



Antimicrobial Activity of Five Flower Extracts Against Three Pathogenic Bacteria

Kandukuri Vasu and M. A. Singara Charya

Department of Microbiology, Kakatiya University, Warangal-506 009, A.P., India

Nat. Env. Poll. Tech.
ISSN: 0972-6268
www.neptjournal.com

Key Words:

Antimicrobial activity
Pathogenic bacteria
Flower extracts

ABSTRACT

Five flowering plants dominant in Warangal dist. in Andhra Pradesh region were selected for their antimicrobial activity against *Pseudomonas aeruginosa*, *Bacillus cereus* and *Staphylococcus aureus*. Among these plants the flowers of *Butea monosperma* were effective. The solvents methanol and ethanol were effective in extraction of antimicrobial compounds from the plant materials. The aqueous extracts of the plants were not so effective while petroleum ether is failed to extract any antimicrobial compounds from the plants.

INTRODUCTION

Herbal medicine represents one of the most important fields of traditional medicine all over the world. To promote the proper use of herbal medicines and to determine their potential as sources for new drugs, it is essential to study medicinal plants, which have folklore reputation in a more intensified way (Jigna & Sumitra 2007). Over the past 20 years, there has been an increased interest in the investigation of natural materials as sources of new antibacterial agents. Different extracts from traditional medicinal plants have been tested to identify the source of the therapeutic effects. As a result, some natural products have been approved as new antibacterial drugs, but there is still an urgent need to identify novel substances that are active towards pathogens with high resistance (Recio 1989, Cragg et al. 1997). Recently, multiple drug resistance has developed due to indiscriminate use of commercial antimicrobial drugs commonly used in the treatment of infectious diseases (Service 1995) making it a new source of antimicrobial agents with possibly novel mechanisms of action (Hamil et al. 2003, Barbour et al. 2004). Contrary to the synthetic drugs, antimicrobials of plant origin are not associated with many side effects and have an enormous therapeutic potential to heal many infectious diseases (Iwu et al. 1999). Isolation of microbial agents less susceptible to regular antibiotics and recovery of increasing resistant isolates during antibacterial therapy is rising throughout the world, which highlights the need for new principles (Shahidi & Nik 2004). The anti bacterial property of these flowers under study was assayed and communicated.

MATERIALS AND METHODS

Plant material: Five flowering plants (*Nerium odorum*, *Cassia auriculata*, *Butea monosperma*, *Hibiscus rosa sinensis*, *Tagetes lemonii*) were collected from Warangal district, A.P. Fresh flowers were plucked and washed under running tap water, dried under shade and powdered.

Preparation of plant extracts: Two grammes of air dried fine powder was placed in 20 mL of water and solvents in different flasks to prepare pant extracts. The solvents used were methanol, ethanol,

acetone, chloroform and petroleum ether. Flasks were plugged with cotton and then kept on a rotary shaker for 3 days. After 3 days, the contents were centrifuged at 2500 rpm for 15 min. The supernatant was collected and the solvent was evaporated to make higher concentration and it was stored at 4°C in airtight bottles for further studies.

Test organisms: The test organisms used were *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Bacillus cereus*.

Culture media: The media used for bacteria was nutrient agar media (Peptone-1.25 g, NaCl-1.25 g, Beef extract-0.75 g, Agar-5 g, Distilled water-250 mL).

Antimicrobial activity (Agar well diffusion method): The extracts obtained from flowers were studied for antimicrobial activity. A loop full of standard strains *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Bacillus cereus* were inoculated in 30 mL nutrient broth in a conical flask and incubated for 24 hrs to activate the strain. In agar well diffusion method (Perez & Bazerque 1990), the media and the test bacteria cultures were poured into petri dishes. The test strain 0.25 mL was inoculated into the media. Care was taken to ensure proper homogenisation.

The experiment was performed under strict aseptic conditions. After the medium was solidified, a well was made in the plates with sterile borer (6 mm). The extracted compound was introduced into the well and the plates were incubated at 37°C for 24 hrs. All samples were tested in triplicates. Microbial growth was determined by measuring the diameter of the zone of inhibition. Controls without test compound were used and the control and activity was deducted from the test and the results were obtained as zone of inhibition in mm.

RESULTS AND DISCUSSION

The antimicrobial activity of flowers of five plants present in sub-tropical climates of Warangal was screened and the data are presented in Tables 1, 2 and 3. It is evident that the plants behaved differently for different bacteria and altered their efficiency based on the solvent used.

The flowers of *B. monosperma* and *N. odorum* were (+ 5 mm) proved to be very effective against *B. cereus* in methanol extract. *B. monosperma* and *T. lemonii* showed activity of 4 mm against *B. cereus* in ethanol extract. Moderate activity was observed in acetone and chloroform extract. Petroleum ether completely failed to extract any antimicrobial compound from the plants, hence, showed no activity with any strain. *B. monosperma* aqueous extract showed + 3mm activity against *B. cereus*.

Methanol extract of flowers showed maximum 4-5 mm activity against *S. aureus* ethanol extract of *B. monosperma* and *C. auriculata*. *T. lemonii* was potential in controlling *S. aureus* (+ 4mm). Moderate activity (1-3 mm) was observed in acetone and chloroform extracts. Aqueous extract of *H. rosa sinensis* showed + 2 mm activity against *S. aureus*.

The flowers of *N. odorum*, *C. auriculata*, *B. monosperma* and *T. lemonii* showed maximum activity against *P. aeruginosa* in methanol extract. Ethanol extract of *N. odorum* and *C. auriculata* showed activity (+ 4 mm) against *P. aeruginosa*. Moderate activity was observed in acetone extract. Chloroform was not a suitable solvent in the antimicrobial activity. Aqueous extract of *C. auriculata* and *B. monosperma* showed their low to moderate range of inhibition zone (+3 mm).

Similar to the present observations many flowers were tried for their antimicrobial activity against dominant and pathogenic bacteria (Benson 1990, Ahmed et al. 1998, Kudi et al. 1999, Nair et al. 2005). Antimicrobial activity spectra of different plants were screened with their different solvent extractions against variety of pathogenic bacteria and flowers proved to be useful in their applica-

Table 1: Antimicrobial activity of flowers against *Bacillus cereus* (zone of inhibition in mm)

Flowers	Extracts					
	Methanol	Ethanol	Acetone	Chloroform	Pet. ether	Aqueous
<i>Nerium odora</i>	+5.0	+2.0	+1.0	+1.0	-	+1.0
<i>Cassia auriculata</i>	+3.0	+2.0	+2.0	-	-	+1.0
<i>Butea monosperma</i>	+5.0	+4.0	+2.0	+2.0	-	+3.0
<i>Hibiscus rosa sinensis</i>	+3.0	+1.0	-	+2.0	-	-
<i>Tagetes lemonii</i>	+4.0	+4.0	+2.0	+2.0	-	+1.0

Table 2: Antimicrobial activity of flowers against *Staphylococcus aureus* (zone of inhibition in mm).

Flowers	Extracts					
	Methanol	Ethanol	Acetone	Chloroform	Pet. ether	Aqueous
<i>Nerium odora</i>	+4.0	+2.0	+2.0	+1.0	-	+1.0
<i>Cassia ariculata</i>	+4.0	+4.0	+2.0	-	-	+1.0
<i>Butea monosperma</i>	+5.0	+4.0	+2.0	+3.0	-	+1.0
<i>Hibiscus rosa sinensis</i>	+4.0	+1.0	+1.0	+3.0	-	+2.0
<i>Tagetes lemonii</i>	+4.0	+4.0	+1.0	+2.0	-	-

Table 3: Antimicrobial activity of flowers against *Pseudomonas aeruginosa* (zone of inhibition in mm).

Flowers	Extracts					
	Methanol	Ethanol	Acetone	Chloroform	Pet. ether	Aqueous
<i>Nerium odora</i>	+4.0	+4.0	+3.0	-	-	-
<i>Cassia ariculata</i>	+4.0	+4.0	+3.0	-	-	+3.0
<i>Butea monosperma</i>	+4.0	+2.0	+3.0	+2.0	-	+3.0
<i>Hibiscus rosasinensis</i>	+2.0	+2.0	+2.0	-	-	+1.0
<i>Tagetes lemonii</i>	+4.0	+3.0	+2.0	-	-	+1.0

tions (Agarwal et al. 2000). Nayak & Pereira (2006) observed that *Catharanthus roseus* flower extract have wound healing activity in rats. The antimicrobial activities of the leaf and flower essential oils of *Lippia chevalieri* and *Ocimum canum* were evaluated against nine bacteria (Bassole et al. 2005). Antimicrobial activity was observed in leaf and flower extract of six endemic plant species against *Staphylococcus aureus* and *Bacillus subtilis* (Mehlika et al. 2008). Antibacterial substances in *N. indicum* and *H. rosa sinensis* and their efficacy on ten different species of Gram positive and Gram negative bacteria has been investigated by Baqir et al. (1994). Juliana et al. (2004) studied crude methanolic extracts from different parts of the plant (roots, stems, leaves, flowers and fruits) for their antimicrobial activity against pathogenic bacteria. The methanol extract of the *Woodfordia fruticosa* flower exhibited antibacterial activity against *Pseudomonas pseudoalcaligenes*. The extract was more active against Gram negative bacteria as compared to Gram positive (Jigna & Sumitra 2007). Wunwisa & Areeya (2005) evaluated the antimicrobial properties of Thai traditional flower vegetable extracts.

The characterisation and purification of biomolecules of flowers such as *N. odorum*, *C. ariculata*, *B. monosperma*, *H. rosa sinensis* and *T. lemonii* are useful for various purposes including healthcare. Scientific investigations with a view to establish pharmaceutical industries and identifying alternate

new medicinal resources are important. Many of these plant flowers yield exceptionally promising compounds for use in modern drugs.

REFERENCES

- Agarwal, K.K., Ahmad, A., Santha Kumar, T.K., Jain, N., Gupta, V.K., Sushi K and Khanuja, S.P.S. 2000. Antimicrobial activity spectra of *Pelargonium graveolans* L. and *Cymbopogon winterianus* Jowitt oil constituents and acyl derivatives. *J. Med. and Arom. Plant Sci.*, 22: 544-548.
- Ahmed, L., Mohammed, Z. and Mohammed, F. 1998. Screening of some Indian medicinal plants for their antimicrobial properties. *J. Ethnopharmacol.*, 62: 183-193.
- Baqir, S.N., Rafi Shaikh, M., Maleka, F.A. and Dilnawaz, S. 1994. Studies on antibacterial activity of ethanolic extracts from *Nerium indicum* and *Hibiscus rosa sinensis*. *J. Islamic Aca Sci.*, 7: 167-168.
- Barbour, E.K., Al Sharif, M., Sagherian, V.K., Habre, A.N., Talhouk, R.S. and Talhouk, S.N. 2004. Screening of selected indigenous plants of Lebanon for antimicrobial activity. *J. Ethnopharmacol.*, 93: 1-7.
- Bassole, I., Nebie R., Savadogo, A., Ouattara, C. T., Barro, N. and Traore, S.A. 2005. Composition and antimicrobial activities of the leaf and flower essential oils of *Lippia chevalieri* and *Ocimum canum* from Burkina Faso. *Afr. J. Biotech.*, 4: 1156-1160.
- Benson, H.J. 1990. In: Microbiological Applications. 5th edition, p. 134, W.M.C. Brown Publ., U.S.A.
- Cragg, G.M., Newman, D.J. and Snader, K.M. 1997. Natural products in drug discovery and development. *J. Natural Prod.*, 60: 52-60.
- Hamil, F.A., Apio, S., Mubiru, N.K., Bukenya-Ziraba, R., Mosango, M., Maganyi, O.W. and Soejarto, D.D. 2003. Traditional herbal drugs of Southern Uganda, II: Literature analysis and antimicrobial assays. *J. Ethnopharmacol.*, 84: 57-78.
- Iwu, M.W., Duncan, A.R. and Okunji, C.O. 1999. New antimicrobials of plant origin In: Janick, J. (Ed.), Perspectives on New Crops and New Uses. ASHS Press, Alexandria, VA, pp. 457-462.
- Jigna, P. and Sumitra, C. 2007. *In vitro* antibacterial activity of the crude methanol extract of *Woodfordia fruticosa* flower. *Brazilian J. Microbiol.*, 38: 204-207.
- Juliana, B., Valdir, C., Vania, F. N., Mara, R. K. Daniela, E. B. and Alexandre, B. C. 2004. Antimicrobial activity of fractions and compounds from *Calophyllum brasiliense*. *Naturforsch.*, 59: 657-662.
- Kudi, A.C., Umoh, J.U., Eduvie, L.O. and Gefu, J. 1999. Screening of some Nigerian medicinal plants for antibacterial activity. *J. Ethnopharmacol.*, 67: 225-228.
- Mehlaka, B., Umit, B., Fatmagul, G., Kerim, G. and Nazife, Y. 2008. An investigation on the antimicrobial activity of some endemic plant species from Turkey. *Afr. J. Biotech.*, 7: 1-5.
- Nayak, B.S. and Pereira, M.P. 2006. *Catharanthus roseus* flower extract has wound-healing activity in Sprague Dawley rats. *BMC Alt. Med.*, 6: 41.
- Nair, R., Kalariya, T. and Chanda, S. 2005. Antibacterial activity of some selected Indian medicinal flora. *Turk. J. Biol.*, 29: 41-47.
- Parez, C., Paul, M. and Bazerque, P. 1990. Antibiotic assay by agar-well diffusion method. *Acta. Biol. Med. Exp.*, 15: 113-115.
- Recio, M.C. 1989. A review of some antimicrobial compounds isolated from medicinal plants reported in the literature 1978-1988. *Phytotherap Res.*, 3: 117-125.
- Service, R.F. 1995. Antibiotics that resist resistance. *Science*, 270: 724-727.
- Shahidi, B.G.H. and Nik, K.A. 2004. Antibacterial activity of some medicinal plants of Iran against *Pseudomonas aeruginosa* and *P. fluorescens*. *Asian J. Plant Sciences*, 3: 61-64.
- Wunwisa, K. and Areeya, K. 2005. Anti-microbial properties of Thai traditional flower vegetable extracts. *AU J.T.*, 9: 71-74.