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ANTAGONISTIC ACTIVITY OF MARINE ACTINOBACTERIA – A REVIEW

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Abstract

Actinobacteria are Gram positive prokaryotes which produce branching mycelium. Actinobacteria are widely distributed in natural and man-made environments and play an important role in the degradation of organic matter. They are also well known as a rich source of antibiotics and bioactive molecules. Actinobacteria hold a prominent position for their diversity and ability to produce novel substances. The terrestrial soil Actinobacteria has potential biotechnological applications and is a new resource for structurally diverse secondary metabolites. At present, an alternative approach was to make the isolation procedure more selective by adding chemical such as phenol to the soil suspension. Considering the practically useful compounds, today about 130 to 140 microbial products and a similar number of derivatives are applied in human medicine, mostly in chemotherapy and veterinary medicine. The majority of these compounds, except fungal penicillins, cephalosporins and several bacterial peptides and few others, are also produced by Actinobacteria. The high percentage of new compounds derived from new target - oriented screening methods is also of Actinobacteria origin.

Key words: Actinobacteria, Antagonistic activity, Bioactive compounds and Marine ecosystem.

1. Introduction

Marine microorganisms have innumerable new genes and biochemical approaches and available enzymes, antibiotics and other useful molecules. As to biological technology, they are really a huge gold mine. Thus, the combination of biological technology and marine microorganism research will create a mutual benefit prospect. In the nearly 300 new compounds, almost 80 % of them refer to bioactivities, especially some important discoveries of pharmacology and toxicology. But, the microorganisms from mangrove have not yet gained enough recognition until now (Liua, 2008).

Marine environments are largely untapped source for the isolation of new microorganisms with potentiality to produce active secondary metabolites (Baskaran *et al*., 2011). Among such microorganisms, Actinobacteria are of special interest, since they are known to produce chemically diverse compounds with a wide range of biological activities. The demand for new antibiotics continues to grow due to the rapid emerging of multiple antibiotic resistant pathogens causing life threatening infection. Nowadays, considerable progress is being continuing within the fields of chemical synthesis and in the field of engineered biosynthesis of antibacterial

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compounds. So, the nature still remains the richest and the most versatile source for new antibiotics (Kpehn and Carter, 2005).

The Actinomycetes or Actinobacteria are Gram positive, aerobic bacteria that form branching filaments or hyphae and asexual spores. The name "Actinomycetes" was derived from Greek "atkis" (a ray) and "mykes" (fungus) and has features of both bacteria and fungi (Das *et al*., 2008). Although, these are a diverse groups and the Actinobacteria shared many properties. Actinobacteria, when growing on a solid such as agar, the branching network of hyphae developed by Actinobacteria grows both on the surface of the substratum and into it to form a substrate mycelium. Septa usually divide the hyphae into long cells (20 µm and longer) containing several nucleoids. Many Actinobacteria also have an aerial mycelium that extends above the substratum and forms asexual, thin walled spores called conidia or conidiospores on the end of filaments. With a high guanine (G) plus cytosine (C) ratio in their DNA (>55 mol %), which are phylogenetically related from the evidence of 16S ribosomal cataloguing and DNA: rRNA pairing studies (Goodfellow and Williams, 1983).

Most of the bioactive microbial metabolites were isolated from Actinobacteria especially from *Streptomycetes* and also from some rare Actinobacteria. During the last 20 - 30 years, the interest in the marine microflora increased due to the investigation of novel bioactive compounds especially antibiotics and enzymes. As the frequency of novel bioactive compounds obtained from terrestrial Actinobacteria decreases with time, Actinobacteria from diverse environments have been increasingly screened for their ability to produce new secondary metabolites. It has been emphasized that the Actinobacteria from marine sediments may be valuable for the isolation of novel strains which could potentially yield a broad spectrum of secondary metabolites (Ismet *et al.*, 2004).

In the recent years, marine microorganisms have known for antimicrobial, antiviral, antitumour, anticoagulant, antidiabetic and cardio active properties. Antibiotic effect of Actinobacteria has been used in many fields including agriculture, veterinary and pharmaceutical industry. Actinobacteria are well known as secondary metabolite producers and hence of high pharmacological and commercial interest. Waksman (1961) discovered actinomycin from soil bacteria. Then, hundreds of naturally occurring antibiotics have been discovered in these terrestrial microorganisms, especially from the genus *Streptomyces*. Some *Actinobacteria* form branching filaments, which somewhat resemble the mycelia of the unrelated fungi, among which they were originally classified under the older name "Actinobacteria". Most members are aerobic, but a few, such as *Actinomyces israelii*, can grow under anaerobic conditions. Unlike the Firmicutes, the other main group of Gram positive bacteria, they have DNA with a high GC-content, and some *Actinobacteria* species produce external spores. Some types of Actinobacteria are responsible for the peculiar odour emanating from the soil after rain, mainly in warmer climate.

Actinobacteria are ubiquitous in soils, where they usually are present in numbers of 10^5 - 10^6 colony forming units per gram of soil. The majority of Actinobacteria are free living saprophytic bacteria found widely distributed in soil, water and colonizing plants. Actinobacteria population has been identified as one of the major group of soil population, which may vary with the soil type. They have in common that they all are Gram positive and have a high content of guanine plus cytosine in their DNA (>55 mol %). In general, the optimal conditions for their growth are temperatures of 25 – 30 °C (50 °C for the thermotolerant Actinobacteria). Most are aerobic and neutrophilic. The Actinobacteria were initially regarded as minute fungi because of their mycelium - like growth and attention paid to this group rose notably after the discovery of streptomycin by Waksman and Schatz (1943) and were finally recognized as bacteria. Their

morphology, however, varies among the different genera, from cocci and pleomorphic rods to branched filaments that break down into spherical cells or aerial mycelium with long chains of spores (Erikson, 1949).

2. Occurrence of Actinobacteria

Actinobacteria are distributed extensively in every aerial substrate such as terrestrial soils, marine soils, compost, fresh water basins, food stuffs and the atmosphere. Marine Actinobacteria produce different type of bioactive compounds. In the recent years, marine microorganisms have known for antimicrobial, antiviral, antitumor, anticoagulant, ant diabetic and cardio active properties. Antibiotic effect of Actinobacteria has been used in many fields including agriculture, veterinary and pharmaceutical industry. Cohn (1975) first described Actinobacteria, based on secretions of the lachrymal ducts which were later named as *Streptothrix foesteri.* Two years later, Harz (1878) isolated *Actinomyces bovis* from lumpy jaws in cattle. Crook *et al*. (1950) investigated sodium propionate to be an effective fungal inhibitor for isolation of *Streptomyces.*

Dulaney *et al*. (1955) isolated Streptomyces selectively on a Nutrient agar medium containing, cycloheximide 20 units; polymyxin 20 units; subtilin 20 units. They stated that the amounts of antibiotics in the medium were critical and that, where as the selective inhibition of *Streptomyces* and bacteria was a difficult achievement. Corke and Chase (1956) reported cycloheximide as more effective and used in Actinobacteria isolation media at a level of 40 µg per ml of agar.

Grein and Meyers (1958) identified the Actinobacteria species such as *Nocardia, Micromonospora* and *Streptomyces* from marine sediments. Among this 1/5 of total number of isolates, *Nocardia* was more prevalent. Waksman (1959) discussed the distribution of Actinobacteria in natural environment. Their predominant source being terrestrial while the marine source was less exploited.

Nakeeb and Lechevalier (1963) reported that the composition of an Arginine Glycerol Salt medium (AGS) suitable for the selective isolation of aerobic Actinobacteria was given. When soil samples were treated with calcium carbonate and plated on the AGS medium, higher total and relative plate counts of Actinobacteria were obtained than when other media and methods were used.

Nonomura and Hayakawa (1988) isolated Actinobacteria from marine environments such as sediments, water column samples and marine microorganisms using dry heat and selective inhibitors like nalidixic acid.

Jiang and Xu (1990) characterized the populations of soil Actinobacteria. Kurtboke *et al*. (1992) isolated *Saccharomonospora* from a substrate under study revealed the presence of *Saccharomonospora* sp. in the composted material and once the common bacteria were eliminated through their phage susceptibility.

A new species of the genus *Nocardiopsis* named *Nocardiopsis lucentensis* was isolated from a salt marsh soil sample (Yassin *et al*., 1993). An Actinobacteria strain was isolated from soil collected in Yunnan province, China. It was recognized as *Nocardia flavorosea* (Chun *et al*., 1998).

Seong *et al*. (2001) identified various pretreatment procedures and selective media were applied to assess the optimal conditions for the isolation of rare Actinobacteria from soil. Pre-treatment of wet heating for 15 min at 70 ºC and phenol treatment of soil suspension were the most effective methods for the isolation of those microorganisms. Hair Hydrolysate Vitamin Agar (HHVA) was the most suitable medium for the recovery of rare Actinobacteria. Thirty five rare Actinobacteria strains were chosen using selective isolation approaches, then morphological and chemical properties of the isolates were determined. The isolates belonged to one of the following genus, *Micromonospora, Microbispora, Actinoplanes* and *Stretosporangium*. Mincer *et al*.

(2002) reported 212 Actinobacteria from 112 ocean sediment, samples in California. Dhanasekaran *et al*. (2005) isolated 65 Actinobacteria from 32 soil samples collected from Cuddalore, East coastal region of Tamil Nadu. Dhevagi and Poorani (2006) isolated marine Actinobacteria from sediment samples of Parangipettai and Cochin areas of South India.

3. Characteristics of Actinobacteria

Okami (1952) grouped the genus *Streptomyces* on the basis of formation of aerial mycelium. Benedict *et al*. (1955) reported the development of *Streptomyces* colonies on agar plates could be favored over bacteria by selection of the nitrogen source in the medium. They noted that L-arginine was readily attacked by most Streptomycetes and they recommended the use of this amino acid as a replacement for the glycine of glycerol-glycine medium of Lindenbein. Actinobacteria also synthesis and excrete dark pigments, melanin which are considered to be a useful criterion for taxonomical studies. Melanin compounds are irregular, dark brown polymers that are produced by various microorganisms by the fermentative oxidation and have the radio protective and antioxidant properties that can effectively protect the living organisms from ultraviolet radiation. Melanins are frequently used in medicine, pharmacology and cosmetics preparations.

Veiga *et al.* (1983) studied the cellulolytic activity of 36 Actinobacteria strains isolated from marine sediments was investigated by Cellulose - azure method. *Streptomyces* produce potent immunomodulators such as tacrolimus (Umezawa *et al*., 1985) and rapamycin (Vezina *et al*., 1975). This suggests that the secondary metabolites may provide other virulence functions such as the immune suppression that accompanies *Mycobacterium* infections. Thus, these compounds may have therapeutic value because lethal pathogenicity effectors would potentially be turned into life saving compounds.

Settle *et al.* (2005) identified that the Alachlor [2-chloro-N-(methoxymethyl)-N-(2, 6-diethylphenyl) – acetamide] was extremely toxic and highly mobile herbicide that was widely used for pre-emergence control of grasses and weeds in many commercial crops in Brazil. In order to select soil Actinobacteria able to degrade this herbicide, fifty-three *Actinobacteria* sp. were isolated from soil treated with alachlor using selective conditions and subjected to *in vitro* degradation assays. Sixteen isolates were shown to be tolerant to high concentrations of the herbicide and six of these were able to grow and degrade. Though, a definitive taxonomic assignment of alachlor degrading strains was not possible, these data indicate that ability to degrade this pesticide was detected in different Streptomyces taxa.

Lechevalier (1989) pointed out that the difference between the families Actinomycetaceae and Streptomycetaceae cannot possibly be based on fragmentation, Since, variations in this respect can be observed between isolates of the same strains of organisms now designated as *Nocardia* and *Streptomyces.* A new genus *Micropolyspora,* which fragments like the Actinomycetaceae and sporulates like the Streptomycetaceae by forming chains of conidia on aerial hyphae. The genus also forms chains of conidia on the substrate mycelium, located in and on agar media. It was suggested that the family Streptomycetaceae be dropped and that the family Actinomycetaceae be enlarged to include the genera *Actinomyces, Micromonospora, Thermoactinomyces, Waksmania, Micropolyspora, Nocardia* and *Streptomyces. Micropolyspora brevicatenu* was said to represent a novel morphological type easily distinguishable from the previously described forms. The thermophilic organism described in *Pseudonocardia thermophila* was considered being a facultative.

4. Occurrence of Bioactive compounds in Marine sources

Chemical defenses employed by many organisms have proven to be available sources of

novel molecules providing lead compounds for drug discovery. Among the least explored of the planet's chemically defended organisms are the invertebrates, algae, fungi and bacteria of the marine environment. There are greater than 2,00,000 marine animals and microorganism species available for investigation. The marine environment contains about half of the total global species and contains a biodiversity as extensive as all the world's rain forests combined. This biological diversity has resulted in a vast array of chemical diversity. Many marine organisms have a sedentary life style and require a chemical means of defense. As a result, they have the ability to produce or obtain toxic compounds to deter predators and keep competitors at bay. Bioprospecting in the marine environment has only started relatively and recently, but has already yielded thousands of novel compounds. One group studying the marine environment during the past 25 years has isolated over 10,000 compounds from marine invertebrates: algae, bacteria, fungi and protozoa. Less than 0.5 % of marine animals have been examined to determine if they produce compounds that might be used as therapeutic agents against infectious diseases. Improved underwater life - support systems have provided marine scientists new mechanisms for collecting from unexplored regions and depths (Haefner *et al*., 2003).

Actinobacteria are the most economically and biotechnologically valuable prokaryotes. They are responsible for the production of about half of the discovered bioactive secondary metabolites, notably antibiotics, antitumor agents, immunosuppressive agents and enzyme. Because of the excellent track record of Actinobacteria in this regard, a significant amount of effort has been focused on the successful isolation of novel Actinobacteria from terrestrial sources for drug screening programs in the past fifty years. More than 70 % of our planet's surface is covered by oceans and life on earth originated from the sea. As marine environmental conditions are extremely different from terrestrial ones, it was surmised that the marine Actinobacteria species have different characteristics from those of terrestrial counterparts and, therefore, might produce different type of bioactive compounds. Marine Actinobacteria are a group of organisms that have demonstrated great promise as a source of potential new drugs. These organisms account for 10 % of marine snow (dead or dying animals and plants such as plankton, diatoms, fecal matter, sand, soot and other inorganic dust) that settles to the ocean floor. Marine Actinobacteria exhibit very different 16S rRNA sequences compared to their terrestrial counterparts. As a result, marine Actinobacteria produce novel metabolites that may be biologically active and a potential source of new anti-infective drugs (Lam, 2006).

Marine environments are largely untapped source for the isolation of new microorganisms with potentiality to produce active secondary metabolites. Among such microorganisms, Actinobacteria are of special interest, since they are known to produce chemically diverse compounds with a wide range of biological activities (Bredholt *et al.*, 2008). The demand for new antibiotics continues to grow due to the rapid emerging of multiple antibiotic resistant pathogens causing life threatening infection. Although, considerable progress is being made within the fields of chemical synthesis and engineered biosynthesis of antibacterial compounds, nature still remains the richest and the most versatile source for new antibiotics (Baltz, 2006). Traditionally, Actinobacteria have been isolated from terrestrial sources although, the first report of mycelium forming Actinobacteria being recovered from marine sediments appeared several decades ago (Weyland, 1969).

Recently, the marine derived Actinobacteria have become recognized as a source of novel antibiotic and anticancer agent with unusual structure and properties (Jensen *et al.*, 2005). Actinobacteria represent a ubiquitous group of microbes widely distributed in natural ecosystems around the world and especially significant for their role on the recycling of organic matter (Srinivasan *et al.*, 1991). The literatures suggested that the marine sediment sources are voluble for the isolation of novel

Actinobacteria with the potential to yield useful new products (Goodfellow and Haynes, 1984). However, it has been resolved whether Actinobacteria form part of the autochthonous marine microbial community of sediment samples originated from terrestrial habitats and was simply carried out to sea in the form of resistant spores (Ravel *et al.*, 1998).

Microorganisms found in marine environments have attracted a great deal of attention due to the production of various natural compounds and their specialized mechanisms for adaptation to extreme environment (Solingen *et al.*, 2001). Since, marine sediments represent an environment which was markedly different from that associated with soil sample. It was not clear how effective the pre-treatment of such sediments would be for the recovery of bioactive Actinobacteria. Various reports from the East coast of India suggested that soil was a major source of Actinobacteria (Dhanasekaran *et al.*, 2008).

Marine Actinobacteria have been attracting the attention of scientists for more than 50 years. In the early works, the species of *Mycobacterium*, *Actinomyces*, *Nocardia*, *Micromonospora* and *Streptomyces* have been isolated from the marine sediments (Grein and Meiers, 1958). A significant part of these Actinobacteria was found to exhibit antibiotic activity, suggesting that the marine environment can be an interesting source for bioprospecting. Since then, a number of reports have been published describing isolation of marine Actinobacteria species some of these species were found to produce unique compounds such as salinosporamides (Fenical and Jensen, 2006). Those are now in the Phase I clinical trials as anticancer agents. Recently, new species and new genera (*Salinibacterium, Serinicoccus* and *Salinispora*) of marine Actinobacteria have been described by Han *et al*. (2007).

Searching of novel antimicrobial secondary metabolites from marine Actinobacteria was gaining momentum in recent years. The Indian marine environment was rich in biodiversity, especially microorganisms. However, the wealth of marine microflora has not been fully investigated (Ramesh, 2009). Searching for previously unknown microbial strains was an effective approach which would yield biologically novel active substances. It is known that the antimicrobial activity of the metabolic products of aquatic bacterial strains is not weaker than the corresponding activity of soil strains (Sponga *et al*., 1999). In addition, the limited attempts have been made on marine organisms and their metabolites in India (Ramesh and Mathivanan, 2009). Importantly investigation of marine Actinobacteria with reference to bioactive molecule production in India was still at its infancy. Therefore, exploration of marine Actinobacteria for secondary metabolites production is worthy task. The bioactive molecules derived from these Actinobacteria could be used as therapeutic drugs for the treatments of various ailments in human and animals and as agrochemicals for the management of insect pests, diseases and weeds in agriculture, etc. as suggested by Prabavathy *et al*. (2009).

Actinobacteria are representative of terrestrial microorganism and usually are isolated from soils. Compared to terrestrial Actinobacteria, however, very little work has been conducted on marine Actinobacteria. As marine environmental conditions are extremely different from terrestrial ones, it was surmised that marine Actinobacteria have characteristics different from those of terrestrial Actinobacteria and therefore may produce different types of bioactive compounds (Okami *et al.* 1988). Anderson and Wellington (2001) investigated *Streptomycetes*, producers of more than half of the 10,000 documented bioactive compounds, have offered over 50 years of interest to industry and academics. Baltz (2006) reported that antibiotic biosynthetic pathways are distributed at frequencies ranging from a single antibiotic (Streptothricin) at 1 in 10 to 1000 different antibiotics at 1 in 107, a frequency distribution suggesting that only a fraction of existing antibiotics have been discovered.

Marine microbiology is developing strongly in several countries with a distinct focus on bioactive compounds. Analysis of the geographical origins of compounds, extracts, bioactivities and Actinobacteria indicate that 67 % of marine natural products were sourced from Australia, Caribbean, Indian Ocean, Japan, the Mediterranean and Western Pacific Ocean sites (Blunt *et al*., 2007).

Marine Actinobacteria search and discovery is one thing, development of discoveries to end point products is another. With a few reflections on this dilemma and in this context they relate to antibiotics, almost identical arguments are opposite to orphan drugs in general and 'neglected' diseases. There has been much recent comment about the scarcity of new antibiotic entities. Most of the antibiotics in clinical use today have been developed from compounds isolated from bacteria and fungi with members of the Actinobacteria being the dominant source (Pelaez, 2006). Traditionally, most of these antimicrobials have been isolated from soil derived Actinobacteria of the genus *Streptomyces*. However, isolation strategies in recent years have been directed to unexploited environments like marine sources (Sheridan, 2006). Bioprospecting efforts focusing on the isolation and screening of *Actinobacteria* from ocean habitats (Nathan *et al*., 2004) have added new biodiversity to the order *Actinomycetales* and revealed a range of novel natural products of pharmacological value. The existence of marine *Actinobacteria* species physiologically and phylogenetically distinct from their terrestrial relatives was now widely accepted and new taxonomic groups of marine Actinobacteria have been described for at least six different families within the order Actinomycetes. Apart from being phylogenetically distinct from their terrestrial relatives, marine isolates have been shown to possess specific physiological adaptations (e.g., to high salinity/osmolarity and pressure) to their maritime surroundings and many were found to produce novel and chemically diverse secondary metabolites (Feling *et al.*, 2003).

Actinobacteria perform significant biogeochemical roles in terrestrial soils and are highly valued for their unparalleled ability to produce biologically active secondary metabolites. Totally 22,500 bioactive secondary metabolites have been reported, out of which 16,500 compounds show antibiotic activities. Out of the 22,500 total bioactive secondary metabolites, 10,100 (45 %) are reported to be produced by Actinobacteria in which 7630 from *Streptomycetes* and 2470 from rare Actinobacteria. A search of recent literature revealed that atleast 4607 patents have been issued on Actinobacteria related products and processes (Newman and Cragg, 2007).

In the past two decades, however, there has been a decline in the discovery of new lead compounds from common soil-derived Actinobacteria as culture extracts usually yield unacceptably high number of previously described metabolites. The immense biotechnological ability of Actinobacteria had led to exhaustive surveys of cultivars from normal terrestrial habitats and an associated increase in the number of known compounds being rediscovered due to a high rate of redundancy in the strains isolated. For this reason, searching the less or unexploited ecosystems for Actinobacteria might lead to the discovery of novel bioactive compounds including those that can act against drug resistant pathogens (Mincer *et al*., 2002).

Terrestrial Actinobacteria are one of the most efficient groups of secondary metabolite producers. They are responsible for the production of about half of the discovered microbial bioactive secondary metabolites, notably antibiotics, antitumor agents, immunosuppressive agents and enzymes. However, the marine Actinobacteria are also becoming increasingly appreciated as a rich source of novel bioactive agents. Various novel compounds with biological activities including antifungal, antibacterial and antiviral have been isolated from marine Actinobacteria genera: *Streptomyces, Saccharopolyspora, Amycolatopsis, Micromonospora* and *Actinoplanes* (Solanki *et al.*, 2008). Some of the recent compounds are:

Abyssomicin C, a polycyclic polyketide antibiotic produced by a marine Verrucosispora strain. This compound is a potent inhibitor of para-aminobenzoic acid biosynthesis and, therefore, inhibits the folic acid biosynthesis at an earlier stage than the well-known synthetic sulphonamides (Bister *et al.*, 2004). Abyssomicin C is highly active against gram-positive bacteria, including the multi-drug resistant and vancomycin-resistant *Staphylococcus aureus* (Rath *et al.*, 2005). Compounds such as lipoxazolidinone A, B and C isolated from a bacterium of the genus *Marinispora* (strain NPS008920) showed broad spectrum antimicrobial activities similar to those of the commercial antibiotic linezolid.

The crude extract of the Actinobacterial strain isolated from a marine sediment sample collected near La Jolla, California, exhibited strong antibiotic activity. Two products marinopyrroles A and B, were isolated which displayed noteworthy activity against Methicillin resistant *Staphylococcus aureus* (Hughes *et al.*, 2008). Other compounds including frigocyclinone and himalomycins (Maskey *et al.*, 2003) isolated from *Streptomyces* species have also been shown to have antibacterial activity.

5. Antagonistic activity of Actinobacteria

The history of new drug discovery processes shows that novel skeletons have in the majority of cases, come from natural sources. This involves the screening of microorganisms and plant extracts (Shadomy, 1987). Lacey (1978) reported Actinobacteria are of considerable value as producers of antibiotics, and other therapeutically useful compounds. They play a major role in the cycling of organic matter in the soil ecosystem (Yang and Ling, 1989). Franco and Countinho (1991) reported the antifungal antibiotics are extracted using ethyl acetate. Numerous *Streptomyces aureofaciens* strains have been identified as antibiotic producers and the antibiotics generated by this species are macrolides (White *et al*., 2001) and tetracycline (Stryzhkova *et al*., 2002).

Antifungal Actinobacteria were abundant in Orchard soil and lake mud. More than 50 % of antifungal isolates from most soils were classified as genus *Streptomyces*. Actinobacterial isolates that showed strong antifungal activity against *Alternaria mali*, *Colletotrichumgleo sporioides*, *Fusarium oxysporum* and *Rhizoctonia solani* were predominant in pepper - field soils, whereas those against *Magnaporthe grisea* and *Phytophthora capsici* were abundant in radish - field soils (Lee and Hwang, 2002).

Sahin and Ugur (2003) isolated seventy four different *Streptomyces* from soils of Mugla Province. Antimicrobial activity was determined in 45.9 % of the isolates. Fifteen isolates showed strong activity against Coagulase – negative *Staphylococcus*. These isolates were extensively studied for their *in vitro* antimicrobial activity against Gram positive and Gram negative bacteria and yeast.

Basilio *et al*. (2003) reported Actinobacteria based on their method of isolation and their phenotype diversity was determined by total fatty acid analysis. A total of 335 representative isolates were screened for the production of antimicrobial activities under different conditions of pH and salinity against a panel of bacteria, filamentous fungi and yeasts.

Iznaga *et al.* (2004) reported on the capacity of Actinobacteria strains isolated from Cuban soils to produce antifungal agents. The antimicrobial activities were determined by susceptibility disk assay methods. Totally, 586 different Actinobacteria are isolated and 286 produced compounds with antifungal activity. Our screening method indicated the presence of many possible polyene macrolide antibiotics and the important antifungal activity in the soils rich in minerals.

Nathan *et al*. (2004) suggested a unique selective enrichment procedure which resulted in the identification and isolation of two new genera which are marine - derived Actinobacteria. By their study, it was revealed that approximately 90

% of the microorganisms were cultured by using the presented method which was from the prospective new genera, it indicates as a result which is indicative of its high selectivity. From the Bismarck Sea and the Solomon Sea off the coast of Papua New Guinea 102 Actinobacteria were isolated from the subtidal 8 marine sediment. By performing the test for physiological and chemotaxonomic characteristics and with this distinguishing 16S rRNA gene sequences and phylogenetic analysis based on 16S rRNA genes it ultimately provides strong evidence for the two new genera within the family Micromonosporaceae. Biological activity testing of fermentation products from the new marine-derived Actinobacteria showed that it has activities against multi drug - resistant Gram positive pathogens, malignant cells and vaccinia virus replication.

Oskay *et al.* (2004) identified a total of 50 different Actinobacteria strains recovered from farming soil samples collected from Manisa Province and its surrounding. These were then assessed for their antibacterial activity against four phytopathogenic and six pathogenic bacteria. Results indicated that 34 % of all isolates are active against, at least one of the test organisms; *Agrobacterium tumefaciens, Erwinia amylovora, Pseudomonas viridiflova, Clavibacter michiganensis* subsp. *michiganensis, Bacillus subtilis, Klebsiella pneumoniae, Enterococcus faecalis, Staphylococcus aureus, Escherichia coli* and *Sarcina lutea*. Kathiresan *et al*. (2005) reported the marine Actinobacteria are active against the *Salmonella* species, *Escherichia coli, Klebsiella* sp., *Rhizoltonia solani, Pyricularia oryzae, Helminthosporium oryzae* and *Colleto trichumfalcatum.*

Sujatha *et al*. (2005) screened 26 marine sediment samples near 9 islands of the Andaman coast of the Bay of Bengal resulted in the isolation of 88 isolates of Actinobacteria. On the basis of sporophore morphology and structure of the spore chain, 64 isolates were assigned to the genus *Streptomyces,* 8 isolates to the genus *Micromonospora,* 5 to the genus *Nocardia,* 7 to the genus *Streptoverticilium* and 4 to the genus *Saccharopolyspora*. Among 64 *Streptomyces* sp., 44 isolates showed antibacterial activity and 17 isolates showed antifungal activity.

Twenty five strains of Actinobacteria were isolated from samples of water, soil and tree barks collected at two sites located in the North - east of Algeria. Antimicrobial activity was tested using the agar cylinder method against three Gram positive bacteria, three Gram negative bacteria, three yeasts and three filamentous fungi. Among the 25 isolates, 14 (56 %) strains showed an activity against at least one of the test bacteria studied and two (8 %) showed antifungal activity. Ninety three percent of the active strains were identified by the Universal PCR as belonging to the *Streptomyces* genus and 7 % to the *Actinomadura* genus (Kitouni *et al.,* 2005). Dhanasekaran *et al*. (2005) isolated Actinobacteria from the Saltpan regions. 17 Actinobacteria isolated were obtained and were screened for antibacterial activity against, *Escherichia coli, Klebsiella pneumoniae, Staphylococcus aureus, Vibrio cholerae, Salmonella typhi* and *Shigella dysenteriae.*

You *et al.* (2005) isolated 94 Actinobacteria strains from the marine sediments of a shrimp farm, 87.2% belonged to the genus Streptomyces, and others were *Micromonospora* sp. Fifty-one percent of the Actinobacteria strains showed activity against the pathogenic *Vibrio* spp. strains. Thirty-eight percent of marine Streptomyces strains produced siderophores on chrome azurol and CAS agar plates. Seven strains of Streptomyces were found to produce siderophores and to inhibit the growth of *Vibrio* sp. Two of them belonged to the Cinerogriseus group, the most frequently isolated group of Streptomyces. The results showed that Streptomycetes could be a promising source of biocontrol agents in aquaculture.

Augustine *et al*. (2005) isolated 312 Actinobacteria strains from water and soil samples from different regions. These isolates were purified and screened for their antifungal activity

against pathogenic fungi. *Streptomyces albidoflavus* with strong antifungal activity against pathogenic fungi was selected and studied. Ilic *et al*. (2005) isolated 20 different *Streptomyces* isolates from the soils of South Eastern Serbia. Nine isolates showed a strong activity against *Bortrytis cinerea.* These isolates extensively studied for their *in vitro* antimicrobial activity against Gram positive and Gram negative bacteria and yeasts.

Asha Devi *et al*. (2006) reported marine Actinobacteria strains were isolated from coastal water. Out of 10 isolated Actinobacteria species 3 were identified and selected for antimicrobial activity. Out of the 3 Actinobacteria species, *Streptomyces* sp. showed the best level of antibacterial and antifungal effect against selected human pathogens of *Staphylococcus aureus, Pseudomonas aeruginosa, Salmonella typhi, Vibrio cholerae, Klebsiella* sp. and *Aspergillus niger*. Naggar *et al*. (2006) isolated 12 Actinobacteria strains from Egyptian soil. The isolated Actinobacteria strains were then screened with regard to their potential to generate antibiotics. The cultural and physiological characteristics of the strains identified as genus *Streptomyces* which was active *in vitro* against Gram positive, Gram negative representative and *Candida albicans.*

Shantikumar Singh *et al*. (2006) isolated 37 Actinobacteria from the soil sample collected from the Phoomdi in Loktak Lake of Manipur, India. These isolates were screened for their antimicrobial activity. Out of 37 isolates, only 21 showed antimicrobial activities against test microorganism namely *Bacillus subtilis, Staphylococcus aureus, Escherichia coli, Klebsiella pneumoniae, Micrococcus luteus, Mycobacterium phlei, Candida albicans* and *Fusarium moniliforme*. Srivibool and Sukchotiratana (2006) reported that the forty five soil samples were collected from four coastal islands on the east coast of Thailand. 495 isolates of Actinobacteria were found. Preliminary test to search for antimicrobial activity was done with *Bacillus subtilis, Staphylococcus aureus, Staphylococcus aureus, Micrococcus luteus* and *Pseudomonas aeruginosa* and *Escherichia coli*. Fifty eight Actinobacteria sp. were found to be antimicrobial producing strains. From the morphological determination, cell wall diaminopimelic acid and sugars in whole cell hydrolysate studies, among the 58 strains, *Streptomyces* sp. and *Actinomadura* sp. were the predominant genera.

Singh *et al.* (2006) identified a total of 37 Actinobacteria with distinct characteristics were isolated from the soil sample collected from the Loktak lake. These isolates were screened for their antimicrobial activity. Out of 37 isolates, only 21 showed antimicrobial activity against test microorganisms in primary screening process by spot inoculation technique on agar medium. These 21 putative isolates were then subjected to submerge culture and their antimicrobial activity was evaluated. Of these 21 isolates, 12 were found to be active against the test microorganisms namely *Bacillus subtilis, Staphylococcus aureus, Escherichia coli, Klebsiella pneumoniae, Micrococcus luteus, Mycobacterium phlei, Candida albicans* and *Fusarium moniliforme.* Most of these active isolates showed antifungal property. Parungao *et al*. (2007) isolated Actinobacteria from marine, brackish and terrestrial sediments for antimicrobial activity. A total of 54 Actinobacteria isolates were obtained from the various sediment samples collected and were then tested for antagonistic activity against *Escherichia coli, Staphylococcus aureus, Candida utilis* and *Aspergillus niger*. Jeffrey *et al.* (2007) showed the antagonistic activity of Actinobacteria against three strains of pathogenic microbes (*Fusarium palmivora, Bacillus subtilis, Ralstonia solanacearum* and *Pantoaedispersa*). All the strains were chosen due to the reason that these microbes exhibited pathogenic effect towards certain commodity plants. Antimicrobial tests showed that 3, 25, 37 and 35 isolates of Actinobacteria produces antagonistic reaction for *Fusarium palmivora* and *Bacillus subtilis.*

Remya and Vijayakumar (2008) isolated 173 Actinobacteria colonies from 8 different

locations. Antimicrobial activities of isolates were also tested against various bacterial and fungal pathogens. Of 64 isolates, 21 isolates had antimicrobial activity, with 2 isolates showing broad spectrum of antimicrobial effect. Seventy nine Actinobacteria were isolated from soils of Kalapatthar (5545 m), Mount Everest region by Gurung *et al*. (2009). Among all the isolates twenty seven (34.18 %) of the isolates showed an antibacterial activity against at least one test bacteria among two Gram positive and nine Gram negative bacteria in primary screening by the technique of perpendicular streak method. In secondary screening thirteen (48.15 %) showed antibacterial activity. After that the MIC test was done and the Minimum inhibitory concentration (MIC) of antibacterial metabolites of the isolates K.6.3 was 1 mg/ml, and that of isolates K.14.2 and K.58.5 was 2 mg/ml. From each of the metabolites two spots were detected on thin layer chromatography plate which was completely different from the spot produced by vancomycin and the active isolates from primary screening were heterogeneous in their macroscopic, biochemical and physiological characteristics.

Dhanasekaran *et al*. (2009) investigated a source of Actinobacteria in estuary to screen for production of novel bioactive compounds. The presence of relatively large populations of *Streptomyces* in estuary soil samples indicates that it is an eminently suitable ecosystem for Actinobacteria. Actinobacteria count was ranged 12×10^4 cfu/g of soil. The Actinobacteria isolated from these ecosystems are capable of producing antibiotics that strongly inhibit the growth of Gram positive and Gram negative bacteria and yeast like fungi.

Soil sample was collected by Lakshmipathy and Kannabiran (2010) from the coastal region of Tamil Nadu with the aim of isolating Actinobacteria and screen them for antagonistic activity against common bacterial and fungal pathogens. Serial dilution of the soil sample was done and after that subsequent screening of the isolates obtained. Potential strain with significant activity against *Klebsiella pneumoniae*, *Aspergillus flavus* and *Aspergillus niger*. The strain shows chitinolytic activity. By the Chemotaxonomic analysis the isolate belongs to cell wall Type I. The 16 S rRNA partial gene sequence and phylogenetic analysis showed that the strain shared 93 % similarity with *Streptomyces* sp.

A study was done by Pugazhvendan *et al*. (2010) on marine Actinobacteria. Among the 34 strains, 10 potential marine Actinobacteria strains were screened by cross streak method against five fish pathogenic bacteria. The extract was tested by Disc diffusion method against fish bacterial pathogens and the ethyl acetate extract showed a good inhibition range of 6-15 mm in diameter. The most potential Actinobacteria strain was characterized and identified as *Streptomyces* sp.

Thenmozhi and Kannabiran (2010) screened the antifungal activity of the crude extract prepared from the strain *Streptomyces* sp. Primarily eight strains were screened for antifungal activity against three species of *Aspergillus* namely *Aspergillus fumigatus*, *Aspergillus niger* and *Aspergillus flavus*. This search resulted in the isolation of a potential strain. The optimization was done by the production media for the maximum yield of secondary metabolites and the metabolites were extracted using ethyl acetate, it was than lyophilized and screened for antifungal activity against the three *Aspergillus* species by well diffusion method. Maximum zone of inhibition observed was 21 mm for *Aspergillus fumigates* in comparison with the standard antifungal antibiotic Nystatin which shows 20 mm. By using the Hideo 13 Nonomura classification the strain was further identified. The molecular taxonomy and phylogeny revealed that the strain belonged to the genus *Streptomyces* and was designated as *Streptomyces* sp. After this the blast search of the 16S rRNA sequence of the strain with the sequences available in the NCBI data bank exhibits a maximum similarity of 86 % with *Streptomyces longisporoflavus* with the bootstrap value of 100. The 16S rRNA sequence of the

strain *Streptomyces* sp. was submitted to the GenBank.

A total of 13 Actinobacteria strains were isolated from mangrove sediment in which 5 Actinobacteria strains showed potential antibacterial activity against 5 fish pathogens and 8 human pathogens. Actinobacteria *viz*., ACT-1, ACT-4, ACT-9, ACT-10 and ACT-12 were showed broad spectrum of sensitivity against all pathogens. The minimum MIC value of 10 μg ml^{-1} was recorded with ACT-1 against the fish pathogens *Bacillus subtilis*, *Serratia marcescens* and human pathogens *Escherichia coli*, *Proteus vulgaris* and the maximum MIC value of 40 μg ml^{-1} was recorded against the fish pathogens *Vibrio cholera*, *Vibrio parahemolyticus* and all human pathogens. Well diffusion assay reveals that, ACT-1 showed the maximum zone of inhibition against *Bacillus subtilis* and *Escherichia coli* (Ravikumar *et al*., 2011).

Sateesh and Rathod (2011) selected the fifty three rare Actinobacteria strains by using selective isolation approaches, then morphological and chemical properties of the isolates were determined. The isolates belonged to one of the following genus, *Micromonospora*, *Microbispora*, *Actinoplanes* and *Actinomadura*. Later *Micromonospora* and *Actinomadura* were selected for antimicrobial activity. Minimum bactericidal concentration (MBC) of ethyl acetate extract against *Staphylococcus aureus* were 1.20 mg/ml for *Micromonospora* species and 5 mg/ml for *Actinomadura* species. Thin layer chromatography (TLC) of the ethyl acetate extracts were carried out in duplicate using chloroform: methanol (4:1) as solvent system and Tetracycline as reference antibiotic. Under UV light they gave greenish yellow spots with Rf value 0.85 for the antimicrobial from *Actinomadura* species and 0.88 for that from *Micromonospora species*. In bioautography (using *Staphylococcus auras* as test organism) inhibition zones were obtained and they were associated with the yellowish green spots of the chromatogram as detected under UV light. This may indicate the same compounds were responsible for the antibacterial activity of those Actinobacteria isolates.

Gulve and Deshmukh (2011) showed the 107 different Actinobacteria were isolated from marine sediments collected from five coastal sites of Konkan coast of Maharashtra. Ninety Actinobacterial isolates were identified up to generic level. It was found that these Actinobacterial isolates were belonging to *Streptomyces, Micromonospora, Intrasporangium, Saccharopolyspora, Streptosporangium, Rhodococcus, Saccharomonospora* and *Nocardia*. Enzymatic activities of 90 identified Actinobacterial isolates were performed. It was found that out of 90 Actinobacterial 76 (84.44 %), 70 (77.78 %), 65 (72.22 %), 39 (43.33 %), 34 (37.78 %) and 15 (16.67 %) number of Actinobacteria were possessing protease, gelatinase, amylase, lecithinase, cellulose and ureases activity respectively.

Adel Ayari *et al*. (2012) showed the new Actinobacteria isolate *Streptomyces* sp. with antifungal activity. This isolate was identified based on a great variety of morphological, cultural, physiological and molecular characteristics analysis of 16S rDNA sequence. The test of antifungal activity for several pathogens fungi causing invasive Aspergillosis and systemic Candidiasis revealed that the *Streptomyces* sp. was a good moderate antifungal compound producer against *Aspergillus fumigatus* and *Candida albicans*, and had no activity against *Aspergillus flavus, Aspergillus niger, Candida pseudotropicalis* and *Candida tropicalis*.

6. Bioactive compound from Actinobacteria

Natural products have been the sources of most of the active ingredients of medicines. The microbes keep on producing novel metabolites as they move into the diverse ecological units. From the biologically active compounds that have been obtained so far from microbes, 45 per cent are produced by Actinobacteria, 38 per cent by fungi and 17 per cent by unicellular bacteria. Microbes have always been a better resource for getting lead

molecule with novel scaffold to overcome any such limitation of existing drugs. Actinobacteria are the representative of terrestrial microorganisms and usually are isolated from soils. When compared to terrestrial Actinobacteria, however, very little work has been conducted on marine Actinobacteria. Marine environmental conditions are extremely different from terrestrial ones, it was surmised that marine Actinobacteria have characteristics different from those of terrestrial Actinobacteria and therefore, may produce different types of bioactive compounds (Okami *et al.*, 1972).

Actinobacteria in marine and estuarine sediments have not been investigated extensively although their ubiquitous presence in marine sediments has been well documented (Takizawa *et al*., 1993). They were believed to be of terrestrial origin, transported to rivers by rain or irrigation water and finally to the marine environment where they are exposed to water with salt concentrations and temperatures that differ from those of the terrestrial environment. As a result, some metabolic changes may occur in the organisms found respectively that 50 % and 27 % of *Streptomyces* isolated from the marine environment showed antimicrobial activity and these percentages were increased when tests were conducted in the presence of seawater. As mentioned above, the marine environment was considerably different from the terrestrial environment, and therefore, it needs to be explored and exploited for new biological products. So far, microorganisms producing various bioactive compounds have been isolated largely from marine environments because of the sampling difficulty.

Natural products are widely used in human medicine to fight numerous diseases including bacterial, viral and fungal infections, cancer and immune system disorders, etc. Actinobacteria bacteria have proven to be a rich source of biologically active natural products and a number of terrestrial Actinobacteria, especially those belonging to the genus *Streptomyces* sp. are being extensively used for commercial production of different medically important compounds. As the search for producers of novel compounds continues, it becomes apparent that many terrestrial *Streptomyces* sp. isolated from different environments produce the same compounds, probably due to frequent genetic exchange between the species. Therefore, the chance of finding genuinely new biologically active molecules while isolating and screening large libraries of Actinobacteria was greatly reduced (Busti, 2006). The situation was worsened by the fact that *Streptomyces* species are easy to isolate and cultivate and they generally dominate the collections. On the other hand, evidence was being accumulated that rare Actinobacteria which are often very difficult to isolate and cultivate, might represent a unique source of novel biologically active compounds (Baltz, 2006).

At present, more than 80 % of drug substances were natural products or inspired by a natural compounds. The natural products include compounds from plants, microbes and animals. They cover a range of therapeutic indications like anticancer, antimicrobial and antidiabetic (Harvey, 2008). The most striking feature of the genus *Streptomyces* and closely related genera is their ability to produce a wide variety of secondary metabolites. These natural products have been an extraordinary rich source for lead structures in the development of new successful drugs. In addition to the field of antimicrobials, further compounds have been approved as immunosuppressants (FK-506, Rapamycin and Ascomycin), as anticancer compounds (Bleomycin, Dactinomycin, Doxorubicin and Staurosporin), as antifungal compounds (Amphotericin B and Nystatin), as herbicides (Phosphinothricin), in the treatment of diabetes (Acarbose) and as anthelmintic agents (Avermectin and Milbemycin). Of the 12,000 secondary metabolites with antibiotic activity known in 1995, 55 % were produced by *Streptomyces* and additional 11 % by other Actinobacteria (Demain, 2002). According to a mathematical modeling, only 3 % of all antibacterial agents synthesized by *Streptomyces* have been reported (Watve *et al*., 2001). This leaves a vast amount of possible new drugs to be

discovered or generated by novel innovative methods.

7. Conclusion

Actinobacteria have provide many important bioactive compounds of high commercial value and continue to be routinel screened for new bioactive substances. These searches have been remarkably successful and approximately two-thirds of naturally occurring antibiotics, includes many of medical importance have been isolated from Actinobacteria. They are abundant in terrestrial soils, a source of the majority of isolates shown to produce bioactive compounds. Intensive screening programme carried out over the past several decades resulted in the production of known bioactive compounds. However, Actinobacteria are more abundant in terrestrial soils than in marine sediments show varying degrees of salt tolerance and produce spores that are undoubtedly washed in large numbers from shore into sea. Inspite of the fact that they remain active in the marine environments, their role in the production of bioactive compounds is to be studied. It needs a screening of which varieties of the species of Actinobacteria, that could possibly the potential source of bioactive components.

8. References

1) Adel Ayari, Houda Morakchi and Kirane Gacemi Djamila. 2012. Identification and antifungal activity of *Streptomyces* sp. S72 isolated from Lake Oubeira sediments in North- East of Algeria. *African Journal of Biotechnology*, 11(2): 305-311.
2) Anderson, A. S and Wellington, E. M. H., 2001. The taxonomy of *Streptomyces* and related genera. *International Journal of Systematic Evolutionary Microbiology*, 51: 797 – 814.
3) Asha Devi, N. K., Lisa. Jeyarani and Balakrishnan, Z. K. 2006. Isolation and identification of marine actinomycetes and their potential in Antimicrobial Activity. *Pakistan Journal of Biological Sciences*, 9 (3): 470 - 472.
4) Augustine, S. K., Bhavsar, S. P and Kapadnis, B. P. 2005. A non-polyene antifungal antibiotic from *Streptomyces albidoflavus* PU23. *Journal of Bioscience*, 30: 201 – 211.
5) Balagurunathan, R and Subramanian, A. 1992. Antagonistic *Streptomycetes* from marine sediments. *Advanced Bioscience*, 200: 20: 71 - 76.
6) Baltz, R. H. 2006. Marcel Faber Roundtable, Is our antibiotic pipeline unproductive because of starvation, constitution or lack of inspiration. *Journal of Industrial Microbiology and Biotechnology*, 33: 507 - 513.
7) Basilio, A., Gonzalez, I., Vicente, M.F., Gorrochategui, Cabello, Gonzalez, A. and Genilloud, O. 2003. *Journal of Applied Microbiology*, 95 (5): 814 - 823.
8) Baskaran, R., Vijayakumar, R and Mohan, P. M. 2011. Enrichment method for the isolation of bioactive actinomycetes from mangrove sediments of Andaman Islands, India. *Malaysian Journal of Microbiology*, 7 (1): 26-32.
9) Benedict, R. G., Prindham, T. G., Lindenfelser, L. A., Hal, H. H and Jackson, R. W. 1955. Further studies in the evaluation of carbohydrate utilization tests as aids in the differentiation of species of *Streptomyces. Applied Microbiology*, 3: 1 - 6.
10) Bentley, S. D., Chater, K. F., Cerdeno-Tarraga, A. M., Challis, G. L., Thomson, N. R., James, K. D., Harrris, D. E., Quail, M. A., Kieser, H., Harper, D., Bateman, A., Brown, S., Chandra, G., Chen, C. W., Collins, M., Cronin, A., Fraser, A., Goble, A., Hidalgo, J., Hornsby, T., Howarth, S., Huang, C. H., Kieser, T., Larke, L., Murphy, L., Oliver, K., O'Niel, S., Rabbinowitsch, E., Rajanmdream, M. A., Rutherford, K., Rutter, S., Seeger, K., Saunders, D., Sharp, S., Squares, R., Squares, S., Taylor, K., Warren, T., Wietzorrek, A., Woodward, J., Barrell, B. G., Parkhill, J and Hopwood, D. A. 2002. Complete genome sequence of the model Actinomycete

Streptomyces coelicolor, 2: *Nature*, 417: 141 - 147.

11) Bister, B., Bischoff, D., Strobele, M., Riedlinger, J., Reicke, A., Wolter, F., Bull, A. T., Zahner, H. Fiedler, H. P and Sussmuth, R. D. 2004. Abyssomicin C - A polycyclic antibiotic from a marine *Verrucosispora* strain as an inhibitor of the p-aminobenzoic acid/tetrahydrofolate biosynthesis pathway. *Angew Chemical International Education,* 43 (19): 2574 - 2576.
12) Blunt, J. W., Copp, B. R., Hu, W., Munro, M. H. G., Northcote, P. T and Prinsep, M. R. 2007. Marine natural products. *Natural Product Reproduction,* 24: 31 – 86.
13) Bredholt, H., Fjaervik, E., Jhonsen, G and Zotechev, S. B. 2008. Actinomycetes from sediments in the Trondhein Fjrod, Norway: Diversity and biological activity. *Journal of Marine Drugs,* 6: 12 - 24.
14) Busti, E. 2006. Antibiotic-producing ability by representatives of a newly discovered linage of actinomycetes. *Journal of Microbiology,* 152: 675 – 683.
15) Chun, J., Seong, C. N., Bae, K. S., Lee, K. J., Kang, S. O., Goodfellow, M and Hah, Y. C., 1998. *Nocardia flavorosea*. *International Journal of Systematic Bacteriology*, 48: 901 – 905.
16) Cohn, F. 1975. Under Suchungen Uber Bakterien II. Beitr. *Biol Pflanz*, 11, 188 - 204.
17) Corke, C. T. and Chase, F. E. 1956. The selective enumeration of actinomycetes in the presence of large number of fungi. *Canadian Journal of Microbiology,* 1: 12 - 16.
18) Crook, P., Carpenter, C. C and Klens, P. F. 1950. The use of sodium propionate in isolating actinomycetes from soils. *Science*, 112: 656.
19) Das, S., Lyla, P. S and Khan, S. A. 2008. Distribution and generic composition of culturable marine actinomycetes from the sediments of Indian continental slope of Bay of Bengal. *Chinese Journal of Oceanology and Limnology,* 26 (2): 166 - 177.
20) Das, S., Lyla, P. S., and Khan, S. A. 2008. Distribution and generic composition of culturable marine actinomycetes from the sediments of Indian continental slope of Bay of Bengal. *Chinese Journal of Oceanology and Limnology,* 26 (2): 166 - 177.
21) Demain, A. L. 2002. Prescription for an ailing pharmaceutical and drug industries. *Natural Biotechnology*, 20: 331.
22) Dhanasekaran, D., Panneerselvam, A and Thajuddin, N. 2005. Antifungal actinomycetes in marine soils of Tamilnadu. *Geobios,* 32: 37-40.
23) Dhanasekaran, D., Panneerselvam, A and Thajuddin N. 2008. An antifungal compound: 4'phenyl-1-napthyl–phenyl acetamide from *Streptomyces* spp. DPTB16. *Facta Universitatis Series: Medicine and Biology,* 15: 7 - 12.
24) Dhanasekaran, D., Selvamani, S., Panneerselvam, A and Thajuddin. 2009. Isolation and characterization of actinomycetes in Vellar Estuary, Annagkoil, Tamil Nadu. *African Journal of Biotechnology*, 8 (17): 4159 - 4162.
25) Dhanasekaran, D., Sivagami, P., Arunagirinathan, N., Panneerselvam, A and Thajuddin, N. 2005. Screening and identification of antibiotic producing strains of marine *Streptomyces*. *Journal of Microbial World*, 7 (1): 62 - 66.
26) Dhevagi, P and Poorani, E. 2006. Isolation and characterization of actionmycetes from marine sediments. *Journal of Microbial World,* 8 (1): 59 - 65.
27) Erikson, D. 1949. The morphology, cytology and taxonomy of the actinomycetes. *Annual Review of Microbiology*, 3: 23 - 54.
28) Farid, M. A., Enshasy, H. A. E., Diwany, A. I. E and Sayed, A. E. E. 2000. Optimization of the cultivation for Natamycin production by *Streptomyces natalensis*. *Journal of Basic Microbiology*, 3: 157 - 166.
29) Feling, R. H., Buchanan, G. O., Mincer, T. J., Kauffman, C. A., Jensen, P. R and Fenical, W. 2003. Salinosporamide A: a highly cytotoxic proteasome inhibitor from a novel microbial source, a marine bacterium of the new genus

Salinospora. *Chemistry International Journal*, 42: 355 - 357.

30) Fenical, W and Jensen, P. R. 2006. Developing a new resource for drug discovery: Marine actinomycete bacteria. *Nature Chemistry and Biology*, 2: 666 - 673.
31) Franco, M. M. C and Countinho, L. E. L. 1991. Detection novel secondary metabolites. *Critical Reviews of Biotechnology*, 11: 193-276.
32) Goodfellow, M and Haynes, J. A. 1984. Actinomycetes in marine sediments. *In:* Biological, Biochemical and Biomedical Aspects of Actinomycetes. Oritz - Oritz, L., Bojali, C. F. and Yakoleff, V. (eds.). *Academic Press*. New York, London. pp. 453-463.
33) Goodfellow, M and Williams, S. T. 1983. Ecology of actinomycetes. *Annual Reviews in Microbiology*, 37: 189 - 216.
34) Grein, A and Mayers, S. P. 1958. Growth characteristics and antibiotic production of actinomycetes isolates from littoral sediments and materials suspended in sea water. *Journal of Bacteriology*, 76: 457 – 463.
35) Gulve R. M and Deshmukh A. M. 2011. Enzymatic activity of actinomycetes isolated from marine sediments. *Recent Research in Science and Technology*, 3 (5): 80 - 83.
36) Gurung, T. D., Sherpa, C., Agrawal, P. V and Lekhak, B. 2009. Isolation and Characterization of Antibacterial Actinomycetes from Soil Samples of Kalapatthar. *Nepal Journal of Science and Technology*, 10: 173 - 182.
37) Haefner, B., A. Esparis and J. Fabregas. 2003. Drugs from the deep: marine natural products as drug candidates. *Drug Discovery Today*, 8: 536 - 544.
38) Han, L., Huang, X. S., Sattler, I., Fu H. Z., Grabley, S and Lin, W. H., 2007. Two new constituents from mangrove *Bruguiera gymnorrhiza*. *Journal of Asian Natural Product Research*, 9: 327 - 331.
39) Harvey, A. 2008. Strategies for discovering drugs from previously unexplored natural products. *DDT,* 5: 294 – 300.
40) Harz, C. O. 1878. *Actinomyces bovis*, einneuerschimuel in den Grwenben dis Rinder, Dent. *Thiermed*, 5: 125 – 140.
41) Hayakawa, K., Sato, N and Obinata, T. 1991. Dynamic reorientation of cultured cells and stress fibers under mechanical stress from periodic stretching. *Experimental Cell Research*, 268: 104 – 114.
42) Hughes, C. C., Prieto Davo, A., Jensen, P. R and Fenical, W. 2008. The marinopyrroles, antibiotics of an unprecedented structure class from a marine *Streptomyces* sp. *Organization Letters*, 10 (4): 629 - 631.
43) Ilic, S. B., Konstantinovic, S. S and Todorovic, Z. B. 2005. UV/VIS Analysis and antimicrobial activity of *Streptomyces* isolates. *Medicine and Biology*, 12 (1): 44 - 46.
44) Ismet, A., Vikineswary, S., Paramaswari, S., Wong, W. H and Ward, A. 2004. Production and chemical characterization of antifungal metabolites from *Micromonospora* sp. M39 isolated from mangrove rhizosphere soil. *World Journal of Microbiology and Biotechnology*, 20: 523 - 528.
45) Iznaga, Y., Lemus, M., Gonzalez, L., Garmendia, L., Nadal, L and Vallin, C. 2004. Antifungal activity of Actinomycetes from Cuban soils. *Biotechnology Department*, 18: 494 – 496.
46) Jeffrey, L. S. H., Sahilah, A. M., Son, R and Tosiah, S. 2007. Isolation and screening of actinomycetes from Malaysian soil for their enzymatic and antimicrobial activities, *Journal of Tropical Agriculture and Food Sciences*, 35: 159 - 164.
47) Jensen, P. R., Gontang, E., Mafnas, C., Mincer, T. J and Fenical, W. 2005.Culturable marine Actinomycetes diversity from tropical Pacific Ocean sediments. *Applied and Environmental Microbiology*, 7: 1039 - 1048.
48) Jiang, C. L and Xu, L. H. 1990. Characteristics of the populations of soil Actinomycetes in Yannan. *Actinomycetes*, 1 (3): 67 - 74.
49) Kathiresan, K., Balagurunathan, R and Masilamani Selvam, M. 2005. Fungicidal activity of Marine Actinomycetes against

phytopathogenic fungi. *Indian Journal of Biotechnology*, 4: 271 - 276.

50) Kim, C. J., Lee, K. H., Shimazu, A. and Yoo, I. D. 1994. Re-isolation frequency of soil actinomycetes on multiple isolation media. *Journal of Applied Microbiology and Biotechnology*, 22: 329 - 331 (in Korean).
51) Kitouni, M., Boudemagha, A., Oulmi, L., Reghioua, S., Boughachiche, F., Zerizer, H., Handiken, H., Couble, A., Mouniee, D., Boulahrouf, A and Boiron, P. 2005. Isolation of actinomycetes producing bioactive substances from water, soil and tree bark samples of North - East of Algeria. *Journal of Medical Mycology*, 15: 45 – 51.
52) Kpehn, F. E and Carter, G. T. 2005. The evolving role of natural products in drug discovery. *Nature Reviews Drug Discovery*, 4: 206 - 220.
53) Kurtbokle, D. I., Murphy, N. E. and Sivasithamparam, K. 1992. Use of baceriophage for the selective isolation of thermophilic actinomycetes from composted eucalyptus bark. *Canadian Journal of Microbiology*, 39: 46 - 51.
54) Labeda, D. P. 1985. Actinomycete taxonomy: generic characterization, Developments in Industrial Microbiology. *Journal or Industrial Microbiology*, 28: 115 - 121.
55) Lacey, J. 1978. Ecology of actinomycetes in fodder and related substrates, In: M. Mordarski, W. Kurylowicz and J. Jeljaszewicz (ed.) Nocardia and *Streptomyces*: Proceedings of the international symposium on *Nocardia* and *Streptomyces*. Warsaw. October 1976. *Gustav fisher Verlag*, *Stuttgart*: 161 - 168.
56) Lakshmipathy, D and Kannabiran, K. 2010. Isolation and Characterization of Antagonistic Actinomycetes from Marine Soil, *Journal of Microbial and Biochemical Technology*, 2 (1): 001 - 006.
57) Lam, K. S. 2006. Discovery of novel metabolites, *Current opinion in microbiology*, 9: 245 - 251.
58) Lechevalier, H. A. 1989. The Actinomycetes III. A practical guide to generic Identification of Actinomycetes. *Bergey's Manual of systematic Bacteriology*. Williams and Wilkins Company, Baltimore, 4: 2344 - 2347.
59) Lee, J. Y and Hwang, B. K. 2002. Diversity of antifungal actinomycetes in various vegetative soils of Korea. *Canadian Journal of Microbiology*, 48 (5): 407 – 417.
60) Liua, Z., Shi, Y., Zhang, Y., Zhou, Z., Li, W., Huang, Y. 2008. Rodrigues, C.; Goodfellow, M. Classification of related species and the transfer of "*Microstreptospora cinerea*" to the genus *Streptomyces* as *Streptomyces yanii* sp. nov. *International Journal of Systematic Evoltionary Microbiolohy*, 55: 1605 - 1610.
61) Mathis, D., Vence, L and Benoist, C. 2001. Beta–cell death during progression to diabetes. *Nature*, 414: 792 - 798.
62) Merrick, M. J. and Edwards, R. A. 1995. Nitrogen control in bacteria. *Microbiology Reviews*, 59: 604 - 622.
63) Mincer, T. L., Jensen, P. R., Kauffman, C. A., and Fenical, W. 2002. Widespread and persistent populations of a major new marine actinomycetes taxon in ocean sediments. *Applied Environmental Microbiology*, 68: 5005 – 5011.
64) Naggar, M. Y., El-Assar, S. A. and Abdul-Gaward, S. M. 2006. Meroparamycin production by newly isolated *Streptomyces* sp. Strains MAR01: Taxonomy, fermentation purification and structural elucidation. *Journal of Microbiology*, 7 (8): 432 - 438.
65) Nakeeb, M. A. E and Lechevalier, H. A. 1963. Selective isolation of aerobic Actinomycetes. *Applied Microbiology*, 11: 75 – 77.
66) Nathan A. Magarvey, Keller, J. M., Bernan. V., Dworkin, M and Sherman, D. H. 2004. Isolation and Characterization of Novel Marine - Derived Actinomycete Taxa Rich in Bioactive Metabolites. *Applied Environmental Microbiology*, 70 (12): 7520 – 7529.
67) Newman, D. J and Cragg, G. M. 2007. Natural products as sources of new drugs over the last 25 years. *Journal of Natural Products*, 70: 461 - 477.
68) Nolan, R and Cross, T. 1988. Isolation and Screening of actinomycetes, In: Good fellow M, Williams ST, Mordarski M (ed).

Actinomycetesin Biotechnology. *Academic Press, San Diego*, pp. 1-32.

69) Nonomura H. Key. 1974. For classification and identification of 458 species of the *Streptomycetes* included in ISP. *Journal of Fermentation Technology*, 52: 78 - 92.

70) Nonomura, H and Hayakawa, M. 1988. New methods for the selective isolation soil actinomycetes. In: Y. Okami, T. Beppu and Ogawara, (eds). Biology of Actinomycetes'88. *Japan Scientific Societies Press, Tokyo*.

71) Okami, Y and K. Hotta. 1988. Search and discovery of new antibiotics, In: Goodfellow M, Williams ST, Mordarski M (ed). Actinomycetes in Biotechnology. *Academic Press, San Diego*, pp. 33-67.

72) Okami, Y. 1952. Utilization of Nitrogen compounds by *Streptomycetes* and its application to classification, *Japan Journal of Medicinal Science and Biology*, 5: 265 - 275.

73) Okami, Y., Ogubye, H and Okazaki, T. 1972. Studies on marine microorganisms from the sea. *Journal of Antibiotics*, 25: 456 – 460.

74) Okami, Y., Okazaki, T and Hotta, K. 1988. Search and discovery of new antibiotics, In: Goodfellow M, Williams ST, Mordarski M (ed). Actinomycetes in Biotechnology. *Academic Press, San Diego*, pp. 33-67.

75) Oskay, M., Tamer, A. U and Azeri, C. 2004. Antibacterial activity of some actinomycetes isolated from farming soils of Turkey. *African Journal of Biotechnology*, 3 (9): 441 – 446.

76) Pandey, B., Ghimire, P and Agarwal, V. P. 2004. Studies on the antibacterial activity of actinomycetes isolated from the Khumbu region of Mt. Everst. A paper presented in the International conference on the Great Himalayas: Climate, Health, Ecology, Management and Conservation, Kathmandu.

77) Parungao, M. 2007. Screening of Antibiotic-Producing Actinomycetes From marine, Brackish and Terrestrial Sediments of Samal Island, Philippines. *Journal of Research in Science*, Computing and Engineering, 4: 29 - 38.

78) Pelaez, F. 2006. The historical derive of antibiotic from microbial natural product can history repeat. *Journal of Biochemical Pharmacology*, 71: 981 - 990.

79) Prabavathy, V. R., Vijayanandraj, V. R., Malarvizhi, K., Mathivanan, N., Mohan, N., and Mrugesan, K. 2009. Role of actinomycetes and their metabolites in crop protection. In: Agriculturally Important Microorganisms (Vol. I) (Eds).

80) Pridham, T. G and Gottlieb, D. 1948. The utilization of carbon compounds by some Actinomycetes as an aid for species determination. *Journal of Bacteriology*, 56: 107 - 114.

81) Pridham, T. G., Anderson, P., Foley, E., Lindenfelser, L. A., Hesseltine, E. W and Benedict, R. G. 1957. A selection of media for maintenance and taxonomic study of *Streptomyces*. *Antibiotics*, 947 - 953.

82) Pugazhvendan, S. R., Kumaran, S., Alagappan, K. M and Prasad, G. 2010. Inhibition of Fish Bacteriology Pathogens by Antagonistic Marine Actinomycetes, *European Journal of Applied Sciences*, 2 (2): 41 - 43.

83) Ramesh, S. 2009. Marine actinomycetes diversity in Bay of Bengal, India: Isolation and characterization of bioactive compounds from *Streptomyces fungicidicus* MML 1614. Ph.D., thesis, University of Madras, Chennai, India.

84) Ramesh, S and Mathivanan, N. 2009. Screening of marine actinomycetes isolated from the Bay of Bengal, India for antimicrobial activity and industrial enzymes. *World Journal of Microbiology and Biotechnology*, DOI: 10.1007/s11274-009-0113 - 4.

85) Rath, J. P., Kinast, S and Maier, M. E. 2005. Synthesis of the full functionalized core structure of the antibiotic abyssomicin. *Organization Letter*, 7: 3089 - 3092.

86) Ravel, J., Amoroso, M. J., Colwell R. R. and Hill R. T. 1998. Mercury resistant Actinomycetes from the Chesapeake Bay. *FEMS Microbiology Letters*, 62: 172 - 184.

87) Ravikumar, S., Suganthi, P and Mose, F. 2011. Crude bioactive compounds of actinomycetes

from Manarkudy mangrove sediment. *Journal of Pharmacy Research*, 4 (3): 877 - 879.

88) Remya, M and Ramasamy Vijayakumar. 2008. Isolation and characterization of marine antagonistic actinomycetes from west coast of India. *Medicine and Biology*, 15 (1): 13 – 19.

89) Sahin, N and Ugur, A. 2003. Investigation of the Antimicrobial activity of some *Streptomyces* isolates. *Turkey Journal of Biology*, 27: 79 – 84.

90) Sateesh, V and Rathod, J. L. 2011. Selective isolation and antimicrobial activity of rare actinomycetes from mangrove sediment of Karwar. *Journal of Ecobiotechnology*, 3 (10): 48 - 53.

91) Seong, C. N., Chei, J. H and Baik, K. S. 2001. An improved selective isolation of race actinomycetes from forest soil. *Journal of Microbiology*, 39 (1): 17 – 23.

92) Settle, L. D., De Oliveria, V. M and Manfio, G. P. 2005. Isolation and Characterization of alachlor – degrading Actinomyetes. *Antonie Van Leewenhoek*, 87 (2): 81 – 89.

93) Shadomy, S. 1987. Preclinical evaluation of antifungal agents in Recent trends in the discovery, development and evaluation of antifungal agents. New Jercy. *Prous Science*, 8 - 14.

94) Shantikumar Singh, L., Indra Baruah and Bora, Z. T. C. 2006. Actinomycetes of Loktat Habitat: Isolation and screening for Antimicrobial activities. *Biotechnology*, 5 (2): 217 - 221.

95) Shapiro, S. 1989. Nitrogen Assimilation in Actinomycetes and the Influence of Nitrogen Nutrition on Actinomycetes Secondary Metabolism. In: Regulation of Secondary Metabolism in Actinomycetes, Shapiro, S. (Ed.). CRC Press, Boca Raton, Florida, pp: 135-211.

96) Sheridan, C., 2006. Antibiotics an natural. *Natural Biotechnology*, 24: 1494 - 1496.

97) Shirling, E. G. and Gottlieb, D. 1966. Methods for characterization of *Streptomyces* species. *Int*ernational *Journal of Systematic Bacteriology*, 16: 313 - 340.

98) Singh, L. S., Baruah, I and Bora, T. C. 2006. Actinomycetes of Loktak Habitat: Isolation and screening for antimicrobial activities. *Biotechnology*, 5 (2): 217 – 221.

99) Sivakumar, K., Haritha, R., Jagan Mohan, Y. S. Y and V. Ramana. 2011. Screening of marine Actinobacteria for antimicrobial compounds. *Research Journal of Microbiology*, 6 (4): 385 - 393.

100) Solanki, R., Khanna, M. and Lal, R. 2008. Bioactive compounds from marine actinomycetes. *Journal of Indian Microbiology*, 48: 410 – 431.

101) Solingen, P., Dean, V. M., Wilhelmus, A. H. K., Christopher, B., Robertus, B., Scott, D. P. and Brian E. J. 2001. From a novel *Streptomyces* isolated from an East African Soda Lake. *Extremophiles*, 5: 333 - 341.

102) Sponga, F., Cavaletti, L., Lazzarini, A., Borghi, A., Ciciliato, I., Losi, D. and Marinelli, F. 1999. Biodiversity and potentials of marine derived microorganisms. *Journal of Biotechnology*, 70: 65 – 69.

103) Srinivasan, M. C., Laxman R. S and Deshpande, M. V. 1991. Physiology and nutrition aspects of actinomycetes – An overview. *World Journal of Microbial and Biotechnology*, 7: 171 - 184.

104) Srivibool, R and Sukchotiratana, M. 2006. Bioperspectine of actinomycetes isolates from coastal soils. A new source of antimicrobial procedures. *Journal of Science and Technology*, 28 (3): 493 – 499.

105) Stolpe, N and N. Godkeri. 1981. Non-pathogenic members of genus *Pseudomonas*. In: The prokaryotes, Ed. Marthiner*et al*., *Springer Verlag, New York*, pp. 719-741.

106) Stryzhkova, H. M., Kopeiko, O. P., Lavrinchuk, V. L., Bambura, O. L and Matscliukh, B. P. 2002. Spontaneous and induced variability of *Streptomyces aurofaciensch* lortetracycline producer. *Microbial Zentable*, 64: 19 - 23.

107) Sujatha Peela, V. V. S. N., Bapiraju Kurada, and Ramana Terli. 2005. Studies on antagonistic marine actinomycetes from the

Bay of Bengal. *World Journal of Microbiology and Biotechnology,* 21: 583 – 585.

108) Takizawa, M., Colwell, R. R and Hill, R. T. 1993. Isolation and diversity of Actinomycetes in the Chesapeake. *Applied Environmental Microbiology*, 59 (4): 997 - 1002.

109) Thenmozhi, M and Kannabiran, K. 2010. Studies on Isolation, Classification and Phylogenetic Characterization of Novel Antifungal *Streptomyces* sp. VITSTK7 in India. *Current Research Journal of Biological Sciences,* 2 (5): 306 - 312.

110) Umezawa, Y., Miyajima, H., Ayanne, K and Oike, I. K. 1985. In press. Significance of ground water nitrogen discharge into coral reefs at Ishigaki Island, Southwest of Japan. Coral Reefs.

111) Vanajakumar, G., Selvakumar, N and Natarajan, R. 1995. Antagonistic properties of actinomycetes isolated from mollusks of the porto Novo region. *South India*, 267 - 274.

112) Veiga, M., Esparis, A and Fabregas, J. 1983. Isolation of cellulolytic Actinomycetes from the marine sediments. *Applied Environmental Microbiology*, 46: 286 - 287.

113) Vezina, C., Kudelski, A., Sehgal, S. N. 1975. Rifamycin (AY – 22, 989), a new antifungal antibiotic Taxonomy of producing *Streptomyces* and Isolation of the active principle. *Journal of Antibiotics,* 28: 721 – 726.

114) Waksman, S. A and Schatz, A. 1943. Strain Specificity and Production of Antibiotic Substances. Proceedings in National Academy of Science, USA, 29 (2): 74 – 79.

115) Waksman, S. A. 1961. The Actinomycetes classification, identification and description of genera and species. Vol - II, Williams and Wilkins Co., Baltimore, U.S.A.

116) Watve, M. G., Tickoo, R., Jog, M. M and Bhole, B. D. 2001. Antibiotics are produced by the genus *Streptomyces*. *Archives of Microbiology,* 176: 386 – 390.

117) Weyland, H. 1969. Actinomycetes in North Sea and Atlantic Ocean sediments. *Nature,* 223: 858.

118) White, J. D., Hanselman, R., Jackson, R.W., Porter, W. J., Ohba, Y., Tiller, T. and Wang, S. 2001. Total synthesis of Rutamycin B, amacrolide antibiotic from *Streptomyces aurofacins*. *Journal of Organic Chemistry,* 66: 5217 - 5231.

119) Wise, R. 2008. The worldwide threat of antimicrobial resistance. *Current Science,* 95: 181 - 187.

120) Yang, S. S. and Ling, M. Y. 1989. Tetracycline production with sweet potato residue by Solid state fermentation. *Journal of Biotechnology and Bioengineering,* 33: 1021-1028.

121) Yassin, A. F., Galinski, E. A., Wohlfarth Jahnke, K. D., Schaal, K. P. and Truper, H. G. 1993. A new Actinomycetes species *Nocardiopsis lucentensis. International Journal of Systematic Bacteriology*, 43 (2): 266 - 271.

122) You, J., Cao, L., Liu, G., Zhou, S., Tan, H and Lin, Y. 2005. Isolation and Characterization of actinomycetes antagonistic to pathogenic *Vibrio* spp. from near shore marine sediments. *World Journal of Microbiology,* 21 (5): 679 – 682.