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ORIGINAL ARTICLE

Antimicrobial Activity of Soil Borne Microbes against Pathogenic Bacterial Strain

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ABSTRACT

Background: Soil is a rich source of microbes including those that have the ability to impede the growth of pathogenic bacteria.
Objective: The current study was designed to explore the antimicrobial activity of soil borne microbes against pathogenic bacterial strain.
Methodology: This study was conducted in the microbiology laboratory of University of Swabi. The antimicrobial potential of soils was evaluated against five pathogenic strains (Pseudomonas, Staphylococcus aureus, Salmonella, Citrobacter and E.coli) using well diffusion assay. Antimicrobial activity of soils was assessed from the zone of inhibition around the wells. **Results:** Among the pathogenic strains, Citrobacter proved relatively more susceptible towards soils of both lawns in concentration dependent manner. Pseudomonas was susceptible towards the soil of Pharmacy lawn but did not respond to soil of Microbiology Lawn. Growth of other bacterial strains was not hampered by soil of any lawn.
Conclusion: Based on current findings it is inferred that soils of both lawns are rich in microbes that produce secondary metabolites capable of inhibiting growth pathogenic strains.
Keywords: Antimicrobial Activity; Soil Borne Microbes; Pathogenic Bacteria

INTRODUCTION

Naturally occurring free mixtures of mineral and organic particles are present in a loose mixture of soil [1]. Among soil bacteria population bacillus are one of the major groups and are distributed very broadly [2]. The geographical location such as soil temperature, soil type, soil pH, organic matters contents, cultivation, aeration and moisture content can greatly influenced the number and type of bacteria present in a particular soil [3]. Different bacteria are present in soil which have ability to produce antibiotic for example bacitracin, pumulin and gramicidin antibiotics produce by bacillus which are active against Staphylococci, Streptococci, Corynebacter, Streptomyces species which are gram positive bacteria and these bacteria produce tetracycline, chloramphenicol, vancomycin, gentamycin antibiotics which are active against gram negative bacteria. With the passage of time the problem of resistance against the present antibiotics in bacteria increases .To overcome these problems continuous search for novel antibiotics. During last few years a lot of work done that has observed that different microorganism is used for the production of new antibiotics. In soil, water and colonizing plants actinomycetes which are gram positive, free-living and saprophytic bacteria are broadly dispersed. From 22,500 biologically active compounds that have obtained from microbes, 45% are produced by actinomycetes, 38% by fungi, and 17% by unicellular bacteria [4]. Bacitracin, Gramycidin S, polymyxin, and tyrotricidin are important antibiotics which are produced by bacillus spp used in medical treatment [5]. Different bacteria present in soil that play role in nutrient cycle [6]. Present antibiotics are getting resistance by pathogenic microorganism, some of these antibiotics have side effects e.g. nephrotoxicity which are costly. For resisting the action of antibiotics and antibacterial agents bacteria developed various strategies [7] [8]. Fungal and bacterial researches on soil suggest those soil microorganisms are good producer of bioactive components [9]. In 1928 Penicillin was discovered by Alexander Fleming, but impressive antibiotics that can overcome antibiotics resistance worldwide studies are continued [10-12]. Many investigators present drugs resistance in Pakistan as a result cure fails and excessive health care charges [13]. Drug resistance is proceeding due to miss use of word wide [14]. For drug producers and public health physicians this condition has become shocking. Therefore, this study was carried out to assess the antimicrobial activity of soil borne microbes against pathogenic bacterial strain

MATERIALS AND METHODS

This study was conducted in the microbiology laboratory of University of Swabi to find out the soil antimicrobial activity of different pathogenic bacteria. The pathogenic strains of bacteria (Staphylococcus aureus , Salmonella, E.coli, Citrobacter and pseudomonas) were provided by the microbiology laboratory of Abasyn University Peshawar. The soil sample was collected from two different grounds at University of Swabi. The soil samples were collected in sterile bottles and were transferred to laboratory. The samples were stored at room temperature. All the samples were processed for obtaining pure culture by using standard microbiological procdures.

On the basis of colonial morphology, cellular morphology, reaction to Gram's stain, motility test and biochemical tests all bacterial isolates were characterized. Soil Sprinkle Method was used to check the plates for zone of inhibition. Soil bacterial metabolites were prepared by taking 3 sterile test tube and LB broth and pour the broth in test tube. then add the microbiology soil in one test tube and pharmacy soil in 2 test tube leave the 3 test tube uninoculated to check the contamination. Incubate the sample at 37c for 24 hours. Soil Microbial Metabolite were applied by taking 5 sterile plates. Pour the nutrient agar media in Petri plates and allow solidifying. After solidification make wells in Petri plates by using pinch machine. Then inoculate the pathogenic strains in sterile test tubes containing LB broth with the help of sterile inoculating loop to make sample. Then pour the five different pathogenic strain samples on 5 sterile Petri plates having the wells. Apply different concentration of metabolites i.e. 50ul 100um & 150um.icubate at 37c for 24 hours. After incubation zone of inhibition appear around the wells. All the data was collected and analyzed by using IBM SPSS 23.

RESULTS

Soil sample were collected from two different lawns of microbiology and pharmacy, from university of swabi (kpk) to check the

antimicrobial activity of soil against five different bacterial pathogenic strains i.e. (E.coli, salmonella, pseudomonas, staphylococcus aureus and citrobacter) provided by the microbiology lab Abasyn University. Both the soil sample were inoculated in sterile nutrient broth and incubated at 37c for 24 hours. After incubation both the soil samples of 100ul were poured in the appendof tube with help of micropipette and centrifuge at 100rpm for 5 min. After the centrifugation the supernatant of different quantity were applied to petriplates containing the five different test organisms and incubated at 37c for 24hours.on next day it was observed for the zone of inhibition produced by the microbe present in the soil having antimicrobial activity.

Table 1: Antimicrobial activity of soil from microbiology lawn against five test organisms:

s.no.	Test organisms	Supernatant conc. at 50ul	100ul	150ul
1	E.C	+	+	++
2	Cit	+	++	+++
3	Ps	-	-	-
4	S.A	-	-	-
5	Sal	-	-	-

Test organisms (pathogenic bacteria); E.C: E.coli, cit: citrobacter, Ps: pseudomonas, S.A: staphylococcus aureus, sal: salmonella. Zone of inhibition; +++ Very strong activity; ++ Strong activity; +: Weak activity; -: No activity.

Table 1: indicates that the soil activity of microbiology lawn show weak activity against E.coli while it show strong activity against citrobacter and by increasing the conc. its activity increase against it.

Table 2: Antimicrobial activity of soil from pharmacy lawn against five test organisms:

s.no.	Test organisms	Supernatant conc. at 50ul	100ul	150ul
1	E.C	-	-	-
2	Cit	+	++	+++
3	Ps	-	++	+++
4	S.A	-	-	-
5	Sal	-	-	-

Test organisms (pathogenic bacteria); E.C: E.coli, cit: citrobacter, Ps: pseudomonas, S.A: staphylococcus aureus, sal: salmonella. Zone of inhibition; +++ Very strong activity; ++ Strong activity; +: Weak activity; -: No activity.

Table 2 indicates that the soil activity of pharmacy lawn show strong activity against citrobacter and pseudomonas.

Table 3: Volume/concentration-dependent antimicrobial activity of both soils:

		Microbiology			Pharmacy		
S no	Test organisms	50ul	100ul	150ul	50ul	100ul	150ul
1	E.coli	+	+	+	-	-	-
2	Cit	+	+	++	+	++	+++
3	Ps	-	-	-	-	++	+++
4	S.A	-	-	-	-	-	-
5	Sal	-	-	-	-	-	-

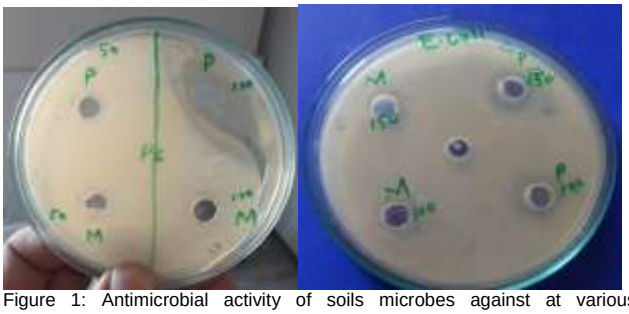

Figure 1: Antimicrobial activity of soils microbes against at various concentration against selected pathogenic bacteria

Test organisms (pathogenic bacteria); E.coli, cit: citrobacter, Ps: pseudomonas, S.A: staphylococcus aureus, sal: salmonella. Zone of inhibition; +++ Very strong activity; ++ Strong activity; +: Weak activity; -: No activity.

Table 3: shows that the soil of microbiology shows weak activity against the E.coli even at 150ul as at the same time it show maximum activity against the citrobacter and its activity affects by increasing the concentration, while the pharmacy soil shows the maximum activity against the citrobacter and pseudomonas and its activity also affects by increasing the conc.

DISCUSSION

Since ancient times soil has been used as antibiotic because it was known to possess the antibiotic producing bacteria which mainly actinomycetes [15]. In a study done by Griffiths et al., in 2003 it was observed that the depth of soil directly influences the population of bacteria, the population of bacteria decreases with increasing the depth [16]. A study done by Lihan et al. in 2014, which was on the antimicrobial activity of soil against ST, Salmonella typhi; SA, Staphylococcus aureus; EC, Escherichia coli; shows remarkable correlation with our own study its antimicrobial activity against these pathogenic bacteria is also null, while that of citrobater shows a high degree of antimicrobial activity [17]. In a study done by Kavitha et al. in 2007 the result shows that the antimicrobial activity of soil against SA, Staphylococcus aureus; EC, Escherichia coli is null while that against Citrobacter is very high and is cited by ++, this study is very much consistent with our own result [18]. A previous study done in Bhopal region of Madhya pardesh in which different bacterial samples were taken and acted against soil and in this result only 12 were found o have shown a antibacterial properties against these strains [19].

A study was conducted by (davis et al.,2018)[20] in which soil was treated against following bacteria E.coli, S.aures, Citrobacter the result showed that Citrobacter showed positive inhibition, E.coli showed negative inhibition and S.aures shows negative inhibition this has been in remarkable resembles with our own results which has been collected from Pharmacy Lawn.

According to the previous which was done on various bacteria including S.aureus, E.coli and Salmonella according to this study the zone of Inhibition for Salmonella, E.coli and S.aureus is maximum while the same zone of inhibition showed a high decrease when the PH of soil is alternated [21].

A study which was put forward by (Oskay et al., 2004)[22] explains that certain bacteria which are treated against soil its antimicrobial activity for these bacteria comes out to be negative, among them is the S.aureus which shows a high negative zone of inhibition and this is correlated to our result which is obtained from our Pharmacy Lawn.

A study was conducted on the Citrobacter bacteria which was treated against soil and the antimicrobial activity of soil for this particular bacteria shows a high positive zone of inhibition, which is in correlation with our result which was obtained from Microbiology Lawn [23].

CONCLUSION

Our study concludes that soils of both lawns are rich in microbes that produce secondary metabolites capable of inhibiting growth pathogenic strains.

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