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International Journal of Current Research
Vol. 9, Issue, 08, pp.55476-55478, August, 2017

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

ISSN: 0975-833X

RESEARCH ARTICLE

ANTIBACTERIAL ACTIVITY OF LEAF EXTRACTS OF SOME HERBS AGAINST SOME CLINICAL ISOLATES

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ARTICLE INFO

Article History:
Received 22nd May, 2017
Received in revised form
06th June, 2017
Accepted 20th July, 2017
Published online 31st August, 2017

Key Words:
Antibacterial, Clinical isolate,
Crude extract, Agar well diffusion,
Zone of inhibition.

ABSTRACT

The antibacterial activity of leaf extract of Chrozophora rottleri, Oxalis corniculata, Parthenium hysterophorus and Solanium xanthocarpum were evaluated in-vitro against some clinical isolates by agar-well diffusion method. Two solvents chloroform and methanol were used for extraction of bioactive compound from fresh leaves. Antimicrobial potential of leaf extract was determined by measuring the zone of inhibition. It was concluded from the results that both chloroform as well as methanol extracts of leaf of tested herbs were quite effective in inhibiting the growth of clinical isolates. Result also revealed that methanol extract has more antibacterial potential than chloroform extract. Methanol extract of Chrozophora rottleri and Oxalis corniculata has very good antibacterial potential against all the clinical isolate. Therefore, the leaf extracts of this plant can be selected for further investigation to determine their therapeutic potential.

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Citation: Jeni R. Patel and Dr. Gohil, T. G., 2017. "Antibacterial activity of leaf extracts of some herbs against some clinical isolates "", *International Journal of Current Research*, 9, (08), 55476-55478.

INTRODUCTION

Majority of the world population depend mainly on various plants for health related problem. It has been estimated that plant drug constitute about 80 % in developing countries like India. In India thousands of species are known to have medicinal value and the use of various parts of medicinal plants to cure disease since ancient times. Plants have variety and huge source of photochemical with proven potential of treating communicable infection with lesser side effects compared to the chemotherapeutic agents. Recently much attention has been paid to biologically active compounds from plants used in the alternative medicine. Antimicrobial compounds of plant origin have tremendous therapeutic potential. There have been lots of intensive studies on antimicrobial activity of plant extracts in last decade. In past few years, it has been increasingly reported by several scholars. (Nair and Chanda S. 2007; Bharath, G. and Farzin, 2011; Iyer, *et al*., 2011) Extraction and phytochemical screening of various plant parts shows antibacterial, antifungal and antioxidant activity (Venkata & *et al*., 2010; Prashant Tiwari & *et al*., 2011; Vaghasiya, and *et al*., 2011). There was a very high rate of infectious diseases in the developing countries like India.

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Pathogens frequently developed resistance to many chemotherapeutic agents and created problems in the treatment of infectious diseases. Because of inadequate availability and high cost of new generation antibiotics, scientists are forced to search for new alternatives i.e. new safer, cheaper therapeutic compound. There is a special need to search and develop new therapeutic agents to combat resistant and emerging pathogens. The main aim of the present study was to evaluate and determine the antibacterial potential leaf extracts of *Chrozophora rottleri, Oxalis corniculata, Parthenium hysterophorus* and *Solanium xanthocarpum* against some clinical isolates.

Chrozophora rottleri locally known as Suryavarti belongs to Euphorbiaceae. It is an annual common waste lands, erect herb with silvery hairs, occurs naturally throughout India. Oxalis corniculata locally known as creeping wood sorrel belongs to Oxalidaceae. It is a one of the most versatile medicinal plants having a wide spectrum of biological activity. It is annual creeping herbs with tiny flowers. Parthenium hysterophorus locally known as carrot grass belongs to Asteraceae. It is an aggressive ubiquitous annual weed. This erect herb also known for its dense growth. All these plant species were possessing useful medicinal properties. In addition, Dipankar C. *et al*., 2011; Patel Seema, 2011; Meruga Srikanth *et al*., 2012; Shelly Rana *et al*., 2016 reported, a good source of phytochemical and antibacterial potential. Considering this facts & figures, it can be claimed that these plants are the valuable sources for new

safer, cheaper therapeutic compound and should be evaluated for their.

MATERIALS AND METHODS

Plant material

Fresh and diseased-free leaves of *Solanium xanthocarpum* Lin., *Oxalis corniculata* Lin. *Parthenium hysterophorus* Lin. and *Chrozophora rottleri* A. Juss. ex Spreng were collected from local site of Vyara, District Tapi, Gujarat, India. The plants were identified with the help of flora of Gujarat and confirmed by Dr. T. G. Gohil & naturalist Dr. Minoo Parabia. The leaves of these plants were washed thoroughly under running tap water and then dry at RT for an hour and then dried in an oven at 55°C for 24 hours. The dried plant material were pulverized to fine powder in a grinder, stored in air tight bottle, labeled and kept in a dark at room temperature.

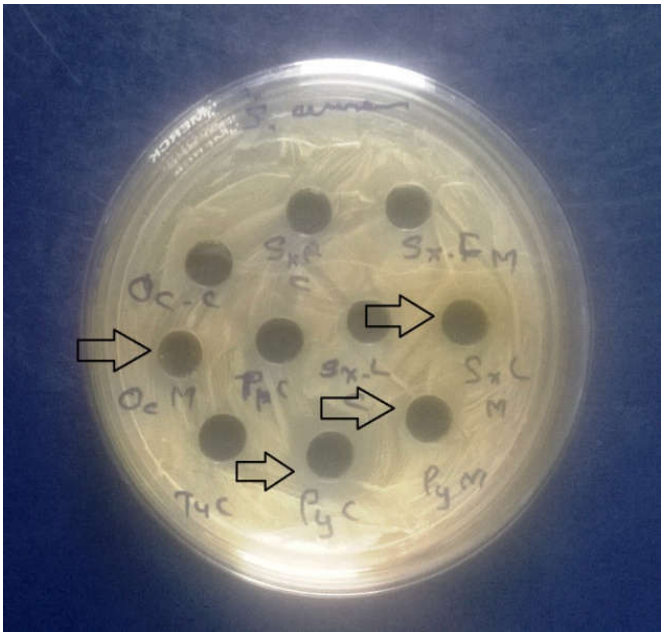
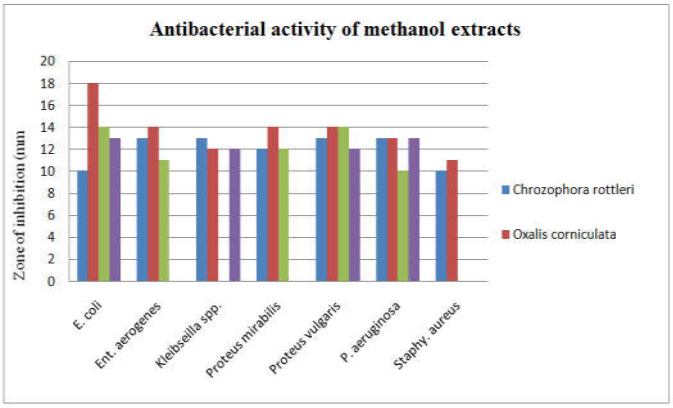


Figure 1. One of the plates showed antibacterial activity against clinical isolates



Note: 8 mm well was loaded with 20 µl of crude extracts (100mg/ml)

Figure 2. Antibacterial activity of methanol extract of leaves of tested Herbs against clinical isolates

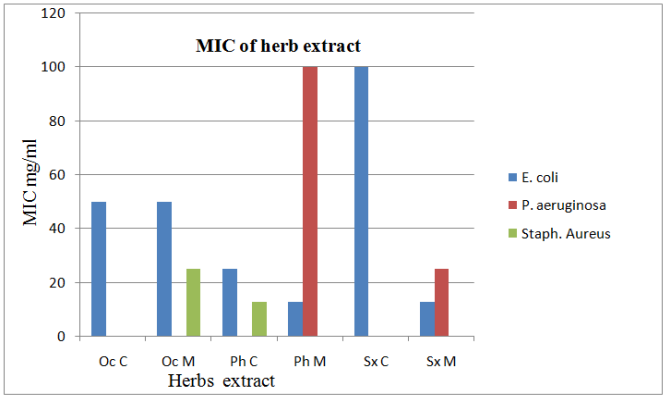


Figure 3. MIC of herb extracts against three most important clinical isolates

Extraction

Extraction of leaves of respective plants was carried out by maceration technique. A 5 gm dried powder was soaked separately in 50 ml of chloroform and methanol in Erlenmeyer flask. The flasks were covered with aluminum foil and allowed to stand in a dark for 72 hrs for extraction. These extracts were filtered through Whatmann filter paper no. 1 and filtrate was evaporated at 55°C in an oven to get dark greenish residue (crude extract), which was stored at 4°C prior to use.

Table 1. The percentage recovery of crude extract of tested Herbs

Sr. No.	Herbs	Specimen code	% Yield of crude extract	
			Chloroform extract	Methanol extract
1	*Chrozophora rottleri*	Cr	117 mg (2.34 %)	258 mg (5.16 %)
2	*Oxalis corniculata*	Oc	86 mg (1.72 %)	354 mg (7.08 %)
3	*Parthenium hysterophorus*	Ph	95 mg (1.9 %)	203 mg (4.06 %)
4	*Solanium xanthocarpum*	Sx	186 mg (4.65 %)	355 mg (7.1 %)

Table 2. Antibacterial activity of chloroform & methanol extract of leaves of tested Herbs by well diffusion method

Clinical isolates	Diameter of zone of inhibition (mm)							
	Cr - C	Cr - M	Oc - C	Oc -M	Ph - C	Ph - M	Sx - C	Sx - M
E. coli	--	10	13	18	16	14	12	13
Enterobacter aerogenes	--	13	--	14	11	11	--	--
Kleibseilla spp.	--	13	--	12	--	--	10	12
Proteus mirabilis	--	12	--	14	--	12	--	--
Proteus vulgaris	14	13	16	14	16	14	--	12
Pseudomonas aeruginosa	11	13	11	13	--	10	19	13
Staphylococcus. aureus	--	10	--	11	17	--	--	--

Note: * 8 mm well was loaded with 20 µl of crude extract (100mg/ml)

These crude extract was further dissolved in DMSO to prepare the stock solution of 100 mg/ml.

Sources of clinical isolates

The antibacterial activity of leaf extract of *Chrozophora rottleri, Oxalis corniculata Parthenium hysterophorus* and *Solanium xanthocarpum* were tested against clinical isolates i.e. *Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa, Proteus vulgaris, Proteus mirabilis, Enterobacter aerogenes and Klebseilla spp.* These bacteria were isolated from urine and pus sample of a patient suffering from urinary tract infection & wound infection by standard microbiological procedure and identified biochemically. All these isolated pathogens were stored in nutrient agar slants at 4°C.

Screening for antibacterial activity

Screening for antibacterial properties of crude extract of all the test plants were tested by agar well diffusion method. A nutrient agar plate was seeded with 100 µl suspension of clinical isolates equivalent to McFarland standard 0.5, spread uniformly over a plate by spread plate technique. After 15-30 minute a sterile stainless steel borer 8 mm in diameter was used to make a well in each plate. These wells were filled with 20 µl of respective crude extract. Then plates were kept at 4˚ C for 1 hour for prediffusion & incubated at 37 ± 1˚ C for 24 hours. After overnight incubation diameter of zone of inhibition were measured in mm.

Determination of MIC

MIC of some herbs extracts were also determined against three most important clinical isolates *Escherichia coli, Staphylococcus aureus and Pseudomonas aeruginosa* by two fold dilution followed by well diffusion method. The stock solution of 100 mg/ml of crude extract of tested herbs which showed antibacterial potential were diluted two fold and then 20 µl of respective diluents were transferred into well in N. agar plates which already seeded with standard inoculums of clinical isolates. Then plates were kept at 4˚ C for 1 hour for prediffusion & incubated at 37 ± 1˚ C for 24 hours. After overnight incubation diameter of zone of inhibition were measured in mm.

RESULTS AND DISCUSSION

5 gm dried powdered were extracted in chloroform and methanol, the percentage recovery of all these four plants crude extracts were given in the Table 1. All the crude extract showed good activity against tested clinical isolates which was shown in the Table 2. and Figure 1. Methanol extract showed significant activity than chloroform extract which was shown in Figure 2. Result showed that methanol extract of *Chrozophora rottleri* and *Oxalis corniculata* were showed significant activity against all the clinical isolates. Result of MIC of herb extract against three most important clinical isolates tested by agar well diffusion method which was shown in Figure 3.

Conclusion

It was concluded from the results that % yield of crude extract was high in methanol extract than chloroform extract. Highest % recovery of crude extract in case of *Solanium xanthocarpum.* Methanol extract showed significant antibacterial activity than chloroform extract. Methanol extract of *Chrozophora rottleri* and *Oxalis corniculata* were showed significant activity against all the clinical isolates. MIC of methanol extract of Ph and Sx was 12.5 mg/ml against *E. coli*, chloroform extract of Ph was 12.5 mg/ml against *Staph. aureus.*

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