

สารต้านอนุมูลอิสระจากเปลือกของต้นเหียงพรัานางแอ *Carallia brachiata* Merr.



นายปรีชา โสวรรณทิพย์

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**FREE RADICAL SCAVENGERS FROM THE BARKS OF
Carallia brachiata Merr.**

Mr.Preecha Sowanthip



**A Thesis Submitted in Partial Fulfillment of the Requirements
for the Degree of Master of Science in Chemistry**

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ในการเสาะหาสารที่มีฤทธิ์ทางชีวภาพ สกัดจากเปลือกต้นเถียงพรัานางแอ (*Carallia brachiata* Merr.) แสดงฤทธิ์ต้านอนุมูลอิสระสูง จึงเลือกศึกษาองค์ประกอบทางเคมีของพืชชนิดนี้ จากการแยกสิ่งสกัดไดคลอโรมีเทนและเอทิลอะซิเตต สามารถแยกสารใหม่ได้หนึ่งชนิดคือ afzalachin-(4→6, 2→O→7)-afzelechin-(4→8, 2→O→7)-afzelechin พร้อมกับของผสมสามชนิดและสารบริสุทธิ์อีกสองชนิด ได้แก่ของผสมเอสเทอร์โซ่ตรง, ของผสม stigmasterol กับ β -sitosterol, ของผสม alkyl *trans*-ferulate ester, *p*-hydroxybenzoic acid และ afzalachin-(4→8, 2→O→7)-epiafzelechin ซึ่งสารใหม่พิสูจน์ทราบโดยวิธีทางสเปกโทรสโกปี

ผลการทดสอบฤทธิ์ทางชีวภาพ พบว่าของผสม alkyl *trans*-ferulate ester, afzalachin-(4→8, 2→O→7)-epiafzelechin และ afzalachin-(4→6, 2→O→7)-afzelechin-(4→8, 2→O→7)-afzelechin แสดงฤทธิ์ต้านอนุมูลอิสระกับ DPPH ด้วยค่า IC_{50} 134.96, 98.86, 83.49 ไมโครกรัมต่อมิลลิลิตร ตามลำดับ afzalachin-(4→8, 2→O→7)-epiafzelechin และ afzalachin-(4→6, 2→O→7)-afzelechin-(4→8, 2→O→7)-afzelechin แสดงฤทธิ์ต้านการเกิดออกซิเดชันเมื่อทดสอบด้วยวิธี ferric thiocyanate assay นอกจากนี้ยังแสดงฤทธิ์สูงต่อการทดสอบด้วยวิธี superoxide dismutase, xanthine oxidase inhibition โดยเทียบกับ BHA และ allopurinol ตามลำดับ

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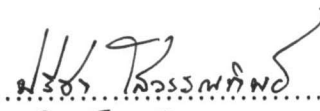
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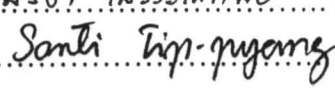
**PREECHA SOWANTHIP : FREE RADICAL SCAVENGERS FROM
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**THESIS ADVISOR : ASSISTANT PROFESSOR SANTI TIP-PYANG,
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In a search for bioactive compounds, the crude extracts from the barks of *Carallia brachiata* Merr. showed significant free radical scavenging activity. This plant was selected for further study on chemical constituents. The fractionation of dichloromethane and ethyl acetate crude extracts led to the isolation of one new compound, afzelechin-(4→6, 2→O→7)-afzelechin-(4→8, 2→O→7)-afzelechin together with three known mixtures and two pure compounds, a mixture of long chain aliphatic esters, a mixture of a stigmasterol and β-sitosterol, a mixture of alkyl *trans*-ferulate ester, *p*-hydroxybenzoic acid and afzelechin-(4→8, 2→O→7)-epiafzelechin. The new compound was elucidated by spectroscopic method.

The biological activities results indicated that a mixture of alkyl *trans*-ferulate ester, afzelechin-(4→8, 2→O→7)-epiafzelechin and afzelechin-(4→6, 2→O→7)-afzelechin-(4→8, 2→O→7)-afzelechin showed significant activity on DPPH with IC₅₀ 134.96, 98.86 and 83.49 μg/ml, respectively. Afzelechin-(4→8, 2→O→7)-epiafzelechin and afzelechin-(4→6, 2→O→7)-afzelechin-(4→8, 2→O→7)-afzelechin also showed antioxidant activity with ferric thiocyanate assay. In addition, afzelechin-(4→8, 2→O→7)-epiafzelechin and afzelechin-(4→6, 2→O→7)-afzelechin-(4→8, 2→O→7)-afzelechin were exhibited strong activities on superoxide dismutase and xanthine oxidase inhibition methods by comparison with BHA and allopurinol, respectively.

Department.....Chemistry.....Student's signature.....

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List of Abbreviations

BHA	= butylated hydroxyanisole
°C	= degree celsius
CC, SiO ₂	= column chromatography using silica gel as adsorbent
CD ₃ OD	= deuterated methanol
CDCl ₃	= deuterated chloroform
COSY	= two – dimensional ¹ H correlation spectroscopy
δ	= chemical shift
d	= doublet
dd	= doublet of doublet
DEPT	= distortionless enhancement by polarization transfer
DMSO	= dimethylsulfoxide
DPPH	= 2,2-diphenyl-1-(2,4,6-trinitrophenyl) hydrazyl radical
DPPHn	= 2,2-diphenyl-1-(2,4,6-trinitrophenyl) hydrazine
EIMS	= electron impact mass spectrometry
FABMS	= fast atom bombardment mass spectrometry
FT	= fourier transform
g	= gram
GC	= gas chromatography
HMBC	= heteronuclear multiple bond connectivity by 2D multiple quantum NMR
HMQC	= ¹ H – detected heteronuclear multiple quantum coherence via direct coupling
Hz	= hertz
IC ₅₀	= inhibition concentration (concentration caused 50% inhibition)
IR	= infrared
<i>J</i>	= coupling constant
kg	= kilogram
m	= multiplet
m.p.	= melting point
m/z	= mass per charge

mg	= milligram
μg	= microgram
ml	= milliliter
mM	= milimolar
MW	= molecular weight
NBT	= nitrobluetetrazolium
nm	= nanometre
NMR	= nuclear magnetic resonance
NOESY	= nuclear overhauser and exchange spectroscopy
ν_{max}	= wave number cause maximum absorption
ppm	= part per million (or mg/g)
q	= quartet
R_f	= retardation factor
s	= singlet
t	= triplet
TLC	= thin layer chromatography
UV	= ultra-violet
w/w	= weight by weight
XA	= xanthine
XOD	= xanthine oxidase

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