

คาบิเรไมด์ สารกลุ่มทรืสออกซาโซลมาโครไลด์ที่ยึดเหนี่ยวแอกดินจากฟองน้ำทะเล
Pachastrissa nux ของไทย และอนุพันธ์กึ่งสังเคราะห์ชนิดเรืองแสงของคาบิเรไมด์ ที

นางสาวชุตินา เพ็ชรประยูร

วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรดุษฎีบัณฑิต

สาขาวิชาเภสัชเคมีและผลิตภัณฑ์ธรรมชาติ

คณะเภสัชศาสตร์ จุฬาลงกรณ์มหาวิทยาลัย

ปีการศึกษา 2547

ISBN 974-53-2125-7

ลิขสิทธิ์ของจุฬาลงกรณ์มหาวิทยาลัย

KABIRAMIDES, THE ACTIN-BINDING TRISOXAZOLE MACROLIDES FROM
THE THAI MARINE SPONGE *PACHASTRISSA NUX*, AND SEMISYNTHETIC FLUORESCENT
DERIVATIVES OF KABIRAMIDE C

Miss Chutima Petchprayoon

A Dissertation Submitted in Partial Fulfillment of the Requirements
for the Degree of Doctor of Philosophy in Pharmaceutical Chemistry and Natural Products

Faculty of Pharmaceutical Sciences

Chulalongkorn University

Academic Year 2004

ISBN 974-53-2125-7

ชุติมา เพ็ชรประยูร : คาบิเรไมด์ สารกลุ่มทริสออกซาโซลมาโครไลด์ที่ยึดเหนี่ยวแอกติน จากฟองน้ำทะเล *Pachastrissa nux* ของไทย และอนุพันธ์กึ่งสังเคราะห์ชนิดเรืองแสงของ คาบิเรไมด์ ซี. (KABIRAMIDES, THE ACTIN-BINDING TRISOXAZOLE MACROLIDES FROM THE THAI MARINE SPONGE *PACHASTRISSA NUX*, AND SEMISYNTHETIC FLUORESCENT DERIVATIVES OF KABIRAMIDE C)

อ. ที่ปรึกษา: ดร. คณิต สุวรรณบริรักษ์, จำนวนหน้า 188 หน้า. ISBN 974-53-2125-7.

จากการศึกษาทางเคมีของฟองน้ำทะเล *Pachastrissa nux* ที่พบในบริเวณเกาะสีชัง อ่าวไทย สามารถแยกสารกลุ่มทริสออกซาโซลมาโครไลด์ที่เคยพบแล้วได้ 2 ชนิด คือ kabiramide C และ D และ สารใหม่ 1 ชนิด คือ kabiramide F การพิสูจน์โครงสร้างทางเคมีของสารที่แยกได้เหล่านี้ ทำได้โดยการ วิเคราะห์ข้อมูลทางสเปกโตรสโคปีจาก MS UV IR และ NMR ร่วมกับการเปรียบเทียบข้อมูลกับเอกสาร ต่างๆ จากนั้นได้นำสาร kabiramide C (KabC) ซึ่งเป็นสารที่แยกได้ในปริมาณสูงมาทำการเปลี่ยนแปลง โครงสร้างเป็น 7-(4-aminomethyl-1H-1,2,3-triazol-1-yl)kabiramide C (AMT-KabC) โดยใช้ปฏิกิริยา Mitsunobu และ 1,3 dipolar cycloaddition เพื่อใช้เป็นสารมัธยันต์ในการสังเคราะห์อนุพันธ์เรืองแสง ของ kabiramide C จำนวน 5 ชนิด คือ อนุพันธ์ tetramethylrhodamine (TMR-KabC), rhodol green (RG-KabC), IC5 (IC5-KabC), dapoxy (DAP-KabC), and fluorescein diester (FDE-KabC) จาก ปฏิกิริยาของสาร AMT-KabC กับอนุพันธ์ N-succinimidyl ester ของสารเรืองแสง

ได้ทำการศึกษาคุณสมบัติของ kabiramide ที่แยกได้ และอนุพันธ์เรืองแสงที่สังเคราะห์ ในการ ยึดเหนี่ยวกับแอกติน โดยใช้เทคนิคต่างๆ ทางฟลูออเรสเซนซ์ ได้แก่ การวิเคราะห์สเปกตรัมการเรืองแสง การวัด fluorescence resonance energy transfer (FRET) การวัด fluorescence anisotropy (FA) และ การวัด iodide quenching พบว่า อนุพันธ์ kabiramide ทุกชนิด ยึดเหนี่ยวกับแอกตินเป็น สารประกอบเชิงซ้อนในอัตราส่วน 1:1 ได้ทดสอบอนุพันธ์เรืองแสงที่สังเคราะห์กับเซลล์ NIH 3T3 พบว่า เฉพาะอนุพันธ์ TMR-KabC และ FDE-KabC เท่านั้นที่สามารถผ่านเข้าสู่เซลล์ และแสดงการเรืองแสงที่ ชัดเจนในบริเวณขอบเซลล์ที่มีการเคลื่อนที่ นอกจากนี้ สาร kabiramide C, D และ F แสดงฤทธิ์ต้านเชื้อ *Candida albicans* โดยให้บริเวณยับยั้งการเจริญของเชื้อขนาดเส้นผ่านศูนย์กลาง 22 มิลลิเมตร ที่ความ เข้มข้น 100 ไมโครกรัมต่อดิสก์

สาขาวิชาเภสัชเคมีและผลิตภัณฑ์ธรรมชาติ
ปีการศึกษา 2547

ลายมือชื่อนิสิต
ลายมือชื่ออาจารย์ที่ปรึกษา

4476952533 : MAJOR PHARMACEUTICAL CHEMISTRY AND NATURAL PRODUCTS

KEY WORDS: KABIRAMIDE / TRISOXAZOLE MACROLIDE / ACTIN-BINDING / SEMISYNTHETIC FLUORESCENCE / *PACHASTRISSA NUX* SPONGE

CHUTIMA PETCHPRAYOON: KABIRAMIDES, THE ACTIN-BINDING TRISOXAZOLE MACROLIDES FROM THE THAI MARINE SPONGE *PACHASTRISSA NUX*, AND SEMISYNTHETIC FLUORESCENT DERIVATIVES OF KABIRAMIDE C. THESIS ADVISOR: KHANIT SUWANBORIRUX, Ph.D., 188 pp. ISBN 974-53-2125-7.

Three trisoxazole macrolides, including two known kabiramides C and D, and a new kabiramide F were isolated from the marine sponge *Pachastrissa nux*, collected from Sichang Island in the Gulf of Thailand. Their identification and structure elucidation were achieved by analyses of MS, UV, IR, 1D-NMR, and 2D-NMR spectral data as well as comparison with the literatures. The major compound, kabiramide C (KabC), was structurally modified to give the key intermediate 7-(4-aminomethyl-1*H*-1,2,3-triazol-1-yl)kabiramide C (AMT-KabC) by using Mitsunobu reaction and 1,3-dipolar cycloaddition reaction. Furthermore, five fluorescent conjugates of kabiramide C, including derivatives of tetramethylrhodamine (TMR-KabC), rhodol green (RG-KabC), IC5 (IC5-KabC), dapoxyl (DAP-KabC), and fluorescein diester (FDE-KabC) were synthesized by coupling AMT-KabC with *N*-succinimidyl esters of the fluorescence dyes.

The steady state fluorescence techniques, including fluorescence emission spectral analysis, fluorescence resonance energy transfer (FRET), fluorescence anisotropy (FA), and iodide quenching were used to determine the actin binding properties of these kabiramide derivatives. All of the isolated and semisynthetic fluorescent kabiramide derivatives bound stoichiometrically to G-actin in 1:1 complex. Five fluorescent kabiramides were further separately introduced to living NIH 3T3 cells. Only TMR-KabC and FDE-KabC showed good permeability through plasma membrane of the cells and expressed intense fluorescence at the protrusion sites. Additionally, kabiramides C, D, and F exhibited equal antifungal activity against *Candida albicans* with inhibition zone of 22 mm at the concentration of 100 µg/disc.

Field of Study Pharmaceutical Chemistry Student's signature.....
and Natural Products Advisor's signature.....

Academic year 2004

ACKNOWLEDGMENTS

I would like to express my appreciation to those who encouraged and assisted me in my research as follows:

Dr. Khanit Suwanborirux, my thesis advisor, of Department of Pharmacognosy, Faculty of Pharmaceutical Sciences, Chulalongkorn University, for his valuable advice, guidance, kindness, and encouragement throughout my research study.

Professor Dr. Gerard Marriott of Department of Physiology, University of Wisconsin-Madison, USA for his warm support, providing research experiences, valuable discussions and suggestions during my residence in USA.

The thesis committee for their suggestions and critical review of my thesis.

The Thailand Research Fund for the 2001 Golden Jubilee (RGJ) Ph.D. Program Scholarship (PHD/0115/2544). Chulalongkorn University for providing a grant for Center of Excellence to Bioactive Marine Natural Product Chemistry Research Unit (BMNCU), and University of Wisconsin at Madison for partial support during my residence in USA.

Associate Professor Dr. Surattana Amnuoypol of Department of Pharmacognosy, Faculty of Pharmaceutical Sciences, Chulalongkorn University for her kind suggestions, helps, and encouragement.

Dr. Prasat Kittakoop of the National Science and Technology Development Agency (NSTDA) for his assistance in ESI-TOF MS experiment.

The members in GM's Laboratory, Department of Physiology, University of Wisconsin-Madison, USA for their kind friendship and helps; Dr. Tomoyo Sakata for NMR measurements, discussion on structure elucidation and suggestion in organic synthesis; Mrs. Subhra Bhattacharya, Mr. Hui Wang, and Ms. Shu Mao for assistance in actin experiments. Ms. Reagan Miller of Department of Chemistry, University of Wisconsin-Madison, USA for her help in NMR, IR, and $[\alpha]$ measurements.

Teachers and members in Department of Pharmacognosy and Department of Pharmaceutical Chemistry, Faculty of Pharmaceutical Sciences, Chulalongkorn University for their kindness and help.

The Pharmaceutical Research Instrument Center and Department of Pharmacognosy, Faculty of Pharmaceutical Sciences, Chulalongkorn University for their support providing equipments during my research study.

Finally, my family, for their love, understanding, encouragement, and support.

CONTENTS

	Page
ABSTRACT (THAI)	iv
ABSTRACT (ENGLISH).....	v
ACKNOWLEDGMENT	vi
CONTENTS.....	vii
LIST OF FIGURES	xi
LIST OF SCHEMES.....	xvi
LIST OF TABLES	xvii
ABBREVIATIONS.....	xix
CHAPTER	
I INTRODUCTION	1
II REVIEW OF LITERATURE.....	3
1. The <i>Pachastrissa nux</i> sponge	3
2. Chemical constituents of the genus <i>Pachastrissa</i>	4
3. Trisoxazole macrolide compounds	7
4. Actin cytoskeleton.....	20
5. The structure of kabiramide C-G-actin complex	24
III EXPERIMENTAL	29
1. Animal material	29
1.1 Sample collection	29
1.2 Identification and characterization of the <i>Pachastrissa nux</i> sponge.....	29
2. Chromatographic techniques.....	31
2.1 Thin-layer chromatography (TLC)	31
2.2 Column chromatography.....	32
2.2.1 Vacuum liquid column chromatography.....	32
2.2.2 Flash column chromatography	32
2.2.3 High performance liquid column chromatography (HPLC).....	33
2.2.4 Gel filtration chromatography.....	33

3. Spectroscopy.....	33
3.1 Ultraviolet (UV) absorption spectroscopy.....	33
3.2 Infrared (IR) absorption spectroscopy.....	34
3.3 Mass spectroscopy (MS).....	34
3.4 Proton and carbon nuclear magnetic resonance (^1H and ^{13}C NMR) spectroscopy.....	34
3.5 Optical rotation.....	35
3.6 Fluorescence spectroscopy.....	35
4. Microscope.....	35
4.1 Confocal microscope.....	35
5. Chemicals for synthesis.....	35
6. Solvents.....	36
7. Biological activities.....	36
7.1 Antifungal activity.....	36
7.2 Cytotoxic activity.....	37
7.3 G-actin binding assay.....	37
7.3.1 Preparation of rabbit muscle acetone powder.....	37
7.3.2 Preparation of actin from muscle acetone powder.....	38
7.3.3 Preparation of prodan-G-actin.....	38
7.3.4 Prodan-G-actin binding experiment.....	39
7.3.5 G-actin binding experiment.....	39
7.3.6 Fluorescence resonance energy transfer (FRET) experiment.....	40
7.3.7 Fluorescence Anisotropy (FA or <i>r</i>) experiment.....	42
7.3.8 Iodide quenching.....	45
8. Fluorescent images in living cells.....	45
9. Extraction and isolation.....	46
10. Synthesis and purification of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl) kabiramide C.....	50
11. Synthesis of fluorescent derivatives of kabiramide C.....	53

11.1 Synthesis of tetramethylrhodamine derivative of 7-(4-amino methyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C (TMR-KabC).....	53
11.2 Synthesis of rhodol green derivative of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C (RG-KabC).....	53
11.3 Synthesis of IC5 derivative of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C (IC5-KabC).....	53
11.4 Synthesis of dapoxyl derivative of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C (DAP- KabC)	54
11.5 Synthesis of fluorescein diester derivative of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C (FDE-KabC)	54
IV RESULTS AND DISCUSSION.....	55
1. Identification and structure elucidation of the isolated kabiramides	55
1.1 Identification of kabiramide C	55
1.2 Identification of kabiramide D	60
1.3 Identification of kabiramide F	64
2. Synthesis of the 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C ...	69
3. Synthesis of the fluorescent derivatives of kabiramide C	82
3.1 TMR-KabC	84
3.2 RG-KabC	85
3.3 IC5-KabC	86
3.4 DAP-KabC	86
3.5 FDE-KabC.....	87
4. Actin-binding properties	88
4.1 Actin-binding properties of the non-fluorescent kabiramides	88
4.2 Actin-binding properties of fluorescent kabiramides.....	89
4.2.1 TMR-KabC.....	89
4.2.2 RG-KabC	91
4.2.3 IC5-KabC.....	92
4.2.4 DAP-KabC	93
4.2.5 FDE-KabC	93
5. Live cell imaging of TMR-KabC and FDE-KabC	93

6. Biological activities	95
6.1 Antifungal activity	95
6.2 Cytotoxic activity.....	95
V CONCLUSION	96
REFERENCES.....	98
APPENDIX	105
VITA.....	166

LIST OF FIGURES

Figure		Page
1	Crystal structure of G-actin.....	20
2	Electron micrograph demonstrates the actin filament decorated with myosin S1 head.....	21
3	Actin filament elongation	21
4	The dendritic-nucleation model for protrusion of lamellipodia	23
5	The structures of natural toxins that bind to actin.....	24
6	Stereo view of the electron density for kabiramide C	25
7	The structure of kabiramide C-G-actin complex.....	26
8	Space-filling representation of the residues on actin that interact with kabiramide C and gelsolin domain 1	27
9	The model of mechanism of action of kabiramide C (KabC)	28
10	The <i>Pachastrissa nux</i> sponge.....	31
11	FRET donor and acceptor spectral profiles	41
12	Schematic drawing of FRET between donor and acceptor.....	41
13	Diagram showing how the four intensity components are measured for determination of the steady state anisotropy	43
14	Schematic drawing of fluorescence anisotropy differences between small and large complexes	44
15	The H,H correlations (bold line) and HMBC correlations (arrow) of kabiramide C	57
16	The structure of kabiramide C.....	57
17	The H,H correlations (bold line) and HMBC correlations (arrow) of kabiramide D	61
18	The structure of kabiramide D.....	61
19	The H,H correlations (bold line) and HMBC correlations (arrow) of kabiramide F.....	65
20	The structure of kabiramide F	66
21	Synthesis of 7-azidokabiramide C.....	69

22	Proposed mechanism of Mitsunobu reaction	70
23	Synthesis of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C	71
24	Proposed mechanism of regioselective 1,4-substituted 1,3-dipolar cycloaddition.....	73
25	Proposed mechanism of deprotection of 7-[4- <i>N</i> -(9 <i>H</i> -fluoren-9-yl)- methoxycarbonyl]aminomethyl-1,2,3-triazol-1-yl]kabiramide C	74
26	Structures of fluorescent dyes.....	82
27	Synthesis of fluorescent kabiramide C probes	83
28	Proposed mechanism of acylation reaction of 7-(4-Aminomethyl- 1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C	84
29	The structure of TMR-KabC.....	85
30	The structure of RG-KabC.....	85
31	The structure of IC5-KabC.....	86
32	The structure of DAP-KabC.....	87
33	The structure of FDE-KabC	87
34	Confocal images of FDE-KabC-G-actin and TMR-KabC-G-actin in actin filament at protrusion site (arrow) of live NIH 3T3 cells.....	94
35	Effect of AMT-KabC on HeLa cells.....	95
36	The UV spectrum of G-actin in G-buffer	106
37	The UV spectrum of prodan-G-actin in G-buffer.....	106
38	The ESI-TOF mass spectrum of kabiramide C.....	107
39	The UV spectrum of kabiramide C in MeOH.....	107
40	The IR spectrum of kabiramide C (film)	108
41	The 300 MHz ¹ H NMR spectrum of kabiramide C in CDCl ₃	109
42	The 75 MHz ¹³ C NMR spectrum of kabiramide C in CDCl ₃	110
43	The 75 MHz DEPT90 and DEPT135 spectra of kabiramide C in CDCl ₃ ..	111
44	The 300 MHz HMQC spectrum of kabiramide C in CDCl ₃	112
45	The 300 MHz H-H COSY spectrum of kabiramide C in CDCl ₃	113
46	The 300 MHz HMBC spectrum (ⁿ J _{HC} = 8 Hz) of kabiramide C in CDCl ₃ ..	114
47	The 300 MHz HMBC spectrum (expanded) of kabiramide C in CDCl ₃	115
48	The ESI-TOF mass spectrum of kabiramide D.....	116

49	The UV spectrum of kabiramide D in MeOH.....	116
50	The IR spectrum of kabiramide D (film)	117
51	The 300 MHz ^1H NMR spectrum of kabiramide D in CDCl_3	118
52	The 75 MHz ^{13}C NMR spectrum of kabiramide C in CDCl_3	119
53	The 75 MHz DEPT90 and DEPT135 spectra of kabiramide D in CDCl_3 ...	120
54	The 300 MHz HMQC spectrum of kabiramide D in CDCl_3	121
55	The 300 MHz H,H COSY spectrum of kabiramide D in CDCl_3	122
56	The 300 MHz HMBC spectrum ($^nJ_{\text{HC}} = 8$ Hz) of kabiramide D in CDCl_3 .	123
57	The 300 MHz HMBC spectrum ($^nJ_{\text{HC}} = 8$ Hz) of kabiramide D in CDCl_3 .	124
58	The ESI-TOF mass spectrum of kabiramide F	125
59	The UV spectrum of kabiramide F in MeOH	125
60	The IR spectrum of kabiramide F (film).....	126
61	The 300 MHz ^1H NMR spectrum of kabiramide F in $\text{DMSO}-d_6$	127
62	The 300 MHz ^1H NMR spectrum of kabiramide F in CDCl_3	128
63	The 75 MHz ^{13}C NMR spectrum of kabiramide F in $\text{DMSO}-d_6$	129
64	The 75 MHz ^{13}C NMR spectrum of kabiramide F in CDCl_3	130
65	The 75 MHz DEPT90 AND DEPT135 spectra of kabiramide F in $\text{DMSO}-d_6$	131
66	The 300 MHz HMQC spectrum of kabiramide F in $\text{DMSO}-d_6$	132
67	The 300 MHz H,H COSY spectrum of kabiramide F in $\text{DMSO}-d_6$	133
68	The 300 MHz HMBC spectrum ($^nJ_{\text{HC}} = 8$ Hz) of kabiramide F in $\text{DMSO}-d_6$	134
69	The 300 MHz HMBC spectrum (expanded) of kabiramide F in $\text{DMSO}-d_6$	135
70	The ESI-TOF mass spectrum of 7-azidokabiramide C	136
71	The UV spectrum of 7-azidokabiramide C in MeOH	136
72	The IR spectrum of 7-azidokabiramide C (CHCl_3).....	137
73	The 300 MHz ^1H NMR spectrum of 7-azidokabiramide C in CDCl_3	138
74	The 75 MHz ^{13}C NMR spectrum of 7-azidokabiramide C in CDCl_3	139
75	The EI mass spectrum 3-(fluoren-9-yl-methoxycarbonyl)aminopropyne .	140

76	The UV spectrum of 3-(fluoren-9-yl-methoxycarbonyl)aminopropyne in MeOH.....	140
77	The IR spectrum of 3-(fluoren-9-yl-methoxycarbonyl)aminopropyne (CHCl ₃).....	141
78	The 300 MHz ¹ H NMR spectrum of 3-(fluoren-9-yl-methoxycarbonyl)aminopropyne in CDCl ₃	142
79	The 75 MHz ¹³ C NMR spectrum of 3-(fluoren-9-yl-methoxycarbonyl)aminopropyne in CDCl ₃	143
80	The ESI-TOF mass spectrum of 7-[4- <i>N</i> -(9 <i>H</i> -fluoren-9-yl-methoxycarbonyl)aminomethyl-1,2,3-triazol-1-yl]kabiramide C.....	144
81	The UV spectrum of 7-[4- <i>N</i> -(9 <i>H</i> -fluoren-9-yl-methoxycarbonyl)aminomethyl-1,2,3-triazol-1-yl]kabiramide C in MeOH.....	145
82	The IR spectrum of 7-[4- <i>N</i> -(9 <i>H</i> -fluoren-9-yl-methoxycarbonyl)aminomethyl-1,2,3-triazol-1-yl]kabiramide C (CHCl ₃).....	145
83	The 300 MHz ¹ H NMR spectrum of 7-[4- <i>N</i> -(9 <i>H</i> -fluoren-9-yl-methoxy carbonyl)aminomethyl-1,2,3-triazol-1-yl]kabiramide C in CDCl ₃	146
84	The 75 MHz ¹³ C NMR spectrum of 7-[4- <i>N</i> -(9 <i>H</i> -fluoren-9-yl-methoxy carbonyl)aminomethyl-1,2,3-triazol-1-yl]kabiramide C in CDCl ₃	147
85	The ESI-TOF mass spectrum of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C.....	148
86	The UV spectrum of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C in MeOH.....	149
87	The IR spectrum of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C (CHCl ₃).....	149
88	The 500 MHz ¹ H NMR spectrum of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C in CDCl ₃	150
89	The 125 MHz ¹³ C NMR spectrum of 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl)kabiramide C in CDCl ₃	151
90	The ESI-TOF mass spectrum of TMR-KabC.....	152
91	The UV spectrum of TMR-KabC in MeOH.....	152
92	The ESI-TOF mass spectrum of RG-KabC.....	153

93	The UV spectrum of RG-KabC in MeOH.....	153
94	The ESI-TOF mass spectrum of IC5-KabC	154
95	The UV spectrum of IC5-KabC in MeOH	154
96	The ESI-TOF mass spectrum of DAP-KabC.....	155
97	The UV spectrum of DAP-KabC in MeOH.....	155
98	The ESI-TOF mass spectrum of FDE-KabC	156
99	The UV spectrum of FDE-KabC in MeOH	156
100	Spectroscopic and actin-binding property of kabiramide C [23]	157
101	Spectroscopic and actin-binding property of kabiramide D [24]	157
102	Spectroscopic and actin-binding property of kabiramide F [55].....	158
103	Spectroscopic and actin-binding property of 7-azidokabiramide C [56].	158
104	Spectroscopic and actin-binding property of 7-[4- <i>N</i> -(9 <i>H</i> -fluoren-9-yl)- methoxycarbonyl)aminomethyl-1,2,3-triazol-1-yl]kabiramide C [57].....	159
105	Spectroscopic and actin-binding property of 7-(4-aminomethyl-1 <i>H</i> - 1,2,3-triazol-1-yl)kabiramide C [58].....	159
106	Spectroscopic and actin-binding properties of TMR-KabC [59].....	160
107	Spectroscopic and actin-binding properties of RG-KabC [60].	161
108	Stern-Volmer plot of iodide quenching behaviors of TMR-KabC, RG-KabC and G-actin complexes in G-buffer as a function of potassium iodide concentrations	162
109	Spectroscopic and actin-binding properties of IC5-KabC [61].....	163
110	Spectroscopic and actin binding property of DAP-KabC [61]	164
111	Spectroscopic and actin binding property of FDE-KabC [62].....	165

LIST OF SCHEME

Scheme		Page
1	Extraction of the sponge <i>Pachastrissa nux</i>	46
2	Fractionation of the crude MeOH extract obtained from the sponge <i>Pachastrissa nux</i>	48

LIST OF TABLES

Table		Page
1	The cytotoxicity of bengamides A and B	6
2	The cytotoxicity against MDA-MB-435 human mammary carcinoma of bengamides A, B, E and F.....	6
3	The bioactivities of kabiramide compounds	9
4	The bioactivities of the halichondramides from the sponge <i>Halichondria</i> sp. and the nudibranch <i>Hexabranhus sanguineus</i>	10
5	The bioactivities of halichondramides from the sponge <i>Chondrosia corticata</i>	13
6	The bioactivities of the jaspisamides	17
7	The bioactivities of the halishigamides	18
8	The ^1H , ^{13}C NMR, and ^1H - ^{13}C long-range correlations in HMBC ($^nJ_{\text{CH}} = 8$ Hz) spectral data in CDCl_3 of kabiramide C.....	58
9	The ^1H , ^{13}C NMR, and ^1H - ^{13}C long-range correlations in HMBC ($^nJ_{\text{CH}} = 8$ Hz) spectral data in CDCl_3 of kabiramide D.....	62
10	The ^1H , ^{13}C NMR, and ^1H - ^{13}C long-range correlations in HMBC ($^nJ_{\text{CH}} = 8$ Hz) spectral data in $\text{DMSO}-d_6$ and CDCl_3 of kabiramide F.....	67
11	The ^1H and ^{13}C NMR spectral data in CDCl_3 of 3-(fluoren-9-yl-methoxycarbonyl)aminopropyne.....	72
12	The UV, IR, specific rotation of 7-azidokabiramide C [56], 7-[4- <i>N</i> -(9 <i>H</i> -fluoren-9-yl-methoxycarbonyl)aminomethyl-1,2,3-triazol-1-yl] kabiramide C [57], and 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl) kabiramide C [58].....	75
13	The ^1H NMR spectral data of 7-azidokabiramide C [56], 7-[4- <i>N</i> -(9 <i>H</i> -fluoren-9-yl-methoxycarbonyl)aminomethyl-1,2,3-triazol-1-yl] kabiramide C [57], and 7-(4-aminomethyl-1 <i>H</i> -1,2,3-triazol-1-yl) kabiramide C [58].....	76

- 14 The ^{13}C NMR spectral data of 7-azidokabiramide C [56], 7-[4-*N* (9*H*-fluoren-9-yl-methoxycarbonyl)aminomethyl-1,2,3-triazol-1-yl] kabiramide C [57], and 7-(4-aminomethyl-1*H*-1,2,3-triazol-1-yl) kabiramide C [58]..... 79

ABBREVIATIONS

%	=	percent or part per hundred
δ	=	chemical shift
τ	=	excited-state lifetime
ϵ	=	molar absorptivity or molar extinction coefficient
λ_{\max}	=	wave length at maximum absorption
ν_{\max}	=	wave number at maximum absorption
$[\alpha]_{\text{D}}^{23}$	=	specific rotation at 23 °C and sodium D line (589 nm)
°C	=	degree Celsius
μg	=	microgram
μl	=	micro liter
μM	=	micro molar
μm	=	micrometer
^{13}C NMR	=	carbon-13 nuclear magnetic resonance
^1H NMR	=	proton nuclear magnetic resonance
2D NMR	=	two dimensional nuclear magnetic resonance
Å	=	angstrom
ADP	=	adenosine diphosphate
Al_2O_3	=	aluminum oxide
Ala	=	alanine
ATCC	=	American Type Culture Collection (Maryland, USA)
ATP	=	adenosine triphosphate
br s	=	broad singlet
c	=	concentration
Ca	=	calcium
CaH_2	=	calcium hydride
cald	=	calculated
CDCl_3	=	deuterated chloroform
CH_2Cl_2	=	dichloromethane

CHCl ₃	=	chloroform
cm	=	centimeter
CO ₂	=	carbon dioxide
Cys	=	cysteine
d	=	doublet
dd	=	doublet of doublets
ddd	=	doublet of doublets of doublets
DEPT	=	distortionless enhancement by polarization transfer
DMEM	=	Dulbecco's Modified Eagle medium
DMSO	=	dimethyl sulphoxide
DMSO- <i>d</i> ₆	=	deuterated dimethyl sulphoxide
dt	=	doublet of triplets
DTT	=	dithiothreitol
EI	=	electron impact ionization
em	=	emission
ESI-TOF	=	electrospray ionization-time of flight
Et ₃ N	=	triethylamine
EtOAc	=	ethyl acetate
EtOH	=	ethanol
ex	=	excitation
FA or r	=	fluorescence anisotropy
FBS	=	fetal bovine serum
Fmoc	=	<i>N</i> -(9 <i>H</i> -fluoren-9-yl-methoxycarbonyl)
FRET	=	fluorescence resonance energy transfer
g	=	gram or earth's gravitational field
Gly	=	glycine
H,H COSY	=	homonuclear (proton-proton) correlation spectroscopy
H ₂ O	=	water
HeLa	=	human cervix carcinoma cells
HMBC	=	proton-detected heteronuclear multiple bond correlation
HMQC	=	proton-detected heteronuclear multiple quantum coherence

HN_3	=	hydrazoic acid
Hz	=	hertz
I or FI	=	fluorescence intensity
IC_{50}	=	50% inhibition concentration
Ile	=	isoleucine
IR	=	infrared
J	=	coupling constant
K_2HPO_4	=	potassium phosphate dibasic
KCl	=	potassium chloride
KH_2PO_4	=	potassium phosphate monobasic
KI	=	potassium iodide
km	=	kilometer
L	=	liter
Leu	=	leucine
M	=	molar
m	=	multiplet
m/z	=	mass to charge ratio
M^+	=	molecular ion
MeOH	=	methanol
Met	=	methionine
mg	=	milligram
MgCl_2	=	magnesium chloride
MgSO_4	=	magnesium sulphate
MHz	=	megahertz
MIC	=	minimum inhibitory concentration
min	=	minute
mL	=	milliliter
mm	=	millimeter
MS	=	mass spectroscopy
NaCl	=	sodium chloride
NaHCO_3	=	sodium bicarbonate

NaN ₃	=	sodium azide
ng	=	nanogram
NIH 3T3	=	mouse fibroblast cells
nM	=	nano molar
nm	=	nanometer
NMR	=	nuclear magnetic resonance
NSS	=	normal saline solution
Phe	=	phenylalanine
PPh ₃	=	triphenylphosphine
ppm	=	part per million
R ²	=	correlation coefficient
RP18	=	reversed phase carbon eighteen
rt	=	room temperature
s	=	singlet
SDA	=	Sabouraud dextrose agar
Ser	=	serine
Si gel	=	silica gel
sp.	=	species
t	=	triplet
THF	=	tetrahydrofuran
Thr	=	threonine
TLC	=	thin layer chromatography
Tyr	=	tyrosine
UV	=	ultraviolet
w or wt	=	weight