Inhibitory activity of Immunobooster Tea against pathogens

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ABSTRACT-Indian medicinal plants manifest miraculous effects in curing a vast range of diseases and disorders among humans and can be better called as "elixirs of life." Currently, there is much growing interest in the use of these medicinal plants as immunomodulators of the complex immune system. Through a number of vast researches conducted in the area, it is being explored that many of the chemicals in the form of alkaloids, flavonoids, terpenoids, polysaccharides, lactones, and glycoside products are responsible to cause alterations in the immunomodulatory properties. Keeping in mind, the tremendous potential of the medicinal plants and their derived drugs, a study is designed to evaluate the inhibitory antibacterial activity of a immunobooster tea formulation against different bacterial strains using methanol, Hexane and water as solvents.

Keywords:Immunity; Immunobooster tea; Immunomodulators, Antimicrobial activity

INTRODUCTION

Traditional medicine research has given high priority to immune boosting medicinal plants due to the prevalence of diseases like HIV/AIDS and other viral diseases which affect the immune system (Devasagayam and Sainis,2002). However, very little information on plants with immune boosting potentials have been documented (Gupta,1994).

Through a number of researches conducted in the area have explored that many of the chemicals in the form of alkaloids, flavonoids, terpenoids, polysaccharides, lactones, and glycoside products are responsible to cause alterations in the immunomodulatory properties(Wadood et al,2013).

The term immunity defines body's natural defense system against a vast array of diseases and disorders. Remarkably sophisticated and advanced among vertebrates, the complex immune system is capable to generate a limitless variety of cells and molecules to arrest enormous spectrum of infections and undesirable substances. Immunomodulators refer to those substances capable of inducing, amplifying, and inhibiting any component or phase of the immune system. Immunostimulators and immunosuppressant are two types of immunomodulators are known for use. In fact, immunopharmacology is a newer branch of pharmacology concerned with immunomodulators(Patil,2012). Administration of immunostimulators as in the case of AIDS and use of immunosuppressor in cases of an exaggerated response of an immune system is appreciating to reconstitute the normal immune system and increase the longevity of life. Immunomodulator intake along with antigen, the process is meant to boost the immune system, and the modulator is known as immune adjuvant(Dutt,2013).

Plants produce a diverse range of bioactive molecules, making them rich source of different types of medicines. Most of the drugs today are obtained from natural sources or semi synthetic derivatives of natural products and used in the traditional systems of medicine. Approximately 20% of the plants found in the world have been submitted to pharmaceutical or biological test and a sustainable number of new drugs introduced in the market are obtained from natural or semi synthetic resources. It has been reported that between the years 1983

and 1994 (Cragg et al., 1999), the systematic screening of antibacterial plant extracts represents a continuous effort to find new immunomodulator compounds with also have the potential to act against bacteria and viruses.

According to World Health Organization (Santos et al., 1995) medicinal plants would be the best source to obtain a variety of drugs. Current advancements in drug discovery technology and search for novel chemical diversity have intensified the efforts for exploring leads from Ayurveda the traditional system of medicine in India.

Ayurvedic system of medicine has its long history of therapeutic potential. The use of plant extracts and phytochemicals both with known antimicrobial properties is of great significance, in the past few years a number of investigations have been conducted world wide to prove antimicrobial activities from medicinal plants (Alonso-Paz et al., 1995;Nascimento et al., 1990).

There is a continuous and urgent need to discover new antimicrobial compounds with diverse chemical structures and novel mechanisms of action for new and re-emerging infectious diseases (Rojas et al., 2003). Therefore,researchers are increasingly turning their attention to folkmedicine, looking for new leads to develop better drugs against microbial infections (Benkeblia, 2004). Reports are available on the use of several plant by-products, which posses antimicrobial properties, on several pathogenic bacteria and fungi (Bylka et al., 2004; Shimpi and Bendre, 2005; Kilani,2006).

The current research in the area to develop plant-derived natural products as potent and safer leads to act as immunomodulators, is gaining much interest. Generation of herbal medicine as multiple-component agent expected to modulate the complex immune process in such a way so as to prevent the infection rather than treatment and cure of the disease. With all these aspects keeping in mind, the present work focuses on an antimicrobial activity of a immunobooster tea formulation.

Material and Methods

Collection of plant material

The immunobooster tea formulation studied consists of a combination of 15 indigenous medicinal herbs were collected from in and around Dehradun (Uttarakhand state, India).The Herbs were identified based on the taxonomical characteristics by Dr.Mayaram Uniyal, Department of Pharmacognosy, The Himalaya Drug Company Dehradun, India. The dried herbs were powdered and used for extraction.

Test microorganisms

The antibacterial activity of the extracts were tested individually on G+ve and G-ve bacterial strains .All bacterial strains were obtained from IMTECH, Chandigarh India. The G+ve strain used was *Staphylococcus aureus MTCC 737 and* G-ve bacterial strains were *E.coli MTCC 1687;Psuedomonas aeruginosa MTCC 1688* , *Salmonella enterica MTCC 3858* and *Candida albicans MTCC 3017.*

Preparation of aqueous extract

The finely powdered plant materials (100 grams) were boiled in 500 ml distilled water till one-fourth of the extract initially taken was left behind after evaporation. The solution was first filtered through double layered muslin cloth and centrifuged at 5000 g for 30 min and the supernatant was filtered through whatman No.1 filter paper under strict aseptic conditions and then the filtrate was collected in fresh sterilized bottles and stored at 4°C until further use.

Preparation of solvent extract

100 grams each of the powdered material was extracted with 500ml of methanol &Hexane separately for 24hrs. The extract were filtered with sterile whatman filter paper No.1 into a

clean conical flasks. The solvent along with the sample was transferred into the sample holder of the rotary flash evaporator for the evaporation of the solvent. The evaporated solvent so obtained was weighed and preserved at 4$_o$C in airtight bottles until further use.

Determination of antimicrobial activity through Agar-well diffusion assay:

Suspension of 24 h cultures of *Escherichia coli, Staphylococcus aureus, Salmonella enterica ,Pseudomonas aeruginosa and Candida albicans* was made in sterile normal saline. Each labeled medium plate was uniformly inoculated with a test organism by using a sterile cotton swab rolled in the suspension to streak the plate surface in a form that lawn growth can be observed. A sterile cork borer of 5mm diameter was used to make wells on the medium. 100 µL of the various extract concentration were dropped into each, appropriate well(Atata et al;2003& Bonjar,2004).Methanol solvent used for extraction apart from water & Hexane was tested for each organism. The inoculated plates were kept in refrigerator for 2 hours to allow the extracts to diffuse into the agar. The agar plates were incubated at 37°C for 24 h. Antimicrobial activity was determined by measuring the diameter of zones of inhibition (mm) produced after incubation. 30 µg of standard antibiotic streptomycin was used as positive control and respective solvents as negative controls.

Results and Discussion

The efficacy of Hexane,methanol and aqueous extracts of immunobooster tea formulation against

pathogenic bacteria showed varied level of inhibition (Table -1). It was revealed from the result that the formulation showed different degrees of inhibition against different microorganisms. The maximum zone of inhibition was observed in the case of *Staphylococcus aureus* (28mm) due to action of methanol extracts followed by *Salmonella enterica*(26mm) followed by *Pseudomonas aeruginosa*(25mm).

The results revealed variability in the bactericidal concentration of each extract for given bacteria. It was clear from the present result that methanol extract exhibited pronounced activity against all the tested four bacteria and *Candida albican*. The highest antibacterial activity as seen with methanol extract might be due to the presence of alkaloids and tannins(Okemo,1996). Broad spectrum activity of methanol extract tended to show that the active ingredients were better extracted with methanol. Earlier studies had also shown the greater antibacterial activity of methanol extracts than other solvent extracts(Aqil et al.2003 & Kannan et al;2009)With least antibacterial activity as seen with other solvent extracts, might be due to loss of some active compounds during extraction process of the sample and lack of solubility of active constituents in the solvent(Sampathkumar,2008).

In spite of this permeability difference between Gram positive and Gram negative bacteria, the methanol extract had a broader spectrum of inhibitory activity. This showed the involvement of more than one active principle of biological significance.This study does not only show the scientific basis for some of the therapeutic uses of these herbs formulation in traditional medicine, but also confirms the fact that ethnobotanical approach should be considered when investigating antimicrobial properties of plants(Iwu,1993 & Adesanya,2005).

Conclusion

On the basis of the results obtained, it can be concluded that methanol can be used for extracting antimicrobial compounds from herbs

and formulations. The present study shows that immunobooster tea extracts possessed the antimicrobial activity against some organisms associated with infections and are highly resistant against antibiotics. Therefore, it suggests that this formulation can be a source of oral drugs to be used in the treatment of opportunistic infections and may be a source for future drug formation.

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