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Lipid Content and Fatty Acid Composition in Taiwan Avocados (*Persea americana* Mill)

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Lipids are the major component in the flesh of avocado fruit. The main objective of this work is to determine the lipid content and composition in Taiwan avocados. The changes of lipid content and fatty acid composition were also investigated in terms of harvest maturity and ripening of avocados. Total lipid content in avocado fruits of seven cultivars, 'Hall', 'Zhang An', 'Ching Jin No.1', 'Ching Jin No.2', 'October Red', 'CASE3' and 'Choquette', ranged from 2.59% to 11.81% in fresh weight. Eight fatty acids were found in the avocado fruits. Concerning the saturated fatty acids, the most abundant in the avocado cultivars investigated were palmitic (C16:0) and oleic acid (C18:1). Palmitic acid levels were found to be the highest of the saturated fatty acid in all samples with the range from 38.37% to 48.32% of the total lipid. Oleic acid content in all avocado cultivars was considerably higher than those of other unsaturated fatty acids. Early- and late-harvested fruits showed higher total lipid content than others. Palmitic acid and oleic acid levels increased and then decreased during harvest time. Fatty acids composition of avocado fruits changed during their ripening.

Key words: avocado cultivar, fatty acid, harvest maturity, lipid, ripening

INTRODUCTION

The avocado fruit is rich in unsaturated fats and vitamins, and the flesh has more energy value than meat of equal weight (Seymour and Tucker, 1993). Lipids are the major component in the flesh of avocado fruit. Research overseas has attributed to flavor, texture and the nutritional characteristics of the fruit to the quantity and quality of lipids. A large number of studies have reported changes in lipid content and composition during growth and maturation of the fruit (Gaydou *et al.*, 1987; Ozdemir and Topuz, 2004; Eaks, 1990). However, studies on this area are still limited in Taiwan.

The market success of traditional avocado producers and exporters in the production areas such as California and South Africa is partly due to the continual acquisition of information of the fruit quality, which allows confidence in the planning of marketing activities. To maintain a differentiated place in the world market, the local avocado industry needs to continue obtaining the knowledge of the features of its product.

Postharvest research represents an important part of the maintenance of consistently high quality fruit delivered to the consumers. Among the different aspects that influence final fruit quality, there is an issue of harvesting fruit at the right stage of maturity. Harvesting only mature avocados enhances good eating characteristics at the end of the ripening process (Lewis, 1978). Thus,

greater understanding of maturity measures such as those for dry matter, and that of how they relate to total lipid content are important to our success. Aside from the issue of minimum maturity, another important aspect is the quality of fruit in late commercial season since there are allegations about rancidity in "over mature" fruit by international competitors.

With rapidly increasing in consumption of avocado worldwide, it becomes valuable fruit with exporting potential. To fulfill the requirements of world market, Taiwan researchers need continuing their research for seasonal changes in the components of the fruit for an accurate determination of quality standards to minimize the possibility of inferior fruit placed on the market. In this work, we examined the effect of harvesting time, variety and ripening on the lipid content and fatty acid composition in each of seven Taiwan avocado cultivars.

MATERIALS AND METHODS

Sample collection

Orchard Location

Avocado fruits (*Persea americana* Mill.) were obtained from local orchards at Chayi County in Taiwan. 'Hass' fruit imported from New Zealand was purchased from a local supermarket.

Fruit Collection

Seven avocado cultivars, 'Hall', 'Zhang An', 'Ching Jin NO.1', 'Ching Jin NO.2', 'October Red', 'CASE3' and 'Choquette', were chosen for the comparison of fatty acid composition. Early, mid, and late season avocado cultivar 'CAES 3' fruit was harvested at some intervals between July 2010 (beginning of the harvest season) and February 2011. These harvests cover the main commercial harvest season in Taiwan. The avocados were har-

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vested early in the morning to avoid water stress, transported overnight at 5°C by courier to Ilan County, and arrived at our laboratory within 24 hours after the harvest.

'CASE3' avocado fruit was harvested every 15 days from early August (early season) to early September (late season). The fruit was prepared to compare the effect of harvest time on the fatty acid content and fatty acid composition. For the determination of the changes of lipid content and fatty acid composition during ripening, 'Choquette' fruit was harvested and held in corrugated carton at 10°C for 17 days until the fruit became fully ripe.

Sample preparation

For the determination of total amount of lipid and fatty acid, fruits were cut in half vertically. The stone and peel of the one-half were removed manually. The remaining mesocarp was then immediately sliced thinly with 0.5–1.0 mm width and randomly mixed. Approximately 30 g of sample in each cultivars was immediately freeze-dried in a freeze-dryer with cooling-trap (KINGMECH, FD12-6P-P, Taiwan) at -59°C for 2 days. Dry weight of the samples were determined, and they were returned to -40°C until use for quantitative lipid extraction and fatty acid analysis. The extraction and analysis in each fruit were replicated three times.

Quantitative determination of total lipids

The Soxhlet extraction technique was used for validation of the method described above and carried out according to AOAC 963.15 with modifications. The Soxhlet extraction method using petroleum ether was carried out manually. Lyophilized mesocarp tissue was ground into powder using a pestle and mortar. The thimble containing 5 g of ground freeze-dried mesocarp from the same sample was placed in the Soxhlet extractor device, and 180 ml of petroleum ether was placed in the round flask. The sample was refluxed for 12 h with the heat adjusted approximately at 80°C. The solvent was removed using a rotary evaporator (Buchi Rotovapor, R114, Switzerland) under vacuum at 40°C. The recovered oil was weighed and stored under nitrogen in capped amber glass vials at -40°C until fatty acid analysis.

Fatty Acid Identification and Quantification

Fatty acid methyl esters (FAMES) were produced according to the method prescribed by AOAC 26.056 with modifications. Methyl esters were prepared from separated fatty acids with methanol-BFs reagent. One μ L of fatty acid methyl esters was injected into a gas chromatograph (Varian model 7400), equipped with a DB-WAX fused silica capillary column (30 m $\times$ 0.25 mm i. d., 0.25 μ m film thickness, J&W SCIENTIFIC) and a flame ionization detector (FID). The temperature was 50°C initially, then increased 10°C per minute to 250°C and held at 250°C for 30 minutes. Injector and detector temperatures were at 250°C. An extra fatty acid component, 12-heneicosenoic methyl (21:1) was added to each sample as an internal standard immediately prior to injection. The detector response was calibrated with a standard fatty acid methyl ester mixture (supplied by Sigma-Aldrich) containing six fatty acids which commonly occur in significant concentrations in avocado fruit: palmitic acid (16:0), palmitoleic acid (16:1), oleic acid (18:1), linoleic acid (18:2), linolenic acid (18:3) and the stearic acid (18:0). The fatty acid peaks in lipid samples were identified by comparison with the retention times of fatty acids in the standard mixture, and the amount calculated as a percentage of the total lipids. Triplicate GC analyses were performed and the results were expressed in GC area percentage as a mean value.

Statistical analysis

Data were subjected to analysis of variance (ANOVA) and means were separated by Fisher's Least Significant Difference (LSD) test at $P < 0.05$ significant level.

RESULTS

The crude fat content and fatty acid composition of the mesocarp of avocado fruits from Chia-yi County were analyzed. The differences of oil quality among cultivars, in different harvest maturity and during ripening were investigated.

Lipid content and fatty acid composition differ among avocado cultivars

Experiments were carried out with seven avocado cultivars including 'Hall', 'Zhang An', 'Ching Jin No.1',

Table 1. Total lipid and fatty acid percentage in fruits of avocado cultivars

Cultivar	Total lipid (%)	Saturated fatty acid (%)	Unsaturated fatty acid (%)
Hall	11.18 $\pm$ 1.17 b	46.22	53.78
Choquette	7.68 $\pm$ 1.30 c	40.69	59.31
CASE	37.00 $\pm$ 1.10 c	47.94	52.06
October Red	7.81 $\pm$ 1.96 c	51.18	48.82
Ching Jin No.1	2.58 $\pm$ 1.19 d	47.50	52.50
Zhang An	2.02 $\pm$ 0.52 d	46.53	53.47
Hass	19.12 $\pm$ 2.41 a	19.61	80.39

Means with different letters within the same row are significantly different ($P < 0.05$).

‘Ching Jin No.2’, ‘October Red’, ‘CASE3’ and ‘Choquette’ varieties. The crude fat contents of seven domestic avocado cultivars ranged from 2.59% to 11.81% based on

fresh weigh (Table 1). The most abundant crude fat content was found in ‘Hall’ avocado. However, it was a significantly low value as compared with ‘Hass’ imported from

Table 2. Fatty acids composition in fruits of avocado cultivars

Cultivar	Fatty acids content (g kg ⁻¹ FW)							
	myristic (C14:0)	palmitic (C16:0)	palmitoleic (C16:1)	stearic (C18:0)	oleic (C18:1)	linoleic (C18:2)	linolenic (C18:3)	arachidic (C20:0)
Hall	0.50 a	48.32 a	18.01 a	0.49 a	27.66 b	11.62 b	0.94 b	0.75 a
Choquette	0.27 c	28.09 c	9.36 cd	0.42 ab	24.34 bc	7.77 c	0.64 cd	0.13 c
CASE	0.39 b	30.27 bc	13.86 b	0.36 ab	13.50 cd	5.99 d	0.53 de	0.20 c
October Red	0.22 c	25.64 cd	6.91 de	0.38 ab	12.27 cd	5.49 d	0.46 ef	0.10 c
Ching Jin No.2	0.37 b	17.34 de	6.13 de	0.23 bc	8.03 d	5.24 d	0.70 c	0.63 a
Ching Jin No.1	0.24 c	14.83 e	4.40 e	0.23 bc	8.93 d	3.53 e	0.35 fg	0.27 bc
Zhang An	0.21 c	10.78 e	4.61 e	0.11 c	5.38 d	2.65 e	0.24 g	0.11 c
Hass	0.53 a	37.25 b	11.95 bc	0.51 a	128.97 a	16.83 a	1.31 a	0.50 ab

Means with different letters within the same row are significantly different ($P < 0.05$).

Table 3. Total lipid and fatty acids percentage of avocado cultivar CASE3 fruit mesocarp during harvest season

Fatty acid	Percentage in indicated harvest season (%)		
	Early season	Mid season	Late season
myristic acid (C14:0)	0.57 b	0.75 a	0.79 a
palmitic acid (C16:0)	45.33 a	45.48 a	44.62 a
palmitoleic acid (C16:1)	20.72 a	18.87 a	18.42 a
stearic acid (C18:0)	0.54 b	0.58 ab	0.62 a
oleic acid (C18:1)	20.43 a	20.69 a	20.61 a
linoleic acid (C18:2)	8.97 b	10.38 a	11.21 a
linolenic acid (C18:3)	0.78 b	0.88 ab	1.06 a
arachidic acid (C20:0)	0.31 a	0.41 a	0.21 a
Saturated	46.75	47.23	46.24
Unsaturated	50.90	50.83	51.30

Means with different letters within the same row are significantly different ($P < 0.05$).

Table 4. Total lipid and fatty acids composition in avocado cultivar Choquette fruit mesocarp unripe and ripe

Fatty acid	Total of fatty acids contained (%)	
	Before-ripening flesh	After-ripening flesh
myristic acid (C14:0)	0.57 b	0.75 a
palmitic acid (C16:0)	45.33 a	45.48 a
palmitoleic acid (C16:1)	20.72 a	18.87 a
stearic acid (C18:0)	0.54 b	0.58 ab
oleic acid (C18:1)	20.43 a	20.69 a
linoleic acid (C18:2)	8.97 b	10.38 a
linolenic acid (C18:3)	0.78 b	0.88 ab
arachidic acid (C20:0)	0.31 a	0.41 a
saturated	46.75	47.23
unsaturated	50.90	50.83
Total lipid (g kg ⁻¹ FW)	76.8a	83.7a

Means with different letters within the same row are significantly different ($P < 0.05$).

New Zealand. 'Zhang An' avocado contained the least crude fat among all cultivars. Eight fatty acids were found in avocado fruits. In domestic avocado, palmitic acid was predominant oil, constituting from 38.37% to 48.32% of total fatty acids. The other fatty acids quantified in descending order of abundance were oleic acid (from 19.96% to 33.17%), palmitoleic acid (from 12.96% to 20.72%), linoleic acid (from 8.97% to 13.12%), linolenic acid from (0.78% to 1.79%), stearic acid (from 0.44% to 0.70%), myristic acid (from 0.37% to 0.94%) and arachidic acid (from 0.18% to 2.64%) (Table 2).

Effect of harvest time on the lipid content and fatty acid composition

In regard to harvest maturity at early, mid and late harvest season, the crude fat contents of 'CASE3' avocado decreased then increased with increase of harvest maturity. The high content of palmitic acid and oleic acid were found in fruit of all stages of the harvest season (Table 3). With increase in the harvest maturity, palmitic acid and oleic acid contents increased then decreased. Although myristic acid, stearic acid and linolenic acid contents were small in amount, they gradually increased up to late season (Table 3).

Changes of lipid content and fatty acid composition during fruit ripening

The changes of total lipid and fatty acid composition before and after ripening were shown in Table 4. Total of lipid contents increased after ripening in 'Choquette' avocado flesh. After ripening, the fatty acid composition was also changed. Palmitic acid and palmitoleic acid contents reduced and the contents of oleic acid and linoleic acid increased. The contents of myristic acid, stearic acid and linolenic acid increased after ripening, while arachidic acid reduced after ripening.

DISCUSSION

The avocados cultivated are classified horticulturally into three races: the West Indian, the Guatemalan and the Mexican. Different races of avocado may have different percentage of fat composition. In this study, the fat content of seven avocado cultivars was analyzed. The avocados used are all from the West Indian race. The lowest fat content is detected in 'Zhang An' (2.59%), the highest is measured in 'Hall' (11.8%). The crude fat content is between 2.59% and 11.8%, as is similar to the result of Thompson *et al.* (1971) and Purseglove (1968), who pointed out that the fat content percentages of the West Indian avocados are between 3% and 10%. Because 'Choquette' and 'Hall' avocados are the hybrid between West Indian and Guatemalan races, the fat content is considered to be higher than the other cultivars from the West Indian race. Wolfe *et al.* (1949) also stated that the fat content of hybrids from West Indian avocado × Guatemalan avocado crosses were higher than the pure strain avocados of West Indian race. The oil content of avocados gradually increase during their development. It is a crucial factor to decide the eating quality and also

an important indicator of fruit ripeness. The fat content of 'Choquette' avocado planted in Venezuela was 5.05% (Gómez López, 1998), while the domestic 'Choquette' had fat content of 7.68%, which was higher than the Venezuelan avocado. The same avocado cultivar grown in different local regions may result in differences in the rate of oil composition. It was inferred from this study that the local climatic conditions are the influencing factors on the rate. In addition, irrigation may also affect the fat content rate of avocado fruit. Shortening the irrigation time interval and increasing the volume of irrigation water will increase the size of avocado fruits and their oil content (Lahav and Kalmar, 1977). In this study, 'Hass' avocado imported from New Zealand was used to refer to regarding domestic Taiwan avocado cultivars. The mean fat content of 'Hass' was 19.12%. The fat content of the Guatemalan race ranges from 8% to 15%, while as to the Mexican race, the fat content is even higher than 30% (Thompson, 1971; Purseglove, 1968). 'Hass' is a hybrid of the Mexican and Guatemalan races; hence the fat content will be higher than the West Indian race. The results of this study agree with those of the literatures above.

The main feature of avocado is in the rich content of fat; some cultivars of avocado can reach up to 40% fat content of their pulp weight (Pearson, 1975). The result of Ozdomir and Topuz (2004) has shown that avocado oil contains a total of seven types of fatty acids, i.e., oleic acid, stearic acid, palmitic acid, palmitoleic acid, linoleic acid, linolenic acid and arachidic acid. Most of the lipids consist of unsaturated fatty acids, among which oleic acid (C 18:1) shows the highest percentage in the lipid composition (Sciancalepore and Dorbessan, 1981; Gaydou *et al.*, 1987; Ozdemir and Topuz, 2004). Oleic acid is a beneficial unsaturated fatty acid and is included in avocado oil in large quantities. It mostly increases during avocado fruit development, while palmitic, palmitoleic and linoleic acids increase only slightly. Linolenic acid, on the other hand, decreases slightly during fruit development (Kikuta and Erickson, 1968). However, the fatty acid content of avocado oil may be affected by various factors. In this study we analyzed seven avocado cultivars and each of the cultivars contains eight types of fatty acids; the highest content rates approximately between 38% and 49% were detected in palmitic acid (C 16:0), followed by oleic acid (C 18:1, about 20.5%), palmitoleic acid (C 16:1, about 19%), linoleic (about 9.5%), linolenic (about 0.8%) myristic (about 0.6%) and stearic (about 0.6%) and arachidic (about 0.4%) acids. These results are different from those of other studies with oleic acid consistently representing the main fraction, followed by palmitic, linoleic, palmitoleic and linolenic acids in decreasing order of abundance (Sciancalepore and Dorbessan, 1981; Gaydou, *et al.*, 1987; Eaks, 1990; Meyer and Terry, 2008; Ozdemir and Topuz, 2004). In this study with New Zealand 'Hass' avocado obtained from a supermarket as a control, the result of lipid content is 19.1%; oleic acid showed the highest percentage of 62.33% in the fatty acid composition, followed by palmitic acid (C 16:0) of 19.30%. These percentages for composition of fatty acids

agree with the result of Ozdemir and Topuz (2004). This suggests that comparison of Taiwan-produced avocados and imported same ones may result in different fatty acid proportion. In olive oil, for example, the factors affecting the quality and chemical composition of the fatty acids are climatic conditions (Bonoli *et al.*, 2004; Morello *et al.*, 2006; Nergiz and Engez, 2000), agricultural practice (Servili *et al.*, 2007), fruit variety (Beltran *et al.*, 2005; Olivares-Lopez *et al.*, 2007), fruit maturity level (Rotondi *et al.*, 2004; Salvador *et al.*, 2001) and extraction technique (Cerretani *et al.*, 2005; Di Giovacchino *et al.*, 2002). The avocado mesocarp is an excellent source of monounsaturated fatty acids, and the research has shown that a highly avocado-enriched diet may contribute to decrease risks of cardiovascular disease, possibly by lowering total and LDL cholesterol levels whilst increasing HDL (Ledesma *et al.*, 1996). Based on the fatty acid composition, all the seven avocado cultivars except 'October Red' are good for health because of their highly unsaturated fatty acid contents (>50% of total lipid).

Before harvest, the fatty acid content and nutrients of avocado fruit will continue to accumulate in the flesh; thus, there is a good correlation between harvest maturity and fruit quality. If the fruits have not reached harvest maturity, they will not ripen properly. Even if they were managed to ripen normally, premature harvesting resulted in poor aroma and flavor because of the reason that the fat content would be too low to soften the flesh. Therefore harvest maturity has a major influence on fat content. In this study, the fat content of 'CASE3' flesh showed the trend of increase firstly and then decrease according to the advance of harvest maturity level. This result is similar to the finding of Ozdemir and Topuz (2004): fat content of 'Fuerte' avocado also revealed the trend of increase at first and then decrease at the later harvest maturity. Regarding the rates of fatty acids composing this avocado oil, Ozdemir and Topuz (2004) have shown that under different harvest maturity level in Turkey, the proportion of fatty acids changes significantly. Except for the result of significant increase of oleic acid, the composition of the other fatty acids significantly decreased with increase in the harvest maturity level. In this study, oleic acid content increased as the harvest maturity level advanced, although this was not significant. Palmitic acid, palmitoleic acid and arachidic acid decreased as the harvest maturity level advanced, and the rates of myristic acid, stearic acid, linoleic acid and linolenic acid significantly increased as the harvesting maturity level proceeded. This result is greatly different from that of the literature mentioned above. The study conducted by Kikuta and Erickson (1968) in California suggested that except for linolenic acid, four fatty acids composing of oleic acid, palmitic acid, palmitoleic acid and linoleic acid showed significant increase; the result is also different from that of this study. It is inferred from the Mediterranean climate in California and Turkey that avocado cultivar difference and climate difference result in the difference in the increasing rates of fatty acids in avocado flesh. As the harvest maturity level proceeds,

there will be changes in the fat composition: the percentage of unsaturated fatty acid will increase while that of saturated fatty acid will decrease (Martinez and Moreno, 1995). This is similar to the finding of this study.

Avocado fruits do not ripen on the tree. They must be harvested at appropriate maturity time, and then their after-ripening will start. It usually takes about 5–7 days after harvest to completely ripen (Seymour and Tucker, 1993). The ripeness of avocado fruits has a significant impact on their quality; unripe avocado flesh will emit a grassy smell. The result in Table 4 shows that the crude fat content in 'Choquette' avocado increases in the flesh of after-ripening fruits compared with that of before-ripening one. The result of Ozdemir and Topuz (2004) also showed that the fat content of 'Hass' avocado increased as the ripening proceeded, as is similar to the result of this study. After 'Choquette' avocado has ripened, there were no significant changes in the remaining fatty acids, except for the significant changes in myristic acid and linoleic acid. This result is similar to the report of Eaks (1990), where there were no significant changes in the rates of fatty acids of after-ripening avocado fruits. As avocado fruits ripe, the lipid content will increase and after the increasing there are no dramatic changes in fatty acid composition in after-ripening fruits.

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