



e-ISSN: 2319-8753 | p-ISSN: 2320-6710

IJIRSET

International Journal of Innovative Research in
SCIENCE | ENGINEERING | TECHNOLOGY

INTERNATIONAL JOURNAL OF INNOVATIVE RESEARCH

IN SCIENCE | ENGINEERING | TECHNOLOGY

Volume 10, Issue 5, May 2021

ISSN INTERNATIONAL
STANDARD
SERIAL
NUMBER
INDIA

Impact Factor: 7.512

9940 572 462

6381 907 438

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Study of Antimicrobial Activity of *Barleria prionitis* on Pathogenic Strains

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ABSTRACT: Antimicrobial analysis of plant products took a distinct place in organic chemistry as well as plant biochemistry in recent years. This investigation was planned to evaluate the antimicrobial activity of *Barleria prionitis* Linn (leaf) extracts in three different solvents viz ethanol, chloroform and petroleum ether. We have tested these extracts against bacteria (*S. aureus*, *E. coli*,) and fungus (*Saccharomyces cerevisiae*). Petroleum ether extract of leaf showed highest antibacterial activity against *S. aureus* (22.66 mm in diameter) followed by ethanol leaf extract against *E. coli* (21.66 mm in diameter). Various extracts of *B. prionitis* were comparable to control antimicrobial agent like antibiotics viz Oflaxacin, Azithromycine. Maximum inhibition shown by Oflaxacin, against *S. aureus* (28.20) followed by against *B. prionitis* (28.40) as same is with antifungal activity shown by different extracts.

KEYWORDS: Leaf extract, Antibiotics, Inhibition, Antimicrobial activity, Solvents.

I. INTRODUCTION

Phytochemical analysis of plant products took a distinct place in organic chemistry as well as plant biochemistry in recent years. It is concerned not only with the variety of organic substances accumulated by plants but also helps to determine chemical structure, distribution and biological function of plant substances.

Thus, advances in our understanding of phytochemistry are directly related to the successful exploitation of known techniques, and the continuing development of new techniques to solve complex problems as they appear. One of the challenges of phytochemistry is to carry out all the above operations on vanishingly small amounts of material. Frequently, the solution of a biological problem i.e. in plant growth regulation, in the biochemistry of plant-animal interactions, or in understanding the origin of fossil plants depends on identifying a range of complex chemical structures which may only be available for study in microgram amounts.

B. prionitis is used as medicinal plants. There are number of important source of medicine, which are being used in the Ayurveda, Unani, Siddha, etc. the world over, since the time immemorial. Approximately 60- 80 % of the world's population still depends on traditional medicines. Several metabolites i.e, medicinal components have been successfully isolated from plants. *B. prionitis* is being used for the treatment of different diseases. In this era the safest and secured way to discover new drugs from natural products^[10] Though the plants contain a large number of compounds of medicinal importance known as secondary metabolites. These are consumed by human beings present in different parts of the plants i.e. the leaves, stems, roots, sap etc.. The secondary metabolites such as saponins, alkaloids, flavonoids, glycosides, anthraquinone, steroids and tannins are obtained from plants. *Barleria prionitis* Linn. (Acanthaceae) commonly found in the tropical Asia, including Malaysia, southern eastern India, Sri Lanka Pakistan, Philippines, and in tropical Africa and Yemen e.t.c.,

II. METHODOLOGY

Collection of plant: -In the present study the plant *B. Prionitis* was collected in early hours of the day, without much disturbance in and around, Arogya Kendra, Bhopal, India. Plants are stored using sterile polythene bag and immediately transferred to the laboratory for further analysis. The plant material was shadow dried for one week, then powdered with the help of mixer grinder and used for preparation of plant extracts.

2) **Extraction:**-Extraction was carried out in different solvent such as petroleum ether, chloroform, Ethanol at 60 °C for 8 hours using the soxhlet extractor. After extraction the extracts were dried at room temperature until extract acquires the solid form. Different organic solvent extracts of *B.Prionitis* were used to screen the following phytochemicals like sterol, reducing sugar, alkaloids, phenolic compounds, flavanoids, tannins, saponins, amino acids, glycosides and terpenoids. Phytochemical tests were carried out in extracts using the standard procedures as described by Sofowara (1993), Trease and Evans (1989) and Harborne (1973). The various extracts of *B.Prionitis* were subjected to phytochemical tests to screen the following chemicals. The phytochemical analysis was conducted using the test developed by Kapoor et al. (2013) and the presence of different phytochemicals in the extract were listed in the table, which would further be used for quantitative investigations. The Antimicrobial activity of plant extract was carried out in different solvents.

3) **Collection of strain:-** The strain of Bacterial pathogens (*S. aureus*, *E. coli*,) and Fungal pathogens like *Saccharomyces cerevisiae* were collected from Biotechnology Department, Sant Hirdaram Girls College, Bhopal.

4) **Preparation of bacterial/fungal strains and culture conditions :-**

The bacterial and fungal cultures were sub cultured in Nutrient broth and Potato Dextrose Broth respectively to prepare fresh subcultures from cultures. The stocks were transferred aseptically under Laminar Air Flow into freshly prepared medium using inoculating loop. The subcultures were then incubated at 37°C (bacteria) for 24 hours and at 28°C (fungus) for 2 days.

Method:

The extracts mentioned above were tested against fungal strain (*Saccharomyces cerevisiae*)^[1] and two gram-negative bacteria (*S. aureus*, *E. coli*,). Antimicrobial screening was done using agar well diffusion method^[3]. For Bacteria we have prepared Nutrient agar medium which was poured in Petri plates and labeled as followed: Extract + solvent as Test, Standard antibiotic (Amoxicillin and ofloxacin) dilution as Reference and Only solvents as Negative control. Triplicates were prepared for both the bacterial under test. Similar setup was prepared for fungus; the medium used was czapedox agar medium. After solidification of medium test micro-organisms were inoculated on the respective mediums via spread plate technique. Then wells were created in the inoculated plates with the help of gel puncture and in each marked plates respective extracts, solvents and antibiotic dilutions were added. The concentration of extract+ solvent used was 10mg/ml. The plates were then incubated at 37°C for 24 hrs in case of bacteria and at 28°C for 2 days in case of fungus for the antimicrobial activity assay. Triplicate plates were prepared for each treatment and the average zone of inhibition excluding well was recorded. The process is modified. (Ref)

III. RESULTS

In the present study, chloroform, petroleum ether ethanol, extracts of leaf of *Barleria prionitis* were evaluated for the presence of phytochemical constituents as well as their antimicrobial activity was tested. It was apparent that standard antibiotics Amoxicillin and ofloxacin shows zone of inhibition against both the bacteria. Negative control of bacteria *S.aureus* showed that it can be inhibited by ethanol as a solvent alone, but it didn't show any inhibition with either petroleum ether or chloroform similar results were observed with *E.coli*. The test plate of bacteria *S.aureus* showed that the extract+ethanol is effective in killing the bacteria in comparison to other two solvents (petroleum ether and chloroform) + extract combinations. In Test plate of bacteria *E.coli* zone of inhibition or affectivity was shown by petroleum ether + extract and Ethanol+extract combination showing that this mixture can effectively kill the bacteria. Antibacterial antibiotics showed no affectivity against fungus the results were negative, whereas in test plate the petroleum ether+ extract combination showed zone of inhibition indicating the antifungal properties of *Barleria prionitis* extract while the other two mixtures show negative results.

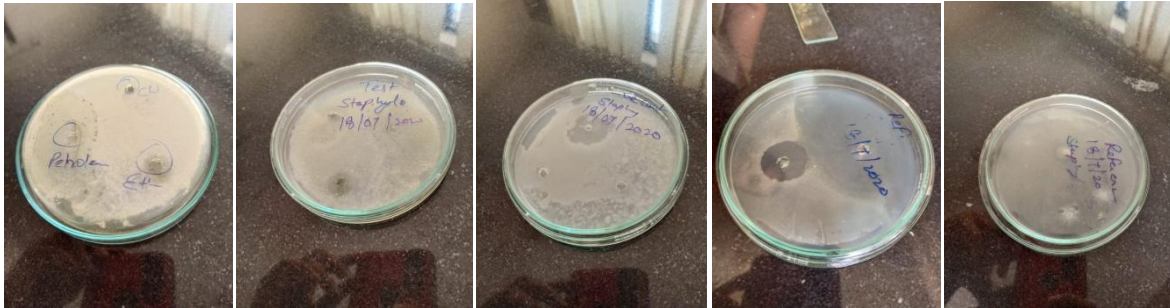


Figure 1a

Figure 1b

Figure 1c

Figure 1d

Figure 1e

Figure 1a and 1b shown the results of positive control or inhibition zone with plant extract against *S. aureus*, Figure 1c shown the results of negative control figure 1d and 1e shown the results of reference against *S. aureus*.

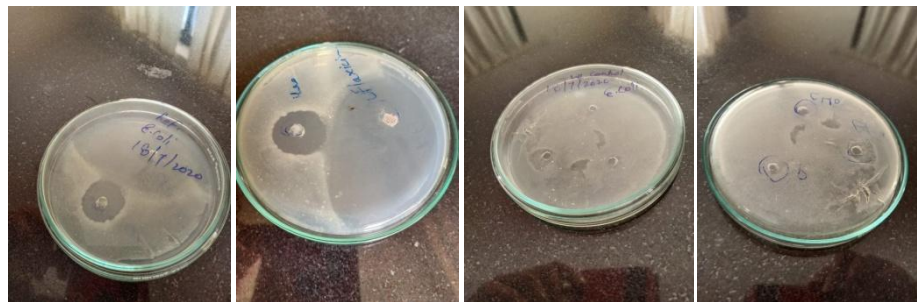


Figure 2a

Figure 2b

Figure 2c

Figure 2d

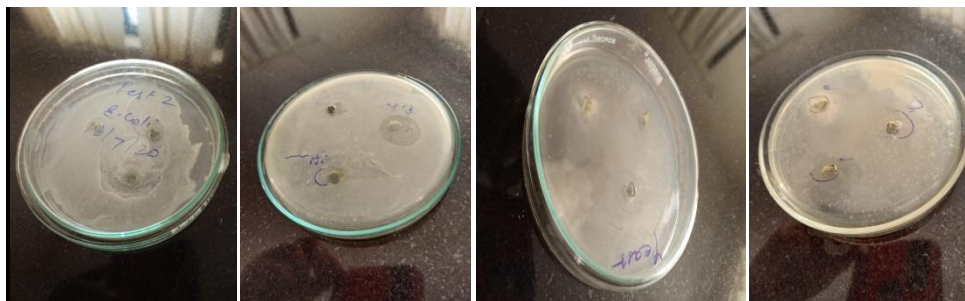


Figure 2e

Figure 2f

Figure 3a

Figure 3b

Figure 2a and 2b shown the results of reference against *E.coli*, Figure 2c and 2d shown the results of negative control, Figure 2e and 2f shown the results of positive control or inhibition zone with plant extract reference against *E.coli*.

Figure 3a and 3b shown the results of positive control or inhibition zone with plant extract against *Saccharomyces cerevisiae*

IV. CONCLUSION

In the present study, Ethanol has been shown as a best solvent to the highest yield for extracted components and antimicrobial effects while petroleum ether and Chloroform were also good solvents to extract secondary metabolite components.



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