

## Antimicrobial activity of *Lawsonia innermis* Linn. on Urinary Pathogens

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### Abstract

Antimicrobial activity of aqueous, ethanol and acetone extracts of leaves of *Lawsonia innermis* Linn. (LI), were tested on clinical isolates of urinary pathogens viz., *Escherichia coli*, *Klebsiella*, *Proteus* and *Pseudomonas* by well diffusion method. Ethanolic and acetone extracts showed greater inhibitory action than the aqueous ones.

**Keywords:** antimicrobial activity, *Henna*, Herbal medicine, *Lawsonia innermis*, urinary pathogens.

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### INTRODUCTION

Herbal medicine is still the mainstay of about 75-80% of the world's population, especially in the developing countries for primary healthcare. According to World Health Organization (WHO) the use of herbal remedies throughout the world exceeds that of conventional drugs by two to three times (Chhetri *et al.*, 2004).

*Henna* plant, *Lawsonia innermis* Linn. (LI), is now the subject of intense scientific study due to its known healing attributes. The henna plant is a glabrous much branched shrub or a small tree with grayish brown bark. Leaves appear opposite, sub-sessile, elliptic or broadly lanceolate, entire, acute or obtuse, 2-3 cm long and 1-2 cm wide. LI plant contains numerous, small white or rose colored and fragrant flowers.

Leaves of *henna* are used as a remedy in skin diseases in the form of paste or decoction against boils, burns, bruises and skin inflammation. It is cultivated in many tropical countries including India, for coloring palms of hands, soles of feet and finger nails and also for personal adornment (Bhuvanewari *et al.*, 2002). They are also used as gargle in sore throat (Chopra, 1958). Crude and ethanolic extracts of LI leaves were found to have analgesic, antipyretic and anti-inflammatory effect in rats (Ali *et al.*, 1998). Leaves bring down the severity of dysenteries and diseases of spleen, lumbago, bronchitis and syphilitic eye infection (Warrier *et al.*, 1995). This paper documents the effects of aqueous, ethanolic and acetone extracts of leaves of *L. innermis* against selected urinary pathogens.

### MATERIALS AND METHODS

#### Collection of Plants

*L. innermis* was collected from in and around Coimbatore, Tamilnadu, India, to evaluate its antimicrobial effects. The plant leaves were washed

thoroughly in tap water followed by sterile distilled water and shade dried at room temperature for 10-15 days.

#### Preparation of Extracts

Dry plant material (5 g) was ground by using mortar and pestle and made into a suspension with 10 ml of sterile distilled water, 2.5 ml of acetone and 50 ml of 70% ethanol, separately. The suspensions were allowed to sediment overnight at room temperature and the extracts were filtered by using filter paper mesh, and stored separately in sterile containers at 4°C (Babu *et al.*, 2002).

#### Sterility Checking

Prior to subjecting the extracts to antibacterial assay they were checked for sterility by inoculating on nutrient agar and incubating at 37°C.

#### Specimen Cultures

Urine samples were collected from various hospitals in and around Coimbatore, Tamilnadu, India. The collected urine samples were subjected to microscopy, biochemical test and cultural characterization for the confirmation of *Escherichia coli*, *Klebsiella*, *Proteus*, and *Pseudomonas* (Cappuccino and Sherman, 1996). The tested bacterial strains were used for further study.

#### Inoculum Preparation

The test organisms were inoculated into nutrient broth (Himedia, Mumbai, India) and incubated at 37°C overnight. The bacterial inoculum size was adjusted to the turbidity of .5 # McFarland standard so as to deliver a final inoculum of approximately 10<sup>5</sup> colony forming units (Cfu/ml). The final standard microbial broth cultures were used for antimicrobial study.

#### Antibacterial Assay

The aqueous, ethanolic and acetone extracts of LI were tested for their antibacterial activity by well diffusion

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**Table 1.** Comparison of the inhibitory effect of plant extracts against urinary pathogens.

S.No.	Concentrations of plant extracts(µl)	Resistance and sensitivity patterns exhibited by isolated pathogens											
		<i>E.coli</i>			<i>Klebsiella</i>			<i>Proteus</i>			<i>Pseudomonas</i>		
		C	A	E	C	A	E	C	A	E	C	A	E
1	10	R	R	S	R	S	S	R	S	S	R	S	S
2	15	R	S	S	R	S	S	R	S	S	R	S	S
3	20	S	S	S	R	S	S	R	S	S	S	S	S
4	25	S	S	S	S	S	S	S	S	S	S	S	S

C - aqueous, A - acetone, E - ethanol extracts of plant under study. S - Sensitive, R - resistant.

method (Chung *et al.*, 1990) against the bacterial strains under study. Broth cultures of bacterial strains were swabbed over Muller-Hinton agar by using sterile cotton buds, and wells were made by using sterile well cutter (6 mm). Extracts of varying concentrations of 10, 15, 20 and 25 µl were aseptically transferred to the wells separately, incubated at 37° C for 24 hours and the diameter of inhibition zone was recorded. Control wells were maintained with sterile distilled water, ethanol and acetone.

**RESULTS AND DISCUSSION**

The clinical conditions caused by urinary pathogens include urinary tract infection, respiratory infections, systemic infections, dermatitis, soft tissue infection, bacteremia and variety of systemic and nosocomial infections.

In the present study aqueous, ethanolic and acetone extracts of *Lawsonia inermis* were tested against urinary pathogens under study. The pathogens were sensitive to all the three extracts of the plant used (Table. 1). Inhibition zone below 5mm diameter is considered as sensitive result. In case of *Klebsiella*, *Proteus* and *Escherichia coli* the inhibition zone was 9 mm for aqueous extracts in maximum concentration. However in case of *Klebsiella* and *Proteus* the inhibition zone was 11mm for ethanolic extracts in maximum concentration whereas *Escherichia coli* and *Proteus* showed a 9mm inhibition zone for acetone extracts. Ethanolic extract was more effective compared to acetone and aqueous extracts. The present study thus showed that *henna* leaf extracts are capable of inhibiting the growth of microorganisms that are involved in urinary tract infections. This finding therefore supports the use of *henna* in the management of urinary tract infections.

Urinary tract infections (UTI) are an exceedingly common problem prompting seven million office visits and one million hospitalizations each year. Advances in the understanding of both host and bacterial factors involved in UTI have led to many improvements in therapy.

In recent years, interest in medicinal plants has increased considerably. Our knowledge on medicinal plants has mostly been inherited traditionally and there is a growing tendency all over the world to shift from synthetic to natural based products including medicinal plants. Plant and plant-based medicaments are the basis of many of the modern pharmaceuticals we use today for our various ailments (Abraham, 1981)

The plant *henna* being a subject of intense scientific study with healing attributes, contains tannic acid, mucilage and gallic acid as constituents; but the main constituent is 2 hydroxy-napthoquinone (lawsone), known to be the major bioactive constituent (Muhammad and Muhammad, 2005).

Further studies need to be undertaken regarding toxicity, safety and absorption pattern of the active ingredients of this plant.

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