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Research Paper

EVALUATION OF ANTIBACTERIAL ACTIVITY OF *Punica granatum*

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Abstract

Plants have been use from thousands of years to conserve food and treat health disease. In Ayurveda many plants have mentioned having potential medicinal values. From mentioned plants *Punica grantum* (pomegranate) shows more medicinal properties. *Punica grantum* (pomegranate) commonly used in India as a traditional medicine for treatment of pathogenic bacteria. The present study investigates comparative study of antibacterial activity of pomegranate rind; leaves extract (Organic & Aqueous) against various enteric pathogens. The pomegranate serves as an ancient, mystical, unique small fruit and it also called as Dalimb. In Ayurveda pomegranate refers as Blood tonic. In addition to historical uses they are used in several system of medicine for verity of alignments. For controlling growth of microorganism; nowadays there has been an increasing interest in extracting relevant natural antimicrobial agents as potent as the chemical antibiotic to be used as alternative approach so pomegranate has taken great attention for its potent antimicrobial activity as bacteria getting resistance to chemical antibiotics. In lieu of above justification; present study aimed at elevating the In vitro antibacterial studies of extract of leaf, rind of pomegranate against enteric bacteria (*Escherichia coli (1692), Staphylococcus aureus (2078), Salmonella typhi (2501), Proteus vulgaris (742), Aerobacter aerogen (10699*) which causes diarrhea and enteric disorders.

Key words: *Punica granatum*, enteric bacteria, Peel extract, Antibacterial activity, Organic solvent.

INTRODUCTION

The antimicrobial substances are those which kill or inhibit growth of microorganism such as bacteria, fungi, protozoa. Antimicrobial substance contains antimicrobial protein which produces by all species of life including bacteria; they enhance the immunity by stimulating adaptive immune system. [1]

The steadily increasing bacterial resistant to existing drug is a serious problem of antibacterial therapy plants have been shown to be good alternative to synthetic chemical antimicrobial drugs and antibiotics as microorganisms shows resistance after some time and these agents shows serious side effects. The WHO (world health organization) indicates that as many as 80% of all population living in world make use of herbal medicines as main source for healthcare. Among the medicinal plants *Punica granatum* commonly known as pomegranate belonging to the family *Punicaceae* has long been estimated as food of medicine and it is one of important ethno medicines plant used in tradition and folklore therapies. It takes in diet after diarrhea. It is used in Siddha; Ayurveda especially for treatment of Gastro Intestinal Disease (GID) In Ayurveda pomegranate is considered "a pharmacy unto itself" and used as anti-parasite

agent. A "Blood tonic" and heal apathies, diarrhea, ulcers [2].The aim of present study deals with finding out Antibacterial activity in different parts of *Punica granatum*. For this purpose peel, leaf, red seed extracts were prepared in aqueous and organic solvents. [3]

MATERIALS AND METHODS

Materials-

Nutrient broth (code- M002-100G), Agar Agar (code- RM666-500G), Methanol (code- 46048 L05), Whatmanns Filter paper No. 1. Chemicals are used from HiMedia Laboratories Pvt. Ltd. Mumbai and s d fine-CHEM Ltd Mumbai.

Method-

Collection of sample-

Plant sample was collected from the Botanical Garden of Shivchhatrapati College N-3 Cidco, Aurangabad, Maharashtra, India.

Authentication of plant-

Plant sample was authenticated in Department of Botany of Dr. Babasaheb Ambedkar Marathawada University, Aurangabad, Maharashtra, India.

Plant extract preparation-

Aqueous and organic extract of peel and leaf of *Punica granatum* was prepared by blending 15gm of peel and leaf in 45ml of distilled water, Methanol (80%) and Ethanol (70%). These crude extract was filtered through two layer of musclin cloth followed by Whatmanns Filter paper No. 1 and stored at -20°C as described by Pradeep [4].

Evaluation of Antibacterial activity-

Evaluation of antibacterial activity of extract was performed by modified Disc Diffusion Assay [5]. 24 hour old culture of selected bacteria (*Escherichia coli (1692), Staphylococcus aureus (2078), Salmonella typhi (2501), Proteus vulgaris (742), Aerobacter aerogen* (10699) was spread on nutrient agar plates. Sterile disc of Whatmanns Filter paper No. 1 having diameter of 0.5cm was placed at middle of plate.5ul respective extract was added on disc with micropipette and plates were incubated at 37°C for 24hours.

All procedures were performed in triplets and antibacterial activity was expressed as zone of inhibition in cm.

RESULTS AND DISCCUSION

Nearly 80% of the world populations depend on the traditional medicine for primary health care, mainly including the use of natural products [4]. Researchers have extensively studied the biological properties of *Punica granatum* and their results showed that this plant is ethno medically valuable [2].

Authentication of plant-

Punica granatum plant was authenticated before extract preparation with Accession number- 0554.

Extract preparation-

The extract was prepared as mentioned by B.V. Pradeep [4] as in Figure -1.

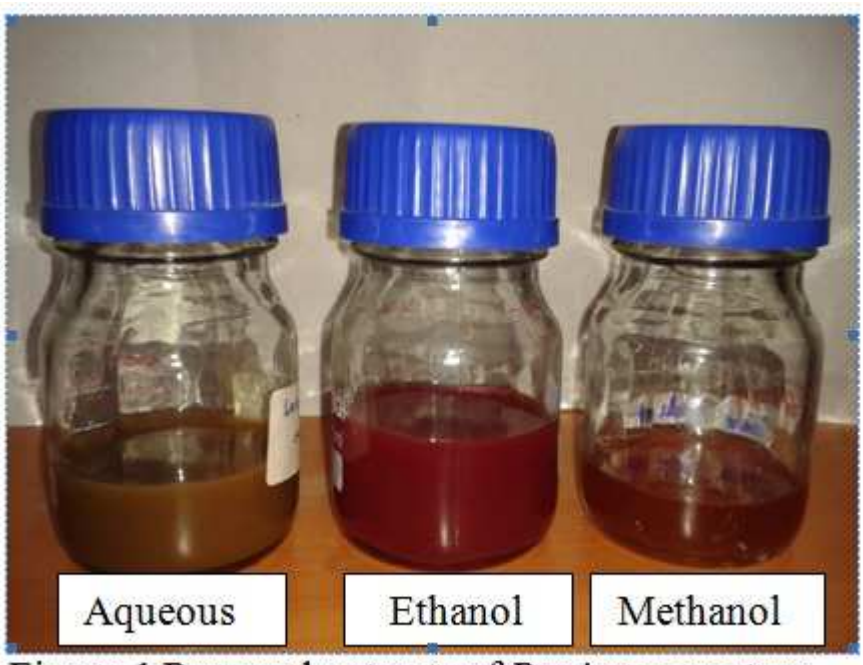
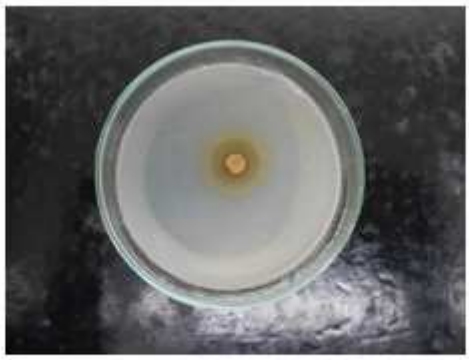


Figure-1 Prepared extracts of *Punica granatum.*

Evaluation of antibacterial activity of *Punica granatum*-

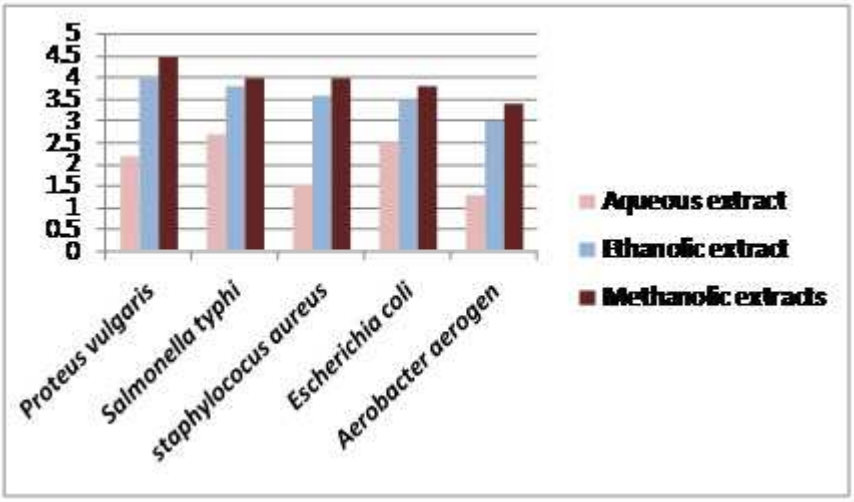
The antibacterial activity of the peel crude extract resulted in clear inhibition zones of 1.3cm for all the strains tested (*Escherichia coli (1692), Staphylococcus aureus (2078), Salmonella typhi (2501), Proteus vulgaris (742), Aerobacter aerogen (10699*) similar results were reported by Saad Sabbar [6]. (Graph -1)

S. Vijyanand [8] reported methanolic extract gives Zone of inhibition 2.8cm where as in our work Methanolic extract gives highest peak of 4.5cm with *Proteus vulgaris (Figure-2)*.



(Figure -2) *Proteus vulgaris* treated with methanolic peel extract.

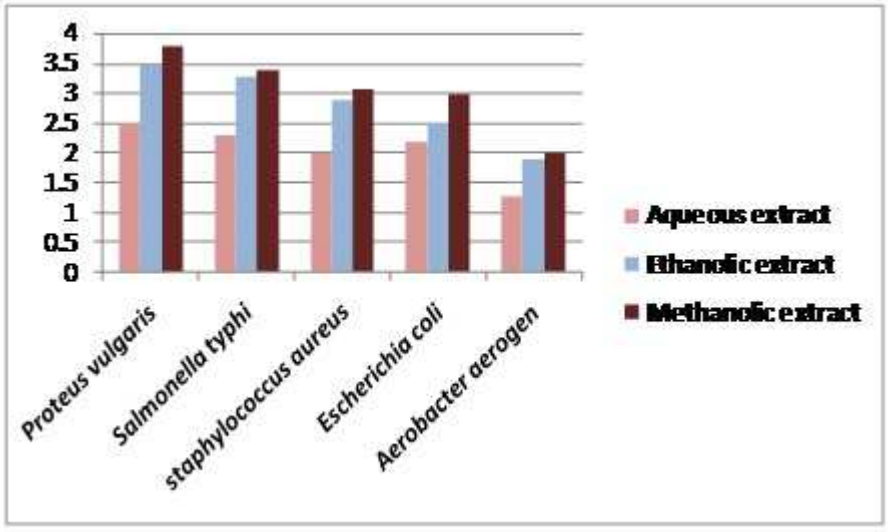
The results of Disc diffusion test indicated that organic extracts of *Punica granatum* peel showed more antibacterial activity (Graph-1) on tested culture of bacteria (*Escherichia coli (1692), Staphylococcus aureus (2078), Salmonella typhi (2501), Proteus vulgaris (742), Aerobacter aerogen*) as mentioned by Pradeep [4]. In current study different plant parts *Punica granatum* were comparatively analyzed for its antibacterial activity. Jayaprakasha [3] have reported antibacterial activity of *Punica granatum* rind and have obtained zone of inhibition in between 1cm and 2.5 cm. in present study zone of inhibition for the same is in between 1.3 to 4.5cm. (Graph -1)



Graph-1 - Comparative study of Aqueous, Methanolic and Ethanolic extracts of *Punica granatum* Peel extracts.

Punica granatum leaf also shows Antibacterial activity [6]. In present study methanolic extract of *Punica granatum* leaf shows zone of inhibition as 3.8cm against *Proteus vulgaris* (742) similar results were reported by Saad Sabbar [6].

Figure-3 *Proteus vulgaris* treated with methanolic peels extract.



Graph-2- Comparative study of Aqueous, Methanolic and Ethanolic extracts of *Punica granatum* Leaf extracts.

CONCLUSION

With the current spread of antibiotic resistance increasing in potent microorganisms there is a need to pay attention to plants and their products as potential antimicrobial agents. The results obtain in present study demonstrate broad spectrum antibacterial activity of *Punica granatum* L. against both Gram positive and Gram negative bacterial strains (*Escherichia coli (1692), Staphylococcus aureus (2078), Salmonella typhi (2501), Proteus vulgaris (742), Aerobacter aerogen* (10699). The disc diffusion bioassay showed that methanolic peel and leaf extracts gives maximum antibacterial activity against *Proteus vulgaris* (742) viz. 4.5cm and 3.8cm respectively.

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