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Research Article

In vitro antibacterial activity of flower extracts of *Quisqualis indica* Linn. against gram-positive and gram-negative bacteria

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ABSTRACT

The present study is designed to investigate the antibacterial activity of flower extracts of *Quisqualis indica* Linn. against gram-positive as well as gram-negative bacterial strains. For this, agar well diffusion assay method was applied to determine the antibacterial activity of the extracts. The flower extracts with different solvents (methanol, ethanol and aqueous) showed remarkable antibacterial activity against the tested bacterial strains. Among the tested bacterial strains, *Micrococcus luteus* was highly susceptible to the flower extract as compared with the *Bacillus subtilis* and *E. coli.* The results also showed that the methanolic extract of *Quisqualis indica* was the most effective as the widest inhibitory zone was observed as compared to the ethanolic as well as aqueous extract. It is inferred from the present investigation that demonstration of antibacterial activity of the studied flower extracts against both gram-positive and gram-negative bacteria may be indicative of the presence of broad-spectrum antibacterial compounds, suggests that there is a scientific basis for its utilization as antibacterial agents in designing and developing new drugs.

Key Words: Antibacterial activity, Agar well diffusion assay, *Quisqualis indica*, Solvents.

INTRODUCTION

Human beings have to depend on nature since his existence for survival. A man using his knowledge has discovered many remedies for aliments from nature, such as plants, mineral materials and animal products. The history of drug is intimately linked with plants from the earliest times and even today plant products have extensive use in ethno-medicine, traditional systems of medicines as well as in the armamentarium of the modern physician[1].

The interest in the study of medicinal plants as a source of pharmacologically active compounds has increased worldwide[2]. Various parts of the plants like root, bark, seeds and leaves have been an important source of medicine for thousands of years. In recent years a predominant interest has been observed in evaluating different plant extracts for their antibacterial properties. The World Health Organization (WHO) estimated that 80% of the populations rely on traditional medicines, mostly plant drugs, for their primary health care needs in developing countries. Scientists in many parts of the world have carried out extensive research and have proven to humanity, the effective use of herbal medicine[1].

The Indian region is very rich in ethnobotanical heritage due to its rich cultural diversity[3,4]. In India, more than 16,000 species of higher plants are available, of which nearly 9,000 are known to be economically useful. The varied topography, tropical climate and heavy rainfall are major causes of the richness and diversity of the medicinal flora of this region [5-7].

Medicinal plants exhibited a rich source of antimicrobial compounds which are used in different countries and are a source of many potent and powerful drugs. The extracts of different parts of a wide range of medicinal plants are used as raw drugs as well as they possess varied medicinal properties.

The local communities and folk healers collected these raw drugs in smaller quantities for local use. Also, many other raw drugs are collected in larger quantities and traded in the market for many herbal industries[8]. There are hundreds of plant species have been tested for antimicrobial activities, while the majority were not evaluated adequately[9]. The incidence of adverse reactions to these plant preparations are relatively lower as compared to the modern conventional pharmaceuticals, coupled with their reduced cost, which is promoting both the consuming public and national health care institutions to consider plant medicines as alternatives to synthetic drugs. Plants with possible antimicrobial activities have been identified by a number of researchers in different parts of the world[10-14.] Much work has been done on ethnomedicinal plants in India[14-17]. It has been suggested that aqueous and ethanolic extracts of plants used in allopathic medicine are potential sources of antiviral, antitumoral and antimicrobial agents[14,18]. The selection of crude plant extracts for screening programs has the potential of being more successful in the initial steps than the screening of pure compounds isolated from natural products.

Quisqualis indica Linn. commonly known as Rangoon creeper is an excellent vine for outdoor gardens belonging to family Combretaceae. It is a strong climber, ligneous vine that can reach from 2.5 m to upto 8 m. This plant is indigenous in Africa, Indo Malaysian region and cultivated all over India. Flowers numerous, pendent, 7.5 cm long, 3.8 cm wide. At first they are white in color, then they become deep red[19]. The present study was planned to evaluate the antibacterial activity of *Quisqualis indica* flower extracts with different solvents (methanol, ethanol and aqueous) against selected gram-positive as well as gram-negative bacterial strains.

MATERIALS AND METHODS

Plant collection and extract preparation

Flower material

Flowers of *Quisqualis indica* Linn. (Figure 1)were collected from my outdoor garden, Rohtak, Haryana, India and brought to the laboratory in polythene bags. Then the flowers were rinsed twice with distilled water and air dried on a clean sheet for one week at room temperature. It was made into small pieces using sharp, sterile scissors and powdered using sterile mortar and pestle.

Preparation of plant extract

Extraction was done at room temperature by the simple extraction method. For this, 5 g dried flower powder was homogenized using a sterile mortar and pestle in 5 ml of solvents (methanol, ethanol and aqueous) separately. The resulting homogenized solutions were incubated at room temperature for overnight for the extraction. Each extract was filtered with Whatman's Filter paper No. 1 and for the complete evaporation of the solvents, filtrate dried in oven. The dried extract was dissolved in solvent Dimethyl sulphoxide (DMSO) in a ratio of 100 mg/ml to determine the antibacterial activity by agar well diffusion assay (AWDA) method.

Source of Microorganisms

Three bacterial strains, including two gram-positive as well as one gram-negative were obtained from: *Bacillus subtilis* (Dr. Bijender Singh, Department of Microbiology), *Escherichia coli* strain DH5α (Dr. Krishan Kant, Department of Microbiology) and *Micococcus luteus* (MTCC106) (Dr. S. K. Tiwari, Department of Genetics) M. D. University, Rohtak, Haryana, India. These bacterial cultures were grown in nutrient broth medium, pH 7.0. Stock cultures were maintained on a nutrient agar slant, pH 7.0 at 4°C until needed. The media components were purchased from Hi-media, Mumbai, India.

Determination of antibacterial activity by agar well diffusion assay (AWDA) method

The antibacterial activity of the crude flower extracts (methanolic, ethanolic and aqueous) of *Quisqualis indica* Linn. against gram-positive as well as gram-negative bacterial strains were evaluated by agar well diffusion assay method[20]. For this, a well (6 mm diameter) was made with the help of a borer in cooled nutrient agar plate, overlaid with soft agar (5 ml), seeded with a target strain (~10^6 cfu/ml). Aliquots (100µl) of the test compound were introduced into the well and the plates were incubated overnight at 37 ^{o}C. The diameters of the inhibition zones were measured in millimeters (mm). For each bacterial strain, the dissolving solvent DMSO was used as negative control and streptomycin (1mg/ml) was used as positive control.

STATISTICAL ANALYSIS

The experiment was carried out in three independent sets, each consisting of 3 replicates. Values shown here represent mean ± standard error of the mean (SEM).

RESULTS AND DISCUSSION

Plants are an important source of potentially useful structures for the development of new chemotherapeutic agents. Researchers are increasingly turning their attention to herbal products, looking for new leads to develop better drugs against multiple drug resistant microbe strains[21]. There is an

urgent need to discover new antimicrobial agents for human and veterinary therapeutic uses, as resistant to current drugs increases in severity and extents[22]. Plants are recognized for their ability to produce a wealth of secondary metabolites and mankind has used many species for centuries to treat a variety of diseases[23].

Many reports are available on the antiviral, antibacterial, antifungal, antihelmintic, antimolluscal and anti-inflammatory properties of plants[24,25]. Some of these observations have helped in identifying the active principle responsible for such activities and in the developing drugs for the therapeutic use in human beings.

There are many literatures reporting the ethno-medicinal values of *Quisqualis indica* Linn., but there is little scientific proof for further using this plant commercially or in a more effective form[26-29]. Therefore, in order to evaluate the antibacterial activity of *Quisqualis indica* Linn. flower extracts against gram-positive as well as gram-negative bacterial strains was assessed by the AWDA method. In the present study, the crude flower extract showed excellent antibacterial activity against both tested two gram-positive and one gram-negative bacterial strains as shown in Table 1. The resulted data shows, *M. luteus* was the most sensitive among all the tested bacterial strains with the zone of inhibition (mm) 30 (methanol), 27 (ethanol), 23 (aqueous). However the gram-negative bacterial strain *E. coli* exhibited less sensitivity as compared to the other two gram-positive bacterial strains (*M. Luteus and Bacillus subtilis*) with the zone of inhibition (mm) 22 (methanol), 20 (ethanol), 16 (aqueous). The present study revealed that the significant antibacterial activity was shown against both tested gram-positive as well as gram-negative bacterial strains. However, methanolic extract showed the greater zone of inhibition as compared with the used standards antibiotic streptomycin against all the tested bacterial strains. In contrast, no inhibition zones were observed against DMSO. The results indicate that the different solvent extracts showed inhibition of growth against tested bacterial strains with to the various degrees.

It is recognized that in the extraction preparation the successful prediction of extracting compounds from plant material is mostly dependent on the type of solvent used, and the traditional practitioners were using the water as a primary solvent. However, the results received from the present study it was observed that the methanolic extract of *Quisqualis indica* Linn., was the most effective as the widest inhibitory zone was noted as compared to the ethanolic as well as aqueous extract as shown in Table 1. This may be due to better solubility of the active compounds in organic solvents[30]. Many studies have been reported that the methanol extracts inhibited the growth of testing bacteria more than aqueous extracts[31-33]. The present study also supports the above observations. In the present investigation only crude flower extracts are used and hence further investigation and identification of components are needed.

CONCLUSION

In the above study, the crude flower extracts of *Quisqualis indica* showed remarkable antibacterial activity against tested two gram-positive (*M. luteus and Bacillus subtilis*) and one gram-negative (*E. coli*) bacterial strains. The data revealed that the methanolic extract of the flower showed better antibacterial activity as compared to the other two solvent extracts, which is attributed to the presence of some active components in the flower extracts. The presentation of a wide spectrum of antibacterial activities of the flower extracts tested in this study may help to discover new chemical classes of antibiotic substances. The evolution of natural antimicrobial agents will help to diminish the negative effects like pollution in the environment, resistance, etc., of synthetic chemicals as well as drugs. It can truly contribute to medical and pharmaceutical practices, yet, there are many more activities waiting for screening the drugs. Further research has to be conducted on the activity of the extracts against a wider range of bacteria and fungi.

COMPETING INTERESTS

The authors declare that they have no competing interests.

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Table 1
Antibacterial activity of flower extracts of *Quisqualis indica* Linn.

Zone of Inhibition (mm)			
Flower extracts in various solvents	***M. luteus***	***Bacillus subtilis***	***E. coli***
Methanol	30	28	22
Ethanol	27	25	20
Aqueous	23	22	16
Streptomycin (+ ve control)	28	26	23
DMSO (- ve control)	0	0	0

Figure 1
***Quiqualis indica* Linn., studied plant**

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