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BIOACTIVE COMPOUNDS FROM
Andrographis paniculata AND *Caesalpinia bonduc*

Miss Nattida Suwanakitti

A Thesis Submitted in Partial Fulfillment of the Requirements
for the Degree of Master of Science Program in Biotechnology

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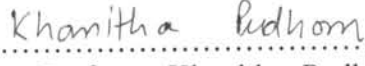
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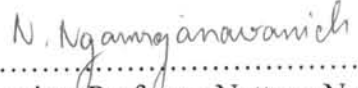
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ณัฐดา สุวรรณกิตติ: สารออกฤทธิ์ทางชีวภาพจากฟ้าทะลายโจรและสวาด
(BIOACTIVE COMPOUNDS FROM *Andrographis paniculata* AND
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จากการแยกสารสกัดจากพืชสมุนไพรไทย ฟ้าทะลายโจร และ เมล็ดสวาด โดยนำสารสกัดเอธิลเอซิเตดและเมทานอลจากฟ้าทะลายโจร มาทำการแยกสารบริสุทธิ์โดยเทคนิคโครมาโทกราฟี ได้สาร 7 ชนิดได้แก่ andrographolide (1), 14-deoxy-11,12-didehydroandrographolide (2), neoandrographolide (3), andrographiside (4), 5,2'-dihydroxy-7,8-dimethoxyflavone (5), 8-O-acetylharpagide (6) และ antirrhinoside (7) และซึ่งสาร iridoid glucoside 6 และ 7 พบเป็นครั้งแรกในพืชชนิดนี้ ส่วนการแยกสารสกัดเอธิลเอซิเตดจากเมล็ดสวาด ได้ cassane diterpenoid ชนิดใหม่ 1 ชนิด คือ caesalpinin Q(17) และสารที่มีรายงานมาก่อนอีก 9 ชนิด ได้แก่ caesalpinin C (8), caesalpinin P (9), 14(17)-dehydrocaesalpin F (10), E-caesalpin (11), bonducellpins C (12), caesalpinin K (13), caesalmin B (14), caesalpinin I (15), และ 2-acetoxycaesaldehyd e (16) การพิสูจน์โครงสร้างของสารที่แยกได้ อาศัยข้อมูลทางสเปกโทรสโกปีและการเปรียบเทียบกับข้อมูลที่มีรายงานมาก่อนหน้านี้ เมื่อนำสารบริสุทธิ์ที่ได้มาทดสอบฤทธิ์ยับยั้งเชื้อมาลาเรียและความเป็นพิษต่อเซลล์มะเร็ง พบว่า สารประกอบที่ 1 และ 5 มีฤทธิ์ยับยั้งเชื้อมาลาเรีย *Plasmodium falciparum* สายพันธุ์ K1 ได้ดี โดยให้ค่า IC_{50} เท่ากับ 17.0 และ 6.46 μ M ตามลำดับ สารประกอบที่ 2 และ 5 มีความเป็นพิษในระดับกลางต่อเซลล์มะเร็ง Hep-G₂ และ SW620 และมีความเป็นพิษในระดับดีมากต่อเซลล์มะเร็ง KATO-3 ส่วนสารประกอบ cassane ทุกตัวที่แยกได้จากเมล็ดสวาดมีฤทธิ์ยับยั้งเชื้อมาลาเรียได้ดีมาก แต่มีความเป็นพิษต่อเซลล์มะเร็งน้อยมากและ/หรือไม่มีความเป็นพิษต่อเซลล์มะเร็ง

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NATTIDA SUWANAKITTI: BIOACTIVE COMPOUNDS FROM
Andrographis paniculata AND *Caesalpinia bonduc*. THESIS
ADVISOR: ASSIT. PROF. KHANITHA PUDHOM Ph.D.84 pp.

In phytochemical investigation of isolated compounds from *Andrographis paniculata* and *Caesalpinia bonduc*, the ethyl acetate and methanol crude extract of *A. paniculata* purified by chromatographic techniques to afford seven known compounds; andrographolide (1), 14-deoxy-11,12-didehydroandrographolide (2), neoandrographolide (3), andrographiside (4), 5,2'-dihydroxy-7,8-dimethoxyflavone (5), 8-*O*-acetylharpagide (6) and antirrinocide (7). Iridoid glucosides 6 and 7 were first isolated from this plant. On the otherhand, the chromatography of the ethyl acetate crude extract of *C. bonduc* led to the isolation of one new cassane diterpenoid, caesalpinin Q (17), along with nine known cassanes; caesalpinin C (8), caesalpinin P (9), 14(17)-dehydrocaesalpin F (10), ϵ -caesalpin (11), bonducellpins C (12), caesalpinin K (13), caesalmin B (14), caesalpinin I (15), and 2-acetoxycasaldehyd (16). The chemical structures of all isolated compounds were established on the basis of spectroscopic data and compared to literatures. Pure isolated compounds were also evaluated for antimalarial and cytotoxic activity. Compounds 1 and 5 showed good antimalarial activity against chloroquine-resistant K1 strain of *Plasmodium falciparum* with IC_{50} values of 17.0 and 6.46 μ M, respectively. Moreover, compounds 2 and 5 exhibited moderate cytotoxicity on Hep-G₂, SW620 cell lines, and showed strong cytotoxicity on KATO-3 cell lines. For cassane diterpenoids isolated from *C. bonduc*, all compounds showed significant antimalarial activity, but exhibited weak or inactive cytotoxicity against all tested cell lines.

Field of study.....Biotechnology.....Student's signature.....*Nattida Suwanakitti*.....
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CONTENTS

	Pages
Thai Abstract	iv
English Abstract	v
Acknowledgements	vi
Contents	vii
List of Figures	x
List of Tables	xiii
List of Schemes	xv
List of Abbreviations	xvi
CHAPTER	
I Introduction	1
1.1 Life cycle of the Plasmodium parasite and its epidemiology.....	1
1.2 Symptom.....	4
1.3 Medicinal plant and malarial activity.....	4
1.4 Botanical Aspects and Distribution.....	6
1.4.1 <i>Andrographis paniculata</i>	6
1.4.1.1 Ethnobotanical of <i>A. paniculata</i>	6
1.4.1.2 Previous studies in chemical constituents of <i>A. paniculata</i>	7
1.4.2 <i>Caesalpinia bonduc</i>	10
1.4.2.1 Ethnobotanical of <i>C. bonduc</i>	10
1.4.2.2 Previous studies in chemical constituents of <i>C. bonduc</i>	11
II Isolation and Characterization of compounds from <i>Andrographis paniculata</i> and <i>Caesalpinia bonduc</i>	15
2.1 Experimental Section	15
2.1.1 Plant material	15
2.1.2 General Experimental Procedure	15

	Pages
2.1.3 Extraction.....	16
2.2 Isolation and Purification.....	18
2.2.1 Isolation and purification of compounds from crude extract of <i>A. paniculata</i>	18
2.2.2 Isolation and purification of compounds from crude extract of <i>C. bonduc</i>	20
2.3 Structural Elucidation.....	23
2.3.1 Compounds isolated from <i>A. paniculata</i>	23
2.3.2 Compounds isolated from <i>C. bonduc</i>	30
III Investigation of Pharmacological Activity of Isolated Compounds	44
3.1 Experimental Section	44
3.1.1 Antimalarial assay.....	44
3.1.2 Cytotoxicity assay.....	44
3.2 Results and Discussion	45
3.2.1 Antimalarial assay of isolated compounds from <i>A. paniculata</i>	45
3.2.2 Antimalarial assay of isolated compounds from <i>C. bonduc</i>	46
3.2.3 Cytotoxicity of isolated compounds from <i>A. paniculata</i> and <i>C. bonduc</i>	49
IV Conclusion	51
References	54
Appendices	59
VITA	84

List of Figures

	Pages
Figure 1.1	Life cycle of malaria.....3
Figure 1.2	<i>Andrographis paniculata</i> Nees.....7
Figure 1.3	The chemical constituents of <i>A. paniculata</i> Nees.....8
Figure 1.4	<i>Caesalpinia bonduc</i> (L.) Roxb.....10
Figure 1.5	The chemical constituents of <i>C. bonduc</i>12
Figure 2.1	Structure of Andrographolide (1).....23
Figure 2.2	Structure of 14-Deoxy-11,12-didehydroandrographolide (2).....24
Figure 2.3	Structure of Neoandrographolide (3).....25
Figure 2.4	Structure of Andrographiside (4).....26
Figure 2.5	Key HMBC and COSY correlations for 5,2'-Dihydroxy-7,8-dimethoxyflavone (5).....27
Figure 2.6	Structure of 8-O-Acetylharpagide (6).....28
Figure 2.7	Key HMBC and COSY correlations for Antirrinoside (7).....29
Figure 2.8	Key HMBC and COSY correlations for Compound 8.....30
Figure 2.9	Key HMBC and COSY correlations for Compound 9.....32
Figure 2.10	Key HMBC and COSY correlations for Compound 10.....33
Figure 2.11	Key HMBC and COSY correlations for Compound 11.....34
Figure 2.12	Key HMBC and COSY correlations for Compound 12.....35
Figure 2.13	Key HMBC and COSY correlations for Compound 13.....37
Figure 2.14	Key HMBC and COSY correlations for Compound 14.....38
Figure 2.15	Key HMBC and COSY correlations for Compound 15.....39
Figure 2.16	Key HMBC and COSY correlations for Compound 16.....41
Figure 2.17	Key HMBC and COSY correlations for Compound 17.....42
Figure 2.18	Selected NOE correlations for Compound 17.....43
Figure 1	The ¹ H-NMR spectrum of Andrographolide (1).....60
Figure 2	The ¹³ C-NMR spectrum of Andrographolide (1).....60
Figure 3	The ¹ H-NMR spectrum of 14-Deoxy-11,12-didehydroandrographolide (2).....61
Figure 4	The ¹³ C-NMR spectrum of 14-Deoxy-11,12-didehydroandrographolide (2).....61

	Pages
Figure 5	The ¹ H-NMR spectrum of Neoandrographolide (3).....62
Figure 6	The ¹³ C-NMR spectrum of Neoandrographolide (3).....62
Figure 7	The COSY spectrum of Neoandrographolide (3).....63
Figure 8	The HMBC spectrum of Neoandrographolide (3).....63
Figure 9	The HSQC spectrum of Neoandrographolide (3).....64
Figure 10	The ¹ H-NMR spectrum of Andrographiside (4).....64
Figure 11	The ¹ H-NMR spectrum of 5,2'-Dihydroxy-7,8- dimethoxy flavone (5).....65
Figure 12	The ¹³ C-NMR spectrum of 5,2'-Dihydroxy-7,8- dimethoxyflavone (5).....65
Figure 13	The HMBC spectrum of 5,2'-Dihydroxy-7,8- dimethoxyflavone (5).....66
Figure 14	The HSQC spectrum of 5,2'-Dihydroxy-7,8- dimethoxyflavone (5).....66
Figure 15	The ¹ H-NMR spectrum of 8- <i>O</i> -Acetylharpagide (6).....67
Figure 16	The ¹³ C-NMR spectrum of 8- <i>O</i> -Acetylharpagide (6).....67
Figure 17	The COSY spectrum of 8- <i>O</i> -Acetylharpagide (6).....68
Figure 18	The HMBC spectrum of 8- <i>O</i> -Acetylharpagide (6).....68
Figure 19	The HSQC spectrum of 8- <i>O</i> -Acetylharpagide (6).....69
Figure 20	The ¹ H-NMR spectrum of Antirrinocide(7).....69
Figure 21	The ¹³ C-NMR spectrum of Antirrinocide(7).....70
Figure 22	The ¹ H-NMR spectrum of Caesalpinins C (8).....70
Figure 23	The ¹³ C-NMR spectrum of Caesalpinins C (8).....71
Figure 24	The COSY spectrum of Caesalpinins C (8).....71
Figure 25	The HMBC spectrum of Caesalpinins C (8).....72
Figure 26	The HSQC spectrum of Caesalpinins C (8).....72
Figure 27	The ¹ H-NMR spectrum of Caesalpinins P (9).....73
Figure 28	The ¹³ C-NMR spectrum of Caesalpinins P (9).....73
Figure 29	The ¹ H-NMR spectrum of 14(17)-Dehydrocaesalpin F (10).....74
Figure 30	The ¹³ C-NMR spectrum of 14(17)-Dehydrocaesalpin F (10).....74

	Pages
Figure 31	The ^1H -NMR spectrum of ϵ -Caesalpin (11).....75
Figure 32	The ^{13}C -NMR spectrum of ϵ -Caesalpin (11).....75
Figure 33	The ^1H -NMR spectrum of Bonducellpins C (12).....76
Figure 34	The ^{13}C -NMR spectrum of Bonducellpins C (13).....76
Figure 35	The ^1H -NMR spectrum of Caesalpinins K (13).....77
Figure 36	The ^1H -NMR spectrum of Caesalmin B (14).....77
Figure 37	The ^{13}C -NMR spectrum of Caesalmin B (14).....78
Figure 38	The ^1H -NMR spectrum of Caesalpinins I (15).....78
Figure 39	The ^{13}C -NMR spectrum of Caesalpinins I (15).....79
Figure 40	The ^1H -NMR spectrum of 2-Acetoxycaesaldekarin e (16).....79
Figure 41	The ^{13}C -NMR spectrum of 2-Acetoxycaesaldekarin e (16).....80
Figure 42	The ^1H -NMR spectrum of Caesalpinin Q(17).....80
Figure 43	The ^{13}C -NMR spectrum of Caesalpinin Q (17).....81
Figure 44	The COSY spectrum of Caesalpinin Q (17).....81
Figure 45	The HMBC spectrum of Caesalpinin Q (17).....82
Figure 46	The HSQC spectrum of Caesalpinin Q (17)82
Figure 47	IR spectrum of Caesalpinin Q (17).....83

List of Tables

		Pages
Table 2.1	The ^1H and ^{13}C NMR spectral data of Andrographolide and Compound 1.....	23
Table 2.2	The ^1H and ^{13}C NMR spectral data of 14-Deoxy-11,12-didehydroandrographolide and Compound 2.....	24
Table 2.3	The ^1H and ^{13}C NMR spectral data of Neoandrographolide and Compound 3.....	25
Table 2.4	The ^1H and ^{13}C NMR spectral data of Androgaphoside and Compound 4.....	26
Table 2.5	The ^1H and ^{13}C NMR spectral data of 5, 2'-Dihydroxy-7,8-dimethoxyflavone and Compound 5.....	27
Table 2.6	The ^1H and ^{13}C NMR spectral data of 8- <i>O</i> -Acetylharpagide and Compound 6.....	28
Table 2.7	The ^1H and ^{13}C NMR spectral data of compound 7.....	29
Table 2.8	The ^1H and ^{13}C NMR spectral data of Caesalpinin C and compound 8.....	31
Table 2.9	The ^1H and ^{13}C NMR spectral data of Caesalpinin P and compound 9.....	32
Table 2.10	The ^1H and ^{13}C NMR spectral data of compound 10.....	33
Table 2.11	The ^1H and ^{13}C NMR spectral data of compound 11.....	34
Table 2.12	The ^1H and ^{13}C NMR spectral data of Bonducellpin C and compound 12.....	36
Table 2.13	The ^1H and ^{13}C NMR spectral data of Caesalpinin K and compound 13.....	37
Table 2.14	The ^1H and ^{13}C NMR spectral data of Caesalmin B and compound 14.....	38
Table 2.15	The ^1H and ^{13}C NMR spectral data of Caesalpinin I and compound 15.....	40
Table 2.16	The ^1H and ^{13}C NMR spectral data of compound 16.....	41

Table 2.17	The ^1H and ^{13}C NMR spectral data of compound 17.....	43
Table 3.1	In vitro antiplasmodial activity of isolated compounds from <i>A. paniculata</i>	45
Table 3.2	In vitro antiplasmodial activity of isolated compounds from <i>C. bonduc</i>	47
Table 3.3	In vitro cytotoxicity of isolated compounds from <i>A. paniculata</i> and <i>C. bonduc</i>	50

List of Schemes

	Pages
Scheme 2.1 The extraction procedure of the whole plant of <i>A. paniculata</i>	17
Scheme 2.2 The extraction procedure of the kernel of <i>C. bonduc</i>	18
Scheme 2.3 The isolation and purification procedure of Andrographolide (1), 14-Deoxy-11,12-didehydroandrographolide (2), Neoandrographolide (3), Andrographoside (4), 5,2'-Dihydroxy-7,8-dimethoxyflavone (5).....	19
Scheme 2.4 The isolation and purification procedure of 8- <i>O</i> -Acetylharpagide (6), Antirrinoside (7).....	20
Scheme 2.5 The isolation and purification procedure of 14(17)-Dehydro caesalpin F (10), 2-Acetoxycaesaldekarin e (16).....	21
Scheme 2.6 The isolation and purification procedure of Bonducellpins C (12), Caesalpinin I (15), Caesalpinin Q (17), Caesalpinin P (9), ϵ -Caesalpin (11), Caesalpinin K (13), Caesalpinin C (8), and Caesalmin B (14).....	22

List of Abbreviations

$^{\circ}\text{C}$	Degree Celsius
$^{13}\text{C-NMR}$	Carbon-13 nuclear magnetic resonance
$^1\text{H-NMR}$	Proton nuclear magnetic resonance
Brd	Broad doublet
brs	Broad singlet
brt	Broad triplet
CDCl_3	Deuterated chloroform
CH_2Cl_2	Dichloromethane
COSY	Correlated spectroscopy
d	Doublet
dd	Doublet of doublet
ddd	Doublet of doublet of doublet
DI water	Deionize water
DMSO	Dimethyl sulfoxide
dt	Double of triplet
EtOAc	Ethyl acetate
g	Gram
HMBC	Heteronuclear multiple bond connectivity
HMQC	Heteronuclear multiple-quantum coherence
hr	Hour
Hz	Hertz
IC_{50}	Median inhibitory concentration
<i>J</i>	Coupling constant
m	Multiplex
<i>m/z</i>	Mass per charge
MeOH	Methanol
mg	Milligram
MHz	Megahertz
MW	Molecular weight
NOESY	Nuclear overhauser effect spectroscopy

s	Singlet
t	Triplet
td	Triplet of doublet
TLC	Thin layer chromatography
w/w	Weight by weight
δ	Chemical shift
μl	Microliter