Research Article

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Evaluation of phytochemical constituents and *In-vitro* antibacterial activity of *Synadium grantii*

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Abstract

In this study we investigated phytochemical screening and *In-vitro* antibacterial activity of hexane, ethyl acetate, ethanol (70%v/v) and methanol extracts of *Synadium grantii*. The results were revealed the presence of steroids, terpinoides, glycosides, tannins, alkaloids, phenols and carbohydrates but the all extracts are do not contain saponins, flavanoids, quinines, amino acids and oils.. The selected plant extracts were produced concentration dependent zone of inhibition against tested bacterial strains. The extracts showed more activity against gram negative organisms than gram positive organisms. The extracts showed better activity at highest concentrations i.e. at 600 and 1200 µg/100µl. Among the four types of *Synadium grantii* extracts, the ethyl acetate extract showed better activity than remaining extracts at 1200 µg concentrations against gram negative bacteria.

Key-Words: *Synadium grantii*, Phytochemical screening, *In-vitro* Antibacterial activity.

Introduction

Medicinal plants are an important therapeutic aid for various ailments and those are a source for generating novel drug compounds for curing many diseases. Scientific experiments on the antimicrobial properties of plant components were first documented in the late 19th century [1]. Now, Antimicrobial drugs have caused a dramatic change not only of the treatment of infectious diseases but of a fate of mankind. Antimicrobial chemotherapy made remarkable advances, resulting in the overly optimistic view that infectious diseases would be conquered in the near future. However, in reality, emerging and re-emerging infectious diseases have left us facing a countercharge from infections. Infections with drug resistant organisms remain an important problem in clinical practice that is difficult to solve. The shortcomings of the drugs available today, propel the discovery of new pharmacotherapeutic agents in medicinal plants[2]. Accordingly, in the present investigation, we tried to determine the antimicrobial activity of *Synadium grantii* in hexane, ethyl acetate, ethanol (70%v/v) and methanol and these extracts were found to be potent antibacterial activity.

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Synadium grantii is a Succulent shrub with milky latex; leaves alternate, simple, and fleshy; flowers small and inconspicuous, in a small cup with a red rim of glands belongs to the family Euphorbiaceae.

Material and methods

Chemicals

Muller Hinton agar media was purchased from Sisco Research Laboratories Pvt Ltd., Mumbai. All the chemicals and reagents used were of analytical grade.

Test Organisms

Eight bacterial species were used. The bacterial species were purchased from National collection of industrial micro organisms (NCIM), Pune. The Bacterial species were maintained in the nutrient broth medium on placing shaker in separate culture tubes for each species separately. Out of eight, four are Gram positive Organisms *(Bacillus pumillis, Bacillus megaterium, Staphylococcus epidermidis, Streptococcus pneumoniae)* four are Gram Negative *(Escherichia coli, Pseudomonas aeruginosa,, Klebsiella pneumonia, Salmonella typhimurium,)*

Culture Media

For Anti bacterial activity Muller-Hinton Agar media (Solid and Broth) was used. For maintaining the bacterial species Nutrient both was used.

Preparation of Extracts

The plant material used in present study was collected from Visakhapatnam, Andhra Pradesh and

authenticated by the taxonomist Dr. Prayaga Murthy Pragada, Depart of Botany, Andhra University. Freshly collected plant material was dried under shade and the dried material was milled to obtain a coarse powder. The powdered material was separately extracted in a Soxhlet apparatus for 6 hrs successively with hexane, ethyl acetate, Hydro-alcoholic (ethanol 70%v/v) and methanol was concentrated to dryness under vacuum by using Rota-vapor.

Phytochemical Analysis

Phytochemical studies were carried out for hexane, ethyl acetate, hydro alcoholic and methanol extracts of *Synadium grantii* to detect the presence of different phytochemical constituents like steroids, terpinoides, tannins, flavanoids, saponins, glycosides, amino acids etc by using standard procedures [3-5].

Antibacterial Activity

The cylinder plate assay of drug potency is based on measurement of the diameter of zone of inhibition of microbial growth surrounding cylinders (cups), containing various dilutions of test compounds (extracts) [6]. The dilutions were prepared at a concentration of 150, 300, 600, 1200 µg/100µl. A sterile borer was used to prepare the cups of 4 mm diameter in the agar medium spread with the micro-organisms and 0.1 ml of inoculums. These cups were spread on the agar plate by spread plate technique. Accurately measured (0.05 ml) solution of each concentration and reference standards were added to the cups with a micropipette. All the plates were kept in a refrigerator at 2 to 8°C for a period of 2 hours for effective diffusion of test compounds and standards. Later, they were incubated at 37°C for 24 hours. The presence of definite zone of inhibition of any size around the cup indicated antibacterial activity.

Results and Discussion

Phytochemical screening

Qualitative phytochemical screening different extracts of *Synadium grantii* revealed the presence of steroids, terpenoids, glycosides, tannins, alkaloids, phenols and carbohydrates but the all extracts are do not contain saponins, flavonoids, quinines, amino acids and oils. The results of phyto chemical screening were showed in the Table 1.

Evaluation of antibacterial activity

Among all the tested *Synadium grantii* extracts ethyl acetate and methanol extracts have shown significant anti bacterial activity as compared to that of hexane, 70% ethanol and standard drug. The extracts showed good zone of inhibition against gram negative bacteria than gram positive bacteria. Hexane and ethyl acetate extracts of *Synadium grantii* had not produced zone of inhibition against some tested bacterial species at a dose of 150 µg/100µl. Ethyl acetate and Methanolic extracts showed good zone of inhibition at a dose of 600 and 1200 µg/100µl compared to other extracts. The 70% ethanol extract showed minimum zone of inhibition. The highest zone of inhibition was shwoed by ethyl acetate extract against *E.coli* and *Pseudomonas aeruginosa* at the dose of 1200 µg/100µl. further study will be carrying out to isolate the active compounds from the extracts through column chromatography. The results of Antibacterial activity were showed in Table 2.

The data clearly indicated that the hexane, ethyl acetate, hydro-alcoholic and methanol extracts of *Synadium grantii* showed good antibacterial activity. Among the all the ethyl acetate extract showed better activity and highest zone of inhibition on *Pseudomonas aeruginosa* at a concentration of 1200 µg/100µl i.e. 16mm.

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Table 1: Phytochemical constituents present in different extracts of *Synadium grantii*

Name of the Phytochemicals	Extracts of *Synadium grantii*			
	Hexane extract	Ethyl acetate extract	Methanol extract	Ethanol (70%)
Phenols	+	+	+	+
Phytosterols	+	+	+	+
Terpenoids	+	+	+	+
Glycosides	+	+	+	+
Saponins	-	-	-	-
Flavonoids	-	-	-	-
Tannins	-	+	+	+
Carbohydrates	+	+	+	+
Alkaloids	+	+	+	+
Amino acids	-	-	-	-
Oils	-	-	-	-
Quinones	-	-	-	-

+=Present, – = Absent

Table 2: Antibacterial activity of *Synadium grantii* extracts

Name of the *Synadium grantii* Extract	dose (µg/cup)	zone of inhibition# (diameter in mm)							
		gram +ve				gram -ve			
		B.p.	*B.m*	*S.e*	*S.p*	*E.c*	*P.a*	*K.p*	*S.t*
Hexane	150	-	-	-	-	-	-	-	-
	300	-	-	-	5	-	-	-	-
	600	-	-	6	6	5	5	6	6
	1200	-	6	8	8	7	8	8	7
Ethyl acetate	150	-	-	-	-	6	7	5	-
	300	-	5	5	6	9	10	7	6
	600	-	7	6	8	12	13	10	8
	1200	-	10	8	10	15	16	12	10
Ethanol (70%)	150	-	-	-	-	-	6	5	-
	300	-	-	5	-	5	8	7	6
	600	-	6	8	7	7	10	9	8
	1200	-	9	10	10	10	13	12	11
Methanolic	150	-	-	5	-	-	-	-	6
	300	-	6	7	-	6	5	6	8
	600	-	7	10	7	8	7	8	10
	1200	-	10	12	9	11	10	11	12
Chloramphenicol	10	14	15	15	14	15	16	16	17
DMSO		-	-	-	-	-	-	-	-

B.a=Bacillus pumillis, B.m=Bacillus megaterium, S.m= Staphylococcus epidermidis, S.p=Streptococcus pneumonia; E.c=Escherichia coli, P.a= Pseudomonas aeruginosa,K.p= Klebsiella pneumonia, S.t=Salmonella typhimurium

-=No activity #Values are the average of triplicate; Includes the cup diameter (4mm)