



ANTI BACTERIAL ACTIVITY OF SOME LOCAL MEDICINAL PLANTS AGAINST *S.AUREUS AUREUS* AND *P.AERUGINOSA*

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ABSTRACT

Six medicinal plants including *Euphorbia serrata*, *Cannabis sativa*, *Trifolium repense*, *Chenopodium album*, *Carthamus oxyacantha* and *Calotropis procera* were collected from the district Swabi (Pakistan). Ethanol and water extracts of these locally collected medicinal plants were tested for their anti-bacterial activity against two pathogenic bacteria i.e., *Staphylococcus aureus* and *Pseudomonas aeruginosa*. It was observed that different concentrations (100, 200, 300 and 400mg/ml) of ethanol extracts of these medicinal plants possess great antimicrobial activity against the two tested pathogens as compared to same concentrations of their hot water extracts. The antimicrobial activity of tested plants may be due to the presence of some compounds such as phenols, flavonoids, alkaloids, tannins, carotenoids and saponins, etc. having antimicrobial and antioxidant activity.

KEYWORDS: Antibacterial, herbs, *Pseudomonas sp.*, *Staphylococcus sp.*

INTRODUCTION

The substance which stops the growth of microorganisms or kills it is called antimicrobial agent such as bacteria. The use of medicinal plants as a source of antimicrobial can be traced back over five millennia. Plants are frequently used in the treatment of many diseases as they are rich source of effective and safe drugs. From various parts of the world there are a lot of published reports on medicinal plants having antimicrobial properties, which showed that plants are still documented as the bedrock for modern medicine to cure infective diseases.^[1]

Medicinal plants show antimicrobial action since they have countless biologically active chemical compounds.^[2] The significance of medicinal plants is place in certain chemical substances that create a particular physiological action on the body of human beings. The most important bioactive compounds of these plants are phenolic compounds, alkaloids, tannins and flavonoids.^[3]

Medicinal plants are commonly used to cure several medical disorders including stomach aches, skin infections and respiratory conditions.^[4,5] They are inexpensive, can be available easily, affordable and are safe for both environment and human use.^[6]

Bacterial pathogens like *S. aureus* causes serious diseases of the bone, skin, soft tissues, lung, brain, blood

or heart. Similarly *P. aeruginosa aeruginosa* causes infectious diseases in healthy persons and those who are hospitalized as a result of other infections or have a weak immune system. This bacterium can cause various human infections like ventilator-associated pneumonia, urinary tract infection, respiratory infection, surgical site infections, ear infections (malignant external otitis, external otitis), soft tissues and skin infections, ocular infections, osteomyelitis and hot tub folliculitis.^[7,8]

Keeping in view the importance of medicinal plants, the present study is designed to find out the antimicrobial activity of extracts of some locally available medicinal plants and the effectiveness of all the plant extracts against *S. aureus* and *P. aeruginosa aeruginosa*.

MATERIALS AND METHODS

Collection of plants

Six medicinal plants including *Euphorbia serrata*, *Cannabis sativa*, *Trifolium repense*, *Chenopodium album*, *Carthamus oxyacantha* and *Calotropis procera* were collected from the local areas of Swabi.

Preparation of hot water and ethanol extracts

With tap water the plants were washed in order to wash out soil and dust particles and air dried for 5 to 7 days. Then plants were grounded into fine powder with the help of pistol and mortar and the powder was stored in reagent bottles.

For ethanol extracts preparation five grams of each of the powdered samples of plant was soaked in 50 ml ethanol in a sterile beaker and kept at room temperature for 24 hours. Similarly for hot water extracts preparation five grams of each of the powdered samples of plant were soaked in 50ml water in a sterile beaker and kept at 50°C for 24 hours. After 24 hours they were filtered by using filter paper and filtrates were kept for evaporation at room temperature. After drying the filtrates were weighted and then re-dissolved in their respective solvents to obtained final concentration of 1ml.

Preparation of dilutions

Reconstituted extracts were diluted to four concentrations i-e., 100, 200, 300 and 400mg/ml and stored in refrigerator.

Antimicrobial activity of the Extracts

Antimicrobial action of plant extracts was checked out by using agar well diffusion assay. The indicator bacteria were spread and allowed to on plates containing nutrient agar media at 37°C. Diameters of the inhibitory zones around the wells formed were measured in centimeters

(cm) and the results were recorded. Data was statistically analyzed by Chi-square test.

RESULTS

Both ethanol and hot water extracts of the tested plants showed activity against *S. aureus* and *P. aeruginosa*. Ethanol extracts of the plants were more active than its hot water extracts. The minimum inhibitory concentration (MIC) values acquired for extracts against the bacterial types were different from plant extracts to the other.

4.1 Antimicrobial activity of ethanol extracts of some medicinal plants against *S. aureus* .

The ethanol extracts of almost all the plants used exhibited antibacterial activity against *S. aureus* . *Cannabis sativa*, *Chenopodium album*, *Calotropis procera* and *Carthamus oxyacantha* extracts were more reactive as compared to the *Euphorbia serrata* and *Trifolium repens* (Table 4.1). Overall there was no significant difference between anti-microbial activities of all tested plants and their concentrations against *S. aureus* (P>0.05).

Table 4.1: Antimicrobial activity of ethanol extracts of some medicinal plants in different concentrations, against *S. aureus* .

S. No	Plants used	100mg/ml	200 mg/ml	300 mg/ml	400 mg/ml
1	<i>Trifolium repens</i>	NI ^a	NI	NI	NI
2	<i>Euphorbia serrata</i>	NI	NI	0.55cm	0.5cm
3	<i>Cannabis sativa</i>	1.4cm	1.05cm	1.15cm	0.6cm
4	<i>Chenopodium album</i>	1.55cm	1.7cm	1.75cm	1.3cm
5	<i>Carthamus oxyacantha</i>	2cm	3cm	3.6cm	4cm
6	<i>Calotropis procera</i>	3cm	3.1cm	1.25cm	2cm

^a NI= No zone of inhibition

4.2: Antimicrobial activity of Hot water extracts of some medicinal plants against *S. aureus*.

Among all the tested plants only the hot water extract of *Carthamus oxyacantha* inhibit the growth of *S. aureus*

sp. It was observed that antibacterial activity increases with the increase in concentration of water extract of this plant (Table 4.2).

Table 4.2: Antimicrobial activity of Hot water extracts of some medicinal plants in different concentrations, against *S. aureus* .

S.No	Plants used	100 mg/ml	200 mg/ml	300 mg/ml	400 mg/ml
1	<i>Trifolium repens</i>	NI ^a	NI	NI ^a	NI
2	<i>Euphorbia serrata</i>	NI	NI	NI	NI
3	<i>Cannabis sativa</i>	NI	NI	NI	NI
4	<i>Chenopodium album</i>	NI	NI	NI	NI
5	<i>Carthamus oxyacantha</i>	1.1cm	1.5cm	1.7cm	2cm
6	<i>Calotropis procera</i>	NI	NI	NI	NI

^aNI= No zone of inhibition

4.3: Antimicrobial activity of ethanol extracts of some medicinal plants against *P. aeruginosa*.

Ethanol extracts of all the tested medicinal plants exhibited great action against *P. aeruginosa*. *Carthamus oxyacantha* and *Calotropis procera* are more active

while other plants are least active (Table 4.3). Overall there was no significant difference between anti-microbial activities of all tested plants and their concentrations (P>0.05).

Table 4.3: Antimicrobial activity of ethanol extracts of some medicinal plants in different concentrations, against *P. aeruginosa*.

S.No	Medicinal Plants	100 mg/ml	200 mg/ml	300 mg/ml	400 mg/ml
1	<i>Trifolium repens</i>	1.45cm	1.55cm	1.61cm	1.65cm
2	<i>Euphorbia serrata</i>	NI	0.2cm	1.1cm	1.1cm
3	<i>Cannabis sativa</i>	NI	1.05cm	1.0cm	0.7cm
4	<i>Chenopodium album</i>	1.1cm	1.05cm	1.3cm	1.4cm
5	<i>Carthamus oxyacantha</i>	0.4cm	0.5cm	0.7cm	1.1cm
6	<i>Calotropis procera</i>	3.2cm	1.6cm	1.3cm	1.9cm

^aNI= No zone of inhibition

4.4: Antimicrobial activity of Hot water extracts of some medicinal plants against *P. aeruginosa*.

In all the tested plants the hot water extract of *Calotropis procera* inhibit the growth of *P. aeruginosa* and show

great antimicrobial activity (Table 4.4). Overall there was no significant difference between anti-microbial activities of different concentrations of extract ($P > 0.05$).

Table 4.4: Antimicrobial activity of Hot water extracts of some medicinal plants against *P. aeruginosa*.

S.No	Plants used	100mg/ml	200mg/ml	300mg/ml	400mg/ml
1	<i>Trifolium repens</i>	NI	NI	NI	NI
2	<i>Euphorbia serrata</i>	NI	NI	NI	NI
3	<i>Cannabis sativa</i>	NI	NI	NI	NI
4	<i>Chenopodium album</i>	NI	NI	NI	NI
5	<i>Carthamus oxyacantha</i>	1.1cm	NI	NI	NI
6	<i>Calotropis procera</i>	2cm	2.45cm	2.5cm	3cm

^aNI= No zone of inhibition

DISCUSSION

As a source of medicinal compounds plants have contained to play a dominant role in the maintenance of the human health since ancient times. According to World Health Organization (WHO) extracts of plants or their active elements are used as a traditional medicine in folk therapies of 80% of the population of the world. More than 50% of all recent clinical medicines are of natural source.^[9]

Among tested plants *Cannabis sativa* showed great antimicrobial activity against both gram +ve and gram -ve bacteria. It is reported that *Cannabis sativa* possess inhibitory action against both gram positive and gram negative bacterium.^[10] *Cannabis sativa* ethanol extracts showed great antimicrobial activity against *S. aureus* which is similar to the findings of Naveed et al.,^[11] Monika et al.^[12] and Borchardt et al.,^[13] while its ethanol extracts did not inhibit the growth of *P. aeruginosa* which is contrary to the findings of Naveed et al.^[11]

The hot water extracts of *Euphorbia serrata* did not inhibit the growth of all the tested organisms, while the ethanolic extracts inhibited the growth *S. aureus*. This is similar to the findings of Daud et al.^[14] who reported that the hot water extracts showed no activity against known bacterial strains, but ethanol extracts of the plant showed antibacterial activity.

The ethanol extracts of *Chenopodium album* showed great antimicrobial activity against *S. aureus* and *P. aeruginosa*. The similar observations were also studied

by Jangir et al.^[15] The aqueous extracts of *Chenopodium album* did not show any antibacterial activity which is contrary to the observations of Singh et al.^[16] who reported that aqueous extracts of *Chenopodium album* performed strongest antibacterial activity in *S. aureus*. Leila et al.^[17] reported that ethanolic extracts of *Chenopodium album* do not have any antimicrobial activity against *S. aureus* and *P. aeruginosa* which is differ from our findings.

The ethanol extracts of *Calotropis procera* inhibit the growth of *S. aureus* and *P. aeruginosa* and water extracts of *Calotropis procera* showed antibacterial activity against *P. aeruginosa* which is similar to the findings of Mako et al.^[18] The ethanol and aqueous extracts of *Calotropis procera* showed higher antimicrobial activity against *S. aureus* which is similar to the findings of Leonard et al.^[19] (2013), Salem et al.^[20] and Muzammal^[21] Water extracts of *Calotropis procera* inhibit the growth of *P. aeruginosa*. Similar observations were reported by Abdulmoniem et al.^[22] The ethanol extracts of *Calotropis procera* inhibit the growth of *S. aureus* and *P. aeruginosa* while its water extracts only inhabit the growth of *P. aeruginosa* which is similar to the observations of Kareem et al.^[23]

According to our observations *Trifolium repens* hot water extracts did not showed any antimicrobial activity against any of the tested organisms but the ethanol extracts of *Trifolium repens* inhibited the growth of *P. aeruginosa* and showed no antimicrobial activity against *S. aureus*. *Carthamus oxyacantha* showed pronounced antimicrobial activity against *P. aeruginosa* and *S.*

aureus while its water extract only inhibit the growth of *S. aureus* only. These plants are not tested against *P. aeruginosa* and *S. aureus* before. Some researchers have suggested that antimicrobial components of the plant extracts (terpenoid, alkaloid and phenolic compounds) may induce cell death or inhibit enzymes necessary for amino acids biosynthesis of microbes.^[24,25] Other researchers attributed the inhibitory effect of these plant extracts enable them to react with protein of microbial cell membrane and mitochondria disturbing their structures and changing their permeability.^[26,27]

CONCLUSION

From this study it is concluded that ethanol extracts of all the tested medicinal plants possess great antimicrobial activity against both *S. aureus* and *P. aeruginosa* as compared to their hot water extracts. As the origin of antimicrobial compounds are natural and it is believed that their effects on the environment are little and can be utilized as biological control agents.

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