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RESEARCH ARTICLE

EFFECT OF DIFFERENT PLANT EXTRACTS AGAINST CARBUNCLE CAUSING PATHOGEN STAPHYLOCOCCUS AUREUS – A COMPARATIVE ACCOUNT

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ABSTRACT

The use of plants in the treatment of carbuncle is common in traditional medicine at Purulia. Based on ethnopharmacological and taxonomic information, antibacterial activities of aqueous, ethanol, acetone, petroleum benzene and chloroform extracts of four medicinal plants were determined by in vitro agar diffusion method against *Staphylococcus aureus* which causes for Carbuncle. The roots, bulb, aerial part, and leaves of four different plants, of different family were studied for antibacterial activity. Powdered materials of all the selected plant parts were extracted with the help of different solvents (water, ethanol, acetone, petroleum benzene and chloroform) using soxhlet apparatus and antibacterial activity was observed against *S aureus*. The significant results were obtained in *U indica* bulbs and *C anthemoides* roots. This study experimentally strengthen the concept of the tribal traditional knowledge to cure carbuncle.

Key words:

Antibacterial activity; Carbuncle;
Staphylococcus aureus;
Ethnomedicinal Plants.

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INTRODUCTION

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 & 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, phenols, terpenoids which is therefore should be utilized to combat the disease causing pathogen (2-4).

With the advancement in Science and Technology, remarkable Progress has been achieved 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 disease known have been treated from these 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). In general, bacteria have the genetic ability to transmit and acquire resistance to drugs, which are utilized as therapeutic agents (9).

The Problem of microbial resistance is growing and the outlook for the use of antimicrobial drugs in the future is still uncertain. Therefore, actions must be taken to reduce this problem, for example, to control the use of antibiotic, develop research to better understand the genetic mechanisms of resistance & to continue studies to develop new drugs, either synthetic or natural. The ultimate goal is to offer appropriate and efficient antimicrobial drugs to the patient.

Over the past twenty years, there has been a lot of interest in the investigation of natural materials as source of new antibacterial agents (10, 11). Until natural Products have been approved as new antibacterial drugs, there is an urgent need to identify novel substances active towards highly resistant pathogens (12, 13). According to WHO the traditional medicine are proven to be efficacious & safe(14) and about 80 % of the world population is dependent on the traditional medicines and a major part of traditional therapies involves the use of plant extracts or their active constituents' Purulia is the good source of medicinal plants in west Bengal. About 85% of the rural Populations of Purulia depends on wild plants for the treatment of carbuncle. Folk herbal remedies used against carbuncle in Purulia, the antibacterial activity of that herbals has been dealt with in the present work. Carbuncle is group of

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boils, which is caused by methicillin-resistant *S. aureus* (15). It is a red swollen and painful cluster of boils that are connected to each other under the skin. Most of the carbuncles are caused by *S. aureus* which inhabits the skin surface, throat and nasal passages. If it is untreated, it may cause serious complications like sepsis and infections in other parts of the body.

In the present Study different plant extracts are used to inhibit the growth, of bacterial strain *S. aureus*. Different solvents are used for this study to determine the effectiveness of plant extracts. From this study it is proved that out of 4 different plant extracts acetone extract of *Cotula anthemoides* root and ethanol extract of *Urginea indica* bulb is more effective.

MATERIALS AND METHOD

Selection of plants

Selection of plants based on ethnomedicinal uses against carbuncle in Purulia district. About four medicinal plants (table 1) their vernacular names, parts used, for the treatment of carbuncle have been presented in the table 1 below. Plants are identified with the help of authentic specimen, books, journals and floras (16-21) and documented in the herbarium of A.M. College, Jhalda.

Table 1 Plants selected for experiments

SL No.	Scientific name	Vernacular name	Parts used
1	<i>Cotula anthemoides</i>	Pisainandi	Whole
2	<i>Urginea indica</i>	Banpiyaj	Bulbs
3	<i>Curculigo orchioides</i>	Talmuli	Roots
4	<i>Ammona squamosa</i>	Atapata	Leaves

Collection of plant samples

Medicinal Plant samples were collected during rainy season, 2013 from Purulia district, West Bengal, India and it is available in any season of year.

Extraction

Plant material extraction process

After collection of plant materials, they are cutted in small pieces and they are dried under shade for 12 days. After drying in finally grinded in powder by grinder machine. Then the powdered material was extracted with acetone, ethanol, petroleum benzene, chloroform, and distilled water using soxhlet Apparatus.



Fig. 1. Preparation of plant extract by using Soxhlet apparatus



Fig. 2. Different Plant Extracts

About 10 grams of powder was loaded in soxhlet extraction unit and exhaustively extracted using 100ml of solvents such as acetone, ethanol, methanol, chloroform, distilled water and chloroform respectively at 60°C for 12 hours. Thereafter, it was filtered with the help of Whatman No.1 filter paper and use for antimicrobial tests.

Bacterial strain and culture conditions

Bacterial sample *Staphylococcus aureus* (*S. aureus* MTCC 3160) were provided by microbiological laboratory and clinical detection center Paschim Medinipur, India. They were cultured in tryptone soybroth or agar (TSB or TSA) in aerobic condition at 37 °C.

Antibacterial Activity Assay

Antibacterial assay was carried out by Agar well diffusion method. Fresh microbial culture of 100µl (10⁶ cells/ml) was spread on Muller Hilton Agar plate with cotton swab. A well of 6mm diameter was punched off into agar medium with sterile cork borer and filled with 100µl of each (100µg/ml) ethanol, acetone, petroleum ether, distilled water and chloroform extracts by using micropipette in each well in aseptic condition. The petriplates were then kept in a refrigerator to allow pre diffusion of extracts for 30 minutes and further incubated in an incubator at 37°C for 30 minutes for 24 hours extroverted position. The antibacterial screening was evaluated by measuring the zone of inhibition. An inhibition zone of 6mm or greater (incubating diameter of well) was considered antibacterial activity. The experiment was done in triplicate and the mean diameter of inhibition zone was calculated. The results showed that the remarkable inhibition of the bacterial growth was against the tested organism (22).

RESULTS

The ethanol extracts of *Urginea indica* bulbs (IZ 15.50±0.25 mm) and acetone extract of *Cotula anthemoides* roots (IZ 13.45±0.20mm) were found to be more active against *S. aureus* compared to other solvent extracts. While minimum zone of inhibition was observed in ethanol extract of *Curculigo orchioides*.



Fig. 3 Picture showing inhibition zone using plant extracts

Table 2 Antibacterial activity of plant extracts. Numbers indicate the mean diameters of inhibition of triplicate experiments \pm standard deviation.

Sl. No.	Plant species	Local name	Plant part extracted	Solvent used	Diameter of inhibition zone (mm)
1	<i>Curculigo orchiooides</i>	Talmuli	Roots	Acetone	NM
				Chloroform	NM
				Ethanol	6.5 \pm 0.2
				PetroleumBenzene	-
				Water	NM
2	<i>Cotula anthemoides</i>	Pisainandi	Aerial parts	Acetone	-
				Chloroform	-
				Ethanol	NM
				PetroleumBenzene	NM
				Water	-
3	<i>Cotula anthemoides</i>	Pisainandi	Roots	Acetone	13 \pm 0.2
				Chloroform	NM
				Ethanol	8.5 \pm 0.5
				PetroleumBenzene	-
				Water	9.5 \pm 0.5
4	<i>Urginea indica</i>	Ban piyaj	Bulbs	Acetone	8.5 \pm 0.5
				Chloroform	NM
				Ethanol	16.5 \pm 0.2
				PetroleumBenzene	NM
				Water	10.5 \pm 0.5
5	<i>Annona squamosa</i>	Atapata	Leaves	Acetone	8.5 \pm 0.5
				Chloroform	-
				Ethanol	11.5 \pm 0.45
				PetroleumBenzene	-
				Water	6.5 \pm 0.25

'-' indicates no growth inhibition'.

'NM' indicates zone of inhibition not measured.

DISCUSSION

Recently, much attention has been directed toward plant extracts and biologically active compounds isolated from popular plant species. The use of medicinal plants plays a vital role in covering the basic health needs in developing countries and these plants may offer a new source of antibacterial, antifungal and antiviral agents with significant activity against infective microorganisms. (23, 24).

The results conclude that some of the extracts of the plant parts used in the present investigation showed significant antibacterial activities against carbuncle causing pathogen. Thus ethanol extract of *Urginea indica* bulbs and acetone extract of *Cotula anthemoides* roots can be used in the treatment of Carbuncle. Further phytochemical analysis and in vivo studies are necessary to corroborate the findings.

CONCLUSION

The results of present investigation clearly indicate that the antibacterial activity vary with the plant parts used. The present investigation data on antibacterial potency of *U. indica* bulbs and *C. anthemoides* roots provide basis for synthesis of novel antibiotics. From our investigation of screening different plant parts, the result obtained confirm the therapeutic potency of plant used in traditional folkloric medicine and suggest that the plant extracts posses compound with antibacterial properties that can be used as antimicrobial agents in new drugs for the therapy of Carbuncle disease caused by *S. aureus*. In addition these results form a good basis for selection of candidate plant parts for further phytochemical and pharmacological

investigation .The most active extracts can be subjected to isolation of the therapeutic antimicrobials and undergo further pharmacological evaluation.

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