

# ANTIFUNGAL ACTIVITY OF WEED'S EXTRACTS AGAINST IMPORTANT PHYTOPATHOGENS

Deepti Khare, Sanjay Vyas, Neha Sharma

Dept.of Microbiology, Govt. Holkar Science College Indore.

## Abstract

Biocontrol measures have emerged as a major breakthrough in the control of a number of diseases. Being Safe, ecofriendly & effective control measure natural pesticides have become the need of the hour in integrated pest control management. Fungicidal properties of the aqueous and ethanolic extract of *Desmodium sp.*, *Tridex sp.*, *Spilanthus sp.*, *Alternanthera sp.* & *Euphorbia hirta* were in vitro tested against *Alternaria solani* (MTCC No 2101) causes early blight of Potato /Tomato & *Fusarium udum* ( MTCC No 2204 ), the casual organism of wilt of arhar. The ethanolic extracts of *Desmodium sp.*, *Spilanthus sp.*, *Tridex sp.*, and *Alternanthera sp.*, showed maximum toxicity against *Alternaria solani* whereas the ethanolic extract of *Tridex sp.* and *Spilanthus sp.*, showed maximum fungal activity against *Fusarium udum*.

**Key words:** -Antifungal activity, Toxicity, Percent inhibition, *Alternaria* , *Fusarium*, weeds.

## 1- Introduction-

Many medicinal plants or botanicals are found to possess various groups of secondary metabolites like alkaloids, terpenoids, flavanoides, quinines, polyacetones and amino acids which inhibit growth of fungi (Veeravel 2002). Among several medicinal plants, some common weed plants are waste land plants found to exhibit different types of biological activity upon varieties of pathogens due to the presence of different types of cardenolides, the secondary metabolite (Rothschild, 1972). Arhar, Potato & Tomato are the important crops of Madhya Pradesh. *Fusarium udum* causes wilt of arhar and *Alternaria solani* is the casual organism of early blight of tomato/ Potato incurring heavy loss to the agriculturists.

Bioactive products of a plant being less persistent in the environment and are safe for mammals and other non target organisms. Botanical pesticides are readily available in many places, often cheaper than the synthetic counterparts & their crude extracts are easy to prepare even by farmers. These are also less likely or slow down the development of resistance or resurgence in pests. Therefore the benefits of natural pesticides have aroused interest in protection of crop plants.

On other hand weeds become a global problem in the agriculture due to their herbicidal resistance. They are unwanted plants growing out of places and out of time (Alagesboopathi and Balu, 2002). If the antifungal ingredient is investigated against test fungi this will be added advantage. Keeping in view of the above facts, an attempt was made to identify antifungal activity in some common weeds against *Fusarium udum* and *Alternaria solani*.

## 2- Material and methods -

### Collection of weeds-

Some of the common weeds such as *Desmodium sp.*, *Tridex sp.*, *Spilanthus sp.*, *Alternanthera sp.* & *Euphorbia hirta* were collected from different locations (Agricultural Fields, Road side area and waste lands). These are easily identified by standard literature (Oomachan & Shrivastav, 1996). The collected plant materials were thoroughly washed and air dried in hot air oven. The dried plants were milled to a fine powder in a grinder and stored in closed container in the dark room at room temperature.

### Preparation of phyto extracts-

**Aqueous extract:** - 10 grams of finally ground powder was extracted with 100 ml with distilled water and it was filtered by using Whatman filter paper no. 1. The filtrate with concentration of 100mg/ml was kept in refrigerator until required (Patel & et al., 2007).

**Ethanolic extract:** -10gms of each plant powder was extracted in 100ml of ethanol. The extracts were then filtered by whatman filter paper no.1. Then solvent is evaporated at room temperature to yield residue, these residues were redissolved in distilled water to obtain 100mg/ml concentration (Patel *et al.*, 2007).

**Bioassay-** The in vitro test was carried out to measure the effects of the aqueous and ethanolic extracts on radial growth of *A.solani* and *F.udum* by Poisoned food technique. 10ml of the aqueous or 10ml of ethanolic extracts of plants and 90ml of sterilized PDA media were thoroughly mixed in conical flasks and 20ml of this mixture was dispensed in each petridish. An agar disc of 6 mm diameter was cut from 7days old culture of both the test fungus and placed at the centre of the petridish. Three replicates were maintained for each treatment. The basal medium without any phyto extract served as control. All the inoculated plates were incubated at  $27\pm 1^{\circ}\text{C}$  for 7 days. The fungi toxicity of the extracts in terms of percentage inhibition of mycelial growth of fungi was calculated by using formula (Singh and Tripathi, 1999).

$$\% \text{inhibition} = \frac{dc - dt}{dc} \times 100$$

Where, dc = Average increase in mycelial growth in control. dt = Average increase in mycelial growth in treatment.

### 3- Results and Discussion -

Results presented in table 1-4 shows that all weeds which were tested against *A.solani* and *F.udum* were found to be effective in ethanolic as well as aqueous extracts. In case of *A.solani*(Table 1) the aqueous extract of *Alternanthera* sp. is most effective (57.1%). There was no radial mycelia growth till 5<sup>th</sup> day of incubation, but later on growth of fungi appeared. Aqueous extract of *Tridex* sp. was found to be least effective. The growth of fungi appeared on 3<sup>rd</sup> day of incubation. Overall the aqueous extract of *Tridex* sp. was able to inhibit the growth of fungi upto 33.3%. The aqueous extract of *Desmodium* sp., *Spilanthus* sp. & *Euphorbia hirta* were also able to inhibit the radial mycelial growth of the test fungi upto 50%.

Interestingly the ethanolic extracts were found to be more effective (Table 2). *Desmodium* sp., *Tridex* sp., *Spilanthus* sp., and *Alternanthera* sp. were found to inhibit the growth of fungi completely. There was no mycelial growth of fungi till 7<sup>th</sup> day of incubation. Whereas the ethanolic extract of *Euphorbia hirta* inhibit the radial mycelia growth upto 58.8%. This weed can able to inhibit the growth till the 2<sup>nd</sup> day of incubation, later on growth of fungi appeared.

Table 3 exhibits effect of aqueous extract of different weeds against *F.udum*. The aqueous extract of *Tridex* sp. shows maximum inhibition (56.8%) whereas the aqueous extract of *Alternanthera* sp. was least effective (40.9%) which is considerable. There was no radial mycelial growth of fungi till 2<sup>nd</sup> day of incubation, but later on the growth of fungi appeared in aqueous extract of all the weeds. Aqueous extract of *Desmodium* sp. & *Euphorbia hirta*, both were able to inhibit the radial mycelial growth upto 43.1%.

The ethanolic extracts of *Desmodium* sp., *Spilanthus* sp., & *Tridex* sp., were found to be more effective against the test fungi (Table 4). But it is interesting to note that in case of *Alternanthera* sp. & *Euphorbia hirta* the aqueous extracts were more effective as compared to their ethanolic extracts. The ethanolic extract of *Tridex* sp. completely inhibits the growth of *F.udum*. There was no growth appeared till 7<sup>th</sup> day of incubation. The range of inhibition appears between 77.5% in case of ethanolic extract of *Spilanthus* sp. to 27.5 % in case of *Euphorbia hirta*. The ethanolic extract of *Desmodium* sp. Inhibits the mycelial growth upto 60.3% and *Alternanthera* sp. shows 34.4 % inhibition.

Name of plant	Radial Mycelial Growth of fungi in cms							Percentage of Mycelial inhibition
	1st	2nd	3rd	4th	5th	6th	7th	
<i>Desmodium sp.</i>	NG	NG	NG	NG	1.3	1.7	2.1	50
<i>Spilanthus sp.</i>	NG	1.5	1.7	1.7	2	2.1	2.3	45.2
<i>Tridex Sp.</i>	NG	NG	1.4	1.8	2.2	2.5	2.8	33.3
<i>Alternanthera sp.</i>	NG	NG	NG	NG	NG	1.3	1.8	57.1
<i>Euphorbia Hirta</i>	NG	NG	1.2	1.5	1.9	2.1	2.3	45.2
<i>Control</i>	1.6	2.1	2.8	3	3.3	3.9	4.2	

\* NG = No  
Growth

Name of plant	Radial Mycelial Growth of fungi in cms							Percentage of Mycelial inhibition
	1st	2nd	3rd	4th	5th	6th	7th	
<i>Desmodium sp.</i>	NG	NG	NG	NG	NG	NG	NG	100
<i>Spilenthus sp.</i>	NG	NG	NG	NG	NG	NG	NG	100
<i>Tridex Sp.</i>	NG	NG	NG	NG	NG	NG	NG	100
<i>Alternathera sp.</i>	NG	NG	NG	NG	NG	NG	NG	100
<i>EuphorbiaHirta</i>	NG	NG	1.6	2.1	2.3	2.5	2.8	58.8
<i>Control</i>	1.6	2.6	4	4.2	4.3	4.4	4.8	

\* NG = No  
Growth

Name of plant	Radial Mycelial Growth of fungi in cms							Percentage of Mycelial inhibition
	1st	2nd	3rd	4th	5th	6th	7th	
<i>Desmodium sp.</i>	NG	NG	1.4	2.2	2.3	2.5	2.5	43.1
<i>Spilenthus sp.</i>	NG	1.5	1.2	1.5	1.8	1.9	2.2	50
<i>Tridex Sp.</i>	NG	NG	1.5	1.5	1.7	1.8	1.9	56.8
<i>Alternathera sp.</i>	NG	NG	2.7	1.9	2.1	2.3	2.6	40.9
<i>Euphorbia Hirta</i>	NG	NG	2.3	1.8	2.2	2.4	2.5	43.1
<i>Control</i>	0.3	2.1	2.4	2.9	3.2	3.8	4.4	

\* NG = NoGrowth

Table: 4 Antifungal Activity of weeds against <i>F. udum</i> in Ethanolic extracts								
Name of plant	Radial Mycelial Growth of fungi in cms							Percentage of Mycelial inhibition
	1st	2nd	3rd	4 <sup>th</sup>	5th	6th	7th	
<i>Desmodium sp.</i>	NG	NG	NG	NG	2.1	2.3	2.3	60.3
<i>Spilenthus sp.</i>	NG	NG	NG	NG	NG	1.2	1.3	77.7
<i>Tridex Sp.</i>	NG	NG	NG	NG	NG	NG	NG	100
<i>Alternanthera sp.</i>	NG	NG	1.3	3.2	3.6	3.6	3.8	34.4
<i>Euphorbia Hirta</i>	1.8	1.8	1.9	2.8	3.2	3.8	4.2	27.5
<i>Control</i>	0.5	2.6	2.5	3.2	3.8	4.4	5.8	

\* NG = No Growth

#### 4- Conclusion –

The present study revealed that both *A.solani* & *F.udum* are less sensitive to the aqueous extracts except *Alternanthera* sp. and *Euphorbia hirta* as compared to the ethanolic extracts. The reason for this could be that all of the identified components from plants, active against micro organisms are aromatic or saturated organic compounds and they are soluble in ethanol (Cowan, 1999). Several authors have also reported that the growth of *Rhizoctoniasolani* was completely inhibited with the leaf extract of *Acacia nilotica*.

Various workers have screened a large number of plants belonging to Angiosperm and Gymnosperms for their fungitoxic properties. Mostly the aqueous extracts of plants have been used to evaluate their fungitoxicity (Thapliyal et al., 2000 and Alagesaboopathi and Balu, 2000, Iqbal et al, 2008). During the course of present assay use of ethanol ensures the extraction of maximum compounds as well as facilitates further purification of active fractions (Dhar et al., 1973). Antifungal activity of *Euphorbia hirta* was also reported against *Aspergillus sp.* in 2013 by Gayathri and Ramesh.

The present investigation is an important step in developing plant based pesticides which are ecofriendly for the management of the plant pathogens and development of commercial formulation of botanicals as well as helpful in the management of weeds too.

#### 5- Acknowledgement -

Authors are grateful to the Principal Govt. Holkar Science College Indore for providing laboratory facilities and acknowledge to M.T.C.C. Institute of microbial technology Chandigarh for providing pure cultures of test organisms.

#### 6- References -

- [1] Alagesboopathi, C. and Balu, S. (2002) Antifungal activity of some species of *Andrographis wallichex Neer* on *Helminthosporium oryzae* journal of Economic and Taxonomic Botany 24:705 -707
- [2] Chandra, S. and Munshi, A.D. (1995). Ecofriendly botanical pesticides. Employment News 23-29 Dec.20 (30), 1-2
- [3] Cowan, M.M. (1999) Plant products as antimicrobial agent. *Clinical microbiology Review* 12: 564-582.
- [4] Dhar, M.L., Dhawan, B.N., Mehrortra, and Roy, C. (1973) Screening of Indian plants for biological activity, part 1. *Indian Journal of Experimental Biology* 6:232-247.
- [5] Iqbal, Y., Khare, D., Mehta, I. and Verma, K.S. (2008) Fungicidal activity of some weeds against *Aspergillus* sp. *Journal of Basic & Applied Mycology*. 7(1-2): 117-118.
- [6] Patel, S., Venugopalan, N. and Pradeep, S. (2007) Screening for antimicrobial activity of Weeds. *The Internet Journal of Microbiology* 6:1-4
- [7] Thapliyal, M., Ghosh, M. and Bennet, S.S.R. (2000) Screening of six medicinal plants for their antifungal protein activity. *Asian Journal of Microbiology, Biotechnology and Environmental Science* 2:215-218.
- [8] Gayathri, A. and Ramesh, KV (2013) Antifungal activity of *Euphorbia hirta* L. inflorescence extract against *Aspergillus flavus*. A mode of action study. *Int. J. Curr. Microbiol. App. Sci.*