

Studies on Soil Streptomyces from Saudia Arabia

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THE OBJECTIVE of the present study was to investigate the characteristics of Streptomyces in some Saudia Arabia soil as well as their biological , biochemical and anti-microbial activities. A total of 129 strains of Streptomyces were isolated from different samples of soil. Cultural and morphological characters showed the dominance of grey aerial mycelium species; 31% of the isolates have antimicrobial activity. All of these isolates decompose starch, coagulate the milk and most of them had about 70% cellulytic activity, whereas gelatin liquefying activity was about 77.5% and 52.5% produced melanin pigments. This means that the isolated Streptomyces produced many enzymes .

Keywords : Soil streptomyces, Antimicrobial activity.

Soil is an ecosystem that contains a diverse community of organisms differentiated by morphology and physiology. Streptomyces is a bacterial genus of the order Actinomycetales, which resemble fungi in their branching filamentous structure (The Colombia Encyclopedia , 2001) .

As the physiological and biochemical characteristics vary between strains , it is difficult for taxonomists to identify unknown Streptomyces species (Pridham *et al.*,1958). A number of chemicals and morphological features are available for other actinomycetes (Kutzner, 1992). Enzymes are one of the most important biologically active compounds, produced by Streptomyces, in addition to antibiotics. Streptomyces species liberate extracellular enzymes (Gupta *et al.* 1995) . The discovery that members of the genus Streptomyces were the source of useful antibiotics directed great attention to this genus. This group of microorganisms are not fully investigated in soil of the Arabian regions. Some investigations were made at few locations of Saudi Arabia (Elwan and Diab, 1976 ; Al-Zahrani, 1983 ; El-Awadi,1985 and Siddiq ,1995) .

Material and Methods

In this study , an attempt has been made to obtain some information about the Streptomyces population in soil in the Jazan district, in the south western region of Saudia Arabia . This district has two different localities ,Jazan at the sea level and Faifa about 6000 feet above sea level. Two different samples of soil were collected aspectically from each locality. A soil sample from a farm and an other from desert at Jazan, and another two samples from a farm and

mountain at Faifa . The atmospheric or weather conditions were recorded for the two locations at the time of sampling .

The soil was collected to depth of 20 cm in sterile bottles. These samples were dried in a 28°C incubator because the heat treatment of the soil reduces the bacterial flora significantly, with no detrimental effects to the growth of *Streptomyces* (Kutzner *et al.*, 1992) .

Suspensions of soils in sterile distilled water (1 gm of soil / 9 ml) , were shaken in a reciprocal shaker at 190 rpm for 30 minutes . Then serial dilutions (10^{-1} - 10^{-6}) were made , with 0.1 ml of each dilution pipetted and spread evenly over the surface of agar plates with a sterile L shaped glass rod. Each measurement was in triplicate. Agar starch- casein, yeast extract malt extract and agar were used for total viable *Streptomyces* count , and incubated at 28 ± 2 °C . The number of colonies were counted after 10 days of incubation for *Streptomyces* .

Colonies that seemed to represent different types were transferred to starch nitrate agar for further purification and characterization . The isolates were classified into seven series based on the colour of mature sporulating aerial mycelium and biochemical characteristics were carried out following the methods described by the International corporation projects for description and of cultures of *Streptomyces*, I S P (Shirling and Gottlieb, 1966). Chemical analysis were carried out in an attempt to correlate the microbial analysis with the soil characteristics .

Antagonistic properties

The anti-microbial potentialities of 7- day old experimental isolates were studied on solid media starch-caein, glucose yeast extract, malt extract and dextrose peptone . The cultures were tested for their antagonistic activity against wide range of microorganisms (Gramnegative and Grampositive bacteria, yeast, and fungi) using the agar disc method .

Physiological properties

Methods and media used for physiological tests were as described by Waksman (1967); Luedemann (1971) and Nera *et al.* (1977). The proteolytic activity was indicated by gelatin liquification and milk coagulation as well as starch hydrolysis and cellulose decomposition (indicators of amylolytic and cellulolytic activities) for 40 isolates which have antagonistic activity .

Results

Table 1 presents the density of *Streptomyces* in the soil samples collected from the different locations at successive depths in relation to chemical analysis of the soil. A total of 129 *Streptomyces* isolates were collected from all samples of the four locations. The highest number of *Streptomyces* colonies was observed in samples collected from agricultural soil, reaching 55 and 36 colonies /gram

dry weight soil from Jazan and Faifa ,respectively. On the other hand , the lowest count was collected from desert and mountain samples reaching 29 and 9 colonies /gram dry weight soil of Jazan and Faifa respectively .

TABLE 1 . Density of Streptomyces in soil samples collected from different locations at 0-20 cm depth in relation to enviromental factors.

Item	Location			
	Jazan		Faifa	
	Farm	Desert	Farm	Mountain
Counts ($\times 10$) /g dry weight soil	55	29	36	9
pH	7	6.5	6.5	6.5
Organic Carbon (g%)	0.20	0.15	0.42	0.11
Rain, mm/year	100-200		529-630	
Atmospheric temprature	43-35		25-20	
Total soluble salt (g%)	0.02	0.01	0.01	0.01

The distribution of these isolates in the different colour series is shown in Table 2. The highest number of isolates was among the grey series followed, in a decreasing order, by the white, green rose and violet, while the lowest series were the blue and yellow (being at the same level). The red, yellow and blue series were rarely isolated, the former being observed in agricultural soil at Jazan location while the blue series was isolated only from mountain soil from the Faifa location .

TABLE 2 . Streptomyces series in soil samples collected from different locations at Jazan region .

Location		% Streptomyces Series						
		Grey	White	Green	Rose	Violet	Blue	Yellow
Jazan	Farm	18.6	15.5	3.88	3.11	1.55	-	-
	Desert	7.75	9.3	3.1	1.55	-	0.775	-
Faifa	Farm	11.63	9.3	4.65	2.33	-	-	-
	Mounten	2.33	1.55	0.775	1.55	-	-	0.775
Total	100%	40.31	35.65	12.405	8.54	1.55	0.775	0.775

The data given in Table 3 show that 31% of all isolates have variable antagonistic activity against the test organisms depending on the type of the media .

TABLE 3 . Antimicrobial activity of 40 Streptomyces isolates against Gram-positive and Gram-negative bacteria and fungi .

Antimicrobial activity , %	Test Organisms						
	<i>E. coli</i>	<i>Salmonella typhi</i>	<i>Staph. aureus</i>	<i>Bacillus subtilis</i>	<i>Fusarium oxysporum</i>	<i>Rhizoctonia solani</i>	<i>Candida albicans</i>
% of 40 Strep. isolates	65	57.5	72.5	62.5	40	17.5	0
% of all Strep. isolates	20.2	17.8	22.5	19.4	12.4	5.4	0

Table 4 shows that all 40 Streptomyces isolates were capable of affecting starch hydrolysis and milk coagulation . On the other hand, gelatin liquefaction varied from positive to even negative. About 77.5% were incapable of decomposing cellulose and about 70% of the isolates have the ability to liquefy gelatin .

TABLE 4 . Enzymatic properties and production of melanin pigments of 40 Streptomyces isolates .

Activity \ Location	Jazan		Faifa		Total
	Farm	Desert	Farm	Mountain	
Starch hydrolysis	11	11	12	6	40
Cellulose decomposition	6	7	10	5	28
Gelatin liquifaction	8	10	10	3	31
Milk coagulation	11	11	12	6	40
Melanin pigments	8	4	7	2	21

General Discussion

This study showed that the percentage of Streptomyces was low but within the range reported by Abussaud and Saadoun (1991) . The highest count of Streptomyces population was in the farm samples from Jazan followed by Faifa location. The lowest was at the mountain sample from Faifa. However, differences in microbial counts in soils might be due to one or more of many factors working alone or in combination such as soil climatic conditions, soil

type , soil treatment , plant type and age ,organic matter in soil , chlorides and carbonate levels . The counts of Streptomyces in agricultural sample from Jazan (Table 1) was more than the same from Faifa and this might be due to the influence of , the content of organic matter , temperature or rain affecting soil moisture .

The grey series represent the highest frequency of isolation in this study. Hamdi *et al.*(1980) and Al-Zahrani (1983) also found that the grey series were the highest series of their isolates. Some Streptomyces species showed wide antimicrobial spectrum, inhibiting the growth of a wide variety of test organisms including bacteria, yeast and fungi, while other species exhibit limited antimicrobial activities, inhibiting the growth of one or few groups of microbes .

The results in this study revealed that, in each location, there were isolates active on Gram-positive bacteria although dominant but less than those active against Gram-negative ones or fungi. The results did not agree with the finding of Kamel and Al-Zahrani (1985), there is no isolates active against yeast. In most cases, the productivity of the isolates depended on medium constituents. About three-quarters of the Streptomyces species were heterotrophic feeders, and they can utilize both simple and complex molecules as nutrients .

The preliminary enzyme studies revealed minor differences between the amylolytic activity and milk coagulation among 40 isolates whereas , cellulose decomposition and gelatin liquifaction varied from very high to weak or negative. Most of the isolates had cellulytic activity (70 %) while 77.5 % were gelatin - liquifying .

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دراسة صفات الستربتوميسيتات في مناطق المملكة العربية السعودية

صالحه الزهراني واسماء الحربي

كلية البنات - جدة - المملكة العربية السعودية

الهدف من هذه الدراسة هو توضيح صفات الستربتوميسيس في بعض المناطق بالمملكة العربية السعودية ، بالإضافة إلى دراسة بعض الأنشطة الحيوية والبيوكيميائية والضد ميكروبية حيث تم عزل حوالي ١٢٩ عزلة من الستربتوميسيتات من أنواع مختلفة من الأراضي. تمت دراسة الصفات المورفولوجية والمزرعية لأربعين عزلة من الستربتوميسيس التي تم عزلها حيث صنفت هذه العزلات إلى ست مجموعات تبعاً للون الميسيليوم الهوائي ووجد أن الميسيليوم الهوائي السائد هو الرمادي وأن جميع العزلات كانت محللة للنشا ومخثرة للحليب بينما كان ٧٠٪ محللة للسليولوز أما العزلات المسيلة للجلاتين كانت حوالي ٧٧,٥٪ بالإضافة إلى أن حوالي ٥٢,٥٪ من العزلات كانت تفرز صبغة الميلانين ، أما النشاط ضد الميكروبي فقد كان ٣١٪ من العزلات لها قدرة تضادية .