

**LEMBAR
HASIL PENILAIAN SEJAWAT SEBIDANG ATAU PEER REVIEW
KARYA ILMIAH: JURNAL ILMIAH**

Judul Publikasi Ilmiah (Artikel)	: Coconut (Cocos nucifera L.) lipids: Extraction and characterization
Jumlah Penulis	: 3 orang
Status Pengusul	: penulis pertama/utama
Identitas Jurnal Ilmiah	: Oriental Journal of Chemistry
a. Nama Jurnal Ilmiah	: ISSN: 0970-020 X
b. Nomor ISBN /ISSN	: Volume 34, Issue 2, 2018
c. Volume, Nomor, Bulan, Tahun	: Oriental Scientific Publishing Company
d. Penerbit	: http://dx.doi.org/10.13005/ojc/340268
e. DOI artikel (jika ada)	: http://www.orientichem.org/category/vol34no2
f. Alamat web jurnal	: Scimago Journal Rank (SJR 0.17, Q4, H-index 18)
g. Terindeks di	: SCOPUS (CiteScore is 0.61)
e. Jumlah Halaman	: 5 (1136-1140) halaman

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☐ Jurnal Ilmiah Nasional/Nasional Terindeks di DOAJ, CABI, COPENICUS**

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Komponen Yang Dinilai	Nilai Reviewer		Nilai Rata-rata
	Reviewer I	Reviewer II	
a. Kelengkapan unsur isi jurnal (10%)	4	2,8	3,4
b. Ruang lingkup dan kedalaman pembahasan (30%)	9	9,6	9,3
c. Kecukupan dan kemutakhiran data/informasi dan metodologi (30%)	10	10,2	10,1
d. Kelengkapan unsur dan kualitas penerbit (30%)	11	10,8	10,95
Total = (100%)	34	33,4	33,7
Nilai Pengusul = (60%)	20,4	20,04	20,22

Reviewer

Prof. Dr. ret nnt Nuryono, M.S.
NIP. 196407141988111001

Unit kerja : Universitas Gadjah Mada Yogyakarta
Jabatan Fungsional : Guru Besar
Bidang ilmu : Kimia

Semarang,
Reviewer I

Prof. Dr. Moh Djaeni, ST, M.Eng
NIP 197102071995121001

Unit Kerja : Universitas Diponegoro Semarang
Jabatan Fungsional : Guru Besar
Bidang Ilmu : Teknik Kimia

Terindex Scopus sampai 2018, saat ini sudah discontinued.
Nilai diturunkan menjadi 31. Nilai pengusul = $60\% \times 31 = 18,60$

Prof. Dr. Moh. Djaeni, ST, M.Eng
PAK UNDIP

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e. DOI artikel (jika ada)	: http://www.orientichem.org/category/vo134no2
f. Alamat web jurnal	: http://www.orientichem.org/category/vo134no2
g. Terindeks di SCOPUS, Scimago Journal Rank (SJR 0.168 (2018), Q4 (2018) H-index 18)	
c. Jumlah Halaman	: 5 (1136-1140) halaman

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Hasil Penilaian Peer Review

Komponen Yang Dinilai	Nilai Maksimal Jurnal Ilmiah			Nilai Akhir yang diperoleh
	Internasional Bereputasi Berimpact faktor	Nasional Terakreditasi	Nasional	
	(40)			
a. Kelengkapan unsur isi jurnal (10%)	4			4
b. Ruang lingkup dan kedalaman pembahasan (30%)	12			9
c. Kecukupan dan kemutakhiran data/informasi dan metodologi (30%)	12			10
d. Kelengkapan unsur dan kualitas penerbit (30%)	12			11
Total = (100 %)	40			34
Nilai Pengusul = (60% x 34) = 20,4				20,4

Catatan Penilaian artikel oleh Reviewer :

- Kesesuaian dan kelengkapan unsur isi artikel:**
Artikel sangat lengkap, dimana analisis tentang ekstraksi dan karakterisasi lipida kelapa (Cocos nucifera L.) disajikan dengan terperinci, disitasi dan dibahas. Topik dan materi sesuai dengan jurnal yang bersangkutan. Tata penulisan tersaji dengan baik.
- Ruang lingkup dan kedalaman pembahasan:**
Artikel ini membahas tentang ekstraksi dan karakterisasi lipida kelapa (Cocos nucifera L.) utamanya adalah dari kelompok fosfolipida. Ekstraksi dilakukan dengan maserasi dan kolom kromatografi dengan berbagai variasi solven untuk mendapatkan hasil terbaik. Karakterisasi hasil dilakukan dengan FTIR dan GCMS. Pembahasan yang dilakukan sangat singkat dan kurang komprehensif karena hanya 3 referensi yang dijadikan benchmark. Data-data pendukung yang digunakan juga kurang detail.
- Kecukupan dan kemutakhiran data/informasi dan metodologi:**
Referensi yang dicitasi dalam artikel ini ada 30 dimana 27 Baru (dalam 10 tahun terakhir). Nilai novelty/kebaruan artikel cukup baik. Metode cukup jelas, namun tidak dilengkapi dengan skema tahapan yang memadai.
- Kelengkapan unsur dan kualitas terbitan:**
Penerbit adalah Oriental Scientific Publishing Company (bereputasi). Jurnalnya termasuk kategori Q4 (2018) dengan nilai SJR 0.168 (2018) dan H-index 18 (2019). Nilai similaritas artikel berdasarkan Turnitin hanya 4%, sehingga orisinalitas sangat baik.

Terindex Scopus sampai 2018, Saat ini sudah discontinued. Nilai diturunkan menjadi 31. Nilai pengusul = $60\% \times 31 = 18,60$

Semarang, November 2019

Reviewer

Prof. Dr. Moh. Djaeni, ST, M.Eng
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Unit Kerja : Universitas Diponegoro
Jabatan Fungsional : Guru Besar
Bidang Ilmu : Teknik Kimia

Prof. Dr. Moh. Djaeni, ST, M.Eng
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g. Terindeks di	: e. Jumlah Halaman : 5 (1136-1140) halaman

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Komponen Yang Dinilai	Nilai Maksimal Jurnal Ilmiah			Nilai Akhir yang diperoleh
	Internasional Bereputasi Berimpact faktor	Nasional Terakreditasi	Nasional	
	(40)			
a. Kelengkapan unsur isi jurnal (10%)	4			2,8
b. Ruang lingkup dan kedalaman pembahasan (30%)	12			9,6
c. Kecukupan dan kemutakhiran data/informasi dan metodologi (30%)	12			10,2
d. Kelengkapan unsur dan kualitas penerbit (30%)	12			10,8
Total = (100 %)	40			33,4

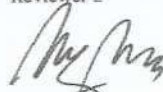
Nilai Pengusul = (60% x 33,4) = 20,04

Catatan Penilaian artikel oleh Reviewer :

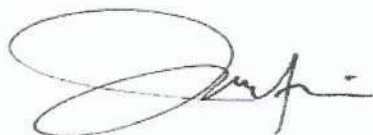
- a. Kelengkapan unsur isi jurnal:**
 Oriental journal of chemistry memuat isi secara lengkap termasuk tim editor bereputasi dari beberapa negara. Jurnal memuat artikel sangat lengkap dan sesuai antara judul, abstract, latar belakang, tujuan, metodologi, hasil dan pembahasan serta kesimpulan, yaitu melaporkan hasil ekstraksi dan karakterisasi lipid kelapa dengan FTIR spectroscopy, TLC dan GCMS. Namun artikel ini disusun kurang teliti, penulisan sitasi tidak konsisten dan gambar kurang jelas. Spektra GCMS tidak dicantumkan dan pembahasan kurang komprehensif. Dari uji similaritas artikel menggunakan Turnitin yang sebesar 4% menunjukkan tingkat orisinalitas yang tinggi.
- b. Ruang lingkup dan kedalaman pembahasan:**
 Ruang lingkup artikel yang membahas tentang hasil ekstraksi dan karakterisasi lipid kelapa kurang luas. Terkait dengan kedalaman pembahasan, hasil kurang mendalam dibahas sehingga nampak singkat. Pembahasan tidak dibandingkan dengan hasil penelitian yang lain.
- c. Kecukupan dan kemutakhiran data/informasi dan metodologi:**
 Pendahuluan dirumuskan sangat singkat dan kurang mengeksplor hasil penelitian sebelumnya yang terkait dengan lipida kelapa. Data eksperimen yang dilaporkan ekstraksi dan karakterisasi lipida kelapa dan kurang menunjukkan adanya kebaruan. Referensi yang disitasi sebanyak 30 dengan 29 (96%) terbit 10 tahun terakhir dan semuanya dari jurnal. Metode yang disajikan tidak ada yang baru dan kurang dilengkapi dengan kuantitas bahan secara detail.
- d. Kelengkapan unsur dan kualitas penerbit:**
 Penerbit adalah Oriental Scientific Publishing Company (bereputasi). Jurnalnya terindeks di SCOPUS, Scimago journal Rank (SJR 0.168 (2018), Q4 (2018) H-index 18). Jurnal kurang teliti dalam editing paper.

Terindeks Scopus sampai 2018, saat ini sudah discontinued. Nilai diturunkan menjadi 31. Nilai pengusul = 60% x 31 = 18,60

Yogyakarta, November 2019
 Reviewer 2



Prof. Dr. rer. nat Nuryono, M.S.
 NIP. 196407141988111001
 Bidang Ilmu/Unit kerja :
 Departemen Kimia pada Fakultas MIPA UGM Yogyakarta



Prof. Dr. Moh. Djaeni, ST, MEng
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Oriental Journal of Chemistry
Volume 34, Issue 2, 2018, Pages 1136-1140

Coconut (*Cocos nucifera* L.) lipids: Extraction and characterization (Article)

(Open Access)

Hudiyanti, D.^a, Al Khafiz, M.F.^b, Anam, K.^a

Save all to author list

^aDepartment of Chemistry, Faculty of Science and Mathematics, Diponegoro University, Jl. Prof. Soedarto, Semarang, SH 50 275, Indonesia

^bChemistry Program, Faculty of Science and Mathematics, Diponegoro University, Jl. Prof. Soedarto, Semarang, SH 50 275, Indonesia

Abstract

View references (30)

Lipids extraction from dried coconut meat is conducted by maceration using chloroform/methanol (2:1,v/v) and partition extraction with n-hexane/87% ethanol (3:1), and column chromatography. Lipids are analyzed using FTIR, GC-MS, and TLC. Results reveal that dried coconut meat has 0.04% phospholipids, 0.22% of neutral lipids, and 0.04% of glycolipids and ceramides. Coconut phospholipids consist of phosphatidylcholine (PC), phosphatidylserine (PS), and/or phosphatidylethanolamine (PE) classes for the hydrophilic parts with dodecanoic acid (C12: 0), hexadecanoic acid (C16:0) and octadecanoic acid (C18: 0) as the lipophilic parts. © 2018 Oriental Scientific Publishing Company. All rights reserved.

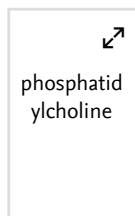
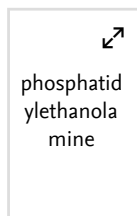
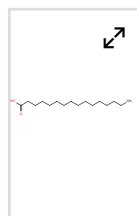
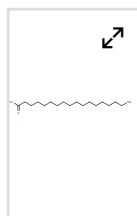
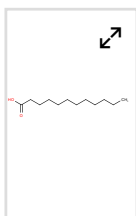
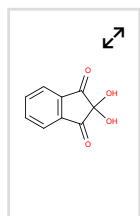
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Hudiyanti, D. , Arya, A.P. , Siahaan, P. (2015) *Oriental Journal of Chemistry*

Study on leakage of sesame (*Sesamum indicum* L.) and coconut (*Cocos nucifera* L.) liposomes

Hudiyanti, D. , Raharjo, T.J. , Narsito, N. (2015) *Oriental Journal of Chemistry*

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Document Type: Article

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Study on leakage of sesame (*Sesamum indicum* L.) and coconut (*Cocos nucifera* L.) liposomes
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Chemical composition and phospholipids content of Indonesian Jack Bean (*Canavalia ensiformis* L.)
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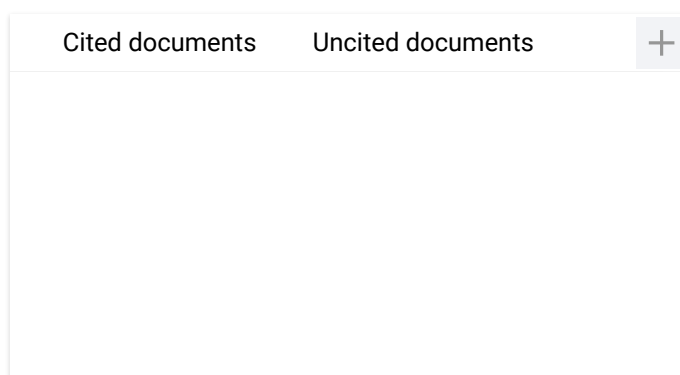
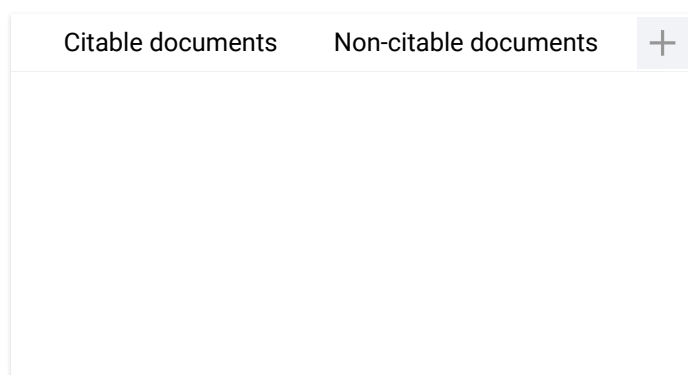
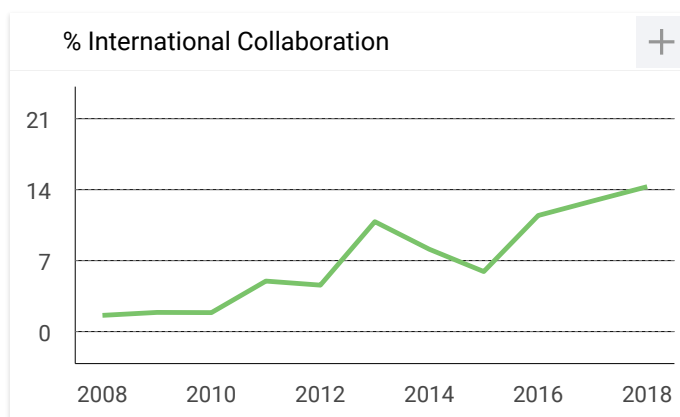
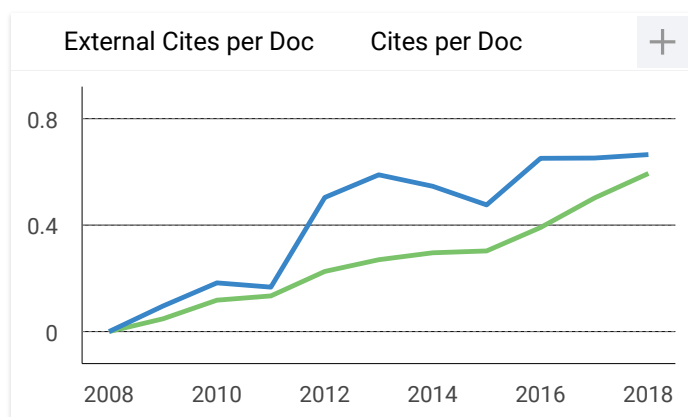
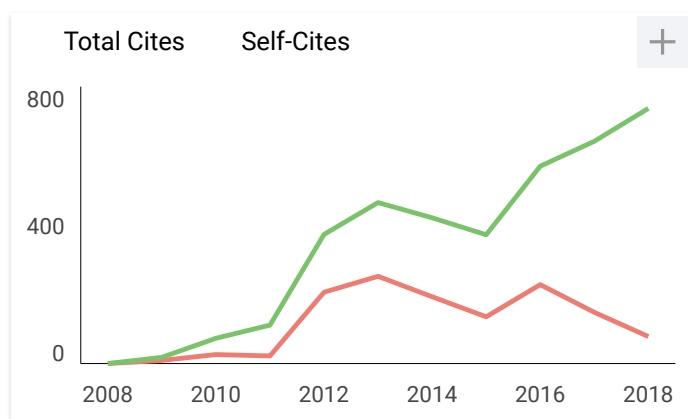
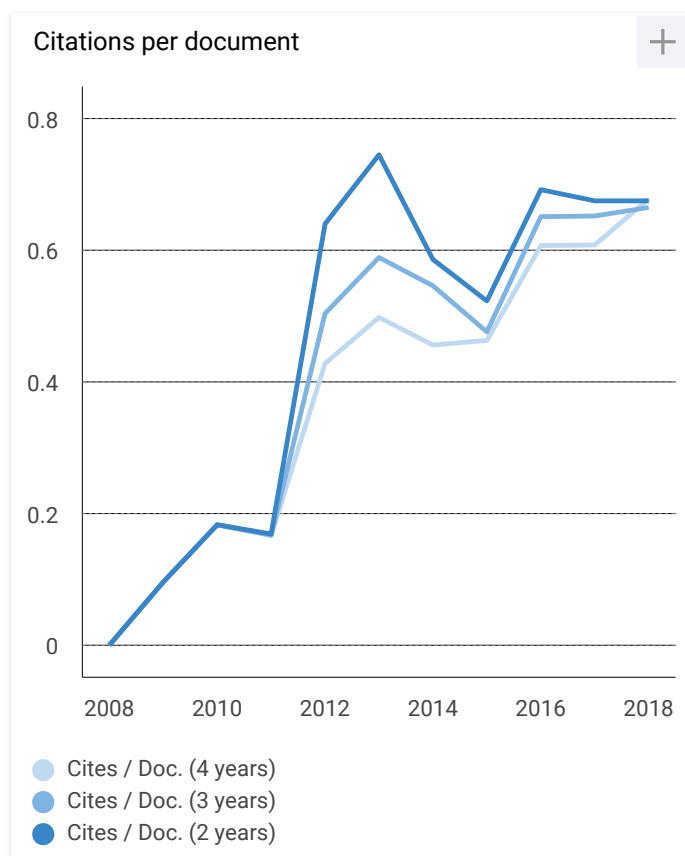
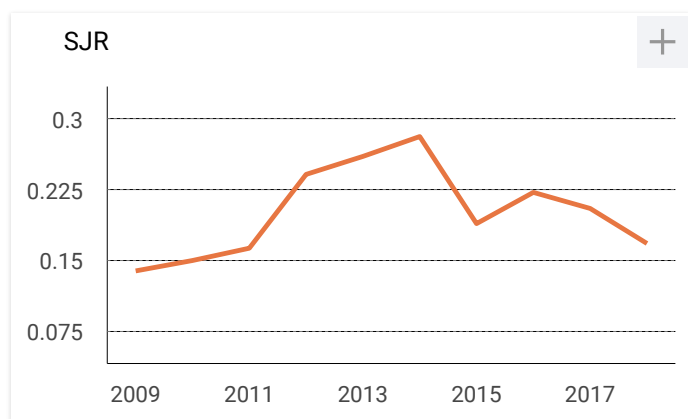
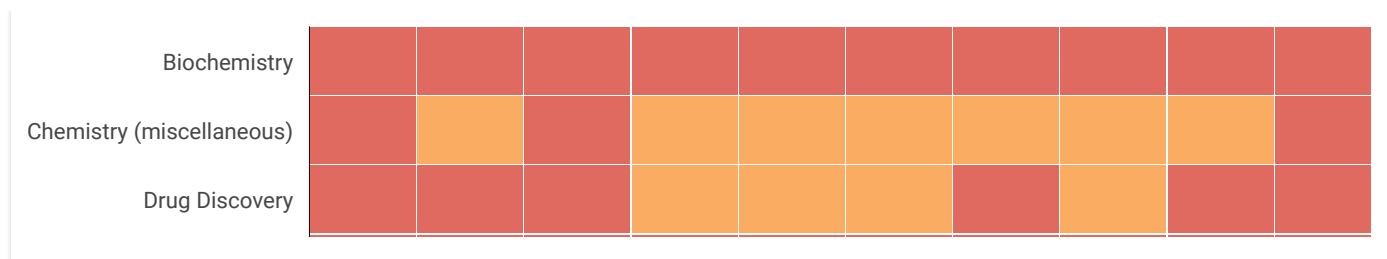
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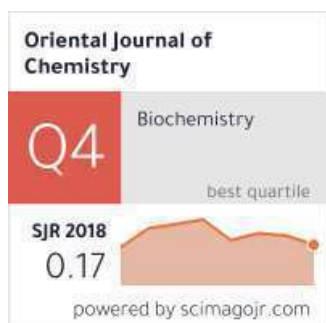
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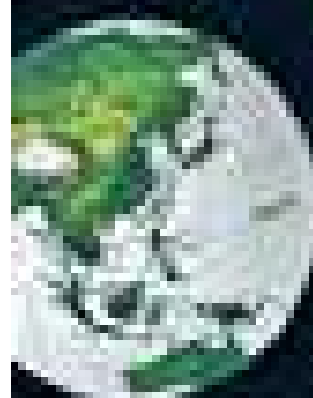
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Silver/oil Nanofluids in Heat Exchanger An Experimental Study on Convective Heat Transfer

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ABSTRACT

In present work, we used two sizes of silver nanoparticles (NPs) with different particle loadings and dispersed them into oil to make nanofluids. The silver/oil nanofluid with different nanoparticles weight fractions of 0.5%, 1% and 2% was prepared by ultrasonic device in two step method. The measurements of heat transfer coefficients were performed. An up to 30% enhancement of the heat transfer coefficient was observed in the laminar flow regime. The heat transfer coefficient is shown to increase with increasing *Reynolds number*. In addition, the heat transfer coefficient decreased with increasing fluid temperature. A constant downward trend of enhancement ratio with respect to *Reynolds number* was specified for the nanofluids discussed in this study.



Synthesis of Silver Nanoparticles in the Presence of Polyethylene Glycol and their Electrochemical Behavior at a Graphite Electrode by Cyclic Voltammetry

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ABSTRACT

The shape, size, optical properties and electrochemical activity of the silver nanoparticles (AgNPs) were studied, as well as their dependence on the amount of the reducing agent, the method of initiation of the formation of nanoparticles and the introduction order of reagents in the reaction mixture. It was determined that in the sols obtained without polyethylene glycol (PEG) AgNPs are predominant with the average size of 40 nm. With PEG the triangular AgNPs predominate with the average size of 20 nm due to the silver ions reduction on the surface of the nuclei. It was determined that AgNPs are most active when obtained with PEG and the molar ratio $[Ag]:[C_6H_5O7^{3-}] = 1:5$. On the anodic branch of the cyclic AgNPs curve the splitting of anodic maximum into two due to AgNPs oxidation was observed. AgNPs obtained in the presence of PEG by light have the most electrochemical activity. The processes of AgNPs oxidation become easier when PEG is present which is caused by the formation of the most stable silver oxides.



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Coconut (*Cocos nucifera* L.) Lipids: Extraction and Characterization

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ABSTRACT

Lipids extraction from dried coconut meat is conducted by maceration using chloroform/methanol (2:1,v/v) and partition extraction with n-hexane/87% ethanol (3:1), and column chromatography. Lipids are analyzed using FTIR, GC-MS, and TLC. Results reveal that dried coconut meat has 0.04% phospholipids, 0.22% of neutral lipids, and 0.04% of glycolipids and ceramides. Coconut phospholipids consist of phosphatidylcholine (PC), phosphatidylserine (PS), and/or phosphatidylethanolamine (PE) classes for the hydrophilic parts with dodecanoic acid (C12: 0), hexadecanoic acid (C16:0) and octadecanoic acid (C18: 0) as the lipophilic parts.

Keywords: coconut, phospholipids, dodecanoic acid, hexadecanoic acid, octadecanoic acid

INTRODUCTION

Phospholipids (PLs) are amphiphilic lipids obtained in cell membranes of all organisms, organized as lipid bilayers. The PLs discovered are mainly glycerophospholipids (GPLs), which are made of fatty acid residues (FAs) attached to a glycerol backbone by ester formation, a phosphate group and a hydrophilic head group (e.g. choline, serine or ethanolamine). Widespread supplies of industrially cultivated PLs are eggs (chicken and fish), bovine milk, soya, rapeseed, sunflower, coconut, sesame (1) and jack bean (2). Each source has a unique composition of distinct phospholipid species and accordingly diverse applications in pharmaceuticals (3), cosmetics (4), nutrition, food (5), and drug delivery (6,7).

In aqueous media, PLs molecules form aggregate structures by self-assembly course of action (8,9). Spherical bilayers known as vesicles or liposomes are among of these structures (10,11). Nowadays liposomes (7,12–14) and related structures (15,16) have been actively developed as carriers of bioactive compounds (13,14,17–19) in various applications (13,20) since their excellent biocompatibility to natural body environment (6,12,21).

The characteristic of liposomes is greatly influenced by the physicochemical properties of PLs constituent, such as their molecular species (22–25). PLs molecular species are determined by their head group (hydrophilic) and hydrocarbon chain types (hydrophobic). These components are diversified based upon their sources (3). PLs acquired from various natural sources mostly have unique head groups and acyl chains compositions. A judicious stage in lipidomic study is lipid extraction with a suitable organic solvent combination (solvent system) preceding to MS analysis. The accomplishment of the lipid extraction by a certain solvent modification depends upon the partitioning of the lipids into the organic phase and therefore on the lipid composition (2,26,27).

Previously Hudiyanti *et al.* (25) obtained that crude coconut PLs contain sphalins groups whilst the lipophilic parts comprise of dodecanoic acid (C12:0) and octanoic acid (C8:0). In this study we take further steps by purifying these crude PLs with column vacuum chromatography using several eluents to learn more information about the chemistry of coconut PLs. Our new finding is that crude coconut PLs also contained phosphatidylcholine groups and the lipophilic components were hexadecanoic acid (C16:0) and 9-octadecenoic acid (C18:1) as well. Further details of this new finding is presented in the following discussions.

MATERIALS AND METHODS

Materials. Dried coconut meat, filter paper, silica gel G60 for column chromatography, chloroform, acetone, methanol, aquabidest, NaCl, the solvent for partition was prepared by partitioning ethanol 87% with n-hexane (1: 1, v/v) and the top layer was mark solvent A while the bottom layer was solvent B, TLC plat of silica gel GF254 was washed with chloroform/methanol (1:1 v/v), then impregnate with 2.3% boric acid in absolute ethanol prior used. Spot reagents for TLC were dragendorff, ninhydrin, and primulin reagents.

Extraction. Polar lipids from coconut meat was isolated by method previously presented by Hudiyanti *et al.* (25). Briefly, this was prepared by the following procedure. Dried coconut

meat powder was macerated with chloroform / methanol (2: 1) for 7 days. The filtrate was washed with a 0.9% of NaCl solution. Chloroform layer was evaporated to acquired coconut lipid extract. After that 10 g of coconut lipid extract was dissolved in 45 ml of solvent A and added to 15 ml solvent B in a separating funnel I. The mixture was shaken for 2 minutes. The bottom layer was transferred into a second separating funnel containing 45 ml solvent A and shaken for 2 minutes. The bottom layer containing polar lipids was collected in a vial for further purification while the upper layer was also kept in a separated vial. Fresh 15 ml solvent B was added to the remaining mixture in the first separating funnel and shaken for about 2 minutes. Then the bottom layer is transferred into the second separating funnel containing of fresh 45 ml solvent A. The mixture then shaken and separated as above. The bottom layer and the upper layer were accumulated into the previous separate containers. The procedure was repeated 4-6 times for each 10 g of coconut lipid extract. The polar lipids containing PLs were purified by column vacuum chromatography as described below.

Purification. A column containing 50 grams of silica gel G60 was prepared. A solution of the polar lipid/silica gel (1:1, w/w) was carefully placed on the silica gel column. For first elution we used chloroform to elute the remaining neutral lipids. Ten ml eluent was employed for each 10 mg of sample. Second eluent was acetone/methanol (9:1, v/v) to elute glycolipids and ceramides (with ratio: 5 ml eluent for 10 mg of sample) and followed by methanol to elute PLs. The coconut lipids was analyzed using FTIR, GCMS, and thin layer chromatography.

RESULTS AND DISCUSSIONS

Extraction of lipids from dried coconut meat resulted in crude lipids extract, 49.03% of dried coconut meat. Furthermore the crude lipids extract contained 0.22% of neutral lipids, 0.04% of glycolipids and ceramides, and 0.04% of PLs.

The FTIR spectra of coconut crude lipids extract, neutral lipids, glycolipids and ceramides and PLs are presented in Figure 1. The major absorption peaks of lipids are evidently detectible in the spectrum. The C-H stretching vibrations can be identified by peaks at 2924 cm^{-1} and 2854 cm^{-1} , C=O stretching of esters at 1744 to 1728 cm^{-1} , CH_2 bending at 1458 cm^{-1} , CH_3 symmetric bending at 1373 cm^{-1} , C-O-C stretching in esters at 1065 - 1250 cm^{-1} , and CH_2 rocking at 718 and 725 cm^{-1} . Furthermore we find on the crude lipids extract and the PLs spectra several absorption peaks related to phosphate and choline groups. The phosphate group is recognized by PO_2^- asymmetric stretching peaks at 1226 cm^{-1} and 1219 cm^{-1} , C-O-P stretching at 1111 cm^{-1} and 1064 cm^{-1} and P-O asymmetric stretching at 887 cm^{-1} .

¹ and 817 cm⁻¹. The choline group is recognised by (CH₃)₃N⁺ asymmetric bending at 1458 cm⁻¹ and (CH₃)₃N⁺ asymmetric stretching at 972 cm⁻¹ and 972 cm⁻¹. Some of these peaks are slightly shifted from the characteristic band of lipids reported by Wolkers (28), Dean *et al.* (29) and Forfang *et al.* (30) it is assumed due to the interfering interaction between neighboring lipids such as acyl chain conformation and formation of hydrogen bonding.

The acyl chains (lipophilic part) of coconut crude lipid extract, neutral lipids, glycolipids and ceramides, and coconut PLs were analyzed by GCMS and presented on table 1. Data on table 1 shows that the composition of acyl chains found in coconut lipids are varied and range from C6 to C18:2. Crude lipids extract which is directly isolated from coconut meat by extraction with chloroform has the most varied acyl chain length and unsaturation compare to others. The variation decrease as the isolation proceed from crude lipids to PLs. Crude lipids extract has 9 types of acyl chains and 2 of them are unsaturated while the PLs has only 3 types and all of them are saturated i.e. C12:0, C16:0 and C18:0. The dodecanoic acid, C12:0, is the most abundant component in all of them. The dodecanoic acid composition in crude lipid extract, neutral lipids, glycolipids and ceramides, and coconut PLs are 45.85%, 48.13%, 68.15%, and 90.45% respectively. The data confirm that dodecanoic acid is the primary acyl chain in all type of lipids in coconut.

Table 1. Acyl chain components in coconut lipids

Acyl chains	Area (%)			
	Crude lipids	Neutral lipids	Glycolipids & ceramides	PLs
C6:0 (hexanoic acid)	0,45	-	-	-
C8:0 (octanoic acid)	6,70	17,92	3,93	-
C10:0 (decanoic acid)	6,10	5,45	0,81	-
C12:0 (dodecanoic acid)	45,85	48,13	68,15	90,45
C14:0 (tetradecanoic acid)	19,61	2,13	-	-
C16:0 (hexadecanoic acid)	10,32	1,16	0,61	0,37
C18:0 (octadecanoic acid)	3,34	10,79	0,91	1,58
C18:1 (9-Octadecenoic acid)	6,34	2,01	-	-
C18:2 (9,12-octadecadienoic acid)	1,29	-	-	-

TLC analysis on coconut PLs using chloroform/methanol 9:1 as eluent we obtain four spots at R_f values 0.115; 0.385; 0.654; and 0.808 respectively, see figure 2. Dragendorff reagent reveals orange spots for R_f value 0.115 and R_f 0.808 which indicates the present of

PLs containing choline groups ($-\text{CH}_2\text{CH}_2\text{N}^+(\text{CH}_3)_3$), figure 3a. The spot at R_f value 0.385 turn to purplish red stain when ninhydrin reagent are used as spotting agent, figure 3b, which indicates the present of PLs containing free amino groups i.e. serine group ($-\text{CH}_2\text{CH}_2\text{N}^+\text{H}_3(\text{COOH})$) or ethanolamine ($-\text{CH}_2\text{CH}_2\text{N}^+\text{H}_3$) groups. These results suggest that coconut PLs contain lipid from phosphatidylcholine (PC), phosphatidylserine (PS), and/or phosphatidylethanolamine (PE) classes.

CONCLUSIONS

Dried coconut meat contain 0.04% phospholipids, 0.22% of neutral lipids, and 0.04% of glycolipids and ceramides. Coconut PLs comprise of phosphatidylcholine (PC), phosphatidylserine (PS), and/or phosphatidylethanolamine (PE) classes with dodecanoic acid (C12: 0), octadecanoic acid (C18: 0) and hexadecanoic acid (C16:0) as the lipophilic parts.

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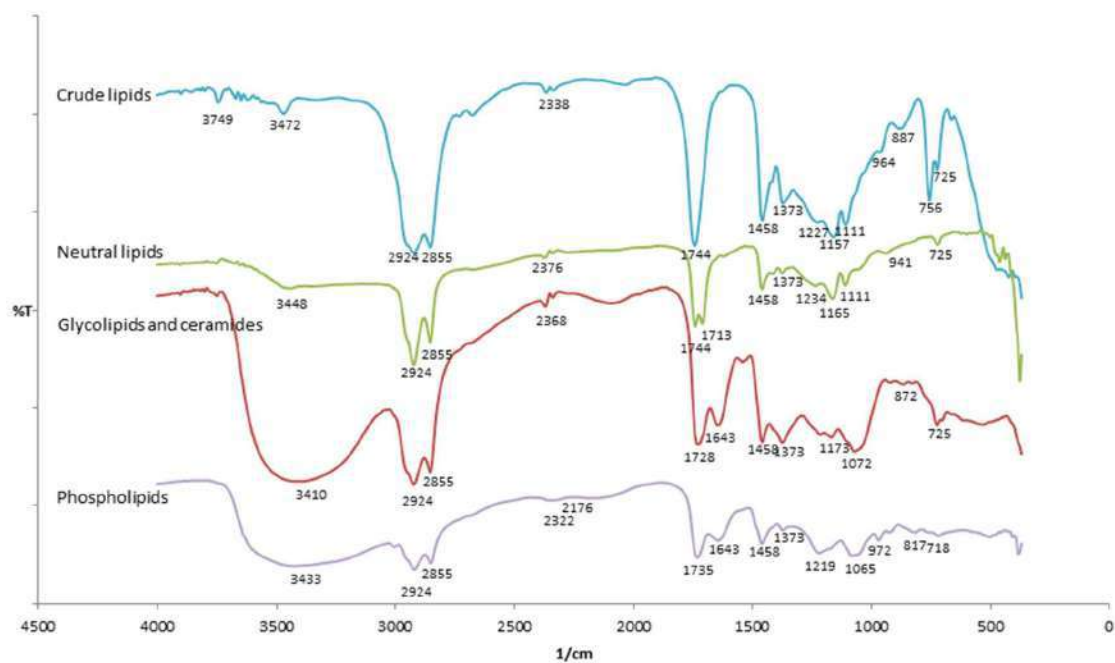


Fig 1. FTIR spectra of coconut lipids

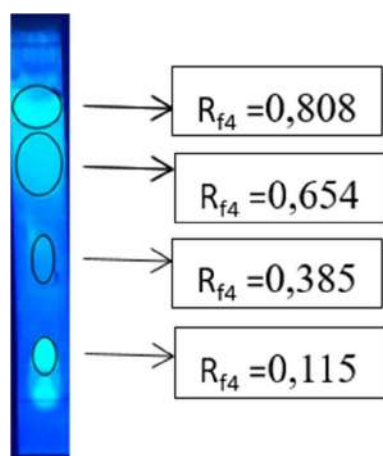


Fig. 2 TLC profile of coconut PLs under UV lamp $\lambda=365$ nm



Fig 3. TLC Profile of coconut PLs (a). Dragendorff stain (b). Ninhydrin stain



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COCONUT (*COCOS NUCIFERA L.*) LIPIDS: EXTRACTION AND CHARACTERIZATION

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ABSTRACT

Lipids extraction from dried coconut meat is conducted by maceration using chloroform/methanol (2:1,v/v) and partition extraction with n-hexane/87% ethanol (3:1), and column chromatography. Lipids are analyzed using FTIR, GC-MS, and TLC. Results reveal that dried coconut meat has 0.04% phospholipids, 0.22% of neutral lipids, and 0.04% of glycolipids and ceramides. Coconut phospholipids consist of phosphatidylcholine (PC), phosphatidylserine (PS), and/or phosphatidylethanolamine (PE) classes for the hydrophilic parts with dodecanoic acid (C12: 0), hexadecanoic acid (C16:0) and octadecanoic acid (C18: 0) as the lipophilic parts.

Keywords: Coconut, Phospholipids, Dodecanoic acid, Hexadecanoic acid, Octadecanoic acid.

INTRODUCTION

Phospholipids (PLs) are amphiphilic lipids obtained in cell membranes of all organisms, organized as lipid bilayers. The PLs discovered are mainly glycerophospholipids (GPLs), which are made of fatty acid residues (FAs) attached to a glycerol backbone by ester formation, a phosphate group and a hydrophilic head group (e.g. choline, serine or ethanolamine). Widespread supplies of industrially cultivated PLs are eggs (chicken and fish), bovine milk, soya, rapeseed, sunflower, coconut, sesame¹ and jack bean². Each source has

a unique composition of distinct phospholipid species and accordingly diverse applications in pharmaceuticals³, cosmetics⁴, nutrition, food⁵, and drug delivery^{6,7}.

In aqueous media, PLs molecules form aggregate structures by self-assembly course of action^{8,9}. Spherical bilayers known as vesicles or liposomes are among of these structures^{10,11}. Nowadays liposomes^{7,12-14} and related structures^{15,16} have been actively developed as carriers of bioactive compounds^{13,14,17-19} in various applications^{13,20} since their excellent biocompatibility to natural body environment^{6,12,21}.



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The characteristic of liposomes is greatly influenced by the physicochemical properties of PLs constituent, such as their molecular species²²⁻²⁵. PLs molecular species are determined by their head group (hydrophilic) and hydrocarbon chain types (hydrophobic). These components are diversified base upon their sources³. PLs acquired from various natural sources mostly have unique head groups and acyl chains compositions. A judicious stage in lipidomic study is lipid extraction with an suitable organic solvent combination (solvent system) preceding to MS analysis. The accomplishment of the lipid extraction by a certain solvent modification depends upon the partitioning of the lipids into the organic phase and therefore on the lipid composition^{2,26,27}.

Previously Hudiyanti *et al.*,²⁵ obtained that crude coconut PLs contain sephaline groups whilst the lipophilic parts comprise of dodecanoic acid (C12:0) and octanoic acid (C8:0). In this study we take further steps by purifying these crude PLs with column vacuum chromatography using several eluents to learn more information about the chemistry of coconut PLs. Our new finding is that crude coconut PLs also contained phosphatidylcholine groups and the lipophilic components were hexadecanoic acid (C16:0) and 9-octadecenoic acid (C18:1) as well. Further details of this new finding is presented in the following discussions.

MATERIALS AND METHODS

Materials

Dried coconut meat, filter paper, silica gel G60 for column chromatography, chloroform, acetone, methanol, aquabidest, NaCl, the solvent for partition was prepared by partitioning ethanol 87% with n-hexane (1: 1, v/v) and the top layer was mark solvent A while the bottom layer was solvent B, TLC plat of silica gel GF254 was washed with chloroform/methanol (1:1 v/v), then impregnate with 2.3% boric acid in absolute ethanol prior used. Spot regents for TLC were dragendorff, ninhidrin, and primulin reagents.

Extraction

Polar lipids from coconut meat was isolated by method previously presented by Hudiyanti *et al.*, (25). Briefly, this was prepared by the following

procedure. Dried coconut meat powder was macerated with chloroform / methanol (2:1) for 7 days. The filtrate was washed with a 0.9% of NaCl solution. Chloroform layer was evaporated to acquired coconut lipid extract. After that 10 g of coconut lipid extract was dissolved in 45 ml of solvent A and added to 15 ml solvent B in a separating funnel I. The mixture was shaken for 2 minutes. The bottom layer was transferred into a second separating funnel containing 45 ml solvent A and shaken for 2 minutes. The bottom layer containing polar lipids was collected in a vial for further purification while the upper layer was also kept in a separated vial. Fresh 15 ml solvent B was added to the remaining mixture in the first separating funnel and shaken for about 2 minutes. Then the bottom layer is transferred into the second separating funnel containing of fresh 45 ml solvent A. The mixture then shaken and separated as above. The bottom layer and the upper layer were accumulated into the previous separate containers. The procedure was repeated 4-6 times for each 10 g of coconut lipid extract. The polar lipids containing PLs were purified by column vaccum chromatography as described below.

Purification

A column containing 50 g of silica gel G60 was prepared. A solution of the polar lipid/silica gel (1:1, w/w) was carefully placed on the silica gel column. For first elution we used chloroform to elute the remaining neutral lipids. Ten ml eluent was employed for each 10 mg of sample. Second eluent was acetone/methanol (9:1, v/v) to elute glycolipids and ceramides (with ratio: 5 ml eluent for 10 mg of sample) and followed by methanol to elute PLs. The coconut lipids was analyzed using FTIR, GCMS, and thin layer chromatography.

RESULTS AND DISCUSSIONS

Extraction of lipids from dried coconut meat resulted in crude lipids extract, 49.03% of dried coconut meat. Furthermore the crude lipids extract contained 0.22% of neutral lipids, 0.04% of glycolipids and ceramides, and 0.04% of PLs.

The FTIR spectra of coconut crude lipids extract, neutral lipids, glycolipids and ceramides and PLs are presented in Fig. 1. The major absorption

peaks of lipids are evidently detectable in the spectrum. The C-H stretching vibrations can be identified by peaks at 2924 cm^{-1} and 2854 cm^{-1} , C=O stretching of esters at 1744 to 1728 cm^{-1} , CH_2 bending at 1458 cm^{-1} , CH_3 symmetric bending at 1373 cm^{-1} , C-O-C stretching in esters at 1065-1250 cm^{-1} , and CH_2 rocking at 718 and 725 cm^{-1} . Furthermore we find on the crude lipids extract and the PLs spectra several absorption peaks related to phosphate and choline groups. The phosphate group is recognized by PO_2^- asymmetric stretching

peaks at 1226 cm^{-1} and 1219 cm^{-1} , C-O-P stretching at 1111 cm^{-1} and 1064 cm^{-1} and P-O asymmetric stretching at 887 cm^{-1} and 817 cm^{-1} . The choline group is recognised by $(\text{CH}_3)_3\text{N}^+$ asymmetric bending at 1458 cm^{-1} and $(\text{CH}_3)_3\text{N}^+$ asymmetric stretching at 972 cm^{-1} and 972 cm^{-1} . Some of these peaks are slightly shifted from the characteristic band of lipids reported by Wolkers (28), Dean *et al.*, (29) and Forfang *et al.*, (30) it is assumed due to the interfering interaction between neighboring lipids such as acyl chain conformation and formation of hydrogen bonding.

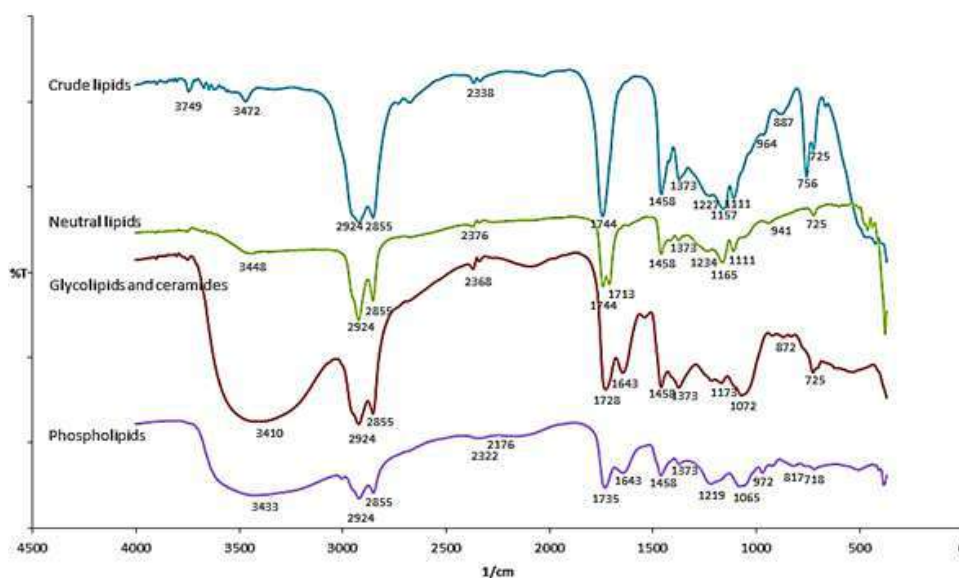


Fig. 1. FTIR spectra of coconut lipids

The acyl chains (lipophilic part) of coconut crude lipid extract, neutral lipids, glycolipids and ceramides, and coconut PLs were analyzed by GCMS and presented on Table 1. Data on Table 1 shows that the composition of acyl chains found in coconut lipids are varied and range from C6 to C18:2. Crude lipids extract which is directly isolated from coconut meat by extraction with chloroform has the most varied acyl chain length and unsaturation compare to others. The variation decrease as the isolation proceed from crude lipids to PLs. Crude lipids extract has 9 types of acyl chains and 2 of them are unsaturated while the PLs has only 3 types and all of them are saturated i.e. C12:0, C16:0 and C18:0. The dodecanoic acid, C12:0, is the most abundant component in all of them. The dodecanoic acid composition in crude

lipid extract, neutral lipids, glycolipids and ceramides, and coconut PLs are 45.85%, 48.13%, 68.15%, and 90.45% respectively. The data confirm that dodecanoic acid is the primary acyl chain in all type of lipids in coconut.

TLC analysis on coconut PLs using chloroform/methanol 9:1 as eluent we obtain four spots at R_f values 0.115; 0.385; 0.654; and 0.808 respectively, see Fig. 2. Dragendorff reagent reveals orange spots for R_f value 0.115 and R_f 0.808 which indicates the present of PLs containing choline groups ($-\text{CH}_2\text{CH}_2\text{N}^+(\text{CH}_3)_3$), Fig. 3a. The spot at R_f value 0.385 turn to purplish red stain when ninhydrin reagent are used as spotting agent, Fig. 3b, which indicates the present of PLs containing free amino groups i.e. serine group ($-\text{CH}_2\text{CH}_2\text{N}^+\text{H}_3(\text{COOH})$) or ethanolamine

($-\text{CH}_2\text{CH}_2\text{N}^+\text{H}_3$) groups. These results suggest phosphatidylcholine (PC), phosphatidylserine (PS), that coconut PLs contain lipid from and/or phosphatidylethanolamine (PE) classes.

Table 1: Acyl chain components in coconut lipids

Acyl chains	Area (%)			
	Crude lipids	Neutral lipids	Glycolipids & ceramides	PLs
C6:0 (hexanoic acid)	0,45	-	-	-
C8:0 (octanoic acid)	6,70	17,92	3,93	-
C10:0 (decanoic acid)	6,10	5,45	0,81	-
C12:0 (dodecanoic acid)	45,85	48,13	68,15	90,45
C14:0 (tetradecanoic acid)	19,61	2,13	-	-
C16:0 (hexadecanoic acid)	10,32	1,16	0,61	0,37
C18:0 (octadecanoic acid)	3,34	10,79	0,91	1,58
C18:1 (9-Octadecenoic acid)	6,34	2,01	-	-
C18:2 (9,12-octadecadienoic acid)	1,29	-	-	-

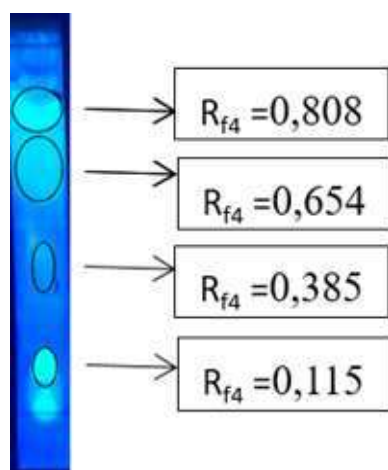


Fig. 2. TLC profile of coconut PLs under UV lamp $\lambda=365$ nm

CONCLUSION

Dried coconut meat contain 0.04% phospholipids, 0.22% of neutral lipids, and 0.04% of glycolipids and ceramides. Coconut PLs comprise of phosphatidylcholine (PC), phosphatidylserine (PS), and/or phosphatidylethanolamine (PE)



Fig. 3. TLC Profile of coconut PLs (a). Dragendorff stain (b). Ninhydrin stain

classes with dodecanoic acid (C12: 0), octadecanoic acid (C18: 0) and hexadecanoic acid (C16:0) as the lipophilic parts.

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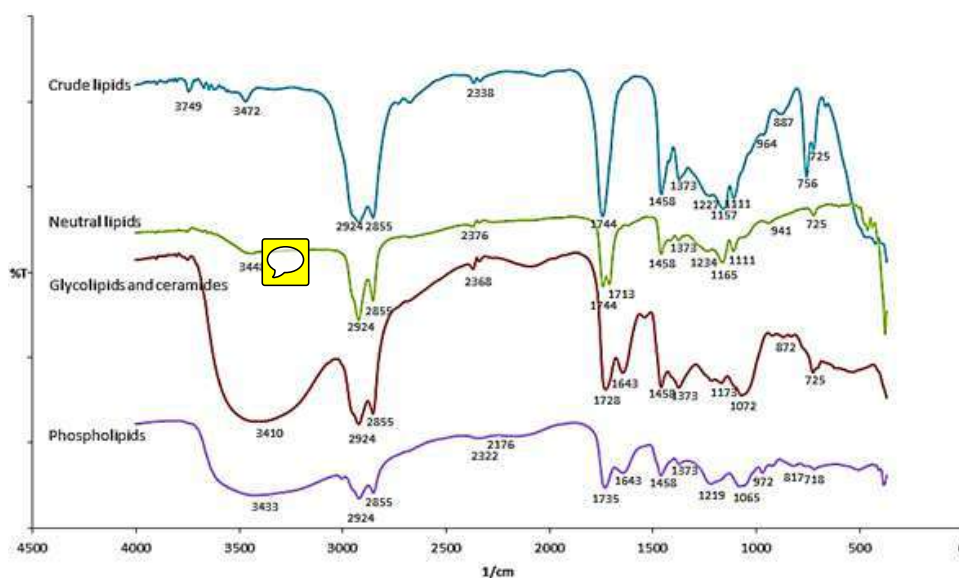


Fig. 1. FTIR spectra of coconut lipids

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($-\text{CH}_2\text{CH}_2\text{N}^+\text{H}_3$) groups. These results suggest that coconut PLs contain lipid from phosphatidylcholine (PC), phosphatidylserine (PS), and/or phosphatidylethanolamine (PE) classes.

Table 1: Acyl chain components in coconut lipids

Acyl chains	Area (%)	Neutral lipids	Glycolipids & ceramides	PLs
	Crude lipids			
C6:0 (hexanoic acid)	0,45	-	-	-
C8:0 (octanoic acid)	6,70	17,92	3,93	-
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C12:0 (dodecanoic acid)	45,85	48,13	68,15	90,45
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C16:0 (hexadecanoic acid)	10,32	1,16	0,61	0,37
C18:0 (octadecanoic acid)	3,34	10,79	0,91	1,58
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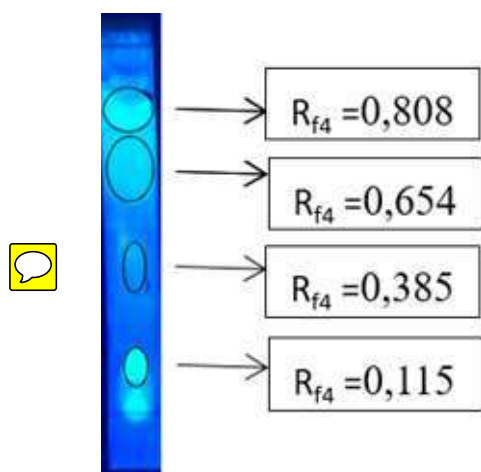


Fig. 2. TLC profile of coconut PLs under UV lamp $\lambda=365$ nm

CONCLUSION

Dried coconut meat contain 0.04% phospholipids, 0.22% of neutral lipids, and 0.04% of glycolipids and ceramides. Coconut PLs comprise of phosphatidylcholine (PC), phosphatidylserine (PS), and/or phosphatidylethanolamine (PE)



Fig. 3. TLC Profile of coconut PLs (a). Dragendorff stain (b). Ninhydrin stain

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