Antibacterial activity of methanolic extract of *Cissus* quadrangularis Linn.

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Abstract

This study was carried out with an objective to investigate the antibacterial potential of methanolic extract of *Cissus quadrangularis* Linn. against ten nosocomial infection causing microorganisms. The preliminary phytochemical analyses of the plant extract were carried out. Antibacterial activity of the extract was done using the agar well diffusion method. The extract exhibited remarkable inhibition of the pathogenic bacterial growth in a dose dependent manner.

Nosocomial infections or Hospitalacquired infections occur worldwide affecting patients in both developed and developing countries. Emergence of antibiotic resistant pathogens, has become a major challenge to patient safety. So it became inevitable to find out an alternative source of drugs to control these pathogenic infections. Plant derived medicines on which 80% of world's population rely for primary health care needs is proved to be the best alternative^{1, 2}. Cissus quadrangularis Linn. (family Vitaceae) known as "Hadjod" or bone setter is used in folk medicine to cure fracture. In Avurveda, it is used for treatment of osteoporosis (weak bones), irregular menstruation, asthma and scurvy. Even the plant was used for upset stomach, hemorrhoids, dyspeptic, peptic ulcer disease (PUD), analgesic

in eye and ear diseases, cancer and in the management of obesity and complications related to metabolic syndrome^{4,9,12}. In this present study an attempt was made to evaluate *in vitro* antibacterial activity of methanolic extract of Cissus *quadrangularis* Linn. against ten nosocomial infection causing microorganisms using the agar well diffusion method.

The dried stems of *Cissus quadrangularis* were grounded into course powder. About 200g of powdered plant stem material was extracted two to three times with methanol at room temperature for 72 h with mass to volume ratio of 1:10 (g/ml). The solvent extracts were evaporated to dryness under vacuum on rotary evaporator between 37°C to 40°C. Extraction yield of the extract was calculated and was

stored at 4°C. Phytochemical analysis of the extract was done for the methanolic extract³. A collection of ten organisms including four Gram-positive (Bacillus subtilis, Methicillin resistant Staphylococcus aureus (MRSA), Staphylococcus epidermidis, Micrococcus luteus) and six Gram-negative organisms (Escherichia coli, Proteus mirabilis, Pseudomonas aeruginosa, Salmonella typhimurium, Klebsiella pneumoniae, and Enterobacter cloacae) used for this study were obtained from Microbiology laboratory of Global Hospital, Hyderabad. Evaluation of antibacterial activity was done following the agar-well diffusion method as reported by Perez et al.,¹⁰.

The methanolic extract yield was 4% and the preliminary phytochemical analysis of *Cissus quadrangularis* extract reported the presence of alkaloids, flavonoids, phenolic

compounds, carbohydrates, proteins, saponins, tannins, lignins and glycosides (Table-1). The extract proved to be very effective against gram negative bacteria compared to gram positive bacteria taken for the present antibacterial activity test. There are few earlier reports of antibacterial activity of this plant but the results differ when compared to the present study done on hospital collected samples. The plant extract showed remarkable activity against Proteus mirabilis, Salmonella typhi, and Micrococcus luteus with 22-24 mm zone of inhibition. While Klebsiella pneumonia, Pseudomonas aeruginosa, Escherichia coli (Fig.1) and Staphylococcus epidermidis where inhibited at 30 mg/ml and high concentration of extract (crude). MRSA and Bacillus subtilis were inhibited only by crude extract exhibiting 20 mm and 15 mm zone of inhibition respectively (Table-2).

 Table-1. Extract yield and phytochemical composition of Methanolic extract of Cissus quadrangularis

Phytochemical tests for compounds	Methanol extract		
Yield* (%)	4		
Colour and physical nature of extract	Green/sticky paste		
Alkaloids	+		
Flavonoids	+		
Phenolic compounds	+		
Steroids	-		
Carbohydrates	+		
Protein	+		
Reducing sugars	-		
Saponins	+		
Tannins	+		
Lignins	+		
Glycosides	+		

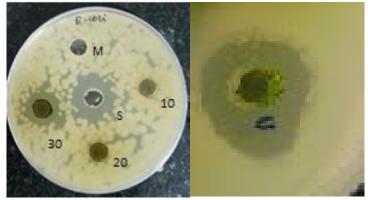
* yield calculated as the ratio of the mass of the obtained methanol extract/mass of the plant powder

+ Presence of constituent

- Absence of constituent



Fig 1. Antibacterial activity of methanolic plant extracts



S: Antibiotic standard; M: Methanol Control; C: Crude extract

	Table-2. Antibacterial activity of methanolic plant extracts						
S .	Micro-organism	Property	Zone of inhibition (diameter in mm)				
No.			Cissus	Cissus quadrangularis Methanolic plant			Antibiotic
			extract (mg/ml)				standard
			Crude	10	20	30	(10mg/ml)
1.	Proteus mirabilis	Gram	24.3±1.52	12.6±0.57	18.3±0.57	20.3±1.52	20.0±0.10
2.	Salmonella typhi	negative	24.6±0.57	12.3±1.15	15.6±0.57	18.3±0.57	12.0±0.10
3.	Enterobacter	bacteria	NIZ	NIZ	NIZ	NIZ	13.0±0.10
	cloacae						
4.	Klebsiella		22.3±1.52	NIZ	15.3±2.08	17.0±1.00	21.3±0.57
	pneumoniae						
5.	Pseudomonas		12.3±1.15	NIZ	NIZ	12.3±1.52	17.0±0.10
	aeruginosa						
6.	Escherichia coli		19.3±0.57	NIZ	NIZ	17.0±2.00	25.3±1.52
7.	Micrococcus	Gram	22.6±0.57	8.6±0.57	12±0.53	20±0.57	$18.0{\pm}1.00$
	luteus	positive					
8.	MRSA	bacteria	20.0±1.00	NIZ	NIZ	NIZ	16.0±1.00
9.	Bacillus subtilis		15.0±2.00	NIZ	NIZ	NIZ	28.0±1.00
10.	Staphylococcus		NIZ	NIZ	NIZ	15±0.57	11.3±1.15
	epidermidis						
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Table-2. Antibacterial	activity of me	thanolic plant	extracts

*Standard drug used: Streptomycin

Each value is Mean \pm Standard deviation (n=3)

NIZ : No inhibition zone

Luseba et al.⁶ reported antibacterial activity of the methanol and dichloromethane extract of Cissus quadrangularis against S. aureus, E. coli, and P. aeruginosa. Antimicrobial activity of leaves7 and root extracts of Cissus quadrangularis has also been reported⁸. Rao and Deshpande¹¹ also reported that C. quadrangularis alcoholic extract of the stem proved to be effective against E. coli. While Kashikar *et al.*,⁵ reported that *E. coli* did not respond to the different solvent extracts of this plant. The present study proves that Cissus quadrangularis have therapeutic efficacy and the methanolic extracts showed good antibacterial activity against nosocomial infection causing organisms. Further chemical characterization of active fraction of the extract against pathogenic microorganisms is under study.

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