

## Characterization Of Actinomycetes Producing Novel Antibiotics

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### ABSTRACT

Actinomycetes that may have good potential of producing novel antibiotics against coal mine soil MMKK2 gram positive bacteria most of the isolates (46%) also showed antimicrobial activity *Streptomyces rochei*. These bacteria are known capable of producing secondary and primary metabolites isolation and characterization of actinomycetes were conducted by serial dilution, pour plate method on GAAM, SCA Medias while characterization was by determining morphological, physiological, ecological and genetic analysis of colonies, cell structure and biochemical tests. The results solved by 16SrDNA sequence analysis showing Antibiotic activity related to the genus *Streptomyces* followed by Microbacterium, Agromyces, Bacillus megaterium, rare antibacterial genera accounted for minor proportions

**Keywords:** Actinomycetes, coal mine soil samples, human pathogens primary and secondary screening.

### INTRODUCTION

*Streptomyces* is the largest antibiotic-producing genus in the microbial world so far. Actinomycetes produce a diverse array of antibiotics including glycopeptides, B lactams, macrolides, tetracyclines etc. During the last 40 years more than 1000 substances and preparations which possess antibiotic properties, i.e., have the capacity to inhibit the growth of and even to destroy various microorganisms in dilute solutions, have been isolated from culture of an actinomycete may therefore, be said to have been isolated in 1940. The genus streptomyces was proposed by Waksman and Henrici (1) for aerobic, spore-forming actinomycetes. Based on these information, laboratory experiments were conducted to study the effect of various nutrients such as nitrogen sources, carbon sources, mineral requirements and optimization improvement, Waksman (2) recognized the natural substrates that are ideal sources for the isolation of Actinomycetes.

In 1900 Beijerinck (3) established that Actinomycetes occur in great abundance in soil actinomycetes have been recognized primarily on their morphological criteria. Actinomycetes are commonly believed to have a role in man's life in particular for his own purpose where they can be used for the protection of producing novel antibiotics the average precipitation is around 65-100mm (4). Actinomycetes, phylogenetically defined as a number of taxa within the high G+C sub division of the gram-positive phylum (5).

### MATERIALS AND METHODS

Dry colonies of Actinomycetes were selected and isolated. The culture of *Streptomyces* sp. MM-KK2 of which microbial activity was studied in the basal medium consisting of yeast extract 10.0g; K<sub>2</sub>HPO<sub>4</sub> 1.0g; MgSO<sub>4</sub> .7 H<sub>2</sub>O 0.5g; CaCl<sub>2</sub> .2H<sub>2</sub>O 0.04g; FeSO<sub>4</sub> 7H<sub>2</sub>O, 0.05g : ZnSO<sub>4</sub> .7H<sub>2</sub>O, 0.005g and distilled water 1000ml. The pH was adjusted to 7.5 and incubated at 28°C for 7 to 14 days.

#### Characterization of the Isolates

Some diagnostic characters of highly active streptomycin strains were determined following the directions given in the probabilistic identification metric of Williams (6) and Bergey's manual of systematic Bacteriology (7). All the microorganisms are obtained from the coal mines of

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**Table-1. Cultural Characteristics of Actinomycetes at different pH Levels (MM-KK-1 & MM-KK-2)**

MM-KK-2				MM-KK-1			
pH	Growth	Aerial Mycelium	Substrate mycelium	pH	Growth	Aerial Mycelium	Substrate mycelium
4	+	Gray	Dark Brown	4	++	Gray	Light Yellow
5	+	Gray	Dark Brown	5	++	Gray	Light Yellow
6	++	White	Yellow	6	-	White	Light Yellow
7	-	Gray	Brown	7	+	White	Light Yellow
8	++	Gray	Light Yellow	8	+	White	Light Yellow
9	+++	Gray	Light Yellow	9	+	White	Light Yellow
10	+++	Gray	Light Yellow	10	+	White	Light Yellow
11	+++	Gray	Light Yellow	11	+	White	Light Yellow

**Table-2. Cultural Characteristics of Actinomycetes at different pH Levels (Bellampally-BPA; Kothagudem-OC)**

BELLAMPALLY-(BPA)				KOTHAGUDEM-OC			
pH	Growth	Aerial Mycelium	Substrate mycelium	pH	Growth	Aerial Mycelium	Substrate mycelium
4	+	Gray	Dark Brown	4	-	Gray	Light Yellow
5	+	Gray	Dark Brown	5	-	Gray	Light Yellow
6	++	White	Yellow	6	-	Gray	Light Yellow
7	-	Gray White	Yellow	7	-	Gray	Brown
8	++	Gray White	Yellow	8	-	Gray	Light Yellow
9	+++	Gray White	Yellow	9	-	Gray	Light Yellow
10	+++	Gray White	Yellow	10	-	Gray	Light Yellow
11	+++	Gray White	Yellow	11	-	Gray	Light Yellow

Andhrapradesh (2005). The microbial interactions were analyzed by the determination of the size of the inhibition zone (8).

## RESULTS & DISCUSSION

The strain MM.KK2 showed grey and brown actinomycete colonies on GAA medium and white powdery form on SCA Medium. The colonies were white, brown powdery form on nutrient agar medium while were light powdery pink form on ISSA medium, Simultaneously the other Actinomycete strains were varied with colony morphologies (Table 1 and 2) of white cottony to grey moderate.

The specific amplification of 16SrDNA fragments of Actinomycetes. The reaction mixture was a template DNA, pH 8.3 and deoxynucleoside triphosphates ideal primer for all studies of actinomycetes because (i) it does not match the 16SrDNA (primer F243) of all actinomycetes and (ii) It matches the 16Sr DNA of a few non actinomycetes. In general the results of the present day study are very encouraging, especially when taken together with easier findings (9-17) as they indicate that relationships between taxonomically complex groups such as the genus *Streptomyces* followed by microbacterium, *Agromyces* and *Bacillus megaterium* can be resolved using appropriate combinations of genotypic and phenotypic data. Antibiotics produced by an

organisms differ in their activity. Streptomycetes are economically important organisms not only for production of useful secondary metabolites but also for their activity as wide spread scavengers in soil and contribution to the fertility and geo-chemical stability of the biosphere.

## Conclusion

This study shows that the test actinomycetes isolates have the potential to act as sources of novel antibiotics against pathogenic microorganisms to humans. The isolates were not able to inhibit intracellular growth of mycelium and filaments in the test organism. Which could form and important input into pharmaceutical industries.

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## Conflicts of Interest

Authors declare that there is no conflict of interests regarding the publication of this paper.

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