

A Study of Antibacterial Activity of the Crude Extracts of *Biophytum sensitivum*

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Received 20/01/2015 Accepted 05/04/2015

Abstract

The leaves of *Biophytum Sensitivum* extracted with ethanol and water in a soxlet extractor were used for the study purpose. The preliminary antibacterial activities of the extracts were determined by measuring the zone of inhibition produced by the extracts against various microorganisms (Enterococcus, Staphylococcus and Klebsiella). The extracts were compared with a positive control Gentamycin and negative control DMSO. All the extracts exhibited marked activities against the tested microorganisms. Ethanol extract exhibited more activity than water extract.

Keywords: *Biophytum Sensitivum*, Antibacterial, Zone of Inhibition, Enterococcus, Staphylococcus, Klebsiella

Introduction

Biophytum sensitivum (Mukkutti) is a perennial herb belonging to the family Oxidaceae. It has very small flowers with wide distribution in India. It is an important flower for the people of Kerala both for its medicinal values and cultural importance. *Biophytum* is used for the treatment of chest complaints, convulsions, cramps, inflammatory tumours, arthritis, back pain, bone spur, leg pains etc. Its ash is mixed with lime juice and given for stomach problems. The leaves are grinded in butter milk and used for treating dysentery. The powdered leaves and seeds were applied on wounds (Bever and Zahnd, 1979). It is a good medicinal herb and is used to clean uterus after delivery. *Biophytum sensitivum* is one of the plants used against snake envenomation.

The administration of methanolic extracts of *Biophytum sensitivum* increases the total White Blood Cell count and stimulate the hematopoietic system by increasing the count of bone marrow cells. The antitumor activities of *Biophytum sensitivum* extract was determined by both in vitro and in vivo methods. In the present study an attempt has been made to reveal that leaf extracts of *Biophytum sensitivum* possess antibacterial activity.

Materials and Methods

Healthy plants of *Biophytum sensitivum* was collected from different areas of Kottarakara of Kollam district (Kerala) during the month of November 2013. Herbarium speci-

men of the study material has been deposited in the department herbarium. Leaves were harvested, washed and dried in shade. Dried samples were powdered and 5g were extracted with ethanol and water in soxhlet extractor. The ethanol and water extract was concentrated under vacuum in a rotary evaporator.

The antimicrobials present in the plant extract were allowed to diffuse out into the medium and interact in a plate freshly seeded with the test organisms. The resulting zones of inhibition will be uniformly circular as there will be a confluent lawn of growth. The diameter of zone of inhibition can be measured in centimeters.

The medium was prepared by dissolving 33.9 of the commercially available Muller Hinton Agar medium (HiMedia) in 1000mL of distilled water. The dissolved medium was autoclaved at 15lbs pressure at 121°C for 15 minutes. The autoclaved medium was mixed well and poured into 100mm petriplates (25-30 mL/plate) while still molten.

One litre of nutrient broth was prepared by dissolving 13g of commercially available nutrient medium (HiMedia) in 1000mL distilled water and boiled to dissolve the medium completely. The medium was dispensed as desired and sterilized by autoclaving at 15lbs pressure (121°C) for 15 minutes. Gentamycin is used as standard antibacterial agent (concentration 20mg/mL).

Petri plates containing 20mL Muller Hinton medium were seeded with 24hrs culture of bacterial strain such as, *Staphylococcus aureus* and *E. coli*. Wells of approximately 10mm was bored using a well cutter and sample of 25, 50 and 100µl concentrations were added. The plates were then incubated at 37°C for 24 hours. The antibacterial activity was assayed by measuring the diameter of the inhibition zone formed around the well (NCCLS, 1993). Gentamycin is used as a positive control.

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Results and Discussion

According to the World Health Organization, infectious diseases are a significant cause of worldwide morbidity and mortality, accounting for approximately 50% of all deaths in tropical countries. Infectious disease hospitalization rates have increased over time and are associated with substantial morbidity, mortality, and economic consequences. Additionally, antimicrobial resistance to antibiotics is emerging as a serious health issue and alternatives to treat infectious diseases in the future need to be developed.

A number of studies have voiced the necessity of developing alternative antimicrobial drugs. Antimicrobial activity of plant extracts were already reported Mehru *et al.*, (2008) and Al-Sieni and Abdul basit (2014). Plant antimicrobials would appear to be an excellent choice. Our study revealed that, Biophytum produced strong antimicrobials and may offer prospective new treatments for bacterial infections. Antimicrobial properties of biophytum were also discovered by Shivakumar and Srinivasan (2012) and Natarajan *et al.*, (2010). The benefit of antimicrobial properties from the plant can only be achieved, however, by using a specific solvent and solvent concentration in extracting the plant materials.

The study of extracts obtained with ethanol warrants future research to determine the active extract constituents at each ethanol level. Antimicrobial properties using ethanolic extracts were already reported by Sule and Agbabiaka (2008). In spite of the number of published scientific articles around the globe that describe the antimicrobial activities of plant extracts, systematic studies conducted on the effects of solvent concentration on the antimicrobial activity

are lacking. Due to the complex nature of the phytochemicals present in a plant extract, the extraction solvent system needs to be considered. The present study provides data on the importance of selection of an appropriate solvent concentration and indicates that ethanol extracts of plants can offer significant potential for the development of novel antibacterial therapies.

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Table 1. Zone of inhibition produced by the *Biophytum sensitivum* extracts against various microorganisms

Sample	Volume of sample (μ L)	Zone of inhibition (cm)
<i>Staphylococcus aureus</i>		
Gentamycin		3.2
Biophytum aqueous	100	1.1
Biophytum ethanol	100	1.6
<i>Enterococcus</i>		
Gentamycin		4
Biophytum aqueous	100	1.4
Biophytum ethanol	100	1.5
<i>Klebsiella</i>		
Gentamycin		3.2
Biophytum aqueous	100	1.1
Biophytum ethanol	100	1.5

Note: 0.1gm sample in 1mL DMSO