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### CHEMICAL CONSTITUENTS AND ANTIMICROBIAL ACTIVITIES OF CERTAIN PLANT PARTS OF *SAUROPUS ANDROGYNUS L*

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#### ABSTRACT

The antibacterial activity of leaves of *Sauropus androgynus* was evaluated on bacteria like *Klebsiella pneumoniae*, *Bacillus cereus*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Escherichia coli* and *Staphylococcus aureus*. The solvents used for the extraction of plants were Petroleum ether, methanol, ethanol, and water. The *in vitro* antibacterial activity was determined by the disc diffusion method. Methanol and ethanol leaf extracts were more effective against pathogenic organisms, than other solvent extracts.



## KEY WORDS

Antibacterial activity, *Sauropus androgynus*, *In vitro*, leaves.

## INTRODUCTION

Many infectious diseases are treated with herbal remedies throughout the history of mankind. Natural products, either as pure compounds or as standardized plant extracts, provide unlimited opportunities for new drug leads, because of the unmatched availability of chemical diversity. There is a continuous and urgent need to discover new antimicrobial compounds with diverse chemical structures and novel mechanisms of action for new and re-emerging infectious diseases.<sup>1</sup> Researchers are increasingly turning their attention to folk medicine, looking for new leads to develop better drugs against microbial infections.<sup>2</sup> In the recent years, secondary plant metabolites are used for the treatment of bacterial infections.<sup>3</sup>

*Sauropus androgynus* L. is commonly known as star goose berry or multivitamin plant, of the family Euphorbiaceae. The plant grows rapidly in hot humid conditions. The species is highly mycorrhizal-dependent, is adapted to acid heavy clay soils. *S androgynus plants have* a high level of vitamin A (carotenoids), especially in freshly picked leaves, as well as high levels of vitamins B and C, protein and minerals.<sup>4</sup> Nutrient content of the leaves is usually higher in more mature leaves. A lignin diglycoside, (-) - isolariciresinol 3 $\alpha$ -O- $\beta$ -apifuranosyl- (1 $\rightarrow$ 2) - O -  $\beta$ -glucopyranoside, and a megastigmane glucoside, sauroposide and papaverine were isolated from the leaves of *Sauropus androgynus*. The aim of the study was to evaluate the leaf extracts of *Sauropus androgynus* L. against several gram positive and gram negative bacteria.

## MATERIALS AND METHODS

*Sauropus androgynus* plants were collected from the gardens of the Presidency

college, Hebbal, Bangalore, Karnataka, India, which were in turn brought from University of Agricultural Sciences, Bangalore. Leaves, stems and roots of these plants were collected, shade dried and powdered, which are used as materials.

### (i) **Preparation of Crude Extracts:**

The powdered plant material (10g) was extracted separately by soaking in, methanol, ethanol, petroleum ether and water for 5days. Each mixture was stirred every 12 hrs using a sterile glass rod. Each extract was passed through a Whatman No.1 filter paper. The filtrate obtained was concentrated in vacuum using a rotary evaporator.

### (ii) **Preliminary Phytochemical Analysis:**

Qualitative phytochemical analysis of the crude extracts of different solvents were carried out using standard methods.<sup>5</sup> Test for the presence of phenolics, tannins, lignins, alkaloids, steroids, saponins, glycosides and triterpenes were carried by standard tests.<sup>5</sup>

### (iii) **Pathogenic bacteria tested for antibacterial activity:**

*Klebsiella pneumoniae* : These are gram negative, non-motile entero-bacteria present in the intestines of humans and animals. It causes urinary tract infections, bronchial and lobar pneumonia wherein part of lung is infected. A marked patchy exudation of lung parenchyma is noticed in broncho-pneumonia.<sup>6</sup>

*Escherichia coli* : These are gram negative, rod shaped bacteria living as an endoparasite in human and animal intestines (enteropathogenic). It causes enteric diseases, haemorrhagic colitis (bloody haemorrhage),



urinary tract infections, pyogenic infections and septicemia.<sup>7</sup>

*Bacillus cereus* : These are gram positive, sporogenous rod shaped bacteria dwelling in water, dust, air and soil. *Bacillus* species other than *Anthrax bacillus* have been implicated in serious infections associated with immunosuppression, and food poisoning. Medical reports and reviews describe cases of bacteremia, meningitis, meningoenzephalitis, pneumonia, endocarditis, urinary tract infections, peritonitis, pleuritis and ocular infections of *Bacillus* species.<sup>8</sup>

*Proteus vulgaris* : These are motile, gram negative, enterobacteria, present in alimentary canal of humans and animals. It invariably causes urinary tract infections.

*Pseudomonas aeruginosa* : These are gram negative, aerobic, non-sporic, motile bacillus present in soil, water, sewage, the mammalian gut and plants. It causes nosocomial infections including metabolic, haematological and malignant diseases. Severe epidemics like diarrhea of infants, ocular infections, urine infections, cystic fibrosis, hot tub and whirlpool-associated folliculitis and osteomyelitis are caused by *P. aeruginosa*. The patients become susceptible to this organism after prolonged treatment with immunosuppressive agents, corticosteroids, antimetabolites, antibiotics and radiation.<sup>9</sup>

*Staphylococcus aureus*: These are gram positive bacteria that cause many skin infections.

#### **Growth media:**

The media used for antibacterial test was Nutrient agar, Nutrient Broth (Himedia Pvt. Ltd. Mumbai, India). Cultures of bacteria were

inoculated into Nutrient Broth (liquid medium) and incubated for 4 hrs and the suspension was checked to provide approximately 10,00,00 CFU/ml.

#### **(iv) Determination of antibacterial activity:**

Susceptibility of the test organisms to all the extracts was determined by employing the standard disc diffusion technique.<sup>10,11</sup> The low cost and simple technique of disc diffusion bioassay is advantageous in the determination of the antimicrobial activity of crude drugs.<sup>12</sup> Whatmann No. 1 filter paper discs of 6 mm diameter, placed in dry Petri dish were autoclaved. The sterile filter paper discs were impregnated with each of the test extracts and shaken thoroughly and kept overnight. Later, the saturated filter paper discs were taken out and dried on the laminar air flow bench and carefully placed over the spread cultures and incubated at 37°C for 24 h for enabling bacterial growth. Paper discs treated with DMF (negative control) and Streptomycin (positive control) served as the control. For each bacterial culture, a control was maintained wherein pure solvents were used instead of the extract. The result was obtained by measuring the zone of the inhibition in centimeters (cm). Each experiment was triplicated and the mean values tabulated.

## **RESULTS AND DISCUSSION**

Phytochemical analysis of *Sauropus androgynus L* revealed the presence of alkaloids, tannins and saponins. The secondary metabolites like flavonoid, steroids etc., were also present in trace amounts (Table.1).



**Table 1**  
**Preliminary screening of secondary metabolites in *Sauropus androgynus L.***

Sl. No.	Tests	PE extract		Methanol extract		Ethanol extract		Aqueous extract	
		S	L	S	L	S	L	S	L
1	<b>Tests for Phenolics</b>								
	a) Phenol test.								
	b) Ellagic acid test.	+	+	+	+	-	-	-	+
	c) Hot water test.	-	-	-	-	-	+	+	-
		-	-	+	+	+	+	+	+
2	<b>Tests for Tannins</b>								
	Sodium chloride test.	-	-	+	+	+	+	+	-
3	<b>Tests for Alkaloids</b>								
	Mayer's reagent test.	+	-	+	+	+	+	-	-
4	<b>Tests for Steroids</b>								
	Salkowski test.	-	-	-	+	-	-	-	-
5	<b>Tests for Saponins</b>								
	Foam test.	-	-	-	-	+	-	+	-
6	<b>Tests for Glycosides</b>								
	Keller-Kiliani test.	-	-	-	+	-	+	-	++
7	<b>Test for Triterpenoids</b>								
	Salkowski test.	+	-	-	-	+	+	-	+

PE: Petroleum Ether; L: Leaf, S: Stem; +: Present; - : Absent

The tannins found in the phytochemical analysis of the extracts may be responsible for the antibacterial activity. Tannins have astringent actions which form the basis for their therapeutic applications, it was found that tannins possess antimicrobial activity.<sup>13,14</sup> The antibacterial activities of *Sauropus androgynus*

*L.* root, stem and leaf extracts against pathogenic bacteria and their potency were assessed by the presence or absence of inhibition zones and the zone diameter. The zones of inhibition diameter (cm) recorded for methanol, ethanol, petroleum ether and aqueous leaf extracts are depicted in Table-2.



**TABLE 2**  
**Antibacterial activity of *Sauropus androgynus L.***

Microorganisms	ME			EE			PE			Aq-E			Strepto mycin
	I	II	III	I	II	III	I	II	III	I	II	III	
<i>Pseudomonas aeruginosa</i>	0.5 ± 0.335	0.3 ± 0.537	0.4 ± 0.494	0.6 ± 0.509	1.0 ± 0.396	0.6 ± 0.427	0.5 ± 0.513	0.9 ± 0.447	0.5 ± 0.473	0.3 ± 0.427	0.3 ± 0.413	0.2 ± 0.296	1.6 ± 0.524
<i>Staphylococcus aureus</i>	1.3 ± 0.423	0.6 ± 0.440	0.5 ± 0.484	1.2 ± 0.352	1.2 ± 0.544	0.5 ± 0.479	0.5 ± 0.536	0.6 ± 0.414	1.0 ± 0.563	0.3 ± 0.467	0.2 ± 0.443	0.5 ± 0.502	1.6 ± 0.492
<i>E.coli</i>	0.4 ± 0.423	0.5 ± 0.386	0.7 ± 0.528	0.9 ± 0.372	0.9 ± 0.414	0.6 ± 0.447	0.8 ± 0.414	1.2 ± 0.297	0.5 ± 0.492	0.5 ± 0.488	0.4 ± 0.324	0.5 ± 0.372	1.7 ± 0.324
<i>Proteus vulgaris</i>	0.7 ± 0.324	0.3 ± 0.455	0.5 ± 0.372	1.0 ± 0.528	1.1 ± 0.528	0.8 ± 0.372	0.6 ± 0.537	0.7 ± 0.296	0.8 ± 0.484	0.7 ± 0.438	0.4 ± 0.517	0.6 ± 0.473	1.7 ± 0.413
<i>Bacillus subtilis</i>	1.2 ± 0.434	0.5 ± 0.386	0.3 ± 0.528	1.3 ± 0.372	0.9 ± 0.414	0.7 ± 0.447	0.8 ± 0.414	0.8 ± 0.297	0.5 ± 0.492	0.6 ± 0.488	0.6 ± 0.324	0.4 ± 0.372	1.7 ± 0.324

I = Leaf, II = stem & III = root

Streptomycin = + ve control for bacteria Data represents an average of 3 experimental sets

This study of the extracts from the medicinal plants showed inhibition of growth of some of the test microorganisms to various degrees. It revealed that the methanolic and ethanolic extract exhibited significant inhibition against *Proteus vulgaris*, *Bacillus cereus* and *Staphylococcus aureus*, and less inhibition was associated with *Klebsiella pneumoniae*, *E.coli* and *Pseudomonas aeruginosa*. The aqueous extract of the leaf showed moderate antibacterial activity on the test organisms compared to the solvent extracts. Leaf extract was more effective than root and stem extract. Generally more antibacterial activity was shown

against gram positive than gram negative bacteria.<sup>15</sup> The effectiveness may be due to the cumulative action(s) of different compounds present in the plant parts.<sup>16</sup> Plant based antimicrobials have enormous therapeutic potential as they can serve the purpose with lesser side effects than that are often associated with synthetic antimicrobials.<sup>17</sup> From the above results it can be concluded that plant extracts have great potential as antimicrobial compounds against microorganisms and that they can be used in the treatment of infectious diseases caused by resistant microorganisms.

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