

## IN VITRO EVALUATION OF ANTI BACTERIAL ACTIVITY OF *EQUISETUM ARVENSE LINN* ON URINARY TRACT PATHOGENES

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### ABSTRACT

The aim of the present study was to evaluate the anti bacterial activity of *Equisetum arvense linn* on selected Urinary tract pathogens. Horsetail (*Equisetum arvense*) is one of the oldest known herb famous for its vast variety of therapeutic properties. Antibacterial activity of ethanolic and aqueous extract of *Equisetum arvense L* was screened against *E.coli*, *Klebsiella pneumonia*, *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Staphylococcus saprophyticus* and *Enterococcus faecalis* using disc diffusion technique. The results of this study showed that both the extracts at different concentrations exhibited anti bacterial activity against the bacterial species tested. The ethanolic extract showed higher degree of activity than aqueous extract when compared with the standards.

**Keywords:** *Equisetum arvense linn*, Disc diffusion technique, Mac Farland's standard, Zone of inhibition.

### INTRODUCTION

Urinary tract infection (UTI) is a condition where one or more parts of the urinary system (the kidneys, ureters, bladder, and urethra) become infected. UTIs are the most common of all bacterial infections and can occur at any time in the life of an individual. Almost 95% of cases of UTIs are caused by bacteria that typically multiply at the opening of the urethra and travel up to the bladder. Much less often, bacteria spread to the kidney from the bloodstream. Urinary tract infection is commonly treated with prescription antibiotics however this approach has some drawbacks. Due to the frequency of antibiotic use for UTI's bacteria develops resistance to the antibiotics, making them less and less effective. Certain herbal remedies relieve urinary tract infections by combating the bacteria, decreasing irritation and healing urinary tract tissues. Some herbs also help prevent future occurrences<sup>1</sup>.

The Horsetail plant is one of the best herbal remedies for UTI. Horsetail has astringent, diuretic, and tissue healing properties that allow it to effectively fight a urinary tract infection. Horsetail has been used for ages by the ancient Romans, Greeks, and native North Americans for curing kidney stones and bladder problems. It was also used for treating prostatitis, urinary incontinence and gonorrhoea in the early 19th century. According to a study, having horsetail tea three times a day has helped people suffering from uric acid kidney stones and urinary tract infection (UTI)<sup>1,2</sup>. Because of horsetail's diuretic activity, the patients' urine discharge increased, enabling them to flush out the kidney stones, also relieving them of the UTI symptoms.

*Equisetum arvense* [family : Equisetaceae] commonly known as the Field Horsetail or Common Horsetail, or Bottle brush is a well known herb; it is a perennial herb growing in moist loamy or sandy soil found in much of the North American continent, as well as in similar climates in Europe and Asia. The morphology of the horsetail herb is very strange and the plant has a creeping, or string like rootstock which gives it its name. The roots at the nodes are turned into numerous hollow stems of two kinds. Horsetail begins growth in two stages, initial growth of the plant is through a fertile and flesh colored stem, this stem can grow to a height of four to seven inches and comes out a cone like spike - this spike contains spores of the plant. The initial stem does not last long and withers away. The second stem is a green and sterile structure reaching a length of eighteen inches in height and crowned by whorls of small branches this is the final shape that the plant will take for its life span<sup>3-5</sup>. Active Compounds of the plant include minerals like silicic acids and silicates, potassium, sulphur, manganese, magnesium; flavonoids: quercetin glycosides; phenolic

acids, alkaloids, equisetonin, phytosterols: cholesterol, isofucosterol, campesterol; tannins<sup>6,7</sup>.

Horsetail possesses diuretic properties, which are believed to be due to equisetonin and flavone glycosides<sup>8</sup>. It is a strong astringent and therefore is used to heal wounds, bleeding gums, sore throat, mouth sores and applied as poultice to sprains or bruises. Important compound found in horsetail plant called silica is proven to promote new hair growth by strengthening hair follicles and increasing blood circulation in the scalp area. Osteoporosis is one among many diseases that horsetail extract benefits<sup>9</sup>. Horsetail herb extract helps body retain calcium more efficiently due to a silica compound and can even help repair bones and cartilage. This is certainly essential for managing joint degeneration conditions or hard to heal bone fractures. Horsetail is known for its anti-inflammatory<sup>10</sup>, anti nociceptive<sup>10</sup>, antioxidant and anti proliferative<sup>11</sup>, antimicrobial<sup>12,13,14</sup>, hepatoprotective<sup>15</sup>, anti diabetic<sup>16</sup>, coagulant and astringent activity

### MATERIALS AND METHODS

#### Plant material

The ethanolic and aqueous extract of *Equisetum arvense linn* was obtained from Green Chem Herbal Extract & Formulations, Bangalore.

#### Test microorganisms

Bacterial strains used were *E.coli* [Gram negative bacilli-GNB], *Klebsiella pneumoniae*[GNB], *Proteus mirabilis*[GNB], *Pseudomonas aeruginosa*[GNB], *Staphylococcus aureus*[Gram positive cocci-GPC], *Staphylococcus saprophyticus*[GPC] and *Enterococcus faecalis*[GPC]. The organisms were obtained from department of Microbiology, Saveetha Dental College and maintained in nutrient agar slope at 4° C.

#### Methodology

The extracts were prepared in the following concentrations in sterile water. 5mg/ml and 10mg/ml and 20mg/ml. 50µl of extract of different concentrations were loaded on sterile filter paper discs measuring 6mm in diameter, so that the concentration of the extract on each disc was 250µg, 500 µg and 1000ug respectively. The discs were dried and kept aseptically.

#### Screening of antibacterial activity [disc diffusion technique]<sup>17</sup>

Broth culture of the bacterial strains compared to Mac Farland's standard 0.5<sup>18-21</sup> were prepared. Lawn culture of the test organisms were made on the Muller Hinton agar [MHA-Hi media M1084] plates using sterile cotton swab and the plates were dried for 15 minutes.

Filter paper discs loaded with different concentrations of the extract were placed on the respective plates. The plates were incubated at 37°C overnight and the zone of inhibition of growth was measured in millimeter diameter<sup>22</sup>. Standard antibiotic discs of Amoxicillin (30mcg/disc) and Ciprofloxacin (30mcg/disc) were used as positive control. All the tests were done in triplicate to minimize the test error.

## RESULT AND DISCUSSION

The antibacterial activity of the extracts (Ethanolic and Aqueous) at different concentrations was screened by disc diffusion technique and the zone of inhibition was measured in mm diameter. The results are given in the table 1.

Both the extracts at different concentration exhibited antibacterial activity against all bacterial strains tested. Ethanolic extract exhibited comparably a high degree of activity than the aqueous extract. The ethanolic extract was more effective against *E.coli*, *Proteus mirabilis* and *Staphylococcus saprophyticus* with a zone of inhibition of 24mm, 23mm and 24 mm diameter (at conc.1000µg.) respectively and was least effective against *Pseudomonas aeruginosa* with zone of inhibition of 11mm (at conc. 1000 µg). Among the other bacterial species studied *Klebsiella pneumoniae* and *Enterococcus faecalis* showed a zone of inhibition of 18mm diameter (at conc. 1000 µg.) and *Staphylococcus aureus* showed inhibition zone of 14mm diameter (at conc.1000 µg).

**Table 1: Anti bacterial activity of Ethanolic and Aqueous Extract of *Equisetum arvense* Linn**

Extract	Conc [µg]	Zone of inhibition [in mm diameter]						
		B1	B2	B3	B4	B5	B6	B7
Ethanolic	250	14	13	9	7	9	12	10
	500	19	17	13	9	11	19	14
	1000	24	23	18	11	14	24	18
	250	9	9	7	-	8	10	9
	500	13	12	10	7	10	16	11
	1000	18	16	14	10	12	20	16
Ciprofloxacin	30mcg/disc	24	21	22	22	23	23	24
Amoxycillin	30mcg/disc	25	23	20	24	25	25	22

B1- *E.coli* , B2- *Proteus mirabilis*, B3- *Klebsiella pneumonia*, B4- *Pseudomonas aeruginosa*, B5 *Staphylococcus aureus*, B6- *Staphylococcus saprophyticus*, B7- *Enterococcus faecalis*.

Urinary tract infections are a serious health problem affecting millions of people each year. It is the second most common infection after respiratory infection. The urinary system consists of the kidneys, ureters, bladder, and urethra. Urinary Tract Infections (UTI) could be of the lower urinary tract encompassing the bladder and urethra or of the upper urinary tract infecting the ureters and kidneys. All areas of the urinary tract above the urethra in healthy humans are sterile, hence urine is normally sterile. Urinary Tract Infections (UTI's) usually occur when bacteria enter the opening of the urethra and multiply in the urinary tract. Although it is much more common in women, because of shorter urethra, it can occur in men also, and can be quite severe. Urinary tract infection is commonly treated with prescription antibiotics however this approach has some drawbacks. Due to the frequency of antibiotic use for UTI's and other infections, bacteria develop resistance to the antibiotics, making them less and less effective. Antibiotics can also affect the immune system, making more prone to more infections.

Herbal therapy for UTI can provide an effective alternative to prescription medications and their side effects. Natural remedies have been used in traditional medicine for thousands of years to support the health of the urinary system. Modern clinical research now supports the use and effectiveness of specific herbs for various health issues. The present study was to evaluate the antibacterial activity of *Equisetum arvense* against bacteria associated with urinary tract infections. The results obtained from our study shows that ethanolic extract has got a very good antibacterial activity against the selected urinary tract pathogens.

## CONCLUSION

The present results therefore offer a scientific basis for traditional use of *Equisetum arvense* linn on urinary pathogens. The use of herbs in folk medicine suggests that they represent an economic and safe alternative to treat infectious diseases. It is clear from the results that, the extract acts as a good source of antimicrobial agent against various bacterial pathogens tested and exhibited broad spectrum of antibacterial activity. The anti-bacterial activities could be enhanced if active components are purified and adequate dosage determined for proper administration.

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