

องค์ประกอบทางเคมีของลำต้นสะค้านหนู

(*Piper aurantiacum* Miq.)

นางสาวมุกอภา มุกดาทอง



วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรมหาบัณฑิต

ภาควิชาเคมี

บัณฑิตวิทยาลัย จุฬาลงกรณ์มหาวิทยาลัย

ปีการศึกษา 2539

ISBN 974-636-660-2

ลิขสิทธิ์ของบัณฑิตวิทยาลัย จุฬาลงกรณ์มหาวิทยาลัย

I 1720245C

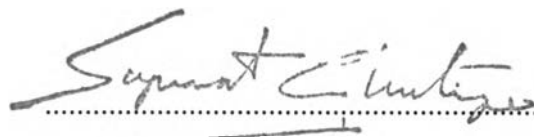
**CHEMICAL CONSTITUENTS OF THE STEM OF
PIPER AURANTIAECUM MIQ.**

Miss Muk-apa Mukdathong

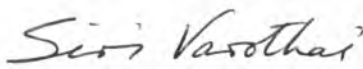
A Thesis Submitted in Partial Fulfillment of the Requirements
for the Degree of Master of Science
Department of Chemistry
Graduate School
Chulalongkorn University
Academic Year 1996
ISBN 974-636-660-2

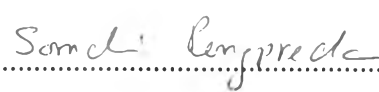
Thesis Title CHEMICAL CONSTITUENTS OF THE STEM OF
Piper aurantiacum Miq.
By Miss Muk-apa Mukdathong
Department Chemistry
Thesis Advisor Assistant Professor Dr.Somchai Pengprecha

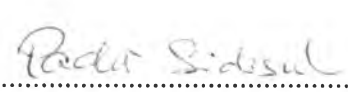
Accepted by the Graduate School, Chulalongkorn University in Partial
Fulfillment of the Requirements for the Master's degree


.....Dean of Graduate School
(Professor Supawat Chutivongse, M.D.)

Thesis committee


.....Chairman
(Associate Professor Siri Varothai, Ph.D.)


.....Thesis Advisor
(Assistant Professor Somchai Pengprecha, Ph.D.)


.....Member
(Professor Padet Sidisunthorn, Ph.D.)


.....Member
(Associate Professor Sophon Roengsumran, Ph.D.)

พิมพ์ต้นฉบับบทความวิทยานิพนธ์ภายในกรอบสี่เหลี่ยมนี้เพียงแผ่นเดียว

มุกอภา มุกดาทอง : องค์ประกอบทางเคมีของลำต้นสะค้านหนู *Piper aurantiacum* Miq. (CHEMICAL CONSTITUENTS OF THE STEM OF *Piper aurantiacum* Miq.) อ.ที่ปรึกษา : ผศ. ดร. สมใจ เพ็งปรีชา , 97 หน้า. ISBN 974-636-660-2.

นำเถาสะค้านหนู (*Piper aurantiacum* Miq.) ที่แห้งและบดละเอียด มาสกัดด้วยตัวทำละลายอินทรีย์ชนิดต่างๆ แล้วทำการแยกองค์ประกอบทางเคมี ด้วยวิธีคอลัมน์โครมาโทกราฟี ได้สารประกอบ 5 ชนิด เมื่อนำมาทำให้บริสุทธิ์ และทดสอบสมบัติทางเคมี สมบัติทางกายภาพ ตลอดจนพิสูจน์สูตรโครงสร้าง โดยใช้ข้อมูลทางสเปกโทรสโกปี พบว่าสารประกอบทั้ง 5 ชนิด คือ β -sitosterol ($C_{28}H_{48}O$, จุดหลอมเหลว $137-138^{\circ}C$, ผลึกรูปเข็มสีขาวเป็นมันวาว), methyl-5-(3',4'-methylenedioxyphenyl)penta-2,4-dienoate (methyl piperate, $C_{11}H_{12}O_4$, จุดหลอมเหลว $142-143^{\circ}C$, ผลึกรูปเข็ม สีเหลือง), endo-1,7,7-trimethylbicyclo[2.2.1]heptan-2-ol-3',4'-hydroxyphenyl)-2'-propenoate (borneol p-coumarate, $C_{19}H_{22}O_3$, จุดหลอมเหลว $158-159^{\circ}C$, ผลึกใสรูปปริซึมสี่เหลี่ยม), 1,3-benzodioxole-5-carboxylic acid ($C_8H_6O_4$, ของแข็งอสัณฐานสีขาว) และ potassium chloride (KCl, จุดหลอมเหลวสูงกว่า $270^{\circ}C$, ผลึกใสรูปลูกบาศก์)

ภาควิชา เคมี
สาขาวิชา เคมี
ปีการศึกษา 2539

ลายมือชื่อนิสิต
ลายมือชื่ออาจารย์ที่ปรึกษา
ลายมือชื่ออาจารย์ที่ปรึกษาร่วม

พิมพ์ต้นฉบับบทความวิทยานิพนธ์ภายในกรอบสี่เหลี่ยมนี้เพียงแผ่นเดียว

C725249 : MAJOR CHEMISTRY

KEY WORD: *Piper aurantiacum* Miq. / CHEMICAL CONSTITUENT

MUK-APA MUKDATHONG : CHEMICAL CONSTITUENTS OF THE STEM OF *Piper aurantiacum* Miq. THESIS ADVISOR : ASSIST. PROF. SOMCHAI PENGPRECHA, Ph.D. pp. ISBN 974-636-660-2.

The dried stems of *Piper aurantiacum* Miq. were extracted with organic solvents. The crude extracts were chromatographed on silica gel by using quick column chromatography and 5 compounds were identified. These compounds were confirmed by chemical reactions, physical testing and spectroscopic methods and were assigned as β -sitosterol ($C_{29}H_{48}O$, m.p. 137-138°C, bright white needle-like crystals), methyl-5-(3',4'-methylenedioxyphenyl)penta-2,4-dienoate (methyl piperate, m.p. 142-143°C, $C_{13}H_{12}O_4$, yellow needle-like crystals), endo-1,7,7-trimethylbicyclo [2.2.1]heptan-2-ol-3'-(4'-hydroxyphenyl)-2'-propenoate (borneol p-coumarate, $C_{19}H_{24}O_3$, m.p. 158-159°C, colourless rectangular crystals), 1,3-benzodioxole-5-carboxylic acid ($C_8H_6O_4$, white amorphous solid) and potassium chloride (KCl, m.p. was over 270°C, colourless cubical crystals).

ภาควิชา.....เคมี

สาขาวิชา.....เคมี

ปีการศึกษา.....2539

ลายมือชื่อนิสิต.....

ลายมือชื่ออาจารย์ที่ปรึกษา.....

ลายมือชื่ออาจารย์ที่ปรึกษาร่วม.....

ACKNOWLEDGEMENT

The author would like to express her sincere appreciation to her advisor, Assistant Professor Dr.Somchai Pengprecha whose advice, suggestions and encouragement have been invaluable to her. She wishes to thank the members of her thesis committee, Associate Professor Dr.Siri Varothai, Professor Dr.Padet Sidisunthorn, and Associate Professor Dr.Sophon Roengsumran for critical comments and advice. She is also grateful to Dr.Tirayut Vilaivan and Dr.Roderick Bates for valuable discussion and guidance.

She is indebted to the Graduate School and the Department of Chemistry, Faculty of Science, Chulalongkorn University for the teacher assistantship and scholarship. She would like to extend her appreciation to the staff of the Science and Technology Research Equipment Centre, for their services on sample analysis.

Further acknowledgement is extended to her friends for their help and encouragement during her graduate studies. Last but not least, she wishes to express her deepest gratitude to her parents. Without their financial and moral support, she could not have completed this work.

CONTENTS

| | Page |
|------------------------------------|-------------|
| Abstract in Thai | iv |
| Abstract in English | v |
| Acknowledgement | vi |
| Contents | vii |
| List of Figures | x |
| List of Schemes | xii |
| List of Tables | xiii |
| List of Abbreviations | xv |

CHAPTER

| | |
|---|----|
| I INTRODUCTION | 1 |
| 1.1 Botanical Description of <i>Piper aurantiacum</i> Miq. | 5 |
| 1.2 Chemical Constituents of Piper Genus..... | 7 |
| 1.3 Pharmacological Activities..... | 18 |
| 1.4 The Objective of This Research..... | 19 |
| II EXPERIMENTAL | 20 |
| 2.1 Plant Materials..... | 20 |
| 2.2 Equipments..... | 20 |
| 2.3 Solvents and Chromatographic Media..... | 21 |
| 2.4 Physical Separation Techniques..... | 22 |
| 2.5 Extraction..... | 23 |

| | Page |
|---|---------------|
| 2.6 Isolation of the Chemical Constituents of the Stem of | |
| <i>Piper aurantiacum</i> Miq. | 25 |
| 2.6.1 Separation of Hexane Extract..... | 25 |
| 2.6.1.1 The Separation of the Eluted Fraction 10-24..... | 28 |
| 2.6.1.2 The Separation of the Eluted Fraction 144-152..... | 29 |
| 2.6.2 Separation of Chloroform Extract..... | 30 |
| 2.6.3 Separation of Ethyl Acetate Extract..... | 31 |
| 2.6.4 Separation of Butanol Extract..... | 32 |
| 2.6.5 Separation of Methanol Extract..... | 33 |
| 2.7 Purification and Properties of the Eluted Compounds..... | 34 |
| 2.7.1 Purification and Properties of Compound I..... | 34 |
| 2.7.2 Purification and Properties of Compound II..... | 35 |
| 2.7.3 Purification and Properties of Compound III..... | 37 |
| 2.7.4 Purification and Properties of Compound IV..... | 37 |
| 2.7.5 Purification and Properties of Compound V..... | 38 |
| III RESULTS AND DISCUSSION..... | 39 |
| 3.1 Structural Elucidation of Compound I..... | 39 |
| 3.2 Structural Elucidation of Compound II..... | 42 |
| 3.3 Structural Elucidation of Compound III..... | 47 |
| 3.4 Structural Elucidation of Compound IV..... | 56 |
| 3.5 Structural Elucidation of Compound V..... | 59 |
| IV CONCLUSION..... | 60 |

| | Page |
|------------------------|-------------|
| REFERENCES..... | 62 |
| APPENDIX..... | 68 |
| VITA..... | 98 |

LIST OF FIGURES

| Figure | Page |
|--|------|
| 1.1 The structural formula of the insecticide and antifeedant from Piper genus..... | 3 |
| 1.2 <i>Piper aurantiacum</i> Miq. | 6 |
| 1.3 The organic compounds from Piper genus..... | 11 |
| A.1 The IR spectrum of Compound I..... | 69 |
| A.2 The MASS spectrum of Compound I..... | 70 |
| A.3 The ¹ H-NMR spectrum of Compound I..... | 71 |
| A.4 The ¹³ C-NMR spectrum of Compound I..... | 72 |
| A.5 The ¹³ C-NMR DEPT-90 spectrum of Compound I..... | 73 |
| A.6 The ¹³ C-NMR DEPT-135 spectrum of Compound I..... | 74 |
| A.7 The GLC chromatograms of standard steroids and Compound I..... | 75 |
| A.8 The IR spectrum of Compound II..... | 76 |
| A.9 The MASS spectrum of Compound II..... | 77 |
| A.10 The ¹ H-NMR spectrum of Compound II..... | 78 |
| A.11 The ¹³ C-NMR spectrum of Compound II..... | 79 |
| A.12 The ¹³ C-NMR spectrum of Compound II..... | 80 |
| A.13 The ¹³ C-NMR spectrum of Compound II..... | 81 |
| A.14 The ¹ H- ¹ H COSY spectrum of Compound II..... | 82 |
| A.15 The ¹ H- ¹ H NOESY spectrum of Compound II..... | 83 |
| A.16 The IR spectrum of Compound III..... | 84 |
| A.17 The MASS spectrum of Compound III..... | 85 |
| A.18 The ¹ H-NMR spectrum of Compound III..... | 86 |

| | Page |
|--|------|
| A.19 The ^{13}C -NMR spectrum of Compound III..... | 87 |
| A.20 The ^{13}C -NMR spectrum of Compound III..... | 88 |
| A.20 The ^{13}C -NMR spectrum of Compound III..... | 89 |
| A.22 The ^1H - ^1H COSY spectrum of Compound III..... | 90 |
| A.23 The ^1H - ^1H NOESY spectrum of Compound III..... | 91 |
| A.24 The HMQC spectrum of Compound III..... | 92 |
| A.25 The HMBC spectrum of Compound III..... | 93 |
| A.26 The IR spectrum of Compound IV..... | 94 |
| A.27 The MASS spectrum of Compound IV..... | 95 |
| A.28 The comparison of fragmentaiton pattern of Compound IV and 1,3-benzodioxole-5-carboxylic acid..... | 96 |
| A.29 The ^1H -NMR spectrum of Compound IV..... | 97 |

LIST OF SCHEMES

| Scheme | Page |
|---|------|
| 2.1 Extraction of the dried stems of <i>Piper aurantiacum</i> Miq. | 24 |
| 3.1 The fragmentation of Compound II..... | 46 |
| 3.2 The fragmentation of Compound III..... | 51 |
| 3.3 The fragmentation of isoborneol p-coumarate..... | 56 |
| 3.4 The fragmentaiton of Compound IV..... | 58 |

LIST OF TABLES

| Table | Page |
|---|------|
| 1.1 The insecticidal and insect antifeedant compounds from Piper genus..... | 2 |
| 1.2 The chemical constituents of some plants in Piper genus..... | 7 |
| 1.3 Pharmacological activities of some medicinal plants in Piper genus..... | 18 |
| 2.1 The results of dried stems extraction of <i>Piper aurantiacum</i> Miq. | 25 |
| 2.2 The results of the separation of the hexane extract by quick column chromatography..... | 26 |
| 2.3 The results of the separation of fraction No.10-24 by chromatotron technique..... | 28 |
| 2.4 The results of the separation of fraction No.144-152 by column chromatography..... | 29 |
| 2.5 The results of the separation of the chloroform extract by quick column chromatography..... | 30 |
| 2.6 The results of the separation of the ethyl acetate extract by quick column chromatography..... | 32 |
| 2.7 The results of the separation of the n-butanol extract by quick column chromatography..... | 33 |
| 2.8 The results of the separation of the methanol extract..... | 34 |
| 2.9 The results of the separation of fraction B by chromatotron technique..... | 35 |
| 3.1 The infrared absorption band assignments of Compound I..... | 39 |
| 3.2 Comparison of the ¹³ C-NMR spectrum of Compound I and β-sitosterol..... | 40 |
| 3.3 The infrared absorption band assignments of Compound II..... | 43 |

| | Page |
|--|-------------|
| 3.4 Comparison of the ^1H -NMR spectrum of Compound II and methyl piperate..... | 44 |
| 3.5 Comparison of the ^{13}C -NMR spectrum of Compound II and methyl piperate..... | 44 |
| 3.6 The infrared absorption band assignments of Compound II..... | 48 |
| 3.7 Comparison of the ^1H -NMR spectrum of Compound III and borneol p-coumarate..... | 49 |
| 3.8 Comparison of the ^{13}C -NMR spectrum of Compound III and borneol p-coumarate..... | 50 |
| 3.9 The One Bond and Multiple Bond Correlation of Compound III..... | 52 |
| 3.10 Comparison of the ^1H -NMR spectrum of borneol p-coumarate and isoborneol p-coumarate..... | 54 |
| 3.11 Comparison of the ^{13}C -NMR spectrum of borneol p-coumarate and isoborneol p-coumarate..... | 55 |
| 3.12 The infrared absorption band assignments of Compound IV..... | 57 |
| 4.1 Chemical constituents of the stem of <i>Piper aurantiacum</i> Miq. | 60 |

LIST OF ABBREVIATIONS

| | |
|---------------------|---|
| br | broad (IR), (NMR) |
| °C | degree celsius |
| CC | column chromatography |
| CHCl ₃ | chloroform |
| ¹³ C-NMR | carbon 13 nuclear magnetic resonance |
| cm | unit of centimetre |
| cm ⁻¹ | unit of wavenumber |
| cont | continue |
| δ | chemical shift |
| d | doublet (NMR) |
| dd | doublet of doublet (NMR) |
| DEPT | distortionless enhancement by polarization transfer |
| EI | electron impact technique in mass spectrometry |
| EtOAc | ethyl acetate |
| Fig. | figure |
| g | gram (s) |
| ¹ H-NMR | proton nuclear magnetic resonance |
| HMBC | heteronuclear multiple bond correlation |
| HMQC | heteronuclear multiple quantum correlation |
| IR | infrared |
| J | coupling constant (NMR) |
| kg | kilogram (s) |
| m | medium (IR) |
| m | multiplet (NMR) |
| M ⁺ | molecular ion in mass spectrum |

| | |
|--------------|----------------------------------|
| MeOH | methanol |
| mg | milligram (s) |
| min | minute (s) |
| ml | millilitre |
| m.p. | melting point |
| M.W. | molecular weight |
| m/z | mass per charge |
| ν_{\max} | wavelength at maximum absorption |
| n-BuOH | n-butanol |
| No. | number |
| NOE | nuclear overhauser effect |
| ppm | part per million |
| q | quartet (NMR) |
| R_f | rate of flow in chromatography |
| s | strong (IR) |
| s | singlet (NMR) |
| t | triplet (NMR) |
| TLC | thin layer chromatography |
| w | weak (IR) |
| wt. by wt. | weight by weight |