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SCREENING OF INDUSTRIAL IMPORTANT SOIL MICROORGANISM

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ABSTRACT

The diversity of soil microorganisms were of great significance and as a factor promoting the early discovery of newer drugs. Most of the industrial important metabolites are obtained from microorganism, especially from soil source of isolates. In the present study, industrial important metabolites antibiotic and amylase screened from two soil sample, which is collected from different places in Padi, Chennai, Tamil Nadu. The crowded plate techniques used for antibiotic producing organism and amylase production identified by using starch agar medium. Both samples for antibiotic screening not found any inhibition of microbial growth which confirms the absence of antibiotic producing organisms. Both samples produces positive reaction for amylase production using starch hydrolysis test. From this study the collected soil not possess any antibiotic producing organism. The isolation and recovery of amylase from that isolated organism may be useful for future studies.

KEYWORDS

Antibiotic activity, Soil microorganisms, Antimicrobial activity and Amylase enzyme.

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INTRODUCTION

Antibiotic resistance is one of the important problems which lead to finding of newer antibiotics in more numbers. Comparison of laboratory synthesis of newer antimicrobial compound is more time consuming and screening of antibiotics (industrial important) from microorganism is ideal. The rapid growth of microorganism and soil is the best source of microorganism, so in our study we screened some industrial important metabolites (Antibiotics and amylase) from soil microorganism promoting the early discovery of antibiotics[1,5,8].

MATERIAL AND METHODS

Collections of soil samples

The soil samples were collected from in an around Chennai, India. The soil samples were dried separately at 37°C for 1 hour in hot air oven. Then they were cooled at room temperature. 1gm of each soil sample was added to a conical flask containing 100 ml of sterile water and few drops of Tween-80 solution. All flasks were shaken for 30 minutes at below room temperature. These flasks were considered as stock cultures[1,4,5].

The reference microorganisms used were *Streptomyces griseus* NCIM 2183 and *Bacillussubtilis* NCIM 5029(I). The selected strains were confirmed for their purity and identity by Grams staining method and by their characteristic biological reactions.

Isolation and screening of antibiotic producing isolates

A series of culture tubes containing 9 ml of sterile water were taken. From the stock culture, 1 ml suspension was transferred aseptically to the 1st tube (10^{-1}), mixed well. From the 1st tube, 1 ml of suspension was transferred into 2nd tube (10^{-2}), mixed well. Similarly, dilutions up to 10^{-5} were made (serial dilution technique). 0.1 ml of suspension from each culture tube was spread on sterile soyabean casein digest medium (SBCD) plates, in Laminar-Air flow bench. The plates were incubated at30°C (±2°C) for 84 hours. The plates were observed intermittently during incubation.

After 72 hours, a clear zone of inhibition around it grown colonies, indicates the antibiotic producing ability. Similarly Prepared serial dilutions a of the soil sample (B) seeded with positive control (standard) of *Streptomyces griseus*[1-5,7, 9].

Screening of amylase producing microorganism

Concisely, 1.0 g of soil sample was mixed with 9.0 ml of 0.9% (w/v) sterile saline and serially diluted up to 10^{-6}. Then a 100µl aliquot of each dilution was plated on starch nutrient agar plates and incubated for 24-48 h at 35°C under static condition in an inverted position.

After the period of incubation, the plates were flooded with Logul's solution and a clear area (starch hydrolysis) surrounding the bacterial growth showed positive reaction for extracellular a-amylase secretion while absence of clear zone around bacterial colonies was considered as a non-secretion of a-amylase enzyme[1,7,8,10, 11].

RESULTS AND DISCUSSION

The Figure No.1 and 2 indicates both serially diluted soil sample I and II shown growth on soybean casein digest agar after 24 hours incubation by crowded plate method. Further increases the duration of incubation period for the production of antibiotic by the microorganism. The zone of inhibition was not found, except the positive control plates. This indicates both samples not possess any antibiotic producing microorganism.

The Figure No.3 and 4 indicates the purified isolates from both soil samples, shown growth on starch agar medium after 24 hours incubation at 37°C.

After addition of iodine solution the colourless area found on the growth of organism and also in positive control. The colourless area formed due to digestion of starch present in the medium by synthesis of amylase enzyme in both samples of test organism and control.

Table No.1: Crowded plate method for screening of antibiotic producing organism

S.No	Sample	Crowded plate method	Antibiotic producing ability
1	Sample I	Absence of clear zone	Absence of antibiotic producing ability
2	Sample II	Absence of clear zone	Absence of antibiotic producing ability
3	Control	Presence of clear zone	Presence of antibiotic producing ability

Table No.2: Starch agar plate method for screening of amylase producing organism

S.No	Sample	Starch agar plate method	Amylase producing ability
1	Sample I	Absence of blue zone	Presence of Amylase producing ability
2	Sample II	Absence of blue zone	Presence of Amylase producing ability
3	Control	Absence of clear zone	Presence of Amylase producing ability

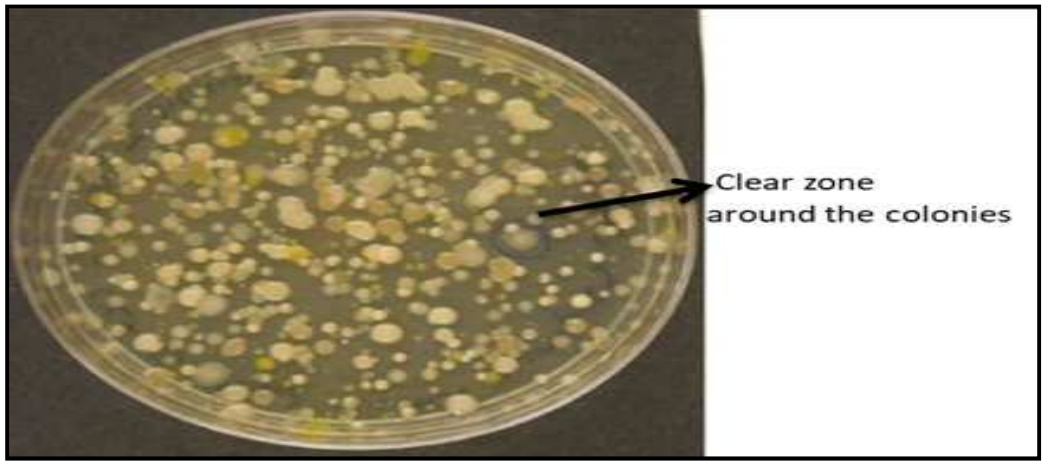

Figure No.1: Crowded plate method- standard organism produced a clear zone of inhibition around the isolated colonies indicates the *Streptomyces griseus* synthesis antibiotic

Figure No.2: Crowded plate method- test organism not produced clear zone of inhibition around the isolated colonies indicates the absence of synthesis antibiotic

Figure No.3: Amylase Test on starch agar media-Sample-I the plate on left side indicates no colour in all the agar surface which is before the addition of iodine solution. The plate on right side indicates after addition of iodine solution which produces no blue colour

Figure No.4: Amylase Test on starch agar media-Sample-II the plate on left side indicates no colour in all the agar surface which is before the addition of iodine solution. The plate on right side indicates after addition of iodine solution which produces no blue colour

CONCLUSION

This study shows that sample collected from this particular area may does not possesses antibiotic producing microorganism. But in both soil samples only industrial important metabolites (extracellular enzyme) amylase producing microorganism is present.

So the further studies may be done by collecting the sample from other area, for possible presence of expected results. The isolation and recovery of amylase from that isolated organism may be useful.

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CONFLICT OF INTEREST

We declare that we have no conflict of interest.

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