

## ANTIMICROBIAL ACTIVITY OF MEDICINAL PLANTS ON URINARY TRACT PATHOGENS

SHRIKANT, SONIA SETHI\*, GUPTA B LAL

Dr. B. Lal Institute of Biotechnology 6E Malviya Industrial Area Malviya Nagar Jaipur. Email :soniakaura198@gmail.com

Received: 20 Dec 2011, Revised and Accepted: 6 Mar 2012

## ABSTRACT

Anti microbial activity of three medicinal plants (*Murraya*, *Azadirachta*, and *Ocimum*) on Urinary tract pathogens were investigated. The methanolic extracts of leaf of all three medicinal plants were the potent antimicrobial agent than ethanolic extract. Methanolic extract of all the three plants inhibited the growth of *Klebsiella*, *Escherichia* and *Serratia* while the ethanolic extract inhibited less. The highest antibacterial activity was found against *Escherichia* with methanolic extract of Leaf and Bark of *Murraya* and Leaf extract of *Azadirachta*. Bark extract of *Azadirachta* showed highest inhibitory activity against *Serratia* with methanolic extract and least with *Escherichia*. *Ocimum* leaf extract possess maximum antibacterial activity against *Serratia* with methanolic extract and least with *Klebsiella*. *Escherichia* was found to be most sensitive than *Klebsiella* and *Serratia*. The highest antibacterial activity was found against all the Urinary tract pathogens with methanolic extract of Leaf and Bark of *Murraya*.

**Keywords:** Medicinal Plants, Antibacterial activity, Methanolic extract, Ethanolic extract.

## INTRODUCTION

Medicinal plants are part and parcel of human society to combat diseases, from the dawn of civilization. Medicinal plants are considerably useful and economically essential. They contain active constituents that are used in the treatment of many human diseases<sup>1</sup>. The plant extracts have been developed and proposed for use as antimicrobial substances<sup>2</sup>. Plants used in traditional medicine contain a vast array of substances that can be used to treat chronic and infectious diseases.

The use of plant extracts and photochemical, both with known antimicrobial properties can be of great significance in therapeutic treatments. In the last few years, a number of studies have been conducted in different countries to prove such efficiency. Many plants have been used because of their antimicrobial traits, which are chiefly due to synthesized during secondary metabolism of the plant<sup>3</sup>.

In modern days, the antioxidants and antimicrobial activities of plant extract have formed the basis of many applications in pharmaceuticals, alternative medicines and natural therapy. Recently extracts of plant have provoked interest as sources for their potential uses as alternative medicines for the treatment of many infectious diseases<sup>4</sup>.

Bacterial resistance to antibiotics represents a serious problem for clinicians and the pharmaceutical industry and great efforts are being made to reverse this trend, and one of them is the widespread screening of medicinal plants from the traditional system of medicine hoping to get some newer, safer, and more effective agents that can be used to fight infectious diseases<sup>5</sup>.

*Azadirachta indica* A. Juss (syn. *Melia azadirachta*) is well known in India and its neighbouring countries for more than 2000 years as one of the most versatile medicinal plants having a wide spectrum of biological activity. Every part of the tree has been used as traditional medicine for household remedy against various human ailments from antiquity<sup>6,7</sup>.

*Murraya koenigii* L. (curry leaf) belonging to family Rutaceae is used as a spice for its characteristic flavour and aroma. It is reported to have anti-oxidant, anti-diabetic, anti-carcinogenic, anti-dysenteric, stimulant, hypoglycaemic and antimicrobial activities<sup>8</sup>. Biologically active carbazole alkaloids are reported to have antimicrobial properties<sup>9</sup>.

*Ocimum sanctum* commonly known as holy basil or Tulsi a herbaceous sacred plant found throughout India. Essential oils of tulsi have antibacterial<sup>10, 11</sup> with emphasis on anti tuberculosis<sup>12</sup>, antifungal<sup>13</sup> and antiviral properties<sup>14</sup>.

## MATERIALS AND METHODS

## Plant material

The plants of *M.koenigii* (MK), *Azadirachta indica*(AI) and *Ocimum sanctum*(OS) were collected from the Local Nursery of Jaipur. Different parts including Leaf, Bark and Roots were separated, washed thoroughly with distilled water, shade dried, powdered using blender and stored.

## Solvent Extraction

After authentication the powdered parts were extracted with methanol, ethanol, petroleum ether and acetone using Soxhlet's apparatus for 12-14 h. The extracts were concentrated, percentage yield calculated and then subjected to preliminary phytochemical analysis.

## Antimicrobial Activity

The *in vitro* screening for antimicrobial study was carried out using selected urinary tract infection (UTI) causing pathogens which includes three gram negative bacteria (*Escherichia coli*, *Klebsiella pneumoniae* and *Serratia marscesns*)

These organisms were identified by following the standard microbiological methods. The antibacterial screening of the extracts were carried out by determining the zone of inhibition using well diffusion method. The strains of microorganisms obtained were inoculated in conical flask containing 100 ml of nutrient broth. These conical flasks were incubated at 37° C for 24 h and were referred to as seeded broth.

Different concentrations of the extracts were prepared by reconstituting with methanol and ethanol. The test microorganisms were seeded into respective medium by spread plate method 10 µl (10 cells/ml) with the 24h cultures of bacteria growth in nutrient broth. One ml of this was used in flooding over nutrient agar plates in the well diffusion method of the *in vitro* antimicrobial sensitivity test.

The plates were left for 5mins after which they were dried at 37° C for 1hour. Four wells, equally distant, were bored round the plate using a sterile cork borer. Various concentrations of the diluted extracts were put inside the wells. Solvents such as Methanol and ethanol were put inside the well in separate petriplates to serve as negative control while Chloramphenicol (1mg/ml) was used as positive control in the separate petriplates. The plates were left free for 1 hour after which there were incubated at 37° C for 24 hours and were examined for zones of inhibition

## Phytochemical Screening

The methanolic extracts of different plants were used as samples for qualitative phytochemical screening for tannins, alkaloids, glycosides, terpenoid, steroid and flavonoids following the standard procedures of Trease and Evans<sup>15</sup>, 1989.

Table 1: The phytochemical screening of the Plant extracts.

Phytoconstituents	OS	AI	MK
Carbohydrate	-	-	+
Tannin	+	+	-
Alkaloid	+	-	+
Flavonoids	-	+	-
Steroid	+	-	-
Glycoside	+	-	-

(+): Present; (-): Absent

Table 2: Antimicrobial activity of methanol and ethanol extracts of plants against UTI Pathogens

Bacteria	Organic solvent	Leaf MK (mm)	Bark MK (mm)	Leaf AI (mm)	Bark AI (mm)	Leaf OS (mm)	Chloramphenicol
<i>Klebsiella</i>	Methanol	27	24	26	25	21	29
	Ethanol	19	15	13	14	13	
<i>Escherichia</i>	Methanol	29	29	18	12	20	25
	Ethanol	23	20	21	10	18	
<i>Serratia</i>	Methanol	22	24	22	20	18	23
	Ethanol	16	12	12	17	16	

Values are statistically significant at (p<.05)

## RESULTS AND DISCUSSION

The phytochemical analysis of the leaf powder and various extracts gave the results as depicted in Table-1.

Results obtained from the susceptibility testing of the organisms revealed that the tested three medicinal plants extracts possess potential antibacterial activity against *Klebsiella*, *Escherichia* and *Serratia*. When tested by agar diffusion method the methanolic extract was the potent antimicrobial agent than ethanolic extract. Methanolic extract inhibited *Klebsiella*, *E.coli* and *Serratia* while the ethanolic extract inhibited less (Table-2).

The highest antibacterial activity was found against *Escherichia* (29mm) with methanolic extract of Leaf and Bark of *Murraya* (Table-2).

*Azadirachta* leaf extract possess maximum antibacterial activity against *Klebsiella* (26mm) with methanolic extract (Table 2). Bark extract of this plant showed highest inhibitory activity against *Klebsiella* (25mm) with methanolic extract and least with *Escherichia* (12mm) (Table 2).

*Ocimum* leaf extract possess maximum antibacterial activity against *Klebsiella* (22mm) with methanolic extract and least with *Serratia* (Table 2).

*Klebsiella* was found to be most sensitive than *Escherichia* and *Serratia*. The highest antibacterial activity was found against all the Urinary tract pathogens with methanolic extract of Leaf and Bark of *Murraya*.

Plants are important source of potentially useful structures for the development of new chemotherapeutic agents. The first step towards this goal is the *in vitro* antibacterial activity assay<sup>16</sup>. Many reports are available on the antiviral, antibacterial, antifungal, anthelmintic, antimolluscal and anti-inflammatory properties of plants<sup>17, 18</sup>. Some of these observations have helped in identifying the active principle responsible for such activities and in the developing drugs for the therapeutic use in human beings.

However, not many reports are available on the exploitation of antifungal or antibacterial property of plants for developing commercial formulations for applications in crop protection. The phytochemical research based on ethno-pharmacological information is generally considered an effective approach in the discovery of new anti-infective agents from higher plants<sup>19</sup>.

Knowledge of the chemical constituents of plants is desirable, not only for the discovery of therapeutic agents, but also because such

information may be of value in disclosing new sources of such economic materials as tannins, oils, gums, precursors for the synthesis of complex chemical substances.

In addition, the knowledge of the chemical constituents of plants would further be valuable in discovering the actual value of folkloric remedies<sup>15</sup>. Chemically constituents may be therapeutically active or inactive. The ones which are active are called active constituents and the inactive ones are called inert chemical constituents<sup>20</sup>.

## REFERENCES

1. Stary F and Hans S: The National guides to medical herbs and plants. Tiger Books 1998 Int. Plc. UK.
2. Del Campo J, Amiot MJ and Nguyen C: Antimicrobial effect of Rosemary extract. J. Food Protect 2000; 63: 1359-1368.
3. Prusti A, Mishra, SR, Sahoo S and Mishra SK: Antibacterial Activity of Some Indian Medicinal Plants *Ethnobotanical Leaflets* 2008; 12: 227-230.
4. Acharya S, Dash GK, Mondal S and Dash SK: Antioxidative and Antimicrobial study of *Spondias Mangifera* Willd Root, IJPPS. 2010; 2:68-71.
5. EL-Mahmood AM, Doughari JH and Ladan N: Antimicrobial screening of stem bark extracts of *Vitellaria paradoxa* against some enteric pathogenic microorganisms. Afr. J. Pharm. Pharmacol 2008; 2(5): 089-094.
6. Chopra RN, Nayer SL and Chopra IC: Glossary of Indian Medicinal Plants, CSIR, New Delhi 1956.
7. Chatterjee A and Prakashi S (eds): The treatise on Indian Medicinal Plants 1994, vol. 3, p. 76.
8. Ningappa MB, Dinesha R, Srinivas L: Antioxidant and free radical scavenging activities of polyphenol-enriched curry leaf (*Murraya koenigii* L.) extracts. Food Chemistry 2008; 106, 720-728.
9. Ramsewak RS, Nair MG, Strasburg GM, De Witt DL and Nitiss JL: Biologically active carbazole alkaloids from *Murraya koenigii*. Journal of Agricultural and Food Chemistry 1999, 47, 444-447.
10. Singh S, Malhotra M and Majumdar DK: Antibacterial activity of *Ocimum sanctum* L. Fixed oil. Indian J. Exp. Biol 2005; 43: 835-837.
11. Mishra P and Mishra S: Study of antibacterial activity of *Ocimum sanctum* extract against gram positive and gram negative bacteria. Am. J. Food Technol 2011; 6: 336-341.
12. Farivar TN, Fard AHM, Zahedani SS, Naderi M and Moud BS: Anti-tuberculosis effect of *Ocimum sanctum* extracts in invitro and macrophage culture. J. Med. Sci 2006; 6: 348-351.
13. Geeta DMV, Kedlaya R, Deepa S and Ballal M: The activity of *Ocimum sanctum* against the Enteric pathogens. Ind. J. Med. Sci 2001; 55: 434-438.

14. Parida MM, Pandya G, Bhargava and Jana A: Assessment of invitro antiviral activity of certain indigenous plants against polio virus type 3. Indian J. Virol 1997; 13:101-105.
15. Trease GE and Evans WC: A Text book of Pharmacognosy, 13th edition 1989; 83, 685. Bailliere Tindall Ltd., London, ISBN: 0702013617
16. Mojab Faraz, Kamalinejad Mohammed, Ghaderi Naysaneh, Reza Hamid Validipour, Iranian Journal of Pharmaceutical Research 2003;2:77.
17. Tona L, Kambu K, Ngimbi N, Cimanga K and Vlietinck AJ: Antiamoebic and phytochemical screening of some Congolese medicinal plants. J. Ethnopharmacol 1998; 61: 57-65.
18. Govindarajan R, Vijayakumar M, Singh M, Rao CHV, Shirwaikar A, Rawat AKS and Pushpangadan P: Antiulcer and antimicrobial activity of *Anogeissus latifolia*. J. Ethnopharmacol 2006; 106: 57-61.
19. Behera SK and Misra MK: Indigenous phytotherapy for genito-urinary diseases used by the Kandha tribe of Orissa, India. J. Ethnopharmacol 2005; 102: 319-325.
20. Duraipandiyan V, Ayyanar M and Ignacimuthu S: Antimicrobial Activity of Some Ethnomedical Plants Used by Paliyar Tribe from Tamil Nadu, India. BMC complementary and alternative medicine 2006; 635.
21. Iyengar MA: Study of Crude Drugs. 8th ed., Manipal Power Press, Manipal, India. 1995;2.