



## ANTIFUNGAL PROPERTIES AND PHYTOCHEMICAL SCREENING OF PLANT EXTRACTS AGAINST PLANT PATHOGENIC FUNGI

SAILY DASS AND \*SHONALI CHATURVEDI

\*Department of Botany,  
Ewing Christian College, Allahabad

*Eucalyptus globules* Linn., *Euphorbia hirta* Linn, *Carica papaya* Linn., *Allium sativum* Linn., *Calotropis procera* Linn belong to different families' viz. Mrytaceae, Euphorbiaceae, Caricaceae, Liliaceae and Asclepidaceae. In the present study, we evaluated the presence of various secondary metabolites such as alkaloids, flavonoids, phenols, steroids, triterpenoids, tannins, saponins. The antifungal activity of the leaf extracts of *Allium sativum*, *Eucalyptus globules*, *Carica papaya*, *Calotropis*, *Euphorbia hirta* against plant pathogenic fungi (*Fusarium oxysporum*, *Rhizoctonia solani* and *Pythium aphanidermatum*) by food poisoned technique *in vitro*, the aqueous extracts of selected plants showed significant inhibition.

**Key words:** Antifungal activities, phytochemical screening, plant extracts.

Plants provide abundant resources of antimicrobial compounds and have been used for centuries to inhibit microbial growth. Plant based natural constituents can be derived from any parts of the plant like bark, leaves, flowers, roots, fruit, seeds etc. (Gordon and David 2001). The beneficial medicinal effects of plant materials typically result from the combination of secondary products present in the plant. The medicinal action of plants is unique to particular plant species or groups are constituent with this concept as the combination of secondary products in particular plant is taxonomically distinct (Wink 1999). It is clear that the plant kingdom harbors an inexhaustible source of active ingredients. The demand on plant based therapeutic is increasing in both developing & developed countries due to growing recognition that they are natural products, non-narcotics, easily biodegradable producing minimum environmental hazards, having no side effects. The screening of plant extracts & plant products for antimicrobial activity has shown that higher plants represent potential source novel antibiotics prototypes (Afolayan 2003).

### MATERIALS AND METHODS

#### Preparation of plant extracts

The fresh leaves of selected plants were washed thoroughly under tap water followed by sterilized water. The leaves were air dried, after that leaves were grinded with the help of pestle and mortar by taking (1:1 weight/volume). One gram of leaf was added in 1ml distilled water separately for each plant extract and filtered through muslin cloth and centrifuged for 15 minutes at 3000rpm. The final extracts were collected separately and stored in of refrigerator until further needed.

#### Phytochemical screening of leaf extracts:

The phytochemical components of *Carica papaya*, *Euphorbia hirta*, *Eucalyptus globules*, *Allium sativum*, *Calotropis procera* were screened by using methods of Brindha *et al.* (1997). The components qualitatively analyzed were alkaloids, flavonoids, phenols, steroids, triterpenoids, and tannins.

#### *In vitro* assay of plant leaf extracts

The test plant pathogenic fungi (*Fusarium*, *Rhizoctonia* and *Pythium*) were

**Table 1:** Qualitative Analysis of leaf sample of certain plants mg /g

Phytochemicals	<i>Allium</i>	<i>Carica</i>	<i>Eucalyptus</i>	<i>Calotropis</i>	<i>Euphorbia</i>
Alkaloids	-	-	+	-	-
Flavonoids	+	+	+	+	+
Phenolic	+	-	+	-	+
Saponins	-	+	+	+	+
Steroids	+	+	+	+	-
Tannins	-	+	+	+	+
Triterpenoids	-	+	-	+	+

isolated from the infected portion (root, stem) of the diseased plant parts, showing specific disease symptoms. Antifungal activity of leaf extract was tested by poisoned food technique method adopted (Nene and Thapliyal 1979). PDA was prepared in flask and required concentration of leaf extract was added and then sterilized. The plant extract along with medium was poured into Petri plates, and then disc of 0.5 cm of the fungal pathogen was cut with the help of sterilized cork borer and transferred aseptically into the Petri dishes containing medium with certain amount of plant extract. Suitable checks were maintained. The fungal colony diameter was measured every 24 hrs of interval for 5-7 days. Observations were recorded on the mycelia growth of test pathogen. Each treatment was replicated five times.

**Table 2:** *In vitro* efficacy of plant extract at 25% concentration against test pathogens.

Extracts	Name of plants	Radial growth h (cm)		
		Pythium	Rhizoctonia	Fusarium
	<i>C. papaya</i>	3.7	-	3.5
Aqueous	<i>E. globules</i>	3.1	3.5	3.1
	<i>E. hirta</i>	3.6	-	-
	<i>A. sativum</i>	2.8	3.0	2.5
	<i>C. procera</i>	3.4	3.3	2.9
	Control	4.2	4.2	4.2
	<i>C. papaya</i>	3.5	-	3.0
Solvent (Chloroform)	<i>E. globules</i>	3.0	3.4	2.4
	<i>E. hirta</i>	3.5	3.3	2.7
	<i>A. sativum</i>	2.6	2.9	2.2
	<i>C. procera</i>	3.3	3.0	2.8
	Control	4.2	4.2	4.2

**Table 3.** *In vitro* efficacy of plant extract at 50% concentration

Extracts	Name of plants	Radial growth (cm)		
		Pythium	Rhizoctonia	Fusarium
	<i>C. papaya</i>	3.5	3.2	3.3
Aqueous	<i>E. globules</i>	2.9	3.1	3.0
	<i>E. hirta</i>	3.4	3.0	3.2
	<i>A. Sativum</i>	2.6	2.8	2.3
	<i>C. procera</i>	3.1	3.1	2.7
	Control	4.2	4.2	4.2
	<i>C. papaya</i>	3.4	3.2	3.2
Solvent (Chloroform)	<i>E. globules</i>	2.7	2.8	2.8
	<i>E. hirta</i>	3.2	2.7	3.0
	<i>A. sativum</i>	2.5	2.6	2.2
	<i>C. procera</i>	3.2	3.3	2.5
	Control	4.2	4.2	4.2

## RESULTS AND DISCUSSION

### Phytochemical analysis

Presence of some important secondary metabolites viz; alkaloids, phenols, flavonoids, saponins, tannins, steroids and triterpenoids in leaves extracts were confirmed after performing specific qualitative tests.

### Antifungal activity of leaf extracts

The antifungal activity of *Carica papaya*, *Euphorbia hirta*, *Allium sativum*, *Calotropis procera* and *Eucalyptus globules* leaves were assessed using the food poisoned technique. The result showed that aqueous and chloroform leaf extracts possess antifungal activity against plant pathogenic tested fungi (*Fusarium*, *Pythium* and *Rhizoctonia*). The leaf extracts of *Allium sativum* and *Eucalyptus globules* in aqueous and solvent (concentrations 100 %) exhibits relatively higher reduction in mycelial growth of test pathogens as compared to 25 & 50% concentration. (Table 3).

In the present study aqueous extract of *Alliums sativum* showed maximum reduction in mycelia growth at all the concentrations compared to chloroform. Among the five selected plant extract, the minimum mycelial growth was recorded with the bulb extract of *Allium sativum* at all the plant concentration (25, 50, and 100%).

**Table 4.** *In vitro* efficacy of plant extract at 100% concentration

Extracts	Name of plants	Radial growth (cm)		
		<i>Pythium</i>	<i>Rhizoctonia</i>	<i>Fusarium</i>
	<i>C. papaya</i>	1.5	1.8	2.0
Aqueous	<i>E. globules</i>	0.9	2.1	1.8
	<i>E. hirta</i>	2.0	2.7	1.5
	<i>A. s ativum</i>	0.2	0.6	1.9
	<i>C. procera</i>	2.1	1.7	1.3
	Control	4.0	4.0	4.0
	<i>C. papaya</i>	1.3	1.9	1.7
Solvent (Chloroform)	<i>E. globules</i>	0.7	2.0	1.6
	<i>E. hirta</i>	1.9	2.6	1.9
	<i>A. sativum</i>	0.3	0.8	0.6
	<i>C. procera</i>	2.2	1.6	0
	Control	4.0	4.0	4.0

The difference in the inhibitory effect of various plant products may be due to qualitative and quantitative differences in the antifungal principles present in them. Antifungal properties of leaf, bulb and oil extracts of *Allium sativum* have been reported by many workers (Singh and Singh 1980). Amonkar and Banerji (1971) have reported that the active principle of *Allium sativum* is a mixture of diallyl-disulphide and disallyl - trisulphide. El- Sayeed *et al.* (1985) obtained that steroidal alkaloids like (saponins from *Eucalyptus* sp. were responsible for fungi toxicity. Similarly, Batish *et al.* (2008) reported that *Eucalyptus* has been found to possess a wide spectrum of biological activity against fungi, bacteria, insects, mites and weeds and provide simple, inexpensive and environmentally

friendly alternative pest control. Successive isolation of botanical compound for plant material is largely dependent on the type of solvent used in the extraction procedure. In the present study plant extract by chloroform provided less consistent antifungal activity compared to those extracted by water.

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