REGULAR ARTICLE

ANTIMICROBIAL ACTIVITY OF IMPORTANT INDIAN MEDICINAL PLANTS AGAINST PYOGENIC INFECTION

B. Bagyalakshmi¹, D. Sridhar², P. Ponmurugan¹

¹Department of Biotechnology, K.S. Rangasamy college of Technology, Tiruchengode-637 215, TamilNadu, India ²Department of Microbiology, Maharaja college for women, Perundurai-638 052 TamilNadu, India

SUMMARY

The antimicrobial activity of crude extracts of five medicinal plants used in traditional Indian medicine was tested against five important pyogenic bacteria. They are Streptococcus pyogenes, Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa, Streptococcus pneumoniae. Of the five medicinal plants used only three were showed considerable antimicrobial activity against one or more species of microorganisms tested. Among the three solvents used the most effective extract was found to be methanol extraction. The most effective antimicrobial plant was identified as *Glycyrriza glabra* followed by *Dathura metal*, *Coccinia grandis*. Least activity was observed in *Sida spinosa*, and *Lab lab purpureus*.

Keywords: Antimicrobial activity, Medicinal plants, Crude extracts, Pyogenic bacteria.

B. Bagyalakshmi et al. Antimicrobial Activity of Important Indian Medicinal Plants Against Pyogenic Infection. J Phytol 1 (2009) 391-396 *Corresponding Author, *Email*: bagya_lakshmi2005@yahoo.com

1. Introduction

India is a varietal emporium of medicinal plants and is one of the richest countries in the world in regard to genetic resources of medicinal plants. The agro-climatic conditions are favorable for introducing new exotic plant varieties [1]. Many infectious diseases are known to be treated with herbal remedies throughout the history of mankind. In India, Herbal medicines have been the basis of treatment and cure for various diseases in traditional methods practiced such as Ayurveda, Unani and Siddha [2]. Most of the medicinal plants contain Tannins, Gallic acid, Ouinine, flavonoids and alkaloids.

Antimicrobials of plant origin are not associated with many side effects and have an enormous therapeutic potential to heal many infectious disease [3].

The increasing prevalence of multi drug resistant strains of bacteria and the recent appearance as strains with reduced susceptibility to antibiotics leads to the emergence of untreatable bacterial infections and need to the search of new antibiotics[4-6]. The potential of higher plants as source for new drugs is still largely unexplored. Random screening stool in discovering new biologically active molecules has been most productive in the area of antibiotics. Secondary metabolites have been extensively investigated as sources of medicinal agents [7].

On a global basis, at least 130 drugs, all single chemical entities extracted from higher plant or modified further synthetically are currently in use [8-9]. A wide range of medicinal plant parts is used for extract as raw drugs and they posses varied medicinal properties. The different parts used include, root, stem, flower, leaves. Although hundreds of plant species have been tested for antimicrobial properties, the vast majority of have not been adequately evaluated.

Systematic investigation was undertaken to screen the antimicrobial activity of selected medicinal plants against pyogenic infection causing bacteria. The plant selected are fresh leaves of *Sida spinosa, Dathura metal, Coccinia grandis* and *Lab lab purpureus* and root of *Glycyrriza glabra*. They are used for several pharmaceutical purposes like skin disease, scabies [10], analgesic activity and anti inflammatory [11-12].

2. Materials and Methods

Collection of plant materials

Fresh leaves of *Sida spinosa, Dathura metal, Coccinia grandis* and *Lablab purpureus* and roots of Glycyrriza glabra were collected. The leaves were washed thoroughly 2-3 times with running water and once with sterile distilled water. Leaf and root material was then air dried on sterile blotter under shade.

Solvent extraction

Dried leaves of *Sida spinosa, Dathura metal, Coccinia grandis* and *Lablab purpureus* and roots of *Glycyrriza glabra* were dried in shade for five days and then powdered with the help of blender. 25 g of dried powdered leaf and root were mixed with solvents like methanol, ethanol and hexane. Added in Soxhlet extractor for 24- 48 hs. The solvent extracts were concentrated under reduced pressure and preserved at 5° C in air tight bottle until further use.

Growth and maintenance of test microorganisms for antimicrobial studies

Pus samples were collected from patients wound infection. Pyogenic who have identified organisms were isolated and biochemically characterized and as Streptococcus pyogenes, Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa, Streptococcus pneumoniae. The bacterial cultures were maintained on nutrient agar slants for further analysis.

Agar well -diffusion assay

The antimicrobial activity of the extracts was determined by using the agar well diffusion technique [13]. Mueller- Hinton agar plates (Himedia, Mumbai) were seeded with 0.1 ml of overnight culture, allowed to set and well made by sterile cork borer (0.85 cm) and 200 µl of extract was added into each well. Then bacterial plates were incubated at 37° C 24 hs. Microbial growth was determined by measuring the diameter of zone of inhibition. Chloramphenicol antibiotic disc were used as control (Himedia, Mumbai). The experiment was done three times and the mean values are presented.

3. Results and Discussion

Medicinal 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 [14]. Many reports are available on the antiviral, antibacterial, antifungal, antihelminthic, anti inflammatory properties of plants [15-18]. However, not many reports are available on the exploration of plants against pyogenic infection causing bacterium.

Table 1: Characterization of the plant extracts

S.N	No Scientific Name	Common name	Parts used	Extraction 1	Extract dry Weight mg/ml
1	Sida spinosa	Mairmanikkam	Leaf	Methanol ,Ethanol & Hexane	100
2	Dathura metal	Ummattai	Leaf	Methanol,Ethanol & Hexane	100
3	Coccinia grand	is Kovai	Leaf	Methanol ,Ethanol & Hexane	100
4	Lablab purpure	us Avarai	Leaf	Methanol ,Ethano & Hexane	ol 100
5	Glycyrriza glab	ra Atimaduram	Leaf	Methanol ,Ethano & Hexane	1 100

In the present investigation the methanol, ethanol and hexane extracts of leaf/root were prepared and tested against pyogenic infection causing bacterium. The results are tabulated (Table 2, 3 and 4).

Table 2: Antimicrobial activity of Methano	extracts (100 µg /ml) by selected medicinal
--	---

	Zone of Inhibition (mm)							
Bacterial species	G. glabra	C. grandis	D. metal	S. spinosa	L .purpureus	Chlo		
S.aureus	12+0.15	11+0.57	10+0.15	9+0.33	9+ 0.33	16+0.33		
S.pyogenes	11+1.42	10+0.52	9+0.15	10+0.66	4+0.66	14+0.33		
S.pneumoniae	10+ 0.72	9+ 0.00	9+0.15	7+0.33	5+0.33	10+0.33		
P.aeruginosa	10+0.06	8+0.33	11+0.15	4+0.66	4+0.00	11+0.33		
E.coli	9+0.33	10+0.33	8±0.15	6+0.33	6+0.06	10+0.33		

Among the three extracts, methanol extracts showed highest antibacterial activity against all the bacterium used in this study. Methanol extracts of roots of *G. glabra* showed considerable activity against *Staphylococcus aureus, Streptococcus pyogenes and Streptococcus pneumonia* (12mm) and also against gram negative bacterium. Our results correlated with the findings scientists who worked with this plant [19]. The methanol, acetone extracts

did not have activity on gram negative bacterium [20]. Methanol extract can subsequently fractioned and monitored by bioassay leading to the isolation of active fraction by further phytochemical analysis [21].

Table 3: Antimicrobial activity of Ethanol extracts (100 g/ml) by selected medicinal Plants

	Zone of Inhibition (mm)							
Bacterial species	G. glabra	C. grandis	D.metal	S. spinosa	L .purpureus	Chlo		
S.aureus	10+0.33	10+0.52	7+0.66	8+0.42	6+ 0.54	16+0.33		
S.pyogenes	7+1.42	9+0.00	5+0.15	7+0.66	5+0.15	14+0.33		
S.pneumoniae	9+ 0.06	8+ 0.15	4+0.00	8+0.23	5+0.33	10+0.33		
P aeruginosa	10+0.06	6+0.33	6+0.33	4+0.15	6+0.12	11+0.33		
E.coli	8+0.33	9+0.66	9+0.15	3+0.33	7+0.16	10+0.33		

Most of the extracts tested showed varied level of antimicrobial activity. This result supported by the observation of many scientists [22-23]. Least antimicrobial activity was found to be Lablab purpureus against all the bacterium used. Highest activity showed against *Staphylococcus aureus* of methanolic extract (8mm). The methanolic extracts of the roots of *C. majus* reveals high resistance to *Fusarium* [24]. A glycoprotein isolated from *C. majus* exhibit good antimicrobial activity against methicillin resistant and multi resistant enterococci [25].

Table 4: Antimicrobial activity of Hexane extracts (100 g/ml) by selected medicinal

	Zone of Inhibition (mm)						
Bacterial species	G. glabra	C. grandis	D.metal	S. spinosa	L .purpureus	Chlo	
S.aureus	8+0.75	9+0.12	7+0.00	5+0.33	5+ 0.21	16+0.33	
S.pyogenes	5+1.00	7+0.52	8+0.33	7+0.66	3+0.66	14+0.33	
S.pneumoniae	8+ 0.06	8+ 0.33	9+0.15	4+0.16	7+0.00	10+0.33	
P aeruginosa	7+0.33	5+0.42	4+0.66	4+0.33	6+0.06	11+0.33	
E.coli	6+0.03	7+0.12	2+0.42	6+0.43	5+0.43	10+0.33	

Chlo-Chloramphenicol

Hexane extract showed considerable activity against Staphylococcus aureus, Streptococcus pyogenes on *G. glabra* and *C. grandis*. Hexane extracts of leaves of D.metal showed least activity agaist *E. coli* was observed (2 mm). Hexane plant extract also have the same level of antimicrobial activity [26].

It is not surprising that there are differences in the antimicrobial effects of plants due to the phytochemical properties and differences among species. Some of the plant extracts may have contained antibacterial constituents, not in sufficient concentrations so as to be effective. It is also possible that compound may not soluble in methanol or water [27].

The results of present investigation clearly indicate that the antimicrobial activity vary with the species the plants and plant material and solvents used for extraction. In conclusion, of all the five plants used in this study. Roots of *G. glabra* showed highest activity in methanolic extracts followed by *C. grandis, D. metal.* Least activity was observed in the hexane extracts. Thus the study ascertains the value of plants used an ayurveda, which could be of considerable interest to the development of new drugs.

Acknowledgement

The authors are thankful to the Principal and the Management for providing necessary facilities and constant encouragement to carry out this study.

References

- Mahesh, B and S. Satish, 2008. Antimicrobial Activity of Some Important Medicinal Plant Against Plant and Human Pathogens. World J. Agri Sci 4: 839-843.
- 2. 2. Sukhder,1997.Ethanotherapeutics and modern drug development. The Potential Aurvedha.Curr.Sci.73:909-928.

- Iwu, M.W., A.R., Duncan, and C.O. Okunji 1999. New Antimicrobials of Plant Origin. In: Janick J. (ed.): Perspectives on New Crops and New Uses. ASHS Press, Alexandria, 43: 457–462.
- 4. Sieradzki, K., Roberts, R.B., Haber S.W., and A. Tomasz, (1999). The development of vancomycin resistance in a patient with methicillin - resistant *Staphylococcus aureus* infection. N. Engl. J. Med, 340: 517– 523.
- Tomoka N., A. Takashi, T. Hiromu, I. Yuka, M. Hiroko, I. Munekazu, T. Totshiyuki, I. Tetsuro, A. Fujio, I. Iriya., N. Tsutomu, W. Kazuhito, 2002. Antibacterial activity of extracts preparated from tropical and subtropical plants on methicillin-resistant *Staphylococcus aureus*. J. Health Sci, 48: 273– 276.
- Bandow, J.E., H. Brotz, L Leichert , 2003. Proteomic approach to understanding antibiotic action. Antimicrob. Agents. Chemother. 47: 948-955.
- Krishnaraju, A.V., T.V.N. Rao, D. Sundararaju, 2005. Assessment of bioactivity of Indian medicinal plants using Brine shrimp (*Artemia salina*) lethality assay. Int. J. Appl. Sci. Eng. 2: 125-134.
- Newman, D.J., G.M. Cragg and K.M. Snader, 2000. The influence of natural Products upon drug discovery. Nat. Prod. Res, 17: 215-234.
- 9. Westh, H., C. Zinn and V. Rosdahl, 2004. An international multicenter study of antimicrobial consumption and resistance in *Staphylococcus aureus* isolates from 15

hospitals in 14 countries. Microb. Drug Resist. 10: 169-176.

- 10. Perry, L.M. 1980. Mediinal plants of east and south east asia, Attributed properties and uses, MIT Press, London.
- Farrukh, U., H, Shareef., S. Mahmud., S, Ayub Ali., and G. H. Rizwani, 2008. Antibacterial activities of *Coccinia grandis* Pak. J. Bo., 40: 1259-1262.
- Wannang, N. N., H. C. Ndukwe and C. Nnabuife, 2009. Evaluation of the analgesic properties of the *Datura metel* seeds aqueous extract. Journal of Medicinal Plants Research, 3: 192-195.
- Bauer, A.W., W.M.M. Kirby, J.S. Sherris, and M. Turck, 1966. Antibiotic Susceptibility testing by a standard single disc method. Am. J. Clin. Pathol. 45, 493-496.
- Tona, L., K. Kambu, N. Ngimbi, K. Cimanga and A.J. Vlietinck, 1998. Antiamoebic and phytochemical screeining of some Congolese medicinal plants. J. Ethanopharmacol, 61: 57-65
- Kumaraswamy, Y., P.J. Cox, M. Jaspars, L. Nahar and S.D. Sarker, 2002. Screening seeds of Scottish plants for antibacterial activity. J. Ethanopharmacol, 83:73-77.
- Stepanovic, S., N. Antic, I. Dakic and M. Svabic vlahovic, 2003. In vitro antimicrobial activity of propilis and antimicrobial drugs. Microbiol. Res., 158: 353-357.
- Bylka, W., M. Szaufer-Hajdrych, I. Matalawskan and O. Goslinka, 2004. Antimicrobial activity of isocytisoside and

extracts of *Aquilegia vulgaris* L. Lett. Appl. Microbiol., 39: 93-97.

- Behera, S.K. and M.K. Misra, 2005. Lab lab purpureus Indigenous phytotherapy for genito-urinary diseases used by the Kandha tribe of Orissa, India. J. Ethnopharmacol., 102: 319- 325.
- 19. Kambizi, L. and A.J. Afolayan, 2008. Extracts from Candida albicans and Neisseria gonorrhoea Aloe ferox and Withania somnifera inhibit African J. Biotechnol. 7: 12-15.
- Afolayan, A.J., D.S. Grierson, L. Kambizi I. Madamombe and P.J. Masika, 2002. In vitro antifungal activity of some South African, medicinal plants. S. Afri. J. Bot., 68: 72-76.
- Raghavendra, M.P., S. Satish and K.A. Raveesha, 2006. Phytochemical analysis of antibacterial activity of Oxalis corniculata: A known medicinal plant. My. Science, 1: 72-78.
- 22. Govinarajan, R., M. Vijayakumar, M. Singh, C.H.V. Rao, A. Shirwaikar, A.K.S. Rawat and P. Pushpangadam, 2006. Antiulcer and antimicrobial activity of *Anogesissus latifolia*. J. Ethanopharmacol, 106:57-61.
- 23. Samy, R.P. and S. Ignacimuthu, 2000. Antibacteria activity of some folklore medicinal plants used by tribals in western Ghats in India. J. Ethanopharmacol, 69: 63-71.
- Janovská D., K.Kubíková and L. Kokoška, 2003. Screening for antimicrobial activity of some medicinal plants species of traditional Chinese medicine. Czech J. Food Sci. 21: 107–110.

- Fik, E., Gozdzicka-Jozefiak, A., Haertle, T., Mirska, I., Kedzia, W. 1997. New plant glycoprotein against methicillin resistant Staphylococci and enterococci. Acta Microbiol. Pol, 46: 325–327.
- Camporasee, A., M.J. Balick, R. Arrigo, R.G. Esposito, O.N. Morselli, F. Desimone and A.Tubaro, 2003.Screening of anti bacterial activity of medicinal plants from Belize. J. Ethinomedicine, 52: 12-19.
- 27. Stainer, R.Y., J.L. Ingraham, M.L Wheelis, 1986. General Microbiology, 5th ed. London: The MacMillan Press Ltd.