



E-ISSN: 2278-4136  
P-ISSN: 2349-8196  
JPP 2014; 3(4): 83-85  
Received: 02-09-2014  
Accepted: 12-10-2014

**S. Sahoo**

University Department of  
Pharmaceutical Sciences, Utkal  
University, Vani Vihar,  
Bhubaneswar-751004, Odisha,  
India.

**B. K. Rout**

University Department of  
Pharmaceutical Sciences, Utkal  
University, Vani Vihar,  
Bhubaneswar-751004, Odisha,  
India.

**S. K. Mekap**

University Department of  
Pharmaceutical Sciences, Utkal  
University, Vani Vihar,  
Bhubaneswar-751004, Odisha,  
India.

**N. K. Dhal**

<sup>1</sup>Institute of Minerals and  
Materials Technology,  
Bhubaneswar – 751 013, Odisha,  
India.

**Correspondence:**

**S. Sahoo**

University Department of  
Pharmaceutical Sciences, Utkal  
University, Vani Vihar,  
Bhubaneswar-751004, Odisha,  
India.

## Antimicrobial activity of *Acalypha godseffiana* against selected urinary tract infection causing pathogens

S. Sahoo, B. K. Rout, S. K. Mekap and N. K. Dhal

### Abstract

The aerial parts of *Acalypha godseffiana* Mast, belonging to family Euphorbiaceae was investigated to evaluate *in vitro* antimicrobial activity of petroleum ether, chloroform, methanolic and aqueous extracts against urinary tract infection causing pathogens consisting of *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli*, *Klebsiella pneumonia*, *Proteus mirabilis*, *Candida albicans*, *Candida tropicalis*, *Candida krusei* and *Aspergillus niger*. The major UTI causing bacterial and fungal pathogens under investigation were tested by the disc diffusion assay method and the minimum inhibitory concentration was evaluated. Methanolic extract exhibited a significant and broader spectrum of inhibition followed by petroleum ether, aqueous and chloroform extract against bacterial pathogens. The results of antifungal activity revealed that aqueous and methanol extract inhibited fungal pathogens significantly with a broader spectrum of inhibition when compared to chloroform and petroleum ether extract. An attempt has been made to compare the activity of extracts with most potent standard antibiotics effective against selected UTI causing pathogens under test.

**Keywords:** *Acalypha godseffiana*, *in vitro* antimicrobial activity, disc diffusion assay, MIC, UTI

### 1. Introduction

*Acalypha godseffiana* Mast, belonging to family Euphorbiaceae [1, 2, 3] is a fast growing, evergreen, bushy shrub, with green leaf blades, creamy-white margins, pinkish tinge on young developing leaves, origin from pacific Islands [4]. In folklore the roots of the plant are used as cathartic and leaves were reported to have laxative and vulnerary [5] activity. The present study is intended to determine the antimicrobial activity of the plant against selected UTI causing pathogens.

### 2. Materials and Methods

The plant was collected from the State Govt. Nursery, Unit-IV of Bhubaneswar, Odisha in the early morning during the month of September (rainy season). The plant was authenticated in the herbarium of Institute of Minerals and Materials Technology, Bhubaneswar, Odisha. The aerial parts were shade dried for 4 days and dried aerial parts grinded by a mechanical grinder and the coarse powder obtained was taken for extraction in petroleum ether followed by chloroform, methanol and water by using Soxhlet assembly for 48 h each. The extracts were concentrated to dryness under vacuum. The percentage yield of extracts was found to be 1.2%, 1.2%, 1.5% and 2% of petroleum ether, chloroform, methanol and aqueous extracts respectively. *In vitro* screening was carried out using selected UTI causing pathogens, which include two gram positive bacteria (*Staphylococcus aureus*, *Enterococcus faecalis*) and three gram negative bacteria (*Escherichia coli*, *Klebsiella pneumonia* and *Proteus mirabilis*). Antifungal activity of *A. godseffiana* extracts were screened against *Candida albicans*, *C. tropicalis*, *C. krusei* and *Aspergillus niger*. The bacterial strains were obtained from the University Department of Pharmaceutical Sciences, Utkal University, Vani Vihar, Bhubaneswar, Orissa. The extracts were screened for their antimicrobial activity by disc diffusion assay method [6, 7].

The petroleum ether, chloroform, methanolic and aqueous extracts were dissolved in dimethyl formamide (DMF, 6%) which was previously tested for antibacterial and antifungal activity against all test organisms and found to have no antimicrobial activity.

The extracts were made solution at a concentration of 1000, 500, 250 and 62.5 µg/ml and finally sterilized by filtration using 0.45 µm millipore filters.

The sterile discs (6 mm in diameter) were impregnated with the above extract solution to achieve the desired concentration. Nutrient agar and Sabouraud's dextrose agar (SDA, HiMedia Laboratories Ltd., Mumbai.) with DMF without extracts served as control sets throughout the study. The density of the bacterial suspension was standardized by standard McFarland method [8]. Disc diffusion assay was carried out by using a suspension of *Candida* species and *A. niger* by spread plate technique on freshly prepared SDA plates. The inoculated bacterial and fungal plates with the plant extract and standard discs on them were incubated at 37 °C for 24 h and 28 °C for 48 h respectively. The zone of inhibition exhibited by different extracts against the test bacteria and fungi by disc diffusion method [6, 7] was read using antibiotic zone reader which is given in Table 1 and Table 2 respectively. The minimum inhibitory concentrations (MIC) of the extracts were determined by two fold serial dilution assay [9] which is shown in Table 3.

Antimicrobial activity of *A. godseffiana* extracts were compared with ciprofloxacin (25 µg/disc) as standard antibacterial and clotrimazole (10 µg/disc) and terbinafine (25µg/disc) (HiMedia Laboratories Ltd.,Mumbai.) as broad spectrum antifungal agent. A comparison of plant extracts with the most effective standards against bacterial and fungal pathogens causing UTI is given in Table 1 and 2 respectively.

**Table 1:** *In vitro* antibacterial activity of *A. godseffiana* extracts against UTI causing pathogens by disc diffusion method

Conc. (µg/disc)	Zone of inhibition (in mm)				
	1	2	3	4	5
pe A	13	18	15	17	17
B	12	15	13	16	15
C	10	13	12	13	14
D	08	09	10	10	12
CH A	10	14	10	13	16
B	09	09	08	11	14
C	07	10	07	09	12
D	-	10	-	-	10
ME A	12	16	13	15	18
B	10	12	12	13	14
C	09	13	11	12	13
D	08	10	09	08	09
AQ A	15	12	16	15	18
B	12	10	14	12	16
C	07	09	12	11	15
D	-	07	09	07	10
CF	28	26	23	25	21

- indicates no zone of inhibition.

A, B, C & D indicates the concentrations of the extracts at 1000, 500, 250 and 62.5 µg/disc respectively.

1. *E. coli*; 2. *P. mirabilis*; 3. *E. faecalis*; 4. *S. aureus*; 5. *K. pneumonia*.

PE, CH, ME and AQ stands for petroleum ether, chloroform, methanolic (95%) and aqueous extracts respectively.

CF indicates ciprofloxacin (25 µg/disc) standard antibiotic.

### 3. Results and Discussions

All extracts of *A. godseffiana* Mast. at 1000 µg/disc showed optimum activity against all tested UTI causing pathogens. However, results of the disc diffusion method as indicated in Table 1, Methanolic extract inhibited significant and broader spectrum of inhibition followed by petroleum ether, aqueous

and chloroform extract against the UTI pathogens. Methanolic extract showed maximum inhibition (18 mm) against *K. pneumonia* and minimum (12 mm) against *E. coli*. Chloroform extract showed highest inhibition (16 mm) against *K. pneumonia* and minimum (10 mm) against *E. coli* and *E. faecalis*. Petroleum ether showed maximum inhibition (18 mm) against *P. mirabilis* and minimum (13 mm) against *E. coli*. Aqueous extract showed highest inhibition (18 mm) against *K. pneumonia* and minimum (13 mm) against *P. mirabilis*.

**Table 2:** *In vitro* antifungal activity of *A. godseffiana* extracts by disc diffusion method

Microorganisms	Zone of inhibition (in mm)					
	PE	CH	ME	AQ	CL	TF
1	09	17	15	17	11	16
2	07	10	18	10	22	12
3	12	15	16	13	13	17
4	11	09	17	12	22	-

- Indicates no zone of inhibition.

PE, CH, ME and AQ stands for petroleum ether, chloroform, methanolic and aqueous extracts respectively.

1. *Candida albicans*, 2. *C. tropicalis*, 3. *C. krusei* and 4. *Aspergillus niger*.

CL and TF stands for clotrimazole (10 µg/disc), terbinafine (25 µg/disc) respectively.

**Table 3:** The MIC Values of *A. godseffiana* Extracts against the Micro Organisms Tested by Two Fold Serial Dilution Assay

Microorganisms	MIC			
	Pet ether	Chloroform	Aqueous	Methanol
<i>S. aureus</i>	500	1000	250	500
<i>P. mirabilis</i>	62.5	500	500	250
<i>K. pneumonia</i>	125	1000	125	500
<i>E. faecalis</i>	250	500	125	125
<i>E. coli</i>	250	250	62.5	62.5
<i>C. albicans</i>	1000	500	500	250
<i>C. tropicalis</i>	n.t	1000	100	500
<i>C. krusei</i>	500	250	500	250
<i>A. niger</i>	500	1000	250	250

MIC - Minimum Inhibitory concentration (µg/ml)

n.t - MIC not found at the tested concentration.

The results of antifungal activities are indicated in Table 2, showed that the methanolic extract exhibit maximum and minimum zone of inhibition (18 and 15 mm) against *C. tropicalis* and *C. albicans* respectively. The chloroform and aqueous extracts showed similar zone of inhibition (17 and 10 mm) against *C. albicans* and *C. tropicalis* respectively, whereas the petroleum ether extract showed maximum and minimum (12 and 07 mm) inhibition against *C. krusei* and *C. tropicalis* respectively. *A. niger* was inhibited by *A. godseffiana* extracts with significant inhibition shown by methanol (17 mm), followed by aqueous, petroleum ether and chloroform extracts (12, 11 and 09 mm) respectively.

MIC against the tested microorganisms, as indicated in Table 3 showed that methanolic and aqueous extracts inhibit microbial growth at comparative lesser concentrations than chloroform extract.

#### 4. Conclusion

The findings of the present investigation revealed that the methanolic extract of aerial parts of *Acalypha godseffiana* was found to be more potent as compared to other three extracts against various UTI causing pathogens and fungal strains. Further investigation needs for isolation and characterization of active phytoconstituents from extracts which may yield few more compounds with greater antimicrobial potential.

#### 5. Acknowledgements

The authors are thankful to the HOD, U.D.P.S., Utkal University, Bhubaneswar for providing laboratory facility. We express our gratitude to the officials of State Government Nursery, Unit IV, Bhubaneswar, Odisha for providing information regarding the plant.

#### 6. References

1. The Wealth of India. Publication and Information Directorate. Vol 1, CSIR, Hillside Road, New Delhi-12, 1935, 47.
2. Quattrochi U. CRC – World Dictionary of Plant Names. Vol 1, CRC Press llc, 2000, 17.
3. The Macquarie Dictionary of Trees and Shrubs, Macquarie Library Pvt. Ltd. 1986, 15.
4. Kapoor SL, Sharga AN. House Plants, Vatika Prakashan. C-166, Nirala Nagar, Lucknow-226020, 1.
5. Kirtikar KR, Basu BD. Indian Medicinal Plants, Vol 3, Lalit Mohan Basu 49, Leader Road, Allahabad, India, 1935, 2261.
6. Indian Pharmacopoeia. The Controller of Publication, Vol 2, New Delhi 1996, 100.
7. Sahoo S, Panda PK, Mishra SR, Parida RK, Ellaiah P, Dash SK *et al.* Antibacterial activity of *Barringtonia acutangula* against selected urinary tract pathogens. Indian J of Pharm Sci 2008; 70(5):677-679.
8. Mc Farland J. Standardization of bacterial Culture for disc diffusion assay. J Amer Med Assoc 1987; 49:1176.
9. Rath CC, Dash SK, Mishra RK. A comparative note on antimycotic activity of turmeric leaf essential oil against *Candida albicans* and *Cryptococcus neoformans*. J Essential Oil Bearing Plants 1999; 2(2):106-111.