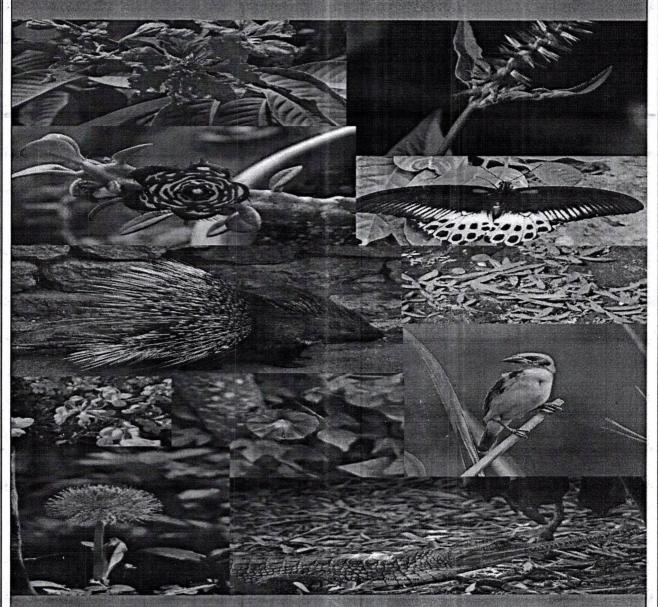
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# CONSERVATION OF WILD TAXA: PRESENT SCENARIO



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## Conservation of Wild Taxa: Present Scenario

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## STUDIES ON GROWTH OF MACROPHOMINA PHASEOLINA WITH EFFECT OF DIFFERENT SOLVENT CONCENTRATIONS OF RHIZOME EXTRACT OF ZINGIBER OFFICINALE

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#### **ABSTRACT:**

Macrophomina phaseolina (Tassi) Goid is a soil borne fungus causes root rot diseases to Sarpagandha (Rauwolfia serpentina). The fungus infects the root and lower stem of over 500 plant species and is widely distributed in the United States (Wyllie, 1988). The efficacy of Zingiber officinale rhizome extract against Macrophomina phaseolina was studied by using aqueous, Methanol and Acetone solvents at different concentrations i.e., 1.00, 2.00, 3.00, 4.00 and 5.00 % for their antifungal efficacy.

**KEYWORDS:-** *Macrophomina phaseolina*, Sarpagandha, *Zingiber officinale*, Methanol, Acetone, etc.

#### **MATERIALS AND METHODS:**

In order to study of antifungal activity of Zingiber officinale leaf extract on Macrophomina phaseolina. Locally available rhizome of Zingiber officinale used i.e., rhizome of Zingiber officinale was tested by poisoned food technique in vitro as used by Shiva et.al, (2008) and Francis Borgio, et.al, (2008) to know their inhibitory effect on the growth of Macrophomina phaseolina.

#### 1) Preparation of aqueous plant part extracts: entleancentrations i.e., 100 200:3.00 4 00

Healthy fresh leaves, rhizomes and bulbs were collected from locally available these different plants, washed thoroughly with fresh water and finally rinsed with sterile distilled water.

Fifty gram of leaves, rhizomes and bulbs of different plants were cut into small pieces and grinded in a grinder by adding 50 ml sterile distilled water. Extracts thus, obtained were

1) Preparation of aqueous plant part extracts

filtered through double layered muslin cloth in 150 ml flasks and plugged. The concentration of extract was considered as 10 %. The extracts then autoclaved at pressure 15 lbs for 20 minutes. Potato Dextrose Agar (PDA) medium was prepared and sterilized at 15 lbs pressure for 20 minutes. The sterilized extracts were considered as standard plant extracts and used for the testing of antifungal activity.

The different concentrations were prepared i.e. 1.00, 2.00, 3.00, 4.00, 5.00, 6.00, 7.00, 8.00, 9.00 and 10.00 percent. The 10 ml extracts of different concentrations were individually added in 10 ml melted, cooled and sterilized PDA at the time of pouring in the petriplates and incubated at room temperature. After solidification, a 5 mm disc of actively growing 7 days old pure culture of Macrophomina phaseolina was incubated aseptically in the centre of plate. Three repetitions were made for each treatment. Medium without phytoextracts served as control. The observations of fungal growth in diameter were observed and recorded and percent growth inhibition was also worked out as per the method given by.

The extracts then autoclaved at pressure 15 lbs for 201

2) Preparation of Methanolic plant part extracts:

Healthy fresh leaves of Ashwagandha (Withania somnifera L.) was taken, washed thoroughly with fresh water and finally rinsed with sterile distilled water and dried.

Fifty grams leaves of Ashwagandha (Withania somnifera L.) were cut into small pieces and grinded in a grinder to make fine powder and then extracted in 50 ml Methanol. Extracts thus obtained were filtered through double layered muslin cloth in 150 ml flasks and plugged. The extracts then autoclaved at pressure 15 lbs for 20 minutes. Potato Dextrose Agar (PDA) medium was prepared and sterilized at 15 lbs pressure for 20 minutes. The sterilized extract was considered as standard plant extract and used for the testing their antifungal activity.

The different concentrations were prepared i.e. 1.00, 2.00, 3.00, 4.00, 5.00, 6.00, 7.00, 8.00, 9.00 and 10.00 percent. The 10 ml extracts of different concentrations were individually added in 10 ml melted, cooled and sterilized PDA at the time of pouring in the petriplate and incubated at room temperature. After solidification a 5 mm disc of actively growing 7 days old pure culture of Macrophomina phaseolina was inoculated aseptically in the centre of plate. Three repetitions were made for each treatment. Medium without phytoextracts served as control. The fungal growth in diameter were observed and recorded and percent growth inhibition was also calculated as per the procedure given by Syeda Fakehha et.al. (2012).

#### 3) Preparation of Acetone plant part extracts:

Healthy fresh leaves, rhizomes and bulbs were taken, washed thoroughly with fresh water and finally rinsed with sterile distilled water and dried.

steadized FDA at the time of pouring

Fifty gram of dried leaves, rhizomes and bulbs of different plants were cut into small pieces and grinded in a grinder to make fine powder and then extracted in 50 ml Acetone. Extracts thus, obtained were filtered through double layered muslin cloth in 150 ml flasks and plugged. The extracts then autoclaved at pressure 15 lbs for 20 minutes. Potato Dextrose Agar (PDA) medium was prepared and sterilized at 15 lbs pressure for 20 minutes. The sterilized extracts were considered as standard plant extracts and used for the testing of antifungal activity. The different concentrations were prepared i.e., 1.00, 2.00, 3.00, 4.00, 5.00, 6.00, 7.00, 8.00, 9.00 and 10.00 percent. The 1 ml extracts of different concentrations were individually added in 15 ml melted, cooled and sterilized PDA at the time of pouring in the petriplates and incubated at room temperature. After solidification a 5 mm disc of actively growing 7 days old pure culture of *Macrophomina phaseolina* was incubated aseptically in the centre of plate. Three repetitions were made for each treatment. Medium without phytoextracts served as control. The observations of fungal growth in diameter were observed and recorded and percent growth inhibition was also worked out. Khajista, et.al., (2013).

The sclerotial formation was determined by observing number of sclerotia per plugged. The extract manual autoclayed at pressure 15 lbs for 20 minutes. Pattern Dextrose Again microscopic field.

#### **Experimental results AND DISCUSSION:**

The efficacy of Zingiber officinale L. against Macrophomina phaseolina with aqueous extract treatment treated at different concentrations i.e., 1.00, 2.00, 3.00, 4.00 and 5.00 % for their antifungal efficacy using standard Poisoned Food Technique.

The treatment of 1 % concentration of aqueous solution shows 11.34 to 81.75 %, at 2% concentration gives 18.77 to 83.15 %, at 3 % concentration shows 23.25 to 85.38 %, at 4 % concentration gives 28.85 to 90.00 % and at 5 % concentration gives 28.94 to 93.50 % inhibition of the pathogen growth with water solvent viz. recorded at 1 to 7 days of incubation period. The efficacy of Zingiber officinale, at 5 % concentration gives maximum inhibition of growth of pathogen. The increase in concentration gives maximum inhibition of pathogen i.e. microscopic field.

93.50 % at 5 % concentration on 7 days of incubation as mentioned in table 1

The efficacy of Zingiber officinale L. against Macrophomina phaseolina by using methanol extract was evaluated at different concentrations i.e., 1.00, 2.00, 3.00, 4.00 and 5.00 % for growth control by using standard Poisoned Food Technique as shown in table 2.

Zingiber officinale efficacy at 1 % concentration shows 12.75 to 80.77 % in 1 to 7 days of incubation periods, at 2% concentration gives 21.22 to 89.20 %, at 3 % concentration shows The treatment of 1% concentration of agreeus solution shows 11.3 to 81.75 % at 2% 25.35 to 87.95 %, at 4 % concentration gives 27.90 to 90.55 % and at 5 % conc. gives 28.98 to

92.47 % inhibition of the growth of the pathogen with methanol solvent viz. recorded at 1 to 7 days of incubation period. The efficacy of *Zingiber officinale*, at 5 % conc. gives maximum inhibition of growth of pathogen with increase in incubation period.

For the control of growth of *Macrophomina phaseolina*, *Zingiber officinale L*. rhizome acetone extract was tested at different concentration percentage i.e., 1.00, 2.00, 3.00, 4.00 and 5.00 % for their antifungal efficacy by using standard Poisoned Food Technique as shown in table 3.

Zingiber officinale efficacy at 1 % concentration shows 14.25 to 81.90 %, at 2% concentration gives 19.49 to 86.10 %, at 3 % concentration shows 24.85 to 86.90 %, at 4 % concentration gives 27.76 to 88.35 % and at 5 % concentration gives 28.75 to 90.27 % inhibition of the growth of the pathogen with acetone solvent viz. recorded at 1 to 7 days of incubation period. The efficacy of Zingiber officinale, at 5 % concentration gives maximum

Table -1: Effect of aqueous rhizome extract of Zingiber officinale on growth of Macrophomina phaseolina.

Incubation	Percent inhibition    Concentration (%)							
Period								
(Days)	1.00	2.00	3.00	4.00	5.00			
concentration	11.34 gives-19.49 to-	18.77 56.10 % at 3	23.25	2885 shows /4 85 to	28.94			
- recent	(6.51)	(10.81)	(13.44)	(16.76)	(16.82)			
2	20.30	26.35	33.45	35.50	36.89			
<b>2</b>	(11.71)	(15.27)	(19.54)	(20.79)	(21.64)			
inhibi <b>g</b> on of p	36.72	41.27	48.32	49.75	51.87			
30,000	(21.54)	(24.37)	(28.89)	(29.83)	(31.24)			
Table4-1: E1	51.82	50.95	55.25	58.50	64.34			
4	(31.20)	(30.62)	(33.53)	(35.80)	(40.04)			
5	62.22	67.39	68.39	72.42	79.90			
5	(38.47)	(42.67)	(43.14)	(46.90)	(53.92)			
(D <b>6</b> vs)	70.45	72.34	76.80	82.26	85.65			
0.47	(45.20)	(46.33)	(50.81)	(56.48)	(60.12)			
7	81.75	83.15	85.38	90.00	93.50			
′	(55.90)	(57.14)	(60.32)	(64.17)	(69.26)			

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S.E ±	3.07	2.95	3.51	2.78	3.24
C.D at 5%	9.44	9.08	1.08	8.56	9.96

Figures in parenthesis are ARCSIN transformed value.

Table -2: Effect of methanolic rhizome extract of Zingiber officinale on growth of Macrophomina phaseolina.

Incubation		Percent inhibition						
Period	Control	Concentration (%)						
(Days)	(Methanol)	1.00	2.00	3.00	4.00	5.00		
1	7.15	12.75	21.22	25.35	27.90	28.98		
,	(4.64)	(7.32)	(12.25)	(14.68)	(16.19)	(16.84)		
2	9.25	24.30	32.35	35.44	36.58	37.85		
_	(5.30)	(14.06)	(18.87)	(20.75)	(21.04)	(22.23)		
3	12.10	43.76	47.39	50.67	50.75	52.45		
	(6.94)	(25.94)	(28.28)	(30.44)	(30.49)	(31.63)		
1.11.14	15.35	57.85	58.67	65.25	69.80	73.74		
- NI	(8.82)	(35.34)	(35.92)	(40.72)	(44.26)	(47.50)		
5	18.44	72.46	74.35	74.36	78.80	79.90		
	(10.62)	(46.43)	(48.46)	(48.57)	(52.59)	(53.85)		
6	21.56	76.46	79.85	79.88	82.65	86.65		
· · · · · · · · · · · · · · · · · · ·	(12.45)	(50.37)	(53.70)	(53.01)	(56.68)	62.16)		
7	22.75	80.77	89.20	87.95	90.55	92.47		
And the control of the same about the same and the same about the	(13.14)	(54.84)	(63.13)	(64.42)	(64.90)	(67.64)		
S.E ±	0.42	2.90	2.97	3.91	3.23	3.76		
C.D at 5%	1.30	8.93	9.15	12.02	9.93	11.58		

Figures in parenthesis are ARCSIN transformed value.

Table -3: Effect of acetone rhizome extract of Zingiber officinale on growth of Macrophomina phaseolina.

Incubation		Percent inhibition						
Period	Control	Concentration (%)						
(Days)	(Acetone)	1.00	2.00	3.00	4.00	5.00		
1	8.15 (5.22)	14.25 (8.19)	19.49	24.85 (14,38)	27.76 (16.11)	28.75 (16.70)		

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2	(5.87)	33.60 (19.63)	40.80 (24.07)	44.48 (26.40)	49.50 (29.66)	50.72 (30.47)	
2	11.55	48.95	52.00	54.75	55.75	57.15	
3	(6.63)	(29.30)	(31.33)	(33.19)	(33.88)	(34.85)	
4	13.00	58.75	62.37	65.35	67.75	68.30	
	(7.46)	(35.97)	(38.58)	(40.80)	(42.64)	(43.47)	
Section 1	16.44	69.35	72.40	74.58	75.45	76.30	t experience of the control of the c
5	(9.46)	(43.90)	(46.38)	(48.22)	(49.60)	(49.72)	
	18.35	75.55	78.75	81.50	85.25	87.35	
6	(10.57)	(49.69)	(52.69)	(55.63)	(59.93)	(62.39)	7 A
makes were suffered	22.10	81.90	86.10	86.90	88.35	90.27	All and the second seco
	(12.76)	(55.77)	(61.33)	(61.76)	(62.07)	(64.52)	
S.E ±	0.42	2.86	3.66	3.41	3.38	3.11	
C.D at 5%	1.30	8,82	11.26	10.50	10.42	9.58	

Figures in parenthesis are ARCSIN transformed value.

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