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Fatty Acid Profiles in the Kernel Oils of *Artocarpus odoratissimus* and *Litsea garciae*

[1]Firus Musfirah Poli and [2]Zaini Bin Assim
[1]Faculty of Applied Sciences, Universiti Teknologi MARA Cawangan Sarawak Kampus Mukah,
KM 7.5 Jalan Oya, 96400 Mukah, Sarawak, Malaysia
firusmusfirah@gmail.com
[2]Department of Chemistry, Faculty of Resource Science and Technology,
Universiti Malaysia Sarawak, Jalan Datuk Mohammad Musa, 94300 Kota Samarahan,
Sarawak, Malaysia

Abstract: Kernels of *A. odoratissimus* and *L. garciae* fruits collected from different areas in Sarawak were extracted and the fatty acid profiles in their kernel oils were studied. Proximate analysis of the kernels such as moisture content, ash content, crude fat and crude fiber were determined. The kernels were extracted using Soxhlet extraction apparatus and the Fatty Acid Methyl Esters (FAME) composition in the kernel oils were analysed using Gas Chromatography-Mass Spectrometry (GC-MS). The number of FAME components found in *A. odoratissimus* kernel oil was higher in comparison to *L.garciae* kernel oil. The major compound in kernel oil of each fruit sample also differs from each other; C12:0 (lauric acid) (28.75±0.09-34.12±1.02%) was the main FAME in *L.garciae* while C18:2n6c (linoleic acid) (37.30±4.62-40.74±6.19%) was the dominant compound in *A. odoratissimus*. This study is significant as it has revealed that the kernel oils of these underexploited indigenous fruits have a great potential to be developed for the applications in nutrition, industrial and cosmetics.

Key words: *Artocarpus odoratissimus*, fatty acid, *Litsea garciae*, kernel oil, proximate analysis, crude fiber

INTRODUCTION

In recent years, there is growing interest in searching for newer sources of edible oils, for example, plant seeds which offer oils with high nutritional, industrial and pharmaceutical importance (Shah *et al*., 2004). Kernel oils generally have broad range of utilization as foods, lubricants, fuel for paraffin lamps, additives for paint formulations, soup ingredient and also in medicinal applications. Some kernel oils have the potential to be developed for oleochemical industries (Ahmad *et al*., 2007). The depletion of world petroleum reserves as a result of increasing energy demands together with environmental concerns has prompted the efforts to discover various alternative sources of petroleum-based fuels. In this context, biodiesel has gained substantial position in public, over the world (Ahmad *et al*., 2007; Shah *et al*., 2004).

Typical vegetable oil feedstock for biodiesel production includes the commodity seed oils, such as soybean, sunflower, rapeseed and canola. These oils are readily converted to the corresponding alkyl esters by base catalyst and offer acceptable fuel properties in modern diesel engines (Schwab *et al*., 1987; Van Gerpen, 2005). Nevertheless, the usage of conventionally grown edible oils leads to alleviate food versus fuel issue. Consequently, the process economics may be improved by exploration of newer and lower cost feedstock, not only due to the high oil costs but also due to their ever-increasing demand (Azam *et al*., 2005).

L. garciae is native to Borneo Island. The English name of *L. garciae* is bagnolo/wuru lilin and the common name differs according to the local language: engkala (Malay Sarawak), Madang enkala/pedar (Iban), Ta'ang (Bidayuh) and pengolaban (Sabah) (Johnny *et al*., 2011; Kueh *et al*., 2000). The fruit of *L. garciae* is round in shape with a unique green stem cap about 4.0- 6.0 cm in length (with the stem cap) and 3.0-6.0 cm in diameter. The thin skin turns to pinkish, bright pink or greenish white when it is ripe, depending on the variety. *A. odoratissimus* on the other hand is in the same genus with jackfruit and breadfruit but it is less popular because the species is native to Borneo and Philippines only (Jagtap and Bapat, 2010). The synonyms of *A. odoratissimus* are *Artocarpus tarap* Becc. and *Artocarpus mutabillis* Becc.. Some local people not only consume the pulp but they also consume the kernel. The kernels are boiled or roasted and are eaten as a snack as they have nutty taste.

Corresponding Author: Firus Musfirah Poli, Faculty of Applied Sciences,
Universiti Teknologi MARA Cawangan Sarawak Kampus Mukah, KM 7.5 Jalan Oya, 96400 Mukah,
Sarawak, Malaysia, firusmusfirah@gmail.com

The objective of this study is to analyse fatty acid compositions in the kernel oils of *L. garciae* and *A. odoratissimus* fruits using Gas Chromatography-Mass Spectrometry (GC-MS). Proximate analysis including moisture content, ash content, crude fat and crude fiber of the fruit kernels were examined according to respective method.

Significance of study: *Artocarpus odoratissimus* and *Litsea garciae* are tropical plants grow within the rainforest in Sarawak and they produce edible fruits that are widely consumed by the local community but the kernels are largely discarded. The information on proximate analysis of the fruit kernels and the fatty acid profiles in the kernel oils are expected to be valuable for future reference. Studies on these kernels are essential in order to discover new sources, whether for human consumption or for industrial purposes.

MATERIALS AND METHODS

Chemicals and solvents: A mixture of FAMEs standard consisting 37 compounds was purchased from Supelco (Pennsylvania, USA). Dichloromethane, sulphuric acid and methanol were from J.T. Baker (Illinois, USA). Petroleum ether was purchased from BDH chemicals (Pennsylvania, USA). Potassium iodide (KI), potassium iodate (KIO_3), sodium hydroxide (NaOH) pellets, Butylated Hydroxytoluene (BHT) and ascorbic acid were purchased from Sigma-Aldrich (Missouri, USA). Hydrochloric acid (HCl) and boric acid were from HmbG Chemicals (Hamburg, Germany). All chemicals were used as received without further purification. All chemicals were used as received without further purification.

Sample collection: The fruit samples were collected from different divisions in Sarawak. The kernels were separated from their pulps, washed with water and allowed to dry before they were measured for moisture and ash contents. Remaining samples were freeze-dried, grounded, kept in air tight container and stored in the freezer prior to analysis.

Proximate analysis: Proximate analysis including moisture content, ash content, crude fat and crude fiber were performed according to standard methods. Moisture content was determined using the direct drying method according to AOAC method 945.32 (Horwitz, 2000). Ash content was determined using the dry ashing method according to AOAC method 930.05 (Horwitz, 2000). Crude fat was determined using Soxhlet extraction according to Tee *et al.* (1996). Determination of crude fiber was carried out according to AOAC method 978.10 (Horwitz, 2000).

Sample extraction: Samples were extracted using Soxhlet extraction apparatus according to the procedure described by Tee *et al.* (1996). Approximately 5 g of dried grounded sample was weighed and placed into an extraction thimble with glass wool covered on top. A total of 250 mL of petroleum ether was filled in a cleaned and dry round bottom flask and 0.05% of BHT was added to prevent oxidation. The flask was placed on the heating mantle and then heated for 8 h. The remaining solvent in the round bottom flask was then evaporated using the rotary evaporator until it was completely dry. The oil extract was diluted with dichloromethane in an air tight vial and kept in the freezer prior to GC-MS analysis.

Fatty acid methylation: In order to enable analysis of fatty acids in the oil extracts with GC-MS, the extracts need to be derivatized into FAMEs. Derivatization of fatty acids was carried out according to the procedure obtained by David *et al.* (2002). Exactly 200 μL of extracted oil was dissolved in 1 mL of hexane in an eppendorf tube. A total of 200 μL of 2 M NaOH in methanol was added into the tube, capped and vortexed for 10 sec. The mixture was then placed in a water bath at 50°C for 20 sec, cooled and vortexed again for 10 sec. The mixture was then centrifuged at 6,000 rpm for 20 sec to obtain a clear separation. These supernatant obtained was then analysed using GC-MS.

Gas chromatography analysis: GC analysis was performed on a Shimadzu gas chromatograph equipped with mass spectrometer model Shimadzu QP2010 PLUS Series. The GC separation was carried out using BPX-5 capillary column (29.5 m×0.25 mm×0.25 μm film thickness). The operating conditions for the gas chromatography were as follows: the column temperature was initially set at 90°C for 5 min and then was increased to 300°C at the rate of 3.5°C/min and held for 10 min. The injection temperature was kept at 260°C with a split ratio of 1:20. Helium gas was used as the carrier gas at a flow rate of 1.0 mL/min. Exactly1 μL of 1,000 μg/mL mixture of FAMEs standard was injected. The identifications of fatty acids in the samples prepared at 1,000 μg/mLwere made by comparison of the retention times with those of the standard FAMEs mixture.

RESULTS AND DISCUSSION

Table 1 shows the proximate compositions in the kernel of *L. garciae* and *A. odoratissimus* fruits that were collected from different districts in Sarawak. The differences in the values between different districts might be due to the environmental factor and growth condition (Demir and Ozcan, 2001). Among the two species, kernel of *L. garciae* collected from Sibu had the highest amount

Table 1: Proximate compositions of kernels from *L. garciae* and *A. odoratissimus* fruits

Variables	Moisture (%)	Ash (%)	Crude fat (%)	Crude fiber (%)
L. garciae (KCH)[a]	41.88±0.37	1.87±0.03	34.29±0.28	17.30±0.10
L. garciae (KSM)[b]	38.08±3.12	1.40±0.10	33.13±1.46	19.64±0.15
L. garciae (SBU)[c]	39.50±0.99	1.32±0.19	37.31±0.53	18.67±0.04
L. garciae (MKH)[d]	43.38±0.14	1.75±0.27	32.09±0.55	15.00±0.09
A. odoratissimus (KCH)[a]	49.32±1.80	1.43±0.07	14.00±0.50	6.95±0.68
A. odoratissimus (KSM)[b]	41.36±0.89	1.13±0.09	14.76±1.02	12.50±1.55
A. odoratissimus (SBU)[c]	45.99±1.60	1.26±0.05	13.22±0.46	11.05±1.57
A. odoratissimus (MRI)[e]	42.29±0.16	1.28±0.03	13.55±0.57	9.20±0.62

[a]KCH, Kuching; [b]KSM, Kota Samarahan; [c]SBU, Sibu; [d]MKH, Mukah; [e]MRI, Miri

Table 2: Fatty acid composition (Percent) in *L. garciae* and *A. odoratissimus* kernel oils

Variables	*L. garciae* (KCH)	*L. garciae* (KSM)	*L. garciae* (SBU)	*L. garciae* (MKH)	*A. odoratissimus* (KCH)	*A. odoratissimus* (KSM)	*A. odoratissimus* (SBU)	*A. odoratissimus* (MRI)
C10:0	4.74	4.52	8.89	6.13	-	-	-	-
C11:0	0.62	0.64	0.66	0.64	-	-	-	-
C12:0	44.79	40.37	38.64	39.10	-	-	-	-
C13:0	0.60	0.62	0.43	0.65	-	-	-	-
C14:0	20.11	18.99	19.59	20.05	-	-	-	-
C16:0	7.02	7.28	6.56	8.07	14.66	14.63	15.74	15.10
C17:0	-	-	-	-	0.07	0.02	0.02	0.07
C18:0	1.17	4.14	1.88	0.86	0.94	0.57	0.19	0.72
C20:0	-	-	-	-	1.48	0.79	2.14	1.38
C22:0	-	-	-	-	7.96	6.18	11.69	5.96
C24:0	-	-	-	-	12.44	12.11	12.46	13.26
Total SFA[a]	**79.05**	**76.56**	**76.22**	**75.50**	**37.55**	**34.30**	**42.24**	**36.49**
C16:1	0.65	0.61	-	0.62	0.02	-	-	0.02
C18:1n9c	13.17	14.99	16.68	15.09	17.94	20.88	11.79	16.82
C20:1	0.62	0.60	0.63	0.63	3.49	2.57	2.75	2.92
C24:1	-	-	-	-	0.91	0.88	0.34	1.12
Total MUFA[b]	**14.44**	**16.20**	**17.31**	**16.34**	**22.36**	**24.33**	**14.88**	**20.88**
C18:2n6c	6.51	7.25	6.49	8.16	37.36	37.30	40.74	40.65
C22:6n3	-	-	-	-	2.72	4.07	2.13	1.98
Total PUFA[c]	**6.51**	**7.25**	**6.49**	**8.16**	**40.08**	**41.37**	**42.87**	**42.63**

[a]SFA, Saturated Fatty Acid; [b]MUFA, Monounsaturated Fatty Acid; [c]PUFA, Polyunsaturated Fatty Acid

of fat which is 37.31%. The highest fiber content was also observed in the kernel of *L. garciae* ranging from 15.00-19.64%.

Table 2 shows the fatty acid percent composition in *L. garciae* and *A. odoratissimus* kernel oils. Kernel oil of *A. odoratissimus* consists higher number of fatty acids compare to *L. garciae* where 13 fatty acids have been successfully identified. The kernel oils of *A. odoratissimus* are rich in unsaturated fatty acids with the total of MUFA and PUFA accounted more than 60% of the total fatty acids. The predominant component of *A. odoratissimus* kernel oils is linoleic acid with the highest percentage of abundance was observed in Sibu sample (40.74±6.19%), followed by Miri (40.65±3.63%), Kuching (37.36±1.39%) and Kota Samarahan (37.30± 4.62%). The significance amount of PUFA in kernel oils (40.08–42.87%) of *A. odoratissimus* are comparable to those in the seed oil of *A. camansi* which was reported at 30.81% by Adeleke and Abiodun (2010). An unusual long chain saturated fatty acid-lignoceric acid (C24:0) has been identified in *A. odoratissimus* kernel oils which accounted for 12.11-13.26% of the total fatty acids. The presence of lignoceric acid has been reported in the seed oils of *Washingtonia filifera* (Nehdi, 2011), *Cucurbita pepo* (Schinas *et al*., 2009) and *Phoenix canariensis* (Nehdi *et al*., 2010) with the percentage of abundance of 0.094, 0.06 and 0.02%, respectively.

In contrast, *L. garciae* kernel oils are rich in SFA with the highest abundance detected in Kuching sample (79.05±0.53%), followed by Kota Samarahan (76.56±0.21%), Sibu (76.22±0.07%) and Mukah (75.50±0.25%). The amount of SFA in kernel oils of *L. garciae* is comparable with the SFA in palm kernel oil (82.1%) as reported by Edem (2002). The SFA identified in *L. garciae* kernel oils are represented mostly by lauric acid and myristic acid with total accounted more than 50%. These acids, particularly lauric acid, have the preventive effect on prostatic hyperplasia development due to their 5α-reductase inhibitory activity (Babu *et al*., 2010; Raynaud *et al*., 2002). The amount of lauric acid in the oils is comparable to lauric acid in virgin coconut oil (45.51%) and coconut oil (43.55%) as reported by Nevin and Rajamohan (2008).

CONCLUSION

This study revealed various composition of fatty acid in the kernel oils for different fruit samples. *L. garciae* kernel oils are rich in SFA while *A. odoratissimus* contain more unsaturated fatty acids than SFA. The fatty acid value of these kernel oils could be justified for cosmetic

and industrial applications. It can also provide a new source and low-cost feedstock which will increase the value of agricultural products. In addition, the high level of SFA and PUFA in kernel oils of *L. garciae* and *A. odoratissimus* respectively, make them desirable in terms of nutrition. Kernel oil of *A. odoratissimus* might be a suitable substitute for PUFA-rich oils such as sunflower and soybean oil while kernel oil of *L. garciae* might be a suitable substitute for virgin coconut oil, coconut oil and palm kernel oil.

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