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Effect of some indian traditional plants on few common pathogens

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ABSTRACT

The antimicrobial activity of ethanolic plant extracts (Ocimum sanctum, Thespesia populnea and Pongamia glabra) were studied against three bacterial and one fungal organisms. The bacterial species include two Gram-positive(Streptococcus sp. and Staphylococcus aureus) and one Gram-negative(Escherichia coli). Aspergillus niger is the fungus taken for the study. In this study, the antimicrobial potency of single plants and the combined extracts, against the above mentioned microorganisms were assessed by well-diffusion technique. The plant extracts were subjected to various preliminary phytochemical analyses to determine the phytochemical constituents present in them. Studies revealed that, the synergism is more marked against Streptococcus spp. and Aspergillus niger especially the combination of Thespesia populnea and Ocimum sanctum is found to have more potent microbicidal activity.

Keywords: synergism, antimicrobial efficacy, ethanolic extracts, phytochemical constituents.

INTRODUCTION

Natural medicines and especially plant-based remedies have for a long time been the main, and often the only drugs, used by doctors and they constituted the basis in the production of pharmaceutical medicines. It is estimated that about 80% of the world population residing in the vast rural areas of the developing and under developed countries still rely mainly on medicinal plants. Medicinal plants are the only affordable and accessible source of primary healthcare for them, especially in the absence of access to modern medical facilities. The use of traditional medicine has increased in developed countries also, mainly due to the failure of modern medicine to provide effective treatment for chronic diseases and emergence of multi-drug resistant bacteria and parasites. The adverse effects of chemical drugs, questioning of the

approaches and assumptions of allopathic medicine, their increasing costs and greater public access to information on traditional medicine have led to an increase in interest in alternative treatments. Plant extracts have become a source of hope that has been used over the centuries almost exclusively based on empirical evidence. Hence, it has become necessary to revisit the importance of these herbal medicines. However, it is often seen that a crude extract is more active compared to the isolated active fractions.

Medicinal plants are important therapeutic aid for various ailments. Scientific experiments on the antimicrobial properties of plant components were first documented in the late 19th century. In India, from ancient times, different parts of medicinal plants have been used to cure specific ailments. More than a hundred species of therapeutically important higher plants are listed and described in the ancient Indian treatise, which have antimicrobial activity. Plants or their extracts are used to treat various diseases including urinary tract infection, enteric fever etc. caused by the bacterial pathogens.

In spite of wide handling of antibiotics in medicine, the search for substances of both plant and microorganism origin continues because better and safer drugs are still in demand to combat gram positive and gram negative bacterial infections. Medicinal plants really act as a boon by overcoming these draw backs as they have a wider spectrum of antimicrobial activity and at the same time they are biologically safe with no side effects. Further, they also gain momentum because of being more economical and easily available. In the present study, an attempt was made to assess the antimicrobial activity of some plant extracts, commonly used in folkloric medicines, against commonest pathological strains. This study is another effort, to scientifically validate the expediency of these plants in folkloric medicine.

MATERIALS AND METHODS

Three plants namely, *Thespesia populnea* (A), *Pongamia glabra* (B) and *Ocimum sanctum* (C) were selected and authenticated. Their antimicrobial efficacy was tested against three bacterial organisms and a fungus. Individual leaf extracts were prepared as per the standard methods. Mixed samples (D, E and F) were obtained by mixing equal proportions of the powdered leaf samples of two different plants. D consists of proportionately equal amounts of A and B, E consists of B and C and D was obtained by mixing A and C. All the above mentioned plant samples were subjected to cold extraction with ethanol. The bacterial strains included two Gram-positive bacteriae (*Staphylococcus aureus* and *Streptococcus spp*.) and one Gram-negative bacterium (Escherichia coli). The fungus, *Aspergillus niger* was also taken for the study. The antibacterial efficacy of the individual and combined extracts was studied by agar well diffusion technique. The plant extracts were subjected to various preliminary phytochemical analyses to determine the phytochemical constituents present in them[1].

RESULTS AND DISCUSSION

The antimicrobial activity shown by the plant samples towards different microbes are expressed as inhibitory zones and the values are tabulated (Tables 1-4).

The preliminary phytochemical analysis of the plant extracts are performed for the identification of the active constituents and the results are shown in Table-5.

The results of Table-1 show that all the plant extracts have inhibitory effect on *Staphylococcus aureus*, which is evident from the zones of inhibition and the values range from 6-18 mm. It is also observed that the inhibitory activity increases with the increasing concentration of extracts. This may be due to the presence of high amount of active constituents, which are soluble in organic solvents [2]. When comparing the antibacterial activities of all the extracts, *Ocimum sanctum*(C) shows maximum inhibition(18 mm) followed by *Pongamia glabra*(16 mm) and *Thespesia populnea(15* mm). High activity of *Ocimum sanctum* may be due to the high amount of eugenol, methyl eugenol [3], flavonoids and tannins [4].

The results of mixed extracts show that sample E (*Ocimum sanctum* and *Pongamia glabra*) has greater inhibition for the bacteria at all concentrations. The increased efficiency may be due to the synergism between the different active constituents that may be present in the both the extracts. Synergism can be due to the individual action of different constituents present in the extracts at multiple target sites [5].

The bactericidal effect of different plant extracts against *Streptococcus spp*. are shown in Table-2. The zone of inhibition for the samples range from 6-16mm. Of the three single plant extracts, sample A has more antibacterial potency for the organism at a higher concentration. The reason may be due to the increased activity of constituents like triterpenoids, alkaloids etc. against the particular organism [6,7].

The inhibitory zone measurements obtained for mixtures range from 7-14mm. All the samples show increasing activity with increasing concentration. The mixture analysis indicates that extracts containing *Thespesia populnea* as one of the constituent, have greater activity. *Pongamia glabra* and *Ocimum sanctum* species have more or less similar bactericidal activity.

The clear zones of inhibition formed against *Escherichia coli* by the different plant extracts are given in Table-3. The values range from 7-14 mm for single plant extracts whereas it lies between 3-14 mm for the mixtures. Comparative analysis of the data shows that all the plant extracts possess more or less similar bactericidal activity against *Escherichia coli*. The antibacterial activity is attributed to some active principles as already mentioned. These components enable them to partition the lipids of bacterial cell membrane and mitochondria, disturbing the cell structures and rendering them more permeable. Extensive leakage from bacteria cells or the exit of critical molecules and ions will lead to their death[8].

When comparing the mixtures, sample E (*Pongamia glabra* and *Ocimum sanctum*) shows higher values ranging from 9-14 mm with increasing concentration whereas D and F show little inhibition for the organism. Extract mixtures containing *Thespesia populnea* show lower activity at all concentrations [9]. This may be because, the ratio of constituents achieved at those concentrations is not optimal for the activity [10].

The values from Table-4 show that all the six samples exhibit significant antimycotic activity towards *Aspergillus niger*. *Thespesia populnea*, show pronounced activity with increasing

concentration of the extract, followed by *Ocimum sanctum* and *Pongamia glabra*. This may be attributed to the amount of triterpenoids present in the extracts [11]. The values obtained for samples D, E and F show that the mixtures are more effective against *Aspergillus niger* when compared to the individual plant extracts which may be due to the synergistic effect of the active constituents present in the mixture .

Phytochemical analysis (Table-5) of the plant extracts reveal the presence of sterols, sugars and alkaloids in all the samples whereas, other components like triterpenoids, flavonoids etc. are found to be present only in few plant extracts. This may be attributed to the solubility of the constituents in the given solvent.

CONCLUSION

The synergistic effect of plant constituents against the organisms revealed that the synergism is more marked against *Streptococcus spp.* and *Aspergillus niger* especially by sample F, which is the combination of *Thespesia populnea* and *Ocimum sanctum.* This study is thus a scientific validation for the traditional therapeutic use of these plants and it suggests that the synergistic studies can be further extended to other plant sources to enhance their curative properties.

Table-1: Zone of inhibition formed by different plant extracts against *Staphylococcus aureus*

S.No.	Name of plant extracts*	Concentration in micro litres (µl)			
		25	50	75	100
1.	A	8	10	12	15
2.	B	7	10	13	16
3.	C	6	8	12	18
4.	D	7	9	11	13
5.	E	7	8	10	15
6.	F	7	10	11	14

A – Thespesia populnea *D – Equal proportions of A and B*
B - Pongamia glabra *E – Equal proportions of B and C*
C - Ocimum sanctum *F - Equal proportions of A and C*

Table-2: Zone of inhibition formed by different plant extracts against *Streptococcus spp.*

S.No.	Name of plant extracts*	Concentration in microlitres(µl)			
		25	50	75	100
1.	A	7	10	13	16
2.	B	7	10	12	14
3.	C	8	10	12	13
4.	D	6	7	9	12
5.	E	7	8	10	11
6.	F	7	8	10	14

A – Thespesia populnea *D – Equal proportions of A and B*
B - Pongamia glabra *E – Equal proportions of B and*
C - Ocimum sanctum *F - Equal proportions of A and C*

Table-3: Zone of inhibition formed by different plant extracts against *Escherichia coli*

S.No.	Name of plant extracts*	Concentration in micro litres (µl)			
		25	50	75	100
1.	A	7	8	10	12
2.	B	8	9	11	13
3.	C	9	10	12	14
4.	D	3	7	9	12
5.	E	9	10	12	14
6.	F	6	7	9	12

A - *Thespesia populnea* D – Equal proportions of A and B
B - *Pongamia glabra* E – Equal proportions of B and C
C - *Ocimum sanctum* F - Equal proportions of A and C

Table-4: Zone of inhibition formed by different plant extracts against *Aspergillus niger*

S.No.	Name of plant extracts*	Concentration in micro litres (µl)			
		25	50	75	100
1.	A	7	10	11	13
2.	B	8	9	10	12
3.	C	8	10	12	12
4.	D	8	10	12	14
5.	E	9	10	13	15
6.	F	8	10	11	13

A - Thespesia populnea *D – Equal proportions of A and B*
B - Pongamia glabra *E – Equal proportions of B and C*
C - Ocimum sanctum *F - Equal proportions of A and C*

Table – 5: Phytochemical constituents present in the different plant extracts

S.No.	**Phytoconstituents**	**Plant Extracts**		
		***Thespesia populnea*(A)**	***Pongamia glabra*(B)**	***Ocimum sanctum*(C)**
1.	Sterols	+	+	+
2.	Triterpenoids	+	_	+
3.	Sugars	+	+	+
4.	Reducing sugars	+	+	+
5.	Amino acids	_	_	_
6.	Flavonoids	+	+	_
7.	Saponin	_	_	_
8.	Catechins	_	_	_
9.	Phenolic compounds	+	_	_
10.	Tannins	_	_	_
11.	Anthroquinone	_	_	_
12.	Alkaloids	+	+	+

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