CHAPTER I

INTRODUCTION

Antibiotics are secondary metabolites, mostly produced by actinomycetes (71.1%), some are produced by fungi (18.2%), e.g. penicillin and cephalosporin, and by bacteria (10.7%), e.g. bacitracin and polymyxin. From all actinomycetes producing antibiotics, 80.2% (3500 antibiotics) produced by *Streptomyces*, 6% (258 antibiotics) produced by *Micromonospora* and 3.7% (156 antibiotics) produced by *Nocardia*, and another actinomycetes producing antibiotics were reported in *Streptoverticillium, Actinoplanes, Actinomadura, Streptosporangium, Saccharopolyspora, Dactylosporangium, Chainia, Nocardiopsis, Ampullariella, Amycolatopsis, Kitasatosporia, Pseudonocardia, Saccharothrix, Microtetraspora, Microellobosporia, Streptoalloteichus, Actinosporangium, Kibdelosporangium, Actinosynema, Planobispora, Microbispora, Planomonospora* and *Saccharomonospora*,etc. (Goodfellow and Williams, 1983; Oki, 1994)

Antibiotics have been extensively used as therapeutic agents, and also in agriculture, and food industry. The actinomycetes provided about two-third (more than 4000) of the naturally occurring antibiotics discovered, including aminoglycosides, anthracyclines, macrolides and tetracyclines (Goodfellow, 1988). There are numerous groups of actinomycetes, for example, *Streptomyces, Micromonospora, Microbispora, Actinomadura, Actinomyces, Actinoplanes and Streptoverticillium*. They are widely distributed in soil and in a variety of habitats, including composts, river mud, and lake bottoms. The best-known genus of antibiotic producing actinomycetes is *Streptomyces*, which is one of the most commonly actinomycetes isolated from soil (Tortora et al., 1995).

Streptomyces is belong to family Streptomycetaceae, order Actinomycetales. Strains in this genus have substrate and aerial mycelium, with vegetative hyphae of average 0.5-2.0 μm in diameter. Substrate mycelium color and soluble pigment are blue, dark, green, red-orange or violet. Spore surface is smooth, hairy, spiny, warty or

rugose. Spore chain morphology is spirals, rectifilexibiles, or retinaculiaperti. Spore colors are blue, gray, green, red, violet, white or yellow. Cell wall contains L-diaminopimelic acid indicated chemotype II. Peptidoglycan typed A3γ. Fatty acid pattern 2C. The major menaquinone was MK-9, and phospholipids type PII. Glucose, glycerol, mannitol, fructose and sucrose are present in cell hydrolysates. The range of G+C content of the DNA was 69-78 mol%. Colony on agar media is granular, powder, velvety, or floccuse. Optimal pH is in the range of 6.5-8.0. Growth occurs normally between 25°C and 30°C but not above 50°C (Goodfellow, 1988).

Most of well known commercial antibiotics are produced by *Streptomyces*, for example, chloramphenicol produced by *Streptomyces venezuelae*, cycloheximide produced by *Streptomyces griseus*, cycloserine produced by *Streptomyces orchidaceus*, kanamycin produced by *Streptomyces kanamyceticus*, lincomycin produced by *Streptomyces lincolnensis*, neomycin produced by *Streptomyces fradiae*, nystatin produced by *Streptomyces nourisei*, streptomycin produced by *Streptomyces griseus*, erythromycin produced by *Streptomyces erythreus*, tetracycline produced by *Streptomyces* rimosus (Oki, 1994), luisol A and B produced by *Streptomyces* sp. CNH-370 and arenaric acid produced by *Streptomyces* sp. CNH-248 (Cheng, *et. al.* 1999).

In addition, *Micromonospora* strains also produced secondary metabolites with diverse chemical structures, and biological activities, e.g. gentamycin, sagamicin, megalomicin, mycinamicin, halomicin, mutamicin. *Micromonospora* strains are aerobic, gram-positive, mesophilic, non-motile actinomycetes which produced single non-motile spore directly from substrate hyphae in generally indicated orange color. Cell wall contained *meso*-diaminopimelic acid. The predominant menaquinones were MK-9, MK-10, and MK-12. The range of G+C content of the DNA was 71-73 mol% (Glasby, 1993).

Recently, Tamura and Sakane (2005) isolated *Asanoa iriomotensis* sp. nov. from mangrove soil in Japan. Takeuchi and Hatano (2001) isolated three strains, *Agromyces lutedus* sp. nov., *A. rhizospherae* sp. nov. and *A. brachium* sp. nov. from the mangrove rhizosphere. Kullama and et. al. (2000) isolated and screened the actinomycetes from mangrove soil in Ranong province area.

Mangrove forest is interesting sources of new secondary metabolites and bioactive compounds because of biodiversity and sophisticated ecological system. The species living in this surrounding are endurable to the changes of salinity and rugose. Spore chain morphology is spirals, rectifilexibiles, or retinaculiaperti. Spore color is blue, gray, green, red, violet, white or yellow. Cell wall contains L-diaminopimelic acid indicated chemotype II. Peptidoglycan typed A3γ. Fatty acid pattern 2C. The major menaquinone was MK-9, and phospholipids type PII. Glucose, glycerol, mannitol, fructose and sucrose are present in cell hydrolysates. The range of G+C content of the DNA was 69-78 mol%. Colony on agar media is granular, powder, velvety, or floccuse. Optimal pH is in the range of 6.5-8.0. Growth occurs normally between 25°C and 30°C but not above 50°C (Goodfellow, 1988).

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rhizosphere. Kullama and et. al. (2000) isolated and screened the actinomycetes from mangrove soil in Ranong province area.

Mangrove forest is interesting sources of new secondary metabolites and bioactive compounds because of biodiversity and sophisticated ecological system. The species living in this surrounding are endurable to the changes of salinity and current between low and high tides. Furthermore, mangrove sediments is rich of organic matters consistent with high sulfur and nitrogen (ประเสริฐ โพธิปักษ์ 2540), which can be used in secondary metabolite production of microorganism. Mangrove rhizosphere is also full of decayed organic matters originated from alluvium (สนิท อักษรแก้ว 2542), with a pH value range of acid to alkaline. Mangrove forests along the inner gulf of Thailand in Samut prakan, Samut sakorn, Samut songkarm and Phetchaburi provinces are lush green and preserved. Therefore, they are an interesting niche environments to isolate a noval antimicrobial producing actinomycetes. The actinomycetes was isolated from both air dried and heat treated mangrove soil to enhance an opportunity of rare actinomycetes isolation. In this present study, we isolated antimicrobial producing actinomycetes from niche environment, mangrove soil.

The objectives of this study :

1. To isolate antimicrobial producing actinomycetes from mangrove soils of the inner gulf of Thailand.
2. To identify the selected actinomycetes based on the phenotypic and chemotaxonomic characteristics including 16S rDNA sequencing.
3. To determine the antimicrobial activity of the selected actinomycetes.