

In Vitro Antimicrobial Activity of *Helicteres isora* Extracts Against Gastrointestinal Pathogen

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Abstract – The problem of drug resistance to human pathogenic bacteria and rising of infectious diseases has been reported from all over the world. This increasing prevalence of multidrug resistant strains of microorganisms and reduced susceptibility to antibiotics raises an urgent need to search for new sources of antimicrobial agents. Human infections, particularly those involving the skin and mucosal surfaces constitute a serious problem, especially in tropical and subtropical developing countries. (Methicillin-resistant *Staphylococcus aureus* (MRSA), *Escherichia coli*, and *Salmonella sp.* were observed to be the most frequent pathogens cause gastrointestinal infection. MRSA gained much attention in the past decade, as it is a major cause of hospital-acquired infections. In this study, different extracts of *Helicteres isora*, was screened for the antimicrobial activity against the multidrug resistant pathogens. Results are encouraging indicating the potential antibacterial activity against the test organisms and further validate the traditional use of *H. isora* in abdominal cramps and gastrointestinal infections.

Key word: *In Vitro* Antimicrobial Activity-*Helicteres isora*, Organic solvents extracts-Drug resistant bacteria

Introduction

Infectious disease are the world's leading cause of premature deaths, killing almost 50 000 people every day. Infections due to variety of bacterial etiologic agents such as

pathogenic *Escherichia coli*, *Salmonella* spp., and *Staphylococcus aureus* are most common. In recent years drug resistance to human pathogenic bacteria has been commonly reported from all over the world (Piddock and Wise, 1989; Singh et al 1992 and Mulligen et al;1993). With the continuous use of antibiotics microorganism have become resistant. In addition to this problem, antibiotics are sometimes associated with adverse effects on host which include hypersensitivity, immune suppressant and allergic reactions (Lopez et al.2001 and Idsoet al.1968). This has created immense clinical problems in the treatment of infectious diseases (Davis 1994). Therefore, there is a need to develop alternative antimicrobial drugs for the treatment of infectious diseases; one approach is to screen local medicinal plants for possible antimicrobial properties. Plant materials remain an important recourse to combat serious diseases in the world. According to WHO (1993), 80% of the world's population is dependent on the traditional medicine and a major part of the traditional therapies involves the use of plant extracts or their active constituents. Yet a scientific study of plants to determine their antimicrobial active compounds is a comparatively new field.

Since ancient times, herbs and their essential oils have been known for their varying degrees of antimicrobial activity (Shelef 1983; Zaika 1988; Beuchat and Golden 1989 and Juven et al.1994). In

recent times, the search for potent antibacterial agents has been shifted to plants. Most plants are medicinally useful in treating disease in the body and in most of cases the antimicrobial efficacy value attributed to some plants is beyond belief. Claims of effective therapy for the treatment of dysentery, diarrhea, respiratory disorders, skin diseases, syphilis, fever, leprosy, eye diseases and kidney and urinary disorders by traditional herbalist in India have prompted our interest in the scientific investigation of such herbal medications (Mukherjee, 1953; Chopra et al; 1956; Kritkar and Basu, 1980; Anonymous, 1986 and Nadkarni, 1989). Conservative estimates suggest that about 10% of all flowering plants on earth have at one time, been used by local communities throughout the world but only 1% have gained recognition by modern scientists. There are about 120 plant-based drugs prescribed worldwide and they come from just 95 plant species. Approximately 250,000 species of flowering plants and only 5000 have had their pharmaceutical potential assessed. The treatment of infectious diseases with antimicrobial agents continues to present problems in modern-day medicine with many studies showing a significant increase in the incidence of bacterial resistance to several antibiotics (Kunin 1993). Due to increased resistance of many microorganisms towards established antibiotics, investigation of the chemical compounds within traditional plants has become desirable (Anonymous 1986). There are many published reports on the effectiveness of traditional herbs against Gram-positive and Gram-negative microorganisms, basic health needs in the developing countries. The WHO reported that 80% of world populations rely chiefly on traditional medicines/herbs for primary healthcare have steadily increased worldwide in the recent years.

Helicteres isora belongs to family Sterculiaceae is a sub-deciduous shrub or

small tree of having spreading habit with stem 1-5 inches in diameter, reaching a height of 5-15 feet. The species is native to Asia and Australia. It occurs, throughout India, from Jamuna eastwards to Nepal, Bihar and Bengal and southern India and Andaman Islands. It occurs as undergrowth, especially as a secondary growth in forests. The fruits are astringent, acrid, refrigerant, demulcent, constipating, stomachic, vermifuge, vulnerary, haemostatic and urinary astringent. They are useful in vitiated conditions of pitta ophthalmitis, colic, flatulence, diarrhea, dysentery, verminosis, wounds, ulcers, hemorrhages, epistaxis and diabetes. The fruit extract of possess weak anti-HIV (I) activity. The fruits were also found to possess significant antispasmodic activity. **Promotes relief from abdominal spasm & pain.** Promotes wellbeing from intestinal infestations & loose motions. Supports as antioxidant and blood purifier. (Chunekar KC, 2010; Sharma PV. 2012 and Satake et al, 1999) Keeping in view this study is designed to evaluate the antimicrobial activity of *Helicteres isora*.

Material and Methods

Collection of plant materials: *Helicteres isora* fruits were collected from the The Himalaya Drug Company Dehradun India. The collected plant material was identified by the department of Pharmaco., The Himalaya Wellness Company Dehradun. Fruits were washed under the running tap water 2-3 times and once with sterile distilled water and dried under shade and then homogenized to fine powder and stored in air tight container till further use.

Preparation of solvent extraction: The method of Alade and Irobi, (1993) was adopted for preparation of plant extracts with little modifications. The dried 25 g powdered fruit soaked separately in 100 ml Hexane, methanol, and aqueous. Each solvents were kept in separate flasks with powdered sample were kept in a rotating shaker for 3 days. The extracts were filtered through whatman Filter paper No.1

and the extracts were reduced to half of its original volume. The organic solvents were concentrated in vacuum using rotary evaporator, while aqueous extract was dried using water bath.

Culture media: The media used for antibacterial test was Soyabean casein digest agar/broth of Hi Media Pvt. Ltd. Bombay, India.

Inoculum: The bacteria were inoculated into soyabean casein digest agar /broth and inoculated and incubated at 37 °C for 4 h and the suspension was checked to provide approximately 10⁵ CFU/ml

Microorganisms: The antibacterial activity of the extract was tested individually on G+ve and G-ve bacterial strains .All bacterial strains were obtained from Microbiologics Cooper avenue north,st.cloud,MN 56303.The G+ve strain used was *Staphylococcus aureus* ATCC 6538 and G-vebacterial strains were *E. coli* ATCC 6538; and *Salmonella spp.* NCTC 6017.

Determination of antibacterial activity: The agar well diffusion method (Perez et al; 1990) was modified. Soyabean casein digest agar (SCDA) was used for bacterial cultures. The culture medium is inoculated with the microorganisms suspended in Soyabean casein digest broth. A total of 8mm diameter wells were punched into agar and filled with plant extracts and solvent blank s(distilled water, hexane and methanol as the case may be).Standard antibiotic was simultaneously used as positive control. The plates were then incubated at 37°C for 18 h. The antibacterial activity was evaluated by

measuring the inhibition zone diameter observed. Wells were filled with 0.1 ml of 20 mg/ml concentration of each sample (2 mg/well).Bioactivity was determined by measuring Diameter of Inhibition Zones (DIZ) in mm.

Results and Discussion

The antimicrobial activities of the extracts were evaluated against 3 test microorganisms including one G+ve bacteria, three G-ve bacteria. Their potency was assessed by diameter of zone of inhibition. Among all the tested extracts hexane extract was found to have maximum zone of 21mm against *Staphylococcus aureus* (Table-1 Plate-1) followed by *E.coli* (18mm), and *Salmonella Spp.*(16mm).

The significant antimicrobial effect of *Helicteres isora* against all the pathogen confirmed that the compound present in the crude extract are responsible for the effective antimicrobial activity.

Conclusion

The traditional therapeutic indications of *Helicteres isora*studied appear to have a fairly good degree of correlation with their antimicrobial activity. The herb *Helicteres isora*appear to have broad spectrum of action and it could be useful in antiseptic, disinfectant formulations and in chemotherapy .The antibacterial activities of the herb is particularly noteworthy,considering the importance of these organisms in gastrointestinal infections.

Test organism	Diameter of zone of inhibition (mm)				
	Hexane extract	Methanol extract	Aqueous extract	Acetone extract	Ciprofl oxacin
<i>E. coli</i>	18	16	NAD	17	28
<i>Staph. aureus</i>	21	19	16	18	30
<i>Salmonella spp.</i>	16	15	12	15	28

Table – 1 Antibacterial activity of different extract of *Helicteres isora*

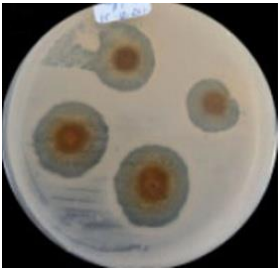


Plate-1 Antibacterial activity of fruit extracts of *Helicteres isora*

Disclaimer Statement

Authors declare that no competing interest exists. The products used for this research are commonly used products in research. There is no conflict of interest between authors and producers of the products.

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