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Antifungal activity of the Carica papaya important food and drug plant

Mohit Kumar¹, Mohammed Faheem, ^{*}Satyapal singh¹, Anwar Shahzad² and Ashok K Bhargava

¹Department of Biotechnology, Singhania University, Pacheri Bari, Raj., India ²Plant Biotechnology Laboratory, Department of Botany, Aligarh Muslim University, Aligarh, India ³Deptt of Botany, M. S. College, Saharanpur, U.P., India

ABSTRACT

The present investigations are being carried out to evaluate the antifungal medicinal properties of Carica papaya. The effects of different concentrations of alcoholic extract of Carica papaya (root, shoot and seed) on the radial growth of plant against the pathogenic fungi viz. Aspergillus niger, Aspergillus flavus, Candida albicans and Microsporum fulvum. That with the increase in concentrations the rate of growth inhibition also increases. Observation further shows that like root extract growth is also inhibited in the presence of shoot and seed alcoholic extract under culture medium. further shows that the growth of these fungi inhibits more in presence of higher concentrations as compared to lower concentrations of extract.

Keywords: Carica papaya, Aspergillus flavus, Candida Albicans, Calatropis procera, Microsporum fulvum

INTRODUCTION

It is a native of West Indies or Mexico. The *papaya* or papaw now forms an important food and drug plant in West Indies, Sri Lanka, India, Malaysia, USA and Hawai Islands. Papaya plants may reach heights of 9 m, and are thus described as giant herbs [1]. Later, it was introduced in China as an Indian plant. The papaya is a tall herb attaining a height of about 25 feet. The cylindrical stem is fleshy with a crown of large palmately lobed leaves at the top. The plant is dioecious. It grows almost throughout the country except at low temperature. A dry and warm climate is necessary. It grows best on loamy soils. The plant is mainly propogated by seeds. The plant flowers in about four months after planting and fruits are ready for harvest in another six months. The fruit is a many seeded berry. The important varieties are Washington, Honey Dew (Madhubindu), Singapore and Ceylon. Co-1 is a selection made in Tamil Nadu from variety Ranchi of Bihar. The papaya is a nutritive fruit containing a small amount of proteins and the same amount of minerals consisting mainly of iron, calcium and phosphorus, vitaminA and C and is rich in the enzyme papain. This enzyme is obtained from green fruits as a latex which is later dried and purified. Papain is said to be useful in the treatment of skin blemishes, diphtheria and even cancer. In industry it is used in the clarification of beer, in tanning and the manufacture of chewing gum. The ripe fruit and the seeds are considered to have medicinal properties against disorders of liver, spleen and the digestive tract.

MATERIALS AND METHODS

Plants were collected from district Saharanpur & Shiwalik belt of Uttar Pradesh as well as from Garhwal hills of Uttranchal, India. Identification of plants was done through herbarium available in the deptt of Botany, M.S. College, Saharanpur & Forest Research Institute, Dehradun. List of plant prepared. The plant material were

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collected from wild areas of Saharanpur district and also of Garhwal Himalaya. The method was followed based on work of with some modifications [2].

For the preparation of *Carica papaya* plant extracts 5 gm of fresh plant part was washed 2-3 times with distilled water & than surface sterilized with 90% alcohol. Subsequently the plant material was grounded in 50 ml of distilled water & alcohol separately for aqueous and alcoholic extracts, respectively. The alcoholic extracts were kept for 24 hrs. at room temperature to evaporate the alcohol. In the remaining residue, 50 ml of distilled water added. The macerates were squeezed through double layered Muslin cloth & filtered through filter paper. After filtration, the aliquot was centrifuged at 5000 rpm for 30 minute. The supernatants were filtered through whatmann no. 1 filter paper & then sterilized by passing through 0.2 micron disposable filters. The various concentrations of extract made & thus obtained were used in studies. initially treated with 0.1% HgCl₂ solution for sterilization and subsequently washed thoroughly with sterile distilled water and grounded in motor & pistil with 50% methanol. The homogenized liquid was filtered and centrifuged at 3000 to 5000 rpm. The supernatant was used as test extract & make up into 20 ml using 50% methanol. Further, the extract was diluted into different concentrations, i.e. 10%, 25%, 50%, 75%, 20 ml of SDA (Sabouraud Dextro Agar) culture medium with 5 ml of the above concentration of the extract was poured in sterile petiplates and allowed to solidify. Then the test fungus was inoculated at the centre of the medium and incubated at room temperature 25 °C ± 2 °C. Replicates and controls were mainained through out the study. The diameter of the fungal growth was measured on 5th and 7th day.

To study the effects of antifungal alcoholic extract and aquous extract of above selected plants two sets of culture media were prepared separately for control and treatment. In the test sets of neutral pH 7, requisite amount of the experimental material were mixed and then added into the sterilized Sabouraud dextrose agar (SDA) medium of respective pH level. In the control set of each experimental set, the same volume of distill water (in place of experimental material) was mixed in appropriate amounts when ever found necessary.

Mycelia discs of 5 mm diameter, were cut from the periphery of 7 day old culture of the test organisms were aseptically inoculated upside down on the surface of the SDA medium in plaster. Inoculated petri plates were incubated at 25° C $\pm 2^{\circ}$ C and observation were recorded at 5^{th} and 7^{th} day. Fungal growth taken as measurement parameter. The absence of Fungi denoted antifungal property of fungicidal nature.

Percentage of mycalial growth inhibition on different pH levels were calculated using following equation:

% growth inhibition Colony diameter in control – colony diameter in treated sets x 100 Colony diameter in control

 $I = (C-t) \times 100$

Test fungi isolated and were used for in vitro studies. The culture was purified by hyphal tip technique. The stock culture of the test fungus was maintained on SDA medium at 25 degree $C \pm 1^{\circ}$ Degree C.

Effect of different concentration of aquous and alcoholic extract against the fungus was studied employing techniques.

Poisoned food technique Poisoned food technique used to assess the antifungal activity of selected plant extracts [3]. A series of double strength of test plant extracts viz. 10%, 25%, 50% and 75% were prepared using sterile distilled water. 30 ml of test extract was poured into 100 ml conical flask containing 30 ml sterilized melted SDA of double concentration. 30 ml of this mixed medium was then poured in each petriplate aseptically. The partiplates were inoculated with previously maintained 7 days old culture. 5 ml mycalial disc was cut with sterilized cork borer and transferred aseptically in the centre in inverted position. All petriplate including control and experimental were incubated at 25 ° C \pm 1 ° C for 7 days. After 7 days of incubation, observation were recorded and percent inhibition of radial growth was calculate using following formula:

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

where, I = percent inhibition, C = radial growth in check in mm/cm and T = radial growthin treated set in mm/cm

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RESULTS

Aspergillus flavus.

Table 1 shows the effects of different concentrations of alcoholic extract of *Carica papaya* on the radial growth of *Aspergillus flavus*.

Table shows that the alcoholic extract of root, shoot and seed is inhibitory to the growth of this fungi. Table further shows that with the increase in concentrations the rate of growth inhibition also increases. Thus, 10%, 25%, 50% and 75% root extract of *Carica papaya* retards radial growth of this fungi in culture medium by 8.4%, 20.9%, 29.2% and 45.7% of the control respectively at 7th day of growth. Observation further shows that like root extract growth is also inhibited in the presence of shoot and seed alcoholic extract under culture medium. Result further shows that the growth of this fungi inhibits more in presence of higher concentrations as compared to lower concentrations of extract.

Candida Albicans

Table 1 the effect of various alcoholic extract concentrations of the plant parts on the radial growth of *Candida albicans* in culture medium. Result shows that the growth is inhibited by alcoholic extract concentration and this inhibition rate increases with the increase in doses of plant part extract. Thus, radial growth of these fungi in 10%, 25%, 50% and 75% root extract concentration is 93.1%, 89.6%, 82.7% and 62.0% of the control respectively. Result further shows that the growth is inhibited more in 75% shoot and seed extract concentration as compared to 10% alcoholic extract concentration. Thus, in 10% shoot and seed extract concentration the radial growth of this fungi was 96.6% and 93.5% of control respectively, at 7th day, while, these values in 75% shoot and seed concentrations are 51.7% and 54.8% of the control respectively on 7th day.

Calatropis procera

Table 1 the effect of different concentrations of alcoholic extracts of various plant parts of *Calatropis procera*. Result shows that the radial growth of *Candida albicans* is affected by various concentration of alcoholic extract of plant parts. Observation further shows that with the increase in concentration of this medicinal plant part the rate of inhibition of fungal growth also increases. Thus, in 10%, 25%, 50% and 75% alcoholic concentration of root the radial growth is 96.1%, 88.4%, 57.6% and 53.8% of the control respectively at 7th day of growth. Result further shows that like root extract, shoot and seed extract also inhibits radial growth of fungi, however, this inhibition is more in higher concentration as compared to lower concentration of various plant parts of *Calatropis procera*.

Days of Study	Diameter of Growth (Cm.) A. Niger			Diameter of Growth (Cm.) A. Flavus			Diameter of Growth (Cm.) C. Albicans			Diameter of Growth (Cm.) M. Fulvum		
	Root	Shoot	Seed	Root	Shoot	Seed	Root	Shoot	Seed	Root	Shoot	Seed
Growth in Control 0% extract												
5 th	2.0	2.5	2.4	1.7	1.6	1.5	1.4	1.7	1.5	1.8	1.6	1.4
7 th	4.0	4.3	4.3	2.4	2.3	2.3	2.8	2.7	2.9	2.2	2.2	2.0
Growth in 10% alcohalic extract												
5 th	1.5	2.2	1.9	1.5	1.4	1.4	1.3	1.1	1.4	1.6	1.4	1.3
7^{th}	2.6	3.1	3.1	2.2	2.1	2.1	2.7	2.5	2.8	2.0	2.0	1.9
Growth in 25% alcohalic extract												
5 th	1.2	1.1	1.3	1.3	1.1	1.2	1.1	1.0	1.2	1.4	1.1	1.1
7 th	2.1	2.6	2.7	1.9	2.0	1.8	2.5	2.4	2.5	1.8	1.8	1.8
Growth in 50% alcohalic extract												
5 th	1.0	0.9	1.0	1.1	1.0	1.0	0.9	0.9	1.1	1.2	0.9	1.0
7 th	2.0	1.4	2.0	1.7	1.8	1.6	1.7	1.3	2.0	1.6	1.7	1.6
Growth in 75% alcohalic extract												
5 th	0.7	0.8	0.6	0.8	0.7	0.8	0.8	0.8	0.9	0.9	0.8	0.8
7 th	1.1	1.1	1.0	1.3	1.3	1.4	1.6	1.2	1.5	1.2	1.4	1.4

 TABLE – 1 Effect of different concentration of alcoholic extracts of plant parts of Carica Papaya on growth performance of A. Niger, A. Flavus, C. Albicans and M. Fulvum.

Microsporum fulvum

Table 1 shows the effect of alcoholic extract concentrations of various plant parts of a plant *Mentha piperita* on the radial growth of *Candida aebicans*. Result shows that various concentrations of alcoholic extract of this plant also inhibit the growth of this fungi. Observation shows that with the increase in the concentration of alcoholic plant part

extracts like root extract, shoot extract and rhizome extract the rate of inhibition increases. Thus, in 10% root extract the growth is 90% of the control whereas in 75% root extract concentration the growth is 43.3% of the control. Result further shows that shoot and rhizome extract also causes increase in inhibition rate like root extract.

DISCUSSION

The present investigations are being carried out to evaluate the antifungal medicinal properties of *Carica papaya* plant against the pathogenic fungi viz. *Aspergillus niger, A.flavus, Candida albicans* and *Microsporum fulvum*. Fungal infections comprise an important faction of diseases occurring not only in plants and animals but also in human beings. Moulds and yeasts are so widely distributed in human environment that human beings are instantly exposed to them. Fortunately, because of the relative resistance of human beings and comparatively non pathogenic nature of fungi, most of these exposures do not lead to over infection. However, fungi are gaining importance with respect to increased incidence of chronic, often fatal, mycoses in immune compromised patients [4]. The fungi present in soil, water and air constitute exogenous fungal opportunists. The roster of opportunistic fungal species continues to increase. However, some of the common ones include *Aspergillus funigates, A.niger, A.terreus, A.flavus, Absida, Candida albicans, Cryptococous neoformis, Microsporum fulvum, Mucor, Rhizomucor, Rhizopus*, and *Torulopsis globrata* [5]. To find suitable drug for the management of fungal diseases is difficult because fungi, like human beings, are eukaryotes. Many of the cellular and molecular processes are similar, and still a number of chemicals are reported to have antifungal activity [3]. These include the derivatives of quinazolinone [6], coumarin [7], thiazolidinone [8], thiadiazole [9], Thiazole [10], Pyridine [11] and Sydnone [12].

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