



Antibacterial and Antifungal Activity of *Opuntia Dillenii* (Cactaceae) Fruit Extract

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Received : 01.11.2012 Revised on : 05.2.2013 Accepted : 01.03.2013

Abstract

The antimicrobial activity of plant extracts and phytochemicals was evaluated with antibiotic susceptible and resistant micro organisms. Methanolic extract of *Opuntia dillenii* (OD) was tested for its antibacterial (plate hole method) activity against fourteen different bacterial strains. From the results, it is deduced 100 % of methanolic extract were active in concentration of 1000 µg/ml, 93 % active in concentration of 500 µg/ml, 87.5 % active in concentration of 250 µg/ml, 50 % active in concentration of 125 µg/ml and no activity in lowest test concentration of 62.5 µg/ml. Antifungal activity against six different fungal organisms were studied and the results were indicated that the 1000 µg/ml & 500 µg/ml showed good activity in all fungus (100%). The methanolic extracts in concentration of 250 µg/ml (83%) and 125 µg/ml (33 %) showed moderate activity in all tested microorganisms. Oxy tetracycline and Amphotericin B with concentration of 1000 µg/ml were used as a standard drug for antibacterial and antifungal studies respectively.

Keywords : Antibacterial ; Antifungal ; *Opuntia dillenii*.

1. INTRODUCTION

Since antiquity, man has used plants to treat common infectious disease, and some of the traditional medicines are still included as part of the habitual treatment of various diseases. Scientific interest in medicinal plant has burgeoned in recent times due to increased efficiency of new plant derived drugs and rising concerns about the side effects of modern medicines. The continuing emergency of drug resistant organisms and the increasing evolutionary adaptations by pathogenic organisms to commonly use anti-microbial have reduced the efficacy of antimicrobial agents correctly in use. Therefore the search for new drugs novel

sources such as plants continues to be necessary. To date, plants continue to be major sources of commercially consumed drugs. Even most synthetic drugs have their origin from natural plant products (1). *Opuntia dillenii* (Cactaceae) is a shrub found in tropical regions of India. The Cactus fruits, cladodes or flower infusions have been traditionally used as folk medicine to treat other ailments such as ulcer, allergies, fatigue and rheumatism and as a diuretic agent.

However so far *Opuntia* has shown its efficacy as antioxidant (Yingkun Qiu et al. 2002), hypotensive and antihyperlipidemic (Rubeena Saleem et al. 2005), antispermatic effect (Gupta et al. 2002). Therefore, the present study was undertaken for the first time to investigate antibacterial and antifungal activity of methanolic extract of *Opuntia dillenii*.

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2. MATERIALS AND METHODS

2.1 Plant material

The plant material was collected during April - June from tropical areas of western Ghats regions of Erode and Dharmapuri districts of Tamilnadu. The plant material were identified by Mr. Satyanarayanan, Joint Director at Botanical survey of India (BSI) Coimbatore, India and a voucher specimen was deposited in Herbarium of laboratory of Botany, Tamilnadu Agricultural University, Coimbatore, Tamilnadu, India.

2.2 Preliminary phytochemical screening

The preliminary phytochemical screening of *Opuntia dillenii* was carried out for the decoction of various phytoconstituents using standard procedures (5). The following solvents were used for the study, Chloroform, Ethyl acetate, Methanol, Ethanol and water. The preliminary phytochemical screening of methanol extract reveals the presence of Alkaloids, Flavonoids, Tannins, Glycosides and Triterpenoids.

2.3 Preparation of crude extract

Weighed quantity of coarsely powdered fruit of *Opuntia dillenii* were placed in maceration flask and added with sufficient quantity of methanol. Complete maceration takes place for about 24hrs, with occasional shaking during first 6hrs. After 24hrs, the menstruum was collected, evaporated to obtain the dried extract (3).

2.4 Antibacterial activity

Antibacterial study (plate hole diffusion or agar well diffusion) (6) assay was used to determine the growth inhibition of bacteria by plant extracts. Bacteria were maintained at 4°C on nutrient agar plate before use. A total of 25ml of Muller Hinton (MH) agar was prepared and poured in to sterile petri dishes which were previously inoculated with 0.2 ml of different bacterial species. A well was prepared in the plates with the help of sterile cork-borer (6mm). Four holes per plates were made in to the MH agar containing the bacterial culture.

A total of 0.2 ml of methanol extract of *O.dillenii* was poured in to the wells with concentrations as 1000µg/ml, 500µg/ml, 250µg/ml, 125µg/ml, 62.5µg/ml. The plates were incubated overnight at 37°C for 24 hours. The results obtained were compared with standard antibiotic oxytetracycline (1000µg/ml).

2.5 Antifungal activity

Saubourads dextrose agar medium (SAD) was prepared and 25ml of each was poured in to sterile petri dishes with different species of fungus (7). Using a sterile cork borer (6mm diameter) four holes per plate were made and a total of 0.2 ml of plant extracts was poured in to the wells. The plates were incubated at 28°C for 36 to 48hrs and the zone diameter was then recorded. Amphotericin B (1000µg/ml) was used as a standard antifungal drug. 3.

Table 1. Results of preliminary phytochemical screening.

Plant extract	Extract fractions	Flavonoids	Tannins	Alkaloids	Glycosides	Resins	Steroids
<i>Opuntia dillenii</i>	a	+	-	-	-	-	-
	b	++	-	++	-	-	-
	c	+++	++	++	+	-	-
	d	++	+	+	-	-	-

a - Chloroform
b - Ethyl acetate
c - Methanol
d - Water

+++ = Excess amount
++ = Presence
+ = Trace amount
- = Absence

Table 2. Antibacterial zone diameter of methanolic extract of *Opuntia dillenii* fruit.

Microorganisms	1000 µg/ml	500µg/ ml	250µg/ml	125µg/ml	62.5µg/ ml	Oxytetracycline (1mg/ml)
<i>Bacillus licheniformis</i> (NCIM 2468)	20	14	10	08	0	26
<i>Brevibacterium leuteum</i> (ATCC 15830)	16	14	07	0	0	24
<i>Escherichchiae coli</i> (ATCC 15830)	17	12	09	07	0	24
<i>Flavobacterium devorans</i> (NCIM 2581)	17	10	0	0	0	22
<i>Klebsiella pneumonia</i> (ATCC 11229)	17	10	07	0	0	22
<i>Micrococcus flavum</i> (NCIM 2984)	14	10	07	0	0	18
<i>Micrococcus leuteum</i> (ATCC 9341)	16	11	08	06	0	20
<i>Rhodococcus terrae</i> (NCIM 5126)	18	12	10	08	0	25
<i>Salmonella typhi</i>	20	15	11	08	0	28
<i>Shigella boydi</i> (ATCC 8700)	16	12	07	0	0	23
<i>Shigella flexneri</i> (NCIM 4924)	12	0	0	0	0	25
<i>Shigella sonnei</i> (ATCC 29930)	16	10	08	06	0	22
<i>Staphylococcus faecalis</i> (ATCC 8043)	16	12	08	0	0	28
<i>Staphylococcus aureus</i> (ATCC 29213)	19	13	10	06	0	25

3. RESULTS

3.1 Preliminary photochemical screening

The chemical tests used for the study were Shinoda test (flavonoid), Phlorotannins test (tannins), Wagners test (alkaloids), and Salkowskii test (glycosides) Among these the methanolic extract was found to contain high amount of flavonoids as shown in Table 1. The preliminary phytochemical screening of methanolic extract reveals the presence of alkaloids, flavonoids, tannins, triterpenes, gums and mucilage.

3.2 Antibacterial activity

From the phytochemical screening, the methanolic extract showed high amount of flavonoids when compared to other extracts. So we selected the methanolic extract of *Opuntia dillenii* fruit for antibacterial screening. The fifteen Bacterias were used for antibacterial screening.

Various concentrations of methanolic extract were used are (1000µg/ml, 500µg/ml, 250µg/ml, 125µg/ml, 62.5µg/ml) to test the antibacterial activity. From the results of antibacterial screening 100% of methanolic extract were active in concentration of 1000µg/ml, 93% active in concentration of 500µg/ml, 87.5% active in concentration of 250µg/ml, 50% active in concentration of 125µg/ml and no activity in lowest test concentration of 62.5µg/ml. Antibacterial activity were shown in the above table 2.

3.3 Antifungal study

Six strains of fungus were used for study. In this, 62.5 µg/ml showed no activity in all tested microorganisms. The 1000 µg/ml & 500 µg/ml showed good activity in all fungus (100%). The 250 µg/ml (83%) and 125 µg/ml (33%) showed moderate activity in all tested microorganisms. The results are shown in table 3.

Table 3. Antifungal zone diameter of methanolic extract of *Opuntia dillenii* fruits.

S.no	Microorganisms	1000µg/ml	500µg/ml	250µg/ml	125µg/ml	62.5µg/ml	A*
01.	<i>Aspergillus niger</i> (NCIM 1207)	18	10	08	0	0	20
02.	<i>Candida albicans</i> (NCIM 3484)	20	14	11	09	0	24
03.	<i>Monilinia fruticola</i> (NCIM 1011)	15	12	07	0	0	22
04.	<i>Auricularia polytricha</i> (NCIM 1303)	14	10	0	0	0	23
05.	<i>Chaetomella raphigera</i> (NCIM 1231)	15	11	07	0	0	25
06.	<i>Arthrotrys oligospora</i> (NCIM 11246)	18	12	08	06	0	22

A*- Standard Amphotericin-B(1000µg/ml)

4. CONCLUSION

The present work has shown that *Opuntia dillenii* is potentially a good source of antibacterial and antifungal agents which can be used in future for supporting primary health care in India.

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