



UNIVERSITI PUTRA MALAYSIA

**PHYTOCHEMICAL INVESTIGATION AND BIOACTIVITY SCREENING
OF VITEX (VERBENACEAE) AND FICUS (MORACEAE) SPECIES**

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FSAS 2002 7

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By

HASSAN ABDALLA ALMAHY DAFALLA

**Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia,
in Fulfilment of the Requirements for Degree of Doctor of Philosophy**

February 2002



DEDICATION

TO THE SOUL OF MY FATHER AND BROTHER ALMAHY

TO MY MOTHER

TO MY BROTHER AND SISTERS

TO MY WIFE

TO ALL OF THEM WITH LOVE AND GRATITUDE

**Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment
of the requirement for the degree of Doctor of Philosophy**

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February 2002

Chairman: Prof. Dr. Mawardi Rahmani

Faculty: Science and Environmental Studies

The study on *Vitex longisepala* involves extraction, various chromatographic methods and structural determination by spectroscopic techniques such as IR, compounds were also elucidated by comparison with the previous works. works on leaves and bark of the plant yielded cholesterol (51), *para*-hydroxybenzoic acid (52), terpene.

Crude extracts and isolated compounds from two parts of this plant were screened for antimicrobial activity using disc diffusion method and cytotoxic activity by using microtitration method.

compouds exhibited antimicrobial activity against Gram-positive and Gram-negative bacteria. Cholesterol exhibited significant cytotoxic activity against T-lymphoblastic leukemic cell line with IC₅₀ 10 µg/ml.

not show any antimicrobial activity against fungi. The methanol crude extract of the bark failed to show any significant antimicrobial activity, while the petroleum ether and the chloroform crude extracts of the bark exhibited weak antimicrobial activity against *Bacillus cereus*.

The study on *Vitex quinata* involved the same procedure adopted above. Isolation works on the leaves and bark of the plant yielded cholesterol (51), β -sitosterol (57), *para*-hydroxybenzoic acid (52), fructose (53), glucose (58), catechin (55), quercetin (59) and quercitrin (60). However, the bark of the plant yielded a mixture of long-chain compound, fatty acid and unidentified terpene. The crude extracts and isolated compounds of this plant were tested for antimicrobial and cytotoxic activity using disc diffusion and microtitration methods respectively. The crude extracts and pure isolated compounds exhibited positive antimicrobial results against two bacteria organisms and negative results against four fungi. Cholesterol and β -sitosterol gave cytotoxic activity against T-lymphoblastic leukemic cell line with IC₅₀ 10 and 25 $\mu\text{g}/\text{ml}$ respectively.

Detail investigation on the leaves, bark and fruits of *Ficus benjamina* has resulted in the isolation of seven compounds. The structure of these compounds were elucidated by means of spectroscopic methods including by the extensive use of various NMR techniques and also comparison with previous studies. The use of High Field NMR is essential in structural determination of these complex molecules. With

the aids of various NMR experimental techniques and other spectroscopic methods such as IR, UV and MS, the correct structures of the pure isolated compounds were established.

(63),

The presence of bioactive compounds in this plant was detected by the use of antimicrobial organism.

crude plant extracts or pure isolated compounds could be determined. of chloroform and methanol extracts of the leaves of *Ficus benjamina* gave no significant activity while caffeic acid gave IC₅₀ value of 25 µg/ml.

Phytochemical studies on leaves and bark of *Ficus elastica* have resulted in the isolation of emodin (66), mannitol (70) together with long-chain fatty acids. were established based on spectral studies using different spectroscopic methods and on comparison with published data.

**Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai
memenuhi keperluan memperoleh ijazah Doktor Falsafah**

**PENYIASATAN FITOKIMIA DAN SARINGAN BIOAKTIVITI *VITEX*
(VERBENACEAE) DAN *FICUS* (MORACEAE) SPESIES**

Oleh

HASSAN ABDALLA ALMAHY DAFALLA

Februari 2002

Pengerusi : Prof. Dr. Mawardi Rahmani
Fakulti : Sains dan Pengajian Alam Sekitar

Kajian tentang *Vitex longisepala* dijalankan dengan melakukan pengekstrakan, pemencilan menggunakan kaedah kromatografi dan penentuan struktur melalui teknik spektroskopi seperti 1R, NMR termasuk 2D-NMR dan MS. Struktur sebatian telah juga diuraikan secara perbandingan dengan kajian sebelum ini. Pemencilan terhadap daun dan kulit tumbuhan menghasilkan kolesterol (51), asid parahidroksibenzoik (52), fruktosa (53), xilosa (54), katekin (55), genestein (56) dan terpena yang tidak dikenali.

Ekstrak mentah dan sebatian yang dipencarkan daripada dua bahagian tumbuhan ini telah dikesan aktiviti antimikrob dengan menggunakan kaedah peresapan cakera dan aktiviti sitotoksik menggunakan kaedah mikropentitratan. Ekstrak mentah daun dan sebatian tulen yang dipencarkan memperlihatkan aktiviti

antimikrob menentang bakteria Gram-positif dan Gram-negatif. Kolesterol menunjukkan aktiviti sitotoksik yang signifikan menentang sel leukemia T-limfoblas dengan IC_{50} 10 μ g/ml.

Walau bagaimanapun semua sebatian tidak menunjukkan sebarang aktiviti antimikrob menentang kulat.

antimikrob yang signifikan,

mentah kloroform kulit menunjukkan aktiviti antimikrob yang lemah terhadap *Bacillus cereus*.

Kajian terhadap *Vitex quinata* melibatkan prosedur yang sama yang telah digunakan di atas. Kajian pemencilan daun dan kulit tumbuhan menghasilkan kolesterol (51),

glukosa (58),

kulit tumbuhan menghasilkan campuran asid lemak berantai panjang dan terpena yang tidak dikenali.

mendapatkan aktiviti antimikrob dan sitotoksik dengan menggunakan kaedah peresapan cakera dan mikropentitratan.

Ekstrak mentah dan sebatian tulen menunjukkan keputusan antimikrob positif terhadap dua bakteria dan keputusan negatif terhadap empat kulat.

dan β -sitosterol memberi aktiviti sitotoksik terhadap sel leukemia T-limfoblas dengan nilai IC₅₀ masing-masing 10 dan 25 $\mu\text{g/ml}$.

Kajian mendalam terhadap daun, kulit dan buah *Ficus benjamina* telah menghasilkan tujuh komponen. Struktur komponen ini telah diuraikan dengan menggunakan kaedah spektroskopi termasuk pelbagai teknik NMR dan juga perbandingan dengan kajian lampau. Penggunaan NMR Medan Tinggi adalah penting dalam penentuan struktur molekul yang kompleks. Dengan bantuan pelbagai teknik eksperimen NMR dan kaedah spektroskopi lain seperti IR, UV dan MS, struktur sebatian yang dipencil tulen dapat dipastikan sebagai asid sinamik (61), laktosa (62), naringenin (63), quersetin (59), asid kafeik (64) dan stigmasterol (65).

Kehadiran sebatian bioaktif dalam tumbuh-tumbuhan ini telah dikesan dengan menggunakan organisma antimikrob. Dengan bantuan sistem biocerakinan ini aktiviti ekstrak tumbuhan mentah atau sebatian tulen dapat dipastikan. Nilai IC₅₀ ekstrak kloroform dan etanol daun *Ficus benjamina* tidak memberikan aktiviti yang signifikan sementara asid kafeik mempunyai nilai IC₅₀ sebanyak 25 $\mu\text{g/ml}$.

Kajian fitokimia terhadap daun dan kulit tumbuhan ini telah menghasilkan emodin (66), sukrosa (67), laktosa (62), morin (68), rutin (69) dan D-manitol (70) bersama dengan asid lemak berantai panjang. Struktur sebatian ini dipastikan

berasasban kajian menggunakan pelbagai kaedah spektroskopi dan perbandingan dengan data yang telah diterbitkan.

ACKNOWLEDGEMENTS

First and foremost, my heartfelt thanks to Almighty Allah for giving me the strength and willpower to complete this challenging task.

My utmost appreciation and gratitude to Professor: Dr. Mawardi Rahmani Chairman of Supervisory Committee, for his invaluable guidances, constructive comments, advice, suggestions and support which led to the completion of my Ph.D study.

My sincere thanks and appreciation to Associate Professor: Dr. Mohd Aspollah Hj. Sukari and Professor: Dr. Abdul Manaf Ali as members of the supervisory committee, for their constructive comments and encouragement.

I am most grateful to the Government of Sudan, University of Juba and Government of Malaysia under IRPA Grant (51263 and 51473) for the research funding and financial assistance to me throughout the course of the study

I am also grateful to my colleagues, friends and the staff members of the Chemistry Department for always being so willing to render assistance throughout the course of my study. My special thanks also go to Mr. Narzari, Zainudin, Zainal

Zahari, Nordin, Zainal, Zulkifli, Abas, Samsuddin Musa and Rusnani for their assistance on IR, NMR and mass spectra.

My thanks also go to all members of the Faculty of Science who contributed in one way or another towards the completion of my study.

I am indebted to Mrs. Normah Abdalla for her invaluable assistance for her translation of the English Abstract of this study to Malay language.

Lastly, my heartfelt appreciation to my parents, brother, sisters and my wife for their understanding, support and encouragement during the period of my study.

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