



EVALUATION OF ANTIMICROBIAL ACTIVITY OF DIFFERENT SOLVENT EXTRACTS OF SAUSSUREA LAPPA

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ABSTRACT

Antimicrobial efficiency of *S.lappa* (root extracts) was examined using different kinds of solvents [Methanol, chloroform, and water] and tested against five human pathogens like Bacteria: *Bacillus cereus*, *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, Fungi: *C.Albicans* using disc diffusion method to evaluate the antimicrobial activity. The plant extracts showed significant activity against all pathogens except *E.coli* and *P.s*, but the alcoholic extract [methanolic extract] of *S.lappa* showed maximum zone of inhibition against *S.aureus* and *B.subtilis* (Table 2). The Spectrum of activity observed in the present study may be indicative that these plants could be possible source of new and effective herbal medicines to treat infections, hence justified the ethnic uses of *S.lappa* against various infectious diseases.

KEYWORDS: Antimicrobial activity, medicinal plants, Disc diffusion method.

INTRODUCTION

Many of the plant materials used in traditional medicine are readily available in rural areas. Medicinal plants are valuable source of natural active constituents that are used to maintain human health and also used for the treatment of many human diseases.^[1] The use of plant and its products has a long history that began with folk medicine and through the years has been incorporated into traditional and allopathic medicine.^[1] Since antiquity, many plants species reported to have pharmacological properties as they are known to possess various secondary metabolites like glycosides, saponins, flavonoids, steroids, tannins, alkaloids, and terpenes which are therefore, should be utilized to combat the disease causing pathogens.^[2,3,4] With the

advancement in Science and Technology, remarkable progress has been made in the field of medicine with the discoveries of many natural and synthetic drugs.^[5] Antibiotics are undeniably one of the most important therapeutic discoveries of the 20th century that had effectiveness against serious bacterial infections. However, only one third of the infectious diseases known have been treated from the synthetic products.^[6] This is because of the emergence of resistant pathogens that is beyond doubt the consequence of years of widespread indiscriminate use, incessant and misuse of antibiotics.^[7,8] Antibiotic resistance has increased substantially in the recent years and is posing an ever increasing therapeutic problem. One of the methods to reduce the resistance to antibiotics is by using antibiotic resistance inhibitors from plants.^[9,10] Plants are known to produce a variety of compounds to protect themselves against a variety of pathogens. It is expected that plant extracts showing target sites other than those used by antibiotics will be active against drug resistant pathogens.^[11] *Saussurea costus* is frequently used in Korean traditional prescriptions for inflammatory diseases. Its total methanol extract at 0.1mg/ml as a final concentration exhibited more than 50% of inhibition on the cytokine induced neutrophil chemotactic factor (CINC) induction.^[12]

MATERIALS AND METHODS

The plant samples used for this study were collected from Omdurman local market. Different plant Extracts (Methanol, Petroleum Ether and Water) were used for further studies.

Preparation of plant extract

The root of plant sample was collected and washed for 2-3 times with tap water and finally with distilled water, followed by ethanol wash and then allowed to dry at 50°C for overnight. Dry roots were milled to coarse powder. 100gm. of powdered material was macerated in to different solvents by using sterile conical flasks. All the extracts were evaporated and dried by freeze drying technique. All extracts were stored in sterile glass bottles at room temperature until screened^[13]

Preparation of test samples

The dried methanolic, water, and chloroform extracts were respectively dissolved in sterile, dimethyl sulfoxide (DMSO) and sterile water at selected concentrations.^[14]

Microorganisms tested and culture media

The strains of bacteria and fungi were grown in Nutrient Agar Media and Nutrient Agar Broth and the species of different fungi were grown in Sabouraud Dextrose Agar and Sabouraud Dextrose Broth. The concentration of bacterial suspension was adjusted to 10^8 cells/ml, and that fungal suspension was adjusted to 10^7 Cells/ml.^[15]

Media Preparation and Its Sterilization

Disc diffusion method for antimicrobial susceptibility was tested on solid (Agar-agar) media in petri plates.^[16,17] For bacterial assay nutrient agar (NA) (40gm/L) and for fungus PDA (39gm/L) was used for developing surface colony growth. The suspension culture, for bacterial cells growth was done by preparing 2% Lauria Broth (w/v), and for fungus cells growth, 2.4% (w/v) PDB (Potato dextrose broth) was taken for evaluation. All the media prepared were then sterilized by autoclaving the media at (121°C) for 20 min.

RESULTS

Table No1: Antibacterial and antifungal activity of methanolic extract of *S.lappa*{diameter of inhibition zone in mm}.

Conc.	<i>E.coli</i>	<i>P.S</i>	<i>S.aureus</i>	<i>B.Subtilis</i>	<i>C.albicans</i>
100mg/ml	R	R	25	26	17
50mg/ml	R	R	19	24	----
25mg/ml	R	R	18	----	----
12.5mg/ml	R	R	16	----	----
6.25mg/ml	R	R	14	----	----

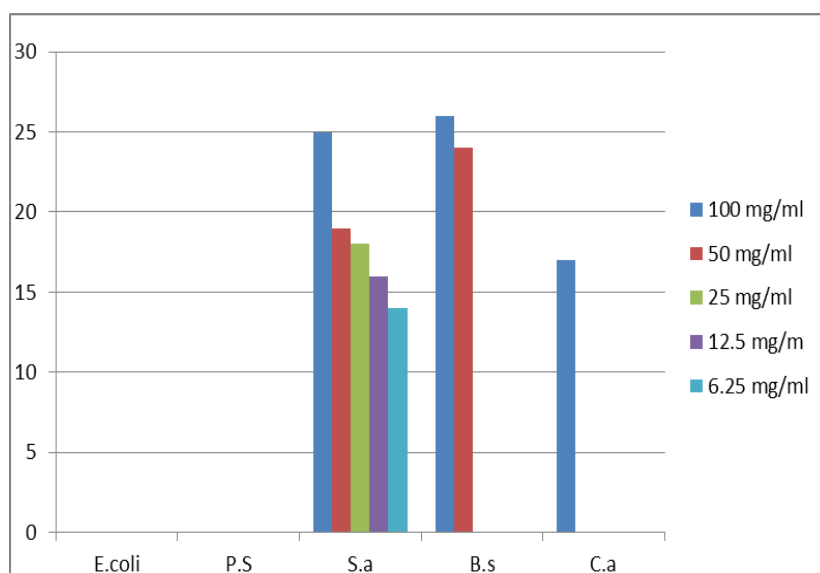


Chart No (1): Antibacterial and antifungal activity of methanolic extract of *S.lappa*. {diameter of inhibition zone in mm}.

Table No (2): Antibacterial and antifungal activity of water extract of *S.lappa* {diameter of inhibition zone in mm}.

Conc.	<i>E.coli</i>	<i>P.S</i>	<i>S.aureus</i>	<i>B.Subtilis</i>	<i>C.albicans</i>
100mg/ml	R	R	18	21	11
50mg/ml	R	R	16	16	----
25mg/ml	R	R	9	----	----
12.5mg/ml	R	R	8	----	----
6.25mg/ml	R	R	----	----	----

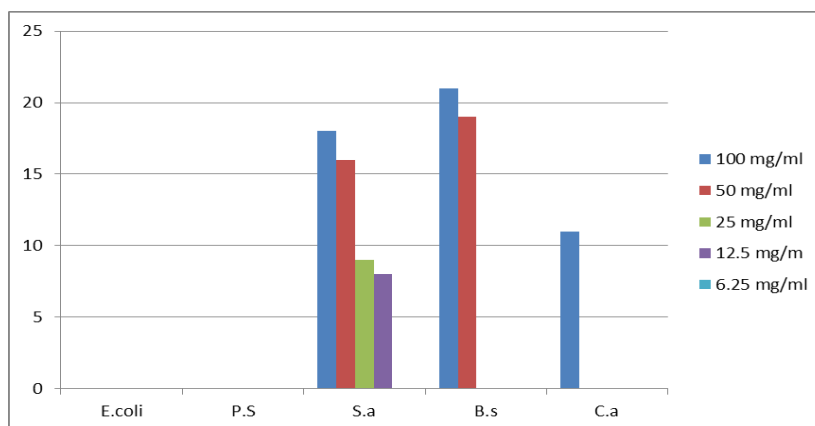


Chart No (2): Antibacterial and antifungal activity of water extract of *S.lappa* {diameter of inhibition zone in mm}.

Table No (3): Antibacterial and antifungal activity of chloroform extract of *S.lappa* {diameter of inhibition zone in mm}.

Conc.	<i>E.coli</i>	<i>P.S</i>	<i>S.aureus</i>	<i>B.Subtilis</i>	<i>C.albicans</i>
100mg/ml	R	R	22	24	11
50mg/ml	R	R	18	19	6
25mg/ml	R	R	16	11	0
12.5mg/ml	R	R	14	0	0
6.25mg/ml	R	R	12	0	0

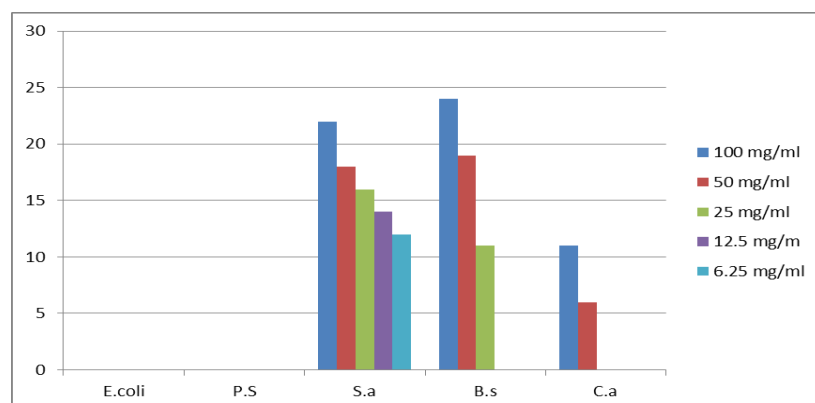


Chart No (3): Three Antibacterial and antifungal activity of chloroform extract of *S.lappa*{diameter of inhibition zone in mm}.

- *P.S: Psuedomonas, S.aureus: Staphylococcus aureus.*
- *B.subtilis: Bacillus subtilis, C.albicans: Candida Albicans.*
- [----]: indicated for unclear result or diameter of inhibition zone.
- [R]: indicated for resistance.
- [S]: indicated for sensitive.
- [M]: indicated for moderate effect.

DISCUSSION

The research for antimicrobials from natural sources has received much attention and efforts have been put to identify compounds that can act as suitable antimicrobial agents to replace synthetic ones. Phytochemicals derived from plant products serve as a prototype to develop less toxic and more effective medicines in controlling the growth of microorganism.^[18,19] These compounds have significant therapeutic application against human pathogens including bacteria, fungi or virus. Numerous studies have been conducted with the extracts of various plants, screening antimicrobial activity as well as for the discovery of new antimicrobial compounds.^[20] In the present investigation, different solvent extracts of *S.lappa* were evaluated for exploration of their antimicrobial activity against certain Gram negative and Gram positive bacteria and fungi which were regarded as human pathogenic microorganism. Susceptibility of each plant extract was tested by disc diffusion method. Our preliminary investigation showed that all Methanol, chloroform and aqueous extracts of *S.lappa* were active against the locally isolated human pathogens like *Staphylococcus aureus*, *Bacillus cereus*, and whereas *Escherichia coli* and *Pseudomonas aeruginosa* were resistant. This result indicates that the extracts of *S.lappa* are active against gram positive bacteria besides their antifungal activity.

Although, the mechanism of the action of these plant constituents is not yet fully known it is clear that the effectiveness of the extracts largely depend on the type of solvent used. The organic extracts provided more powerful antimicrobial activity as compared to aqueous extracts.

In conclusion, of the present investigation the methanol, chloroform and aqueous extracts of *S.lappa* possess significant inhibitory effect against gram positive bacteria with antifungal activity of the tested pathogens.

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