CHAPTER I

INTRODUCTION

Marine organisms such as bacteria, sponges, soft corals, nudibranchs, tunicates etc. are very important sources for the discovery of structurally diverse secondary metabolites. These compounds have interesting biological activities that are potentially useful as (a) new and clinically useful drug, (b) new bioactive lead compounds for the total synthesis and further drug development of more effective analogs, and (c) molecular and biomedical research tools for elucidating sites and mechanisms of biological actions. One important application of the bioactive compounds derived from marine organisms is their use as molecular probes which can be used for investigation of biochemical events (Suwanborirux, 2002; Yeung and Paterson, 2002).

Since 1986, a number of trisoxazole macrolides have been isolated from various marine organisms such as sponges, nudibranchs, and stony corals. This group of compounds consists of ulapualides, kabiramides, mycalolides, halichondramides, halishigamides, and jaspisamides (Roesener and Scheuer, 1986; Kernan et al., 1988; Matsunaga et al., 1989; Kobayashi et al., 1993; 1997; Matsunaga et al., 1998a; Matsunaga et al., 1998b). All compounds in this group share the same basic structure containing a twenty-five membered lactone ring with three consecutive oxazole rings and an eleven-carbon side chain with an *N*-methyl formyl terminal group. These compounds showed various biological activities including antifungal, antileukemic, ichthyotoxic and cytotoxic properties. Several members of the trisoxazole macrolide family exhibited actin monomer sequestering and filament severing activities (Saito et al., 1994; Wada et al., 1998). Recently, some studies reported that kabiramide C and related macrolides bound to G-acin in 1:1 stoichiometry and functioned as unregulated biomimetics of actin filament barbed-end-capping proteins (Klenchin et al., 2003; Tanaka et al., 2003).

These investigations suggested that the optical and chemical probes based on trisoxazole macrolides, typified by kabiramides, could provide an opportunity to develop a new class of optical probes that serves as an indicator of molecular events at the actin filament barbed-end and may lead to new therapies to treat diseases of actin cytoskeleton.

This new group of fluorescent kabiramide probes should exhibit a more selectivity and specific association with the barbed end of actin filaments. While cytochalasin D and its relatives were known to bind to the barbed end of actin filaments, this interaction was reversible and relatively weak. Cytochalasin D also exhibited a filament severing activity which was associated with a weaker binding of the drug to sites within the filament (Forscher and Smith, 1998). Additionally, certain actin probes such as GFP-actin (Choidas et al., 1998) and TMR-phalloidin (Faulstich et al., 1988) are useful in mapping gross changes in actin filament dynamics within a cell but they do not provide specific information on molecular events at the barbed end.

As part of our investigation for bioactive compounds from Thai marine organisms, the grayish black sponge *Pachastrissa nux* was collected in February, 2003 from Sichang Island, Chonburi Province, Thailand at the depth of 5-10 meters. The EtOAc extract from the sponge exhibited remarkable antifungal activity against *Candida albicans* ATCC 10231. Additionally, the ^{1}H NMR spectra of the EtOAc extract showed some characteristic peaks of the trisoxazole macrolide kabiramides in the downfield region, including three singlets of trisoxazole protons and two singlets of *N*-formamide proton in the ratio of 2:1 due to the two geometrical forms (Matsunaga et al., 1986; 1989). Therefore, the objectives of this research are to

1. isolate kabiramides from the Thai marine sponge, *Pachastrissa nux*.
2. elucidate the structures of the isolated kabiramides.
3. synthesize the fluorescent derivatives of kabiramide C.
4. characterize the actin-binding properties of the isolated and synthetic fluorescent derivatives of kabiramide C.