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Antimicrobial activity of crude extract of leaves, roots, and Arial part of Rivea hypocretariformis

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ABSTRACT-*Extraction of different parts (Arial Part, leaves, and roots) of the Rivea hypocretariformis by different solvent like methanol, water, chloroform and n-Hexen and assessed in vitro antimicrobial assay by disc diffusion method. From the observation more polar methanol extracts show more inhibition zone compares to other solvents in all parts (Arial Part, leaves, and roots) of Rivea hypocretariformis. In the all parts of the plant, highest inhibition zone was observed in methanol extract against each bacterial strain. Where S. aureus shows high active zone inhibition and Aspergillus niger show less. Due to presence of major chemical components make it seems to be important for medical purposes and plant contains Potential antibacterial components that may be useful for evolution of pharmaceutical for the therapy of ailments. Also plant extracts can be used for the treatment of infections caused by the strains of the test bacterial organisms.*

Keywords-*Antimicrobial, Rivea hypocretariformis, Extraction, Soxhlet Apparatus, Inhibition zone*

I. INTRODUCTION

Plants are very important in human being. Plants produce numbers of phytochemical compounds which are useful for human. Natural compounds of plants are derived from the phenomenon of biodiversity. Plants produce these compounds to enhance their survival and competitiveness.Due to their biological activities, many plant produce number of chemical compounds, which are used for the treatment of human illnesses or diseases. The survey of the World Health Organization (WHO) indicated that about 70–80% of the world's population relies on traditional medicine based mainly on plant materials for their primary healthcare [1]. The benefits of individual phytochemical supplements are largely unproven, but they are not regulated. High concentrations of some phytochemical may behave like drugs and be toxic, possibly even contributing to cancer cell growth. Rivea hypocrateriformis (Desr.) choisy belongs to family Convolvulaceae phys. *Revia Hypocretariformis* is usually found in tropical region such as India, Pakistan and Afghanistan in the monsoon season. Its regional name is Phang or Phangi. Its vines can be strong and can rise to ground height from two to three meters above on help of wooden base. Its leaves are almost rounded and two to three-inch in diameter. At the time of maturity, the shape of the leaves is higher, which is used to make traditional dishes in India. According to Ayurvedas, its root is also great significance. According to one belief, if the poison of the scorpion spreads in the body, then the poison is removed only by holding the fresh root of the plant [2]. This plant is an effective agent in anti-implantation and pregnancy interruption in female albino rats [3]. This plant is used as psychoactive drug. Where Indians people use these plants as psychedelic drug [4]. Juice of leaves of this plant is taken with cow's milk in rheumatic pain as it has Flavonoids and phenolic acid that are very much rubostic and helpful to humans [5] and also the leaves extract is used for skin disease of hair scalp [6]. It has been reported that leaves, Arial part and Root extracts of this plant show strong antioxidant activity [7].

II. METHODS AND MATERIAL

(A) COLLECTION OF SAMPLE:

Fresh leaves, roots and Arial parts of R. hypocrateriformis were collected from Khavad village, Dist. Mehasana (western region of India) in the month of January-2017. The plant ware authenticated by a taxonomist Dr.K.J.Bhatt from Department of Botany Pramukh Swami science and H.D.Patel Arts College Kadi. All parts were washed well using tap water and twice using distilled water and it was dried in shade for a period of 20-25 days, at an ambient temperatures of 29ºC. After drying, Parts of plant were cut into small pieces. The dried sample was grind properly using a mixer to obtain the powdered form.

Table 1: Data of plant samples

PLANT NAME	PART OF THE PLANT	COLLECTION NO.	ACCESSION NO.
Rivea hypocretariformis	**ARIAL PART**	**RHA**	**RHA-1, RHA-2, RHA-3, RHA-4**
	LEAVES	**RHL**	**RHL-1, RHL-2, RHL-3, RHL-4**
	ROOTS	**RHR**	**RHR-1, RHR-2, RHR-3, RHR-4**

Figure: 1 Images of Rivea hypocretariformis.

(B) CHEMICALS AND MICROBIAL CULTURES

All the chemicals used in this work were purchased from Sigma Aldrich, Merck and Hi-media, Mumbai, India. The reference bacterial strains used in this study were obtained from American Type Culture Collection (ATCC) and Microbial Type Culture Collection (MTCC) Institute of Microbial Technology, Chandigarh, India.

(C) PREPARATION OF PLANT EXTRACT

Parts the plant were dried over ambient temperature and the dried samples were grind properly and dried powder sample was extracted in Methanol at 65ºC, Chloroform 61ºC, n-Hexane 68ºC and Distilled water 100ºC by using Soxhlet apparatus. Further extracts were concentrated by gradually evaporating the respective solvent on hot water bath. The concentrated extract was collected in sterile bottles and kept in a cool and dark place prior to analysis.

(D)ANTIMICROBIAL ACTIVITY OF ALL PARTS OF *R. hypocretariformis*

Antimicrobial activity of four organic extracts viz. aqueous, methanolic, chloroform and n-Hexane of all parts of R. hypocrateriformis were determined by Agar disc diffusion assay according to the Manual of antimicrobial susceptibility testing [8]. To assay, one antibiotic (CIPROFLOXACIN) for bactericidal activity against test strains, *E. coli*, and *S. aureus*, and one antifungal drug (FLUCONAZOLE) assay for antifungal activity against *Aspergillus niger* were spread on separate nutrient agar plates [9]. All four organic extracts (1000µg/ml, 500µg/ml, and 250µg/ml) were loaded separately on sterile discs and allowed to dry and placed on the bacteria inoculated nutrient agar media [10]. Negative control was prepared by loading the discs with solvents and positive control was by Tetracycline. The plates were incubated at 37°C for 24 hr and zone of inhibition around the disc were measured. The experiment was done with three replicates for consistency of the experiment. The organic extracts, Methanol exhibiting maximum antimicrobial activity, were selected for subsequent studies.

III. RESULTS AND DISCUSSION

The antibacterial screening was carried out for four organic solvent plant extract. Antibacterial Activity of plant *Rivea hypocretariformis* is listed in Table 2, 3, and 4.The antibacterial screening of the leaves, highest Inhibition zone was observed in methanol extract in each bacterial strain where *E.coli* showed medium Active zone of inhibition where *S. aureus* show maximum and *Aspergillus niger* show less. in case of n-Hexane and chloroform show less zone of inhabitation while aqueous extracts showed medium Inhibition zone in all bacterial strains. And same observation in the antibacterial screening of the extract of root and Arial part which is clearly shows in table 3 and 4.

Table 2: Antimicrobial activity of crude extract of leaves rivea Hypocretariformis

SAMPLE OF EXTRACT	**ZONE OF INHABINATION (mm) CRUDE EXTRACT OF LEAVES OF Rivea hypocretariformis**								
	S.aureus (GRAM POSITIVE BACTERIA)			*E.coli* (GRAM NEGATIVE BACTERIA)			*Aspergillus niger* (FUNGI)		
	1000µg/ml	**500µg/ml**	**250µg/ml**	**1000µg/ml**	**500µg/ml**	**250µg/ml**	**1000µg/ml**	**500µg/ml**	**250µg/ml**
AQE	22	20	19	18	16	15	17	15	15
MEE	**23**	**22**	**21**	**20**	**18**	**17**	**20**	**19**	**18**
CLE	19	18	16	13	12	11	15	15	14
N-HEXANE	16	14	14	11	9	8	12	10	9
FLUCONAZOLE	0	0	0	0	0	0	22	21	19
CIPROFLOXACIN	24	24	23	23	22	22	0	0	0

Table-3: Antimicrobial activity of crude extracts of roots Rivea hypocretariformis

SAMPLE OF EXTRACT	ZONE OF INHABINATION (mm) CRUDE EXTRACT OF ROOT OF *Rivea hypocretariformis*								
	***S.aureus* (GRAM POSITIVE BACTERIA)**			***E.coli* (GRAM NEGATIVE BACTERIA)**			***Aspergillus niger* (FUNGI)**		
	1000µg/ ml	**500µg/ ml**	**250µg/ ml**	**1000µg/ ml**	**500µg/ ml**	**250µg/ ml**	**1000µg/ml**	**500µg /ml**	**250µg/ ml**
AQE	21	20	20	16	15	15	15	13	12
MEE	**22**	**21**	**20**	**17**	**16**	**16**	**19**	**17**	**17**
CLE	17	17	16	12	11	10	12	11	11
N-HEXANE	14	12	12	10	8	8	10	9	8
FLUCONAZOLE	0	0	0	0	0	0	22	21	19
CIPROFLOXACIN	24	24	23	23	22	22	0	0	0

Table-4: Antimicrobial activity of crude extract of Arial part of rivea Hypocretariformis

sample of extract	ZONE OF INHIBITATION (mm) CRUDE EXTRACT OF ARIAL PART OF Rivea hypocretariformis								
	***S.aureus* (GRAM POSITIVE BACTERIA)**			***E.coli* (GRAM NEGATIVE BACTERIA)**			***Aspergillus niger* (FUNGI)**		
	1000µg/ ml	**500µg/ ml**	**250µg/ ml**	**1000µg/ ml**	**500µg/ ml**	**250µg/ ml**	**1000µg/ml**	**500µg /ml**	**250µg/ ml**
AQE	20	19	17	17	16	16	16	15	15
MEE	**21**	**20**	**20**	**19**	**18**	**18**	**18**	**17**	**17**
CLE	18	17	17	12	10	9	15	14	14
N-HEXANE	15	14	13	10	8	7	11	10	9
FLUCONAZOLE	0	0	0	0	0	0	22	21	19
CIPROFLOXACIN	24	24	23	23	22	22	0	0	0

Aqueous extract [AQE], Methanolic extract [MEE], Chloroform extract [CLE], Antibiotic [AB],

IV. CONCLUSION

Among extracts from different parts of Rivea Hypocretariformis, i.e. Leaves Arial part & roots. Leaves exhibited highest antibacterial activity. Among the three test bacteria *S.aureus* is found to be the most sensitive. And this bacteria is responsible for most common many diseases, hence it is concluded that Rivea hypocretariformis has greater applications of medicinal property. Also the plant contains potential antibacterial components that may be useful for evolution of pharmaceutical for the therapy of ointments and also plant extracts can be used for the treatment of various bacterial infections. This study paves the way for further attention and research to identify the active compounds responsible for the plant biological activity. Further studies should be undertaken to elucidate the exact mechanism of action by which extracts exert their antimicrobial effect.

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