Eucalyptus spp.* response to *Rhizoctonia solani* was shown by Anil Sehajpal, A and et, al, in 2009 [8]. The *Eucalyptus globules* plant is most effective in suppressing the sclerotial germination of *Macrophomina phaseolina* observed by Dubey and Kishore, 1990 [9]. The antifungal activity of *Argemone mexicana* Linn against *Sclerotium rolfsii* was effect least inhibition percentage noticed revealed by 0.2 % with mycelial growth 83.00 mm. In the leaves extracts of *Eucalyptus globulus* Labill the inhibition of the mycelia growth of the *Sclerotium rolfsii* was found to be in between 0% to 31.03% of inhibition, it was recorded as strong percentage of inhibition, because it reached up to 31 percent of inhibition, Whereas *Argemone mexicana* involving 0% to 20.93% percentage of inhibition that is lowered by 10.1 percent by *Eucalyptus globulus* Labill.. But lowest percentage of inhibition was 0% to 17.50% in *Murraya keonigii* that is 3.43 percent lower by *Argemone mexicana* and 13.53 percent by *Eucalyptus globulus* Labill.

V. CONCLUSION

The existing cost of fungicides is not affordable well for the millions of growers particularly in the developing countries like / including India. It also causes environmental pollution which leads to hazardous to human health. So, the search for new antifungal compound for medicinal plant must go on, they are extremely safe and as safe as possible. In this present study we come to concluded as marked inhibitory of *Eucalyptus globulus* Labill, *Argemone mexicana* Linn and *Murraya keonigii* leaves extracts against *Sclerotium rolfsii* was observe. The inhibitory activity are categorize as strong in *Eucalyptus globulus* Labill, moderate in *Argemone mexicana* Linn and least in *Murraya keonigii* among all three medicinal plant leaves extract screening efficacy. The medicinal plants are *Argemone mexicana* Linn, *Eucalyptus globulus* Labill and *Murraya keonigii* (var. curry leaf) evident in this study that they have potential to control of some plant pathogenic fungi and could be consider for developing new fungicides.

VI. ACKNOWLEDGEMENT

Author's grateful to Dr.Sumia Fatima my Guide, Associate Professor and Head of the department of Botany for use full instruction direction. The author's are also express gratitude to principal, of Dr. Rafiq Zakaria College Campus-2

International Journal of Innovative Research in Science, Engineering and Technology

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and Dr. Babasaheb Ambedkar Marathada University, Aurangabad (M.S.) comfort and provided that necessary amenities for this research work.

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