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PHYTOCHEMICAL INVESTIGATION OF DIFFERENT SOLVENT EXTRACTS OF *Berberis lycium* FRUITS

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ABSTRACT

Plants are biosynthetic laboratories for many useful compounds those are playing important role in human health. Ingredients of some medicinal plants are very much effective against bacterial and fungal growth and can be used to minimize their activities. The present work was carried out to study the antimicrobial activity of the aqueous, ethanolic, Dimethyl formamide, propanol-2 and n-hexane extracts of the root, fruit and flower of *Berberis lycium* belonging to the family of Berberidaceae, against different strains of bacteria and fungi.A study was conducted to assess the possible affects of *Berberis lycium* against the selected 5 bacteria and 3 fungi.The study revealed that the fruit extract possessed a good antifungal property in comparison to the other extract of the plant and this can be used in the further study for future in medicines.

Keywords: Antimicrobial activity, Antifungal, Biosynthetic, Berberidaceae, Bacterial strains.

INTRODUCTION

In India, medicinal plants are widely used by all sections of people either directly as folk remedies or in different indigenous systems of medicine or indirectly in the pharmaceutical preparation of modern medicines.According to the WorldHealth Organization (WHO, 1977) "a medicinal plant" is any plant, in which one or more of its organ contains substances that can be used for the therapeutic purposes or which are precursors for the synthesis of useful drugs.[1] Medicinal plants are very useful as they are the easily available source for health purpose in rural and tribal areas.[2]

According to National Health Experts, 2000 different plants are used for medicinal preparations in India alone. Among them only 200 are of animal origin, and 300 of mineral origin, while 1500 drugs are extracted from various plants.[3]The plants are the best source of remedy for curing all the type of infections or diseases. This is the reason that the medicinal plants play a vital role in the health maintenance of the people worldwide. Medicines derived from plants are used from are ancestral time i.e ayurvedic knowledge.

Higher plants products are an important source of therapeutic agents. Many researchers are screening the different biological activities of plant.[4-6] Medicinal herbs constituents greatly vary with the genetic factor, climatic changes, soil and other factors.[7] From recent years infections are increasing as well as the problem of antibiotic resistance.

The aim of this work was to investigate the antimicrobial activity of *Berberis lycium* that has been selected because of their great medicinal relevance. It is well known that in past years, infection rates have increased and antibiotic resistance has become an increasing therapeutic problem.[8,9] Due to the presence of free radicals i.e the antioxidant activity of the plant extracts that can be responsible for many diseases. Diseases like kidney problems, the ageing process, heart stroke and cancer as well.In addition, a greater interest in the antioxidant activity of plant extracts exists because of free radicals (e.g., reactive oxygen species) that can be responsible for several diseases, for example, heart disease, stroke, arteriosclerosis and cancer, as well as the aging process.[10,11]

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EXPERIMENTAL

Extract Procedure

The fruits were collected and were washed thoroughly with running tap water and finally with distilled water. After shade drying, for 10-15 days the fruits were crushed into powdered form with the help of mortar and pesil. Then it was added to soxhlet assembly for extraction against solvents like distilled water, ethanol, n- propanol, DMF, n-hexane.It was heated for about 6 hours at a temperature less than the boiling point of the solvent.The extract was further concentrated by rotary evaporator and residue was stored for a further process where as in case of aqueous media same amount of sample was dissolved in water and boiled, filtered and saved for further process.

Test Organisms

A total 6 microbial culture belonging to 3 bacterial species and 3 fungi were used in this study. The identified microorganisms were obtained from microbiology laboratory Uttaranchal University, Dehradun.

Antimicrobial Activity

For determination of antibacterial activity, the method is given by Kirby–Bauer for antibiotic testing was used in this study. It is also known as **Disc diffusion antibiotic sensitivity testing**. Disc diffusion method uses antibiotic soaked wafers to test whether bacteria is affected by antibiotics or not. Wafers having antibiotic are placed on the agar plate, bacteria are already on the plate and they are kept for incubation. The bacteria are swabbed uniformly across the culture plate. If the antibiotic stops, the bacteria from growing or kills the bacteria. The area around the wafer where bacteria have not grown enough to be visible is known as the zone of inhibition. The size of zone depends upon the amount of antibiotic present on the plate. Stronger antibiotic creates a larger zone of inhibition because a lower concentration of antibiotic is enough to stop the bacterial growth. A filter paper disk containing the compound to be tested is placed on the agar plate. The compound diffuses from the filter paper to the agar. The concentration of the compound increases near to the disk and it keeps on decreasing as the distance from disk increases. If the compound is effective against bacteria at a certain concentration no colonies will grow where the concentration in the agar is greater than or equal to the effective concentration. This zone is known as the zone of inhibition. This helps to estimate the bacterial sensitive to that particular antibiotic. The minimum inhibitory concentration of antibiotic for that bacteria relates to larger zones. Inhibition produced during the test is compared with that produced by the known concentration of a reference compound. This technique can be used to find a particular antibiotic for a specific infection. Inhibition produced by the test is compared with that produced by known concentration of a reference compound. In a similar way, fruit extracts of *Berberis lycium* were tested against various microorganisms i.e. bacteria and fungus.[12]

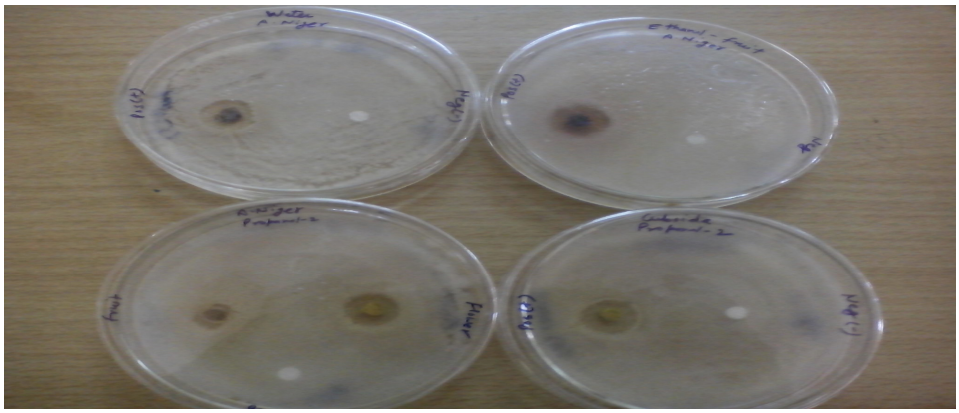

Fig.-1: Zone of inhibition by *Berberislycium*

RESULTS AND DISCUSSION

It is evident from Table-1 that the inhibitory activity was exhibited by the components present in the fruit extract of *Berberis lycium* and not by the solvents used for extraction. Among the different extracts tested against the mixed culture of specific human pathogenic bacteria as seen in Table-2 *A. niger* in ethanolic extract of fruit showed a zone of inhibition of 11mm while Pseudomonas sp. showed the activity of about 14mm and other stains of bacteria like E.coli, and Staphylococcus aureus.

From Table-3 it is evident that the aqueous extract of fruit showed a zone of inhibition of 9mm while Pseudomonas sp. showed a zone of inhibition of about 16mm while Staphylococcus aureus showed 14mm of activity and *E.coli* showed resistant. *A.niger* showed a zone of inhibition of 9mm while *A.cuboida* and *A.fumigatus*.

From the Table-4 it is evident that fruit extract is not effective against selected bacteria. While propanolic extract of fruit showed 5 mm zone of inhibition against fungi *A.fumigatus*. It has shown almost nil activity aginst fungi *A.cuboida and A.niger*. *E.coli*, *Pseudomonas sp*. and *Staphylococcus aureus* showed no activity they were resistant. The medicinal plants are shown to have safety and efficacy in pharmacological activities including as antifungal activity.[13]

In another study done by Singh ,M and his coworkers in 2009[14] observed that alcoholic specially methanolic extract is effective against some selected bacteria and fungi, those results also agree with us that alcoholic extract more effective against selected bacterial strains.

Table-1: Inhibitory activity of 10% root extract *of Berberis lyceum*

Extract	Antimicrobial activity
Aqueous	+
Ethanol	+
Propanol-2	+
n-hexane	-
DMF	-

Table-2: Antimicrobial activity of methanolic extracts of *Berberis lycium*

Microorganisms (Bacteria)	Fruit extract
E.coli sp.	ND
Pseudomonas sp.	14
S.aureus	ND
Microorganism (Fungal)	
A.cuboida	ND
A.niger	11
A.fumigatus	ND

Table-3: Antimicrobial activity of aqueous extracts of *Berberis lycium*

Microorganisms (Bacteria)	Fruit Extract
E.coli sp.	ND
Pseudomonas sp.	16
S.aureus	14
Microorganism (Fungal)	
A.cuboida	ND
A.niger	9
A.fumigatus	ND

Table-4: Antimicrobial activity of Propanolic extracts of *Berberis lycium*

Microorganisms (Bacteria)	Fruit Extract
E.coli sp.	ND
Pseudomonas sp.	ND
S.aureus	ND
Microorganism (Fungal)	
A.cuboida	ND
A.niger	ND
A.fumigatus	5

CONCLUSION

Although this study investigating the in vitro antimicrobial activity, the results showed that the ethanolic extract of fruits from *Berberis lycium* possessed maximum antifungal property in comparison to aqueous and propanolic extract, confirming the great potential of bioactive compounds and are useful for rationalizing the use of this plant in primary health care. This means some component is present in the fruit that is responsible for resistant as well as the activation against bacterial and fungal strains. In vivo data may be helpful in determining the real potential usefulness of this plant for the treatment of infectious diseases.

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