

The *In Vitro* Antibacterial Activity of *Hedyotis Umbellata*

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The *in vitro* antibacterial activity of *Hedyotis umbellata* (aerial parts) was investigated against *Staphylococcus aureus*, *S. citreus*, *Streptococcus faecalis*, *Bacillus subtilis*, *Escherichia coli*, *Klebsiella aerogenes*, *Shigella flexneri*, and *Pseudomonas aeruginosa* at 20 mg/ml, whereas the alcohol extract did not show any promising activity. The ethyl acetate eluate of the chloroform extract showed a zone of inhibition of 10mm against *B. subtilis* and *E. coli*, at 20 mg/ml.

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The roots and leaves of *Hedyotis umbellata* Linn (Rubiaceae), known as Chay root, are considered to be expectorant, and are used in the treatment of asthma, bronchitis, and bronchial catarrh. A decoction of leaves is used as a wash for poisonous bites. The root bark, preferably of a two years old plant, is the source of Chay-root dye, once employed with mordant for imparting red colour to calico, wool, and silk fabrics¹.

1, 2, 3-Trimethoxyanthraquinone, 1,3-dimethoxy-2-hydroxyanthraquinone, 1, 2-dimethoxyanthraquinone, 1-methoxy-2-hydroxyanthraquinone, 1,2-dihydroxyanthraquinone, 1, 3-dimethoxyanthraquinone-2-O-glycoside, and ruberythric acid, were isolated from the roots of the plant².

The plant has been collected in Erode, Tamilnadu, and identified at the Botany Department of CSMDRIA, Chennai. The aerial parts of plant were cut, shade dried, coarsely powdered, and extracted with chloroform by cold percolation method (48h.), and then with ethanol (95%). The extracts were concentrated to dryness *in vacuo*. Chloroform extract was chromatographed over silica gel (100-200 mesh), (Acme) and eluted with hexane, benzene, ethyl acetate, and ethyl acetate-methanol (4:1).

The extracts were dissolved in DMF. The solutions were further diluted to get test solutions of required concentrations. Penicillin (10 Iu/disc, Himedia) for gram positive bacteria, and neomycin (30 µ/disc) for gram negative bacteria, were the standards used.

TABLE 1: ANTIBACTERIAL ACTIVITY OF CHLOROFORM EXTRACT OF HEDYOTIS UMBELLATA

Bacterial strain	Concentration (mg/ml)			
	2.5	5	10	20
<i>S. aureus</i>	2.5	5	10	20
<i>S. citreus</i>	+	+	-	-
<i>S. faecalis</i>	+	+	-	-
<i>B. subtilis</i>	+	+	+	+
<i>E. coli</i>	+	+	+	-
<i>K. aerogenes</i>	+	+	+	+
<i>S. flexneri</i>	+	+	+	-
<i>P. aeruginosa</i>	+	+	+	-

+ No activity, - activity

TABLE 2: ANTIBACTERIAL ACTIVITY OF COLUMN ELUATES OF CHLOROFORM EXTRACT OF H. UMBELLATA AT 20 MG/ML

Bacterial strain	Zone of Inhibition in mm/disc				
	Hexane eluate	Benzene eluate	EtOAc eluate	EtOAc:CH ₃ OH(4:1)	Standard
<i>S. aureus</i>	NS	NS	NS	NS	14
<i>S. citreus</i>	NS	NS	NS	NS	24
<i>B. subtilis</i>	NS	NS	10	NS	18
<i>E. coli</i>	08	08	10	NS	18
<i>S. flexneri</i>	NS	NS	NS	08	22
<i>P. aeruginosa</i>	NS	NS	NS	NS	14

NS - Not sensitive

The antibacterial activity was assayed by streak plate and disc-diffusion methods^{3,4}. Nutrient agar was employed as the medium. The *in vitro* screening was carried out using *S. aureus*, *S. citreus*, *S. faecalis* and *B. subtilis* as gram positive bacteria, and *E. coli*, *K. aerogenes*, *S. flexneri* and *P. aeruginosa*, as gram negative bacteria. The nutrient agar containing the extract in different concentrations of 20,10,5 and 2 mg/ml, was poured into sterile petri dishes, and allowed to set. Different bacterial cultures were inoculated on the surface of the plates. The growth on the surface, means no activity. No growth on the surface, indicates activity of the trial drug at the particular concentration. The extract showing anti-bacterial activity was subjected to chromatography, and disc method was employed for eluates, for determining the activity. The disc diameter was 6 mm.

Results of the screening data were summarized in Tables 1 and 2. The alcohol extract did not show any promising activity, while chloroform extract showed activity against six bacterial strains (Table 1). The activity was noticed at a concentration of 10 mg/ml and above for *S. aureus* and *S. citreus*, and at 20 mg/ml for other bacteria, except *B. subtilis* and *K. aerogenes*. The hexane eluate of the chloroform extract showed a zone of inhibition of 8 mm at 10 mg/ml for *E. coli*. The ethyl acetate eluate was active against *E. coli* and *B. subtilis* at 20 mg/ml, showing a zone of inhibition of 10 mm. and against other strains, it was ineffective. Purification of the eluate yielded ursolic acid. This compound had been shown to be effective against *S. aureus* and *B. subtilis*⁵.

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REFERENCES

1. Anonymous, The Wealth of India, CSIR, New Delhi, 1959, pp 16.
2. Purashothaman, K.K. Saradha Krishnan and Narayanaswami, V.,

1994, 60, 471.

J.Res.Ind. Med.,1972, 7, 37.

3. Burrows, W., Moulder, J.W., and Lewert, R.M. Eds: In Text Book of Microbiology, 18th ed., W.B. Saunders Company, Philadelphia and London, 1963.
4. Kavanagh, F.Ed: In Analytical Microbiology, Academic Press, New York & London, 1963.
5. Richards, R.M.E., Durham, D.G. and Liu X., **Planta Medica**,

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