



## ANTIBACTERIAL ACTIVITY OF *ZALEYA GOVINDIA* AGAINST SOME PATHOGENIC MICROBES OF NORTHERN RAJASTHAN, INDIA

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### ABSTRACT

The in vitro antimicrobial activity of crude ethanolic extracts of the plant parts (leaf, root & stem) of *Zaleya govindia* was investigated. For antibacterial activities test, the extracts were subjected to its effectiveness against both Gram (+ve) and Gram (-ve) bacteria in disk diffusion method. The opportunistic bacterial strains are *Escherichia coli*, *Pseudomonas aeruginosa*, *Streptococcus pyogenes*, *Shigella dysenteriae*, *Vibrio cholerae*, *Bacillus subtilis*, *Salmonella typhi*, *Staphylococcus aureus*, *Klebsiella pneumoniae* & *Agrobacterium rhizogenes*. The zones of inhibition produced by the crude ethanol extracts against few sensitive strains were measured and compared with those of standard antibiotics Streptomycin, Ampicillin, Gentamycin & Tetracycline. The extracts produced greatest inhibitory zone on *S. aureus* one of the major wound infectious bacterial strain. The obtained results provide a support for the use of this plant in traditional medicine as in conjunctivitis, menstrual regulation in female and to prevent wound infections suggest its further investigation.

**Keyword:** Antibacterial, *Zaleya govindia*, Extract, Gram positive, Gram negative, Disk Diffusion method.

### INTRODUCTION

Historically, plants have provided a source of inspiration for novel drug compounds, as plant derived medicines have made large contributions to human health and well-being. Their role is twofold in the development of new drugs: they may become the base for the development of a medicine, a natural blue print for the development of new drugs or; a phyto-medicine to be used for the treatment of diseases.<sup>1</sup> Herbs are staging a comeback and herbal products today symbolize safety in contrast to the synthetics. Moreover, plants used in traditional medicine contain a vast array of substances that can be used to treat chronic and infectious diseases.<sup>2</sup> Antibiotics have been used to treat infectious diseases. Incessant and indiscriminate use of antibiotics has brought in untold misery of antibiotics resistance in many human pathogenic bacteria.<sup>(3,4)</sup> *Zaleya govindia* is a local perennial herb belonging to the family *Aizoaceae* which prefers hard open grounds. These plants grow in abundance in Rajasthan, Punjab and Uttar Pradesh (Distributed in India). Different parts of the plant have been used in Indian Traditional system of medicine for the treatment of conjunctivitis and problems in female to regularize menstruation.<sup>5</sup>

Today plants are the almost exclusive source of drugs for the majority of the world population. People in developing countries utilize traditional medicine for their primary health care needs.<sup>6</sup> Traditional medicine using plant extracts continues to provide health coverage for over 80% of the world's population, especially in the developing world.<sup>7</sup> It has been documented that biological active components such as tannins, saponins and alkaloids are plant metabolites well known for antimicrobial activity. This could be enhanced due to stronger extraction capacity of ethanol.<sup>8</sup> Considering the vast potentiality of plant as a source of new therapeutic agents, higher plants are routinely screened for antimicrobial property in laboratories.<sup>9,10</sup>

Keeping this in view, the present investigation was initiated to find an alternative source of antibacterial compounds to avoid undesirable effect of synthetic drugs. In this study the ethanol extract from *Zaleya govindia* was tested against some pathogens.

### MATERIAL AND METHODS

#### Study Site

The plant material was collected from MD College Sriganganagar, Rajasthan for the evaluation of its antimicrobial effects. The plant parts (root, stem & leaf) were washed under running tap water and dried in hot air oven at 40-50°C. After that the dried plant material was grinded into a fine powder with the help of a suitable grinder. About 30gm of powdered material was extracted by soxhlet apparatus with 200ml 50% ethanol at 20-25°C temperature. The extract thus obtained was concentrated using a vacuum evaporator. The concentrate extracts of various parts of plant were kept in airtight bottles at 4°C in refrigerator for further use.

#### Bacterial Culture

*Escherichia coli*, *Pseudomonas aeruginosa*, *Streptococcus pyogenes*, *Shigella dysenteriae*, *Vibrio cholerae*, *Bacillus subtilis*, *Salmonella typhi*, *Staphylococcus aureus*, *Klebsiella pneumoniae* & *Agrobacterium rhizogenes* were obtained from Department of Microbiology, MD College, Sriganganagar and Rajasthan, India. All test strains were maintained on Nutrient agar slopes (Hi-Media Laboratories Pvt. Ltd. Mumbai) at 37°C & were sub-cultured every two weeks.

#### Testing Antibacterial Activity

Antibacterial tests were carried out using the Disk diffusion method.<sup>11</sup> Nutrient agar media was prepared by adding water to the dehydrated product that contains all the ingredients. Practically all media are available commercially in powdered form.<sup>12</sup> Bacterial inoculums 0.1ml inoculated on solid Nutrient agar media in petriplates. The bacterial inoculum was spreaded by glass spreader until it absorbed fully in agar layer for the development of uniform bacterial growth. Discs of 5mm diameters were made from Whatman filter paper no. 1 with the help of punching machine. The paper discs were dipped in plant extracts, taken aseptically and kept in center of spreaded microorganism cultured petriplate. Standard Antibiotic discs were also kept in same way. The plates were then incubated at 37°C for overnight. After proper incubation inhibition zones were measured by Antibiotic zone scale (PW297, Hi-Media) and expressed in millimeter. Subsequently, the plates were examined for zone of inhibition

and diameter was measured in mm after subtracting disk diameter.<sup>13</sup>

**Table 1: Antibacterial activity of plants**

Microorganisms	50% ethanol	Leaf (mm)	Stem (mm)	Root (mm)	Streptomycin (10 mcg)	Ampicillin (10 mcg)	Gentamycin (10 mcg)	Tetracyclin (30 mcg)
<i>E. coli</i>	0±0	21.3±.88	26.6±.90	29±.57	18.3±.95	21±.52	19±1.15	-----
<i>P. aeruginosa</i>	0±0	13±1.15	9.3±.57	15.6±.72	-----	-----	27±1.10	22±1.20
<i>S. pyogenes</i>	0±0	12.3±1.45	14.6±1.10	17±.52	21.6±.92	18.3±1.15	23.3±.66	22.3±.88
<i>S. dysenteriae</i>	0±0	22±.53	20.6±1.20	35.6±1.45	23.6±.88	2.3±.33	24.3±.57	21.6±1.20
<i>V. cholerae</i>	0±0	16.6±1.20	11.6±.56	20.6±.72	17.6±.56	13.3±.57	28.6±1.20	25.6±1.20
<i>B. subtilis</i>	0±0	18.3±.33	25.3±.95	27.6±.66	16.6±.38	25.6±1.20	24±.57	23.3±1.45
<i>S. typhi</i>	0±0	22.6±1.22	23.3±.66	32.6±1.15	21±1.55	29±.90	26±.88	25±.57
<i>S. aureus</i>	0±0	35.6±.33	32.66±.91	36.6±1.45	21.3±.92	33.3±.66	25.3±.33	29±.76
<i>K.pneumoniae</i>	0±0	10.3±1.33	11±.85	21±1.15	17.3±.88	17.5±1.45	20±1.76	24±1.52
<i>A. rhizogenes</i>	0±0	13.3±.91	14.3±.66	23.6±.89	16.3±1.15	11.3±1.20	24.6±.1.45	23.6±1.10

Result as per shown in Mean±S.E; ----- No inhibition

## RESULTS AND DISCUSSION

The screening of selected plant extracts were done against 10 bacterial species using disk diffusion method. The zone of inhibition greater than 5 mm diameter is found to be having significant against particular bacteria.<sup>14</sup> All three extracts of the plant tested show varying degree of antibacterial activities against the test bacterial species (Table 1). The antibacterial activities of the ethanol extract of various parts of plant compared favorably with that of four standard antibiotics (Streptomycin, Ampicillin, Gentamycin and Tetracycline) and have appeared to be broad spectrum as its activities were independent on gram reaction. The non-activity of the ethanol extract against most bacterial strains investigated in this study is in agreement with previous works which show that aqueous extracts of plant generally showed little or no antibacterial activities.<sup>15-18</sup>

The highest zone of inhibition was shown by root extract against *S. aureus* (36.6mm), *S. dysenteriae* (35.6 mm), *S. typhi* (32.6 mm) and *E. coli* (29 mm). The growth inhibition was moderately active against *B. subtilis* (27.6 mm), *A. rhizogenes* (23.6 mm), *K. pneumoniae* (21 mm) and *V. cholerae* (20.6 mm). The least active inhibition was reported in *P. aeruginosa* (15.6 mm). This zone was much less as in other extract also to bacteria. This bacterium showed no inhibition against Streptomycin and Ampicillin. *S. aureus* is the most common species found in purulent wound.<sup>19</sup> (Sue et al 2004). This bacterial species is found to be more effective in all extracts and showed maximum inhibition zone (33.3 mm) in all antibiotics and especially against Ampicillin. *K. pneumoniae* develops conjunctivitis in eyes. This plant extract also showed antibacterial activity against this. *S. dysenteriae* common in food borne disease showed least inhibition zone against Ampicillin (2.3 mm); Tetracycline (21.6 mm) and showed highest against Streptomycin (23.6 mm). Out of these antibacterial drugs Ampicillin was very least efficient drug against *S. pyogenes*. Lowest inhibition of Streptomycin was performed by *A. rhizogenes*. This bacterium can cause opportunistic infections humans with weakened immune system.<sup>20,21</sup>

The inhibitory effect of extracts of *Z. govindia* against pathogenic bacterial strains can introduce the plant as a potential candidate for drug development for the treatment of ailments caused by these pathogens.

## CONCLUSION

The present study has shown a successful approach in the direction of new antibacterial drug discovery from plant origin. It has revalidated that *Z. govindia* act as a remedy for different microbial diseases traditionally including

Conjunctivitis, prevent wound infections and to regularize menstruation in females.

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