CHAPTER I

INTRODUCTION

When comparing the earth age of 4,600 million with the ocean age of 3,500 to 4,400 million years, in addition, the ocean volume is about $1,370 \times 10^6$ km^3 and covers 71% of the earth's surface which provides space for life more than 300 times of terrestrial. It can be concluded that the ocean is the largest and the oldest habitat in this planet. The ocean water temperature is more constant than the air. Furthermore, water is the universal solvents and can dissolve more substances than others. Therefore, it's used to believe that the earliest organism was formed in the ocean before it migrated onto the dry land. The early objective of the human used the ocean as a source of food producer. Since the exploration of challenger in late 1876, the ocean was of interest in a new biological resource. Particularly, in the mid to late 1900s, the new techniques were developed to investigate life form in the sea (Lali, and Parsons, 1997).

Research on terrestrial bioactive compounds has been done for several decades. In early 1960, the researcher scrutinized the ocean as a new unexplored resources of bioactive compounds. In late 1960s, the development of scuba diving facilities brought a new direction to expand the exploration on marine plants and animals. Over 6,000 compounds were discovered by marine natural product chemists. The target is soft-body organisms such as sponges, soft-coral, tunicates, algae and bryozoan. As similar as primary objectives with searching bioactive substances on terrestrial, the pharmaceutical agents were prospected from marine organisms such as antimicrobial, antitumour, antiviral and anti-inflammatory activities (Davidson, 1995). The examples of bioactive compounds from marine organisms are described below.

1. Dysidazirinne, a novel azacyclopropene, exhibited the activity against gram negative bacteria and showed the activity on antitumor. This compound was isolated from the sponge, *Dysidea fragilis* (Molinski, and Ireland, 1988 cited in Kobayashi, and Ishibashi, 1992).

2. The antivirals, avarol and avarone have been reported to inhibit human immunodeficiency virus were isolated from the sponge, *Disidea avara* (Blunden, 1996)

3. Valdivones A and B, dihydrovaldivone and methoxy ketals have been isolated from the *Alcyonium valdivae*, possessing the antiinflamatory (Benjadol, 1998).

Since the discovery of penicillin in 1929, microorganisms were expected as source of abundance potential bioactive compounds. More than 60 years ago, 30,000- 50,000 natural products were discovered from microorganisms and over 10,000 compounds were bioactive compounds (Fenical, 1993). After the discovery of novel bioactive agents, the development of useful bioactive agents for fermentative industry should aid in investigations. There are several advantages of using microbes for industrial productions, for instant;

1. microorganisms have the advantages in reproducible production in short time and giving large quantities when comparing with higher organisms.

2. production of microorganisms can be magnified more easily than higher organisms.

3. reliable on escape of organism was necessary for the large scale industrial production, microorganisms were cultured in a close fermentation system. It is easier to control microorganisms, that were cultured in a close system while higher organisms have to be cultured in open systems (Okami, 1980).

Until now, more than 100 products have been developed for pharmaceutical and agriculture purposes. Today, the interesting in marine microorganisms have been ridden both academy and industry. The budget was spent more than 9 billions US$ per year for exploring new novel bioactive compounds. The study in bioactive compounds isolated from microorganisms still continues but the discovery rate is decreasing. More than 90% of bioactive compounds were known compounds (Fenical and Jensen, 1993).

Scientists have searched for new bioactive metabolites and considered that marine microorganisms are new bioactive metabolite resources. The ocean has the great extremely environments for examples, pressure, salinity and temperature; to survive in such extremely conditions, marine microorganisms have adapted in physiological and metabolites capabilities. So they offer bioactive metabolites which could not be found in terrestrial organisms (Fenical, 1993). The earliest study on bioactive marine microorganisms concentrated on the development of new pharmaceutical agents (Needham, Andersen, and Kelly, 1991). The examples of bioactive substances which were discovered in marine microorganisms are:

1. A new antibiotic, aplasmomycin was obtained from a strain of *Streptomyces griseus* isolated from shallow sea sediment in Sagami Bay. This compound inhibited growth of gram-positive bacteria including mycobacteria (Okami, Okazaki, and Kitahara, 1976).

2. Macrolactin A, a member of macrolactins group, had been isolated from an unidentified deep sea bacterium. The selective antibacterial agents inhibited B16-F10 murine melanoma cancer cells *in vitro* assays and *Herpes simplex* virus (type I and type II) (Rychnovsky *et al*., 1992).

3. Pseudomic acid (Mupirocin), a new antibiotic produced by *Pseudomonas fluorescens* showed high level of antibacterial activity against Streptococci and Staphylococci, these microorganisms have responsible to infection of the human skin (Sutherland *et al*., 1985).

One of the purposes of the study in marine microorganisms is to understand their ecological metabolites. In the complicated marine habitats, symbiosis is one life form can be found in extremely environments. Symbiotic microorganisms are commonly found in marine organisms, and considered to be great important resources of biologically active natural products (Bell and Carmeli, 1994). This biological active substance was suggested to protect their host from the other decease organisms (Pathirana *et al*., 1991). The symbiosis bacteria especially associated with their host are distinct from that in surrounding seawater. By studying on symbiosis bacteria metabolites, the relationship between marine organisms and bacteria symbionts could be described. As more evidence is obtained, the origin of metabolite that used to believe that it was produced by the host organisms, is responsible by bacteria associated. Several of them were bioactive substances (Fenical and Jensen, 1993). Examples of this case are:

1. Latrunculin A was found in the Fijian sponge *Spongia mycofijiensis* associated with the nudibranch, *Chromodoris lochi.* It was suggested to contain defensive allomone activity (Kakou, Crews, and Bakus, 1987 cited in Kobayashi, and Ishibashi, 1992).

2. The causative toxins of a food poisoning by neosurugatoxin and prosurugatoxin from consumping of the Japanese ivory shell, *Babylonia japonica.* The bacteria associated in the digestive gland of this shell was proved to be a real producer of these toxins (Kosuge *et al*., 1985).

3. The embryo of American lobster *Homarus amesicanus* was protected from the infection of the fungus *Lagenidium callinectes* (the pathogen of many crustaceans) by antifungal substances, 4-hydroxyphenethyl alcohol (tyrosol) which was produced from an unidentified gram-negative epibiont bacteria, SGT-76 (Sofia, Turnes, and Fenical, 1992).

As mentioned above, marine microorganisms are potential resources for searching novel bioactive agents which can be developed to industrial production. *Alteromonas* sp. is one of marine microorganisms that was of interest by chemists. Several bioactive agents were discovered and reported from this genus. These marine bacteria may produce novel bioactive agents.

The main objectives of this investigation are as follows:

1. To isolate bioactive substances from a marine bacterium, *Alteromonas* sp., S9730 and elucidate the chemical structures of the isolated substances.
2. To study antibacterial activities of the isolated substances.