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Effect of Varietal Differences on Composition and Thermal Characteristics of Avocado Oil

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Abstract A study was carried out to compare the characteristics of oils from three Malaysian avocado (Persea americana) cultivars with those of the oil from the Australian Hass avocado variety. Oil samples extracted from matured-avocado fruits were assessed for basic physico-chemical parameters, fatty acid and triacylglycerol (TAG) compositions, and melting and solidification characteristics. In comparison to Hass variety, the oil contents of the local avocado cultivars were significantly lower and found to be mostly in semisolid form. As a common feature, oils of both local cultivars and Hass variety are found to have oleic acid as the most dominant fatty acid. However, there are differences between them with regard to the proportional distributions of palmitic and linoleic acids. While the major TAG of local avocado cultivars were POO, followed by POL, OOO and PPO, the dominant TAG of Hass variety were OOO, followed by PPO, OOL and POL. Due to these differences in fatty acid and TAG distributional patterns, the oils of local avocado cultivars are found to possess iodine value, slip melting point, melting and solidification characteristics, which are completely different from those of the imported Hass avocado variety.

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Malaysian Agricultural Research and Development Institute, P.O. Box 12301, 50774 Kuala Lumpur, Malaysia **Keywords** Avocado oil · DSC · Hass avocado · Lard alternatives · Thermal behavior · Vegetable oils

Introduction

Avocado (Persea americana) is a tropical fruit bearing tree distributed in many parts of the world. Belonging to the family Lauraceae, the tree is believed to have been originated from Mexico. Throughout the world avocado exists in many varietal forms, of which Hass avocado has gained the popularity as the most promising commercial variety due to high yield of rich fruits with excellent outer covering [1]. The stone like exocarp of Hass avocado is an important attribute that could preserve the fruit from insect attack or damage during storage and transport. Apart from its use as an edible fruit, avocado is also a rich source of lipids. Depending on the variety and the growth conditions, the mesocarp of the avocado fruit is found to possess 8-30% oil [2, 3]. The oil of avocado fruit is highly regarded for uses in food, cosmetics and health care products because of some unique, physical and functional properties [4].

Different forms of avocado are also distributed in many parts of Malaysia, although no systematic cultivation could be found yet. Although avocado oil from other parts of the world has been characterized [5–9], past studies on the evaluation of locally available avocado cultivars with respect to their oil composition and thermal characteristics are limited. Information of this kind would be vital not only for product development activities but also for those who are engaged in breeding fruit crops [10]. Hence, the objective of this study is to compare the physico-chemical properties of oil from local avocado cultivars with the imported Hass variety.

Materials and Methods

Materials

For this study, two of avocado cultivars, Avo-1 and Avo-2 having bottled shape were collected from two locations in West Malaysia. A third cultivar Avo-3, which has a round shape was collected from a location in East Malaysia. Imported Hass fruits—Australian Hass were purchased from a local supermarket. Avocado fruits of each variety were cut open, and minced manually into fine pieces after removing the seeds. The minced pieces of the mesocarp were dried in a tray type dryer (Memmert Model UFB 400, GmbH + Co. KG, Germany) for 24 h at 60 °C and subsequently ground into powder using a Warring blender (model 32BL80, Dynamic Corporation of America, New Hartford, CT). All the chemicals used in this study were of analytical grade, unless otherwise specified.

Oil Extraction

Oil extraction from finely ground samples of dried avocado fruits was carried out by the Soxhlet extraction method using petroleum ether (40–60 °C) [11]. The extracted oils were kept in an oven at 60 °C for 1 h to expel solvent before storing at -20 °C. Prior to analysis, the oil samples were removed from frozen storage, and left static at room temperature for 1 h and then warmed at 60 °C until they became completely molten.

Determination of Slip Melting Point (SMP) and Iodine Value (IV)

SMP and IV of the oil samples were determined according to AOCS method Cc.3.25, and AOCS method Cd Id-92, respectively [12].

Color Determination

The liquid oil sample was placed in a 1-in (2.54 cm) cell and its color was determined using a Lovibond Model E tintometer (Lovibond, Salisbury, UK) at 30 °C by achieving the best possible match with the standard color slides of yellow (Y), red (R) and blue (B) and indices [13].

GLC Analysis of Fatty Acid Methyl Esters (FAME)

FAME were prepared by dissolving 50 mg portion of the oil in 0.8 ml of hexane and adding 0.2 ml portion of 1 M solution of sodium methoxide [14], then analyzed on a gas chromatograph (Agilent Technologies, Singapore) fitted with a FID detector. The polar capillary column RTX-5 (0.32 mm internal diameter, 30 m length and 0.25 µm film

thickness; Restex Corp., Bellefonte, PA) was used. The oven temperature was programmed as follows: initial temperature of 50 °C (for 1 min), and programmed to increase to 200 °C at 8 °C/min. Both injector and detector temperatures were maintained at 200 °C throughout the analysis. The carrier gas (helium) flow rate was 1.0 mL/ min and the split ratio was 58:1. The identification of the peaks of the samples was done with reference to a chromatographic profile containing FAME standards (Supelco, Bellefonte, PA). The percentage of fatty acid was calculated as the ratio of the partial area to the total peaks area [15].

HPLC Analysis of TAG Composition

The TAG composition was determined using a Waters Model 510 liquid chromatograph equipped with a differential refractometer Model 410 as a detector (Waters Associates, Milford, MA). The analysis of TAG was performed on a Merck LiChrospher RP-18 column (5 µm) $(12.5 \text{ cm} \times 4 \text{ mm i.d.}; \text{Merck, Darmstadt, Germany})$. The mobile phase was a mixture of acetone:acetonitrile (63.5:36.5) and the flow rate was 1.5 mL/min. The oven temperature was maintained at 30 °C. The injector volume was 10 µL of 5% (w/w) oil in chloroform. Each sample was chromatographed two times, and the data was recorded as area percentages [15]. The identification of the peaks of the samples was done in accordance with the TAG profiles of avocado oil reported previously by Jakab et al. [16] and a set of TAG standards supplied by Sigma-Aldrich Chemicals (Deisenhofen, Germany).

Thermal Analysis by DSC

Thermal analysis was carried out on a Mettler Toledo differential scanning calorimeter (DSC 823 Model) equipped with a thermal analysis data station (STARe software, Version 9.0x, Schwerzenbach, Switzerland). Nitrogen (99.99% purity) was used as the purge gas at a rate of ~20 mL/min. Approximately 4-8 mg of molten sample was placed in a standard DSC aluminum pan and then hermetically sealed. An empty, hermetically sealed DSC aluminum pan was used as the control. The oil/fat samples were subjected to the following temperature program: The sample was held at 70 °C isotherm for 1 min to eliminate the thermal history of the samples, then cooled at 5 °C/min to -90 °C and held for 1 min. The sample was then heated from -90 to 70 °C at the same rate [15].

Determination of Solid Fat Content (SFC) by NMR

SFC was measured using a Bruker Minispec (Model Mq 20) pulse Nuclear Magnetic Resonance (pNMR)

spectrometer (Karlsruhe, Germany), according to AOCS method Cd 16b-93 [12]. The sample in the NMR tube was melted at 70 °C for 15 min, followed by chilling at 0 °C for 60 min, and then held at each measuring temperature for 30 min prior to measurement to eliminate the thermal history of the samples. Melting, chilling and storing of the samples were carried out in pre-equilibrated thermostatted glycol containing baths, accurate to 0.1 °C. SFC measurements were taken at 5 °C intervals over the range of 0-60 °C.

Statistical Analysis

All analyses were carried out in duplicate and the results were expressed as the mean value \pm standard deviation. Data was statistically analyzed by one-way analysis of variance (ANOVA), by using Tukey's test of MINITAB (version 15) statistical package at 0.05 probability level.

Results and Discussion

Basic Physico-chemical Characteristics

The oil content, color, slip melting point and iodine value of the avocado oils extracted from the three local cultivars are compared with those of the Australian Hass variety as shown in Table 1. The oil contents of the three local cultivars are more or less equal [Avo-1 (30.25%), Avo-2 (31.85%) and Avo-3 (33.45%)], but significantly lower than (p < 0.05) that of the Hass variety (54.9%). For the purpose of oil recovery using commercial screw-press oil expellers, the oil contents displayed by the locally grown cultivars are substantial. Avocado oil possessing a dark green color is a common feature, which is said to be due to the presence of pigments like carotene, chlorophyll, etc. [2, 3]. Visual observation indicated that the colors of the oil samples of the local cultivars are little darker than that of the oil from Hass variety. This is in accordance with the measurements of individual color values of yellow, red, and blue obtained from Lovibond tintometer. As for the data presented in Table 1, the lowest values of individual colors are recorded for Hass variety. Among the local cultivars, the color of Avo-3 is lighter than that of Avo-1 and Avo-2 as its values for red and blue are lower in comparison to the other two cultivars.

The physical nature of the lipid is an important aspect with regard to its usage in various consumer products. The three local avocado cultivars are found to yield fats, which are semi solid at room temperature, while the oil of the imported Hass variety remains entirely as a liquid under the same condition. As shown in Table 1, Avo-1, Avo-2, and Avo-3 are found to have SMP at 31, 30, and 27.25 °C, respectively, of which Avo-3 display a value closely similar to that of lard [17]. In fact, SMP values of the local cultivars are found to be in a reverse order with their IVs. As shown in Table 1, IV of Avo-1, Avo-2, and Avo-3 are 82.97, 84.30, and 87.7, respectively and that of Hass variety is 95.41. While the IVs of the local cultivars are generally within the range of those of the avocado oils extracted from ordinary cultivars grown in other parts of Asia [2], the IV of the Hass variety used in this study is comparable to that of the Hass variety grown in the USA, and Japan [5].

Fatty Acid Distribution

The fatty acid distribution of oils from local avocado cultivars are compared with the imported Hass variety as shown in Table 2. In all three local cultivars, oleic is the major fatty acid (43.65-51.22%) followed by palmitic (26.41-30.37%) and linoleic (12.75-17.45%) acids. The proportional distribution of these fatty acids in the local avocado fruit cultivars is some what comparable to those of palm oil and lard as reported in many past studies [18, 19]. However, unlike lard, oils of avocado fruits are generally found to have extremely low (0.27-1.56%) amounts of stearic acid (Table 2). Generally, the Hass variety has the highest abundance of unsaturated fatty acids (84.94%). Although oleic (63.73%) is the most dominant fatty acid of Hass variety, its second most abundant fatty acid was linoleic (15.27%) acid, followed by palmitic (14.8%) acid. However, Hass variety from other parts of the world is

Table 1 Oil content, color, slip melting point (SMP) and iodine value (IV) of oils from different avocados cultivars

Sample	Oil content (%)	Color	SMP (°C)	IV (g I ₂ /100 g)
Avo-1	$30.25 \pm 0.64^{\circ}$	70.0Y + 9.8R + 9.9B	31.00 ± 0.71^{a}	82.97 ± 0.66^{a}
Avo-2	$31.85 \pm 0.49^{\rm b,c}$	70.0Y + 9.6R + 9.9B	30.00 ± 0.71^{a}	84.30 ± 0.14^{a}
Avo-3	$33.45 \pm 0.92^{\rm b}$	70.0Y + 6.5R + 6.9B	$27.25 \pm 0.35^{\rm b}$	87.72 ± 0.23^{b}
Hass	54.90 ± 0.14^{a}	45.0Y + 2.2R + 0.1B	ND	$95.41 \pm 0.15^{\circ}$

Each value in the table represents the mean of two replicates. Means within each column bearing different superscripts are significantly (p < 0.05) different

ND not determined

Fatty acid	Avo-1	Avo-2	Avo-3	Hass
C16:0	$27.63 \pm 0.04^{\circ}$	30.37 ± 0.06^d	26.41 ± 0.06^{b}	14.80 ± 0.03^{a}
C16:1	$4.40\pm0.08^{\rm a}$	$5.22 \pm 0.02^{\circ}$	7.44 ± 0.03^{d}	4.86 ± 0.02^{b}
C18:0	$1.56\pm0.04^{\rm d}$	$1.30 \pm 0.01^{\circ}$	1.03 ± 0.05^{b}	$0.27\pm0.02^{\rm a}$
C18:1	$51.22\pm0.01^{\rm b}$	$43.65 \pm 0.04^{\rm a}$	$51.18 \pm 0.76^{\rm b}$	$63.73 \pm 0.18^{\circ}$
C18:2	$13.82\pm0.03^{\rm b}$	$17.45 \pm 0.04^{\rm d}$	12.75 ± 0.45^{a}	$15.27 \pm 0.16^{\circ}$
C18:3	$1.38\pm0.01^{\rm a}$	2.03 ± 0.01^{b}	$1.20 \pm 0.18^{\mathrm{a}}$	$1.09 \pm 0.04^{\rm a}$
SFA	$29.19 \pm 0.06^{\circ}$	31.66 ± 0.07^{d}	27.44 ± 0.11^{b}	15.07 ± 0.01^{a}
USFA	70.81 ± 0.06^{b}	$68.34 \pm 0.07^{\mathrm{a}}$	$72.57 \pm 0.11^{\circ}$	84.94 ± 0.01^{d}

Table 2 Fatty acid compositions of oils from different avocados cultivars

Each value in the table represents the mean of two replicates. Means within each row bearing different superscripts are significantly (p < 0.05) different

SFA saturated fatty acid, USFA unsaturated fatty acid

Table 3 Triacylglycerol (TAG) compositions of oils from different avocados cultivars

TAG	Avo-1	Avo-2	Avo-3	Hass
LLLn	$1.19 \pm 0.01^{\circ}$	1.87 ± 0.00^{a}	0.90 ± 0.01^{d}	$1.26 \pm 0.00^{\rm b}$
LLL	$0.53 \pm 0.01^{\rm b}$	$0.85\pm0.07^{\rm a}$	$0.42\pm0.02^{\mathrm{b}}$	-
OLL	$2.32 \pm 0.06^{ m a,c}$	$3.23\pm0.02^{\rm a}$	$3.98 \pm 1.20^{\rm a}$	$5.04 \pm 0.01^{a,b}$
PLL	$2.62 \pm 0.01^{\rm b}$	4.21 ± 0.00^{a}	$2.38\pm0.01^{\rm c}$	$2.17\pm0.02^{\rm d}$
OOL	$9.75 \pm 0.02^{\rm b}$	$9.00 \pm 0.03^{\circ}$	7.71 ± 0.12^{d}	20.22 ± 0.11^{a}
POL	$17.99 \pm 0.06^{\rm b}$	$19.29 \pm 0.06^{\rm a}$	$11.05 \pm 0.10^{\rm d}$	$13.20 \pm 0.03^{\circ}$
PPL	$2.45 \pm 0.03^{\circ}$	4.03 ± 0.06^{b}	$9.61 \pm 0.14^{\rm a}$	-
000	$16.08 \pm 0.02^{\rm b}$	$11.42 \pm 0.01^{\circ}$	16.32 ± 0.18^{b}	$29.00\pm0.01^{\rm a}$
POO	27.41 ± 0.01^{a}	$22.76 \pm 0.02^{\circ}$	24.58 ± 0.05^{b}	$22.42\pm0.04^{\rm d}$
PPO	$10.86 \pm 0.01^{\rm b}$	12.43 ± 0.00^{a}	$10.08 \pm 0.01^{\circ}$	$2.80\pm0.01^{\rm d}$
OOS	$1.27 \pm 0.01^{\rm a}$	$0.52 \pm 0.01^{\circ}$	$0.75 \pm 0.02^{\rm b}$	0.41 ± 0.01^{d}
SPO	$0.75 \pm 0.01^{\rm a}$	$0.57 \pm 0.02^{\rm b}$	$0.41 \pm 0.01^{\circ}$	-
PPS	$0.18\pm0.04^{\rm a}$	0.11 ± 0.01^{a}	$0.12\pm0.01^{\mathrm{a}}$	-
Others	6.6 ± 0.05	9.71 ± 0.04	11.69 ± 1.10	3.42 ± 0.01

Each value in the table represents the mean of two replicates. Means within each row bearing different superscripts are significantly (p < 0.05) different

O oleic, P palmitic, L linoleic, Ln linoleic, S stearic

reported to have high oleic acid (45.9–54.5%), followed by palmitic (19.7–20.0%) and linoleic (12.5–5.7%) acids [5, 6]. This anomalous behavior in fatty acid composition could be attributed to the influence of climate and geographical variations.

TAG Composition

The TAG profile of the oils from local avocado cultivars are compared with that of the oil of Hass variety as shown in Table 3. It is clear that the TAG distributional profile of Hass variety is distinctly different from those of the local avocado cultivars. Hass variety has OOO (29%) as the most dominant TAG molecular species followed by POO (22.42%), and OOL (20.22%) while in all three local avocado cultivars (Avo-1, Avo-2 and Avo-3), POO (22.76–27.41%) was the most predominant TAG molecular species. Among the three local cultivars, Avo-1 has the highest content of POO, which is closely comparable to that of lard [17]. According to Jakab et al. [16], the major TAG molecular species of avocado oil from Hungary variety were OOO and POO. Unlike the three local avocado cultivars, trisaturated TAG molecular species such as PPS and SPO have not been detected in Hass variety, but the content of triunsaturated TAG (LLLn, LLL, OLL, OOL, OOO) in it (55.55%) is almost twice as high as those of the three local cultivars (22.76–27.41%). Although POO was the most predominant TAG molecular species of all three local avocado cultivars, there are significant differences in the distribution of other major TAG molecular

species. The second most abundant TAG molecular species of Avo-1, and Avo-2 was POL (11.05–19.29%), while that of Avo-3 was OOO (11.42–16.32%). Likewise, the third most abundant TAG molecular species of Avo-1, Avo-2 and Avo-3 are OOO (16.08%), PPO (12.43%), and POL (11.05%), respectively.

SFC Profile

The SFC profiles of the three local avocado cultivars are compared with that of the imported Hass variety as shown in Fig. 2. The SFC of Avo-1, Avo-2, and Avo-3 at 0 °C are 17.86%, 18.0%, and 13.57, respectively and they tend to become 0% in the range of 45-50 °C. The Hass variety, on the other hand, could be easily distinguishable from the local avocado cultivars for being liquid oil with low solid content (SFC, 0% at 10 °C) in major part of the temperature region. Hence, it could be ideally suited for salad dressing, marinades, and sauces [4]. On the other hand, Avo-1, Avo-2, and Avo-3 being lipids in the semisolid form could useful as base material in the formulation of creams and lotions used for the nourishment of the skin [2, 4]. The observed differences in the solidification behaviors of local avocado cultivars from that of the imported Hass variety could be attributed to the differences in their fatty acid and TAG molecular composition. The oil of the Hass variety is found to have the highest proportion of triunsaturated TAG molecules (Table 3) as well as unsaturated fatty acids (Table 2) when compared to those of the locally grown cultivars. It is usually the TAG molecules esterified with more saturated fatty acids, which gives the solid like nature to lipids [20]. As both Avo-1 and Avo-2 had comparably similar proportions of SFA (Table 2), they displayed similar SFC curves within the range of 0-35 °C. On the other hand, the SFC pattern of Avo-3 within the range of 0–35 °C was significantly (p < 0.05) lower than those of Avo-1 and Avo-2 since its SFA content was fairly lower (Table 2).

Thermal Characteristics by DSC

Thermal characteristics of local avocado cultivars are compared with those of the imported Hass variety as shown in Fig. 2. In Fig 2(X), DSC cooling curves of Avo-1, Avo-2, Avo-3 and Hass are represented by the curves (A), (B), (C) and (D), respectively. From this, it is clear that the cooling profiles of local avocado cultivars are distinctly different from that of the imported Hass variety. For instance, the initial point of crystallization transition of Avo-1, Avo-2, Avo-3, and Hass are $a_1(29.34 \text{ °C})$, $b_1(23.93 \text{ °C})$, $c_1(22.64 \text{ °C})$, and $d_1(-13.67 \text{ °C})$, respectively. While all three local cultivars displayed a highmelting exothermic thermal transition above 0 °C, there is no any distinctly identifiable thermal transition for the Hass variety in this temperature region. This observation corroborates well with the solidification behavior differences of these oils as noticed in Fig. 1. In Fig. 2(Y), DSC melting curves of Avo-1, Avo-2, Avo-3 and Hass are represented by the curves (A), (B), (C) and (D), respectively. Based on the melting profiles also the three local avocado cultivars are distinctly different from the imported Hass variety. As the final melting transition of Avo-1, Avo-2, Avo-3, and Hass are a₅(45.66 °C), b₅(41.37 °C), c₅(41.23 °C), and $d_4(2.30 \text{ °C})$, respectively, two distinguishable regions could be identified in the melting profiles by taking 10 °C as the point of reference. While the melting profiles of all three local cultivars have a high-melting transition above 10 °C, there is no distinctly identifiable endothermic transition in the melting profile of Hass variety (Curve-D) in this temperature region.

The thermal behavior differences between the local cultivars and the imported Hass variety could be mainly due to the differences in their fatty acid and TAG molecular composition as pointed out earlier (Tables 2, 3, respectively). While the imported Hass variety possessed higher proportion of triunsaturated TAG (55.55%), the oils produced from the three locally grown cultivars had significantly lower content of triunsaturated TAG (Table 3). The total contents of di- and trisaturated TAG of the Hass variety was extremely low (2.80%) whereas those of the three locally grown cultivars were found to be in higher proportions (14.24-20.22%). Likewise, the observed thermal behavior differences between Hass variety and local cultivars could also be correlated with the fatty acid compositions of these oils. For instance, the proportions of the saturated fatty acids of Avo-1 and Avo-2 are twice as much as that of Hass variety (Table 1).

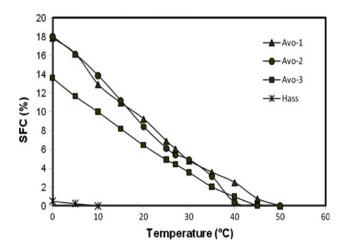


Fig. 1 Solid fat content profiles (SFC) of oils from different avocado cultivars

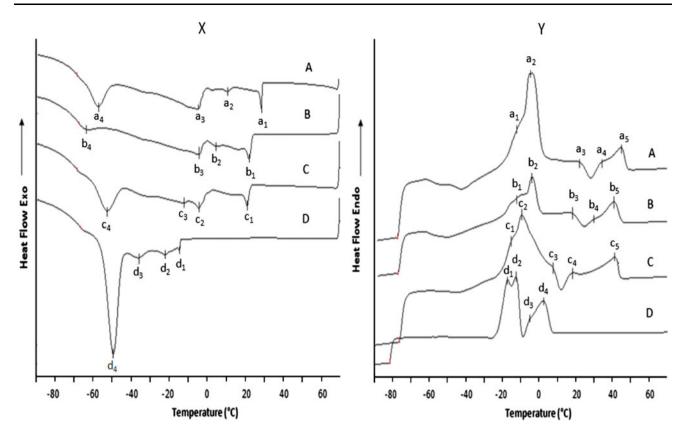


Fig. 2 DSC cooling (X) and melting (Y) profiles of oils from avocado cultivars Avo-1 (*curve A*), Avo-2 (*curve B*), Avo-3 (*curve C*) and Hass (*curve D*)

Conclusions

This study showed that the oils from local avocado cultivars are distinguishable from the oil of the Australian Hass variety based on basic physico-chemical parameters, solidification characteristics, and thermal behavior. When compared to the oils of the three locally grown cultivars, the oil from the imported Hass variety is found to have a higher degree of unsaturation in its fatty acid and TAG compositions. These differences in compositions cause the oil from the Hass variety to remain as a liquid while those from the three local cultivars to be in the semisolid form. The data on the solidification characteristics and thermal behavior suggest that the oils from the locally grown cultivars could be useful as base materials for formulation of creams and lotions.

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