Lipid and Fatty Acid Composition of Mesocarp and Seed of Avocado Fruits Harvested at Northern Range in Japan

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Abstract: The lipid and fatty acid composition of the mesocarp and seed of avocado fruit grown and harvested in Japan, which is located at the northern range of the avocado, was investigated and compared to an imported avocado purchased commercially. The potential of the avocado mesocarp as an agricultural product in Japan was also explored. Total lipids (TL) accounted for approximately 20% of the mesocarp. Further analysis showed that the neutral lipid (NL) fraction accounted for at least 95% of the TL, and almost 90% of NL was triacylglycerol. Monoenoic acids accounted for at least 65% of the total fatty acids, and oleic acid, which is regarded as an especially important functional component of avocado accounted for approximately 50% of the monounsaturated fatty acids. A comparison of the Japanese avocado cultivars and an imported avocado cultivar in the present study revealed no significant differences in the lipid and fatty acid compositions. Therefore, production of avocado fruit, which is rich in various nutritional components, is expected to be increased on a larger number of farms in Japan in the future. It is believed to be necessary to carry out further verification, such as the establishment of a cultivation technique adoptable to Japan, examination of optimal soil and land features, and cultivar selection.

Key words: avocado, fatty acid, oleic acid, agricultural product, Japan

1 INTRODUCTION

Most of the commercially available avocado fruits, which have recently been gaining popularity in Japan, are imported. The current biggest exporter of the avocado fruits available in Japan is Mexico, and the total amount of imports is nearly 30,000 tons. The latitudinal range for tropical avocado fruit is from 40° S to 40° N because of the temperature profile; however, a cold-hardy cultivar can grow at a much lower temperatures. In Japan, which is located at the northern limit of the range for the growth of avocado fruit, the cold-hardy avocado cultivars, Fuerte cultivar and Bacon cultivar, have been grown and harvested on a small scale in warm areas (around 35° N) on the Izu Peninsula and in Wakayama Prefecture. The Plant Protection Law requires imported avocado fruits to be treated at high temperature. Although they need to be ripened after the harvest, avocado fruits are generally harvested in a very unripened condition prior to export to Japan. In addition,

the quality is often deteriorated by cold storage in the domestic distribution system. As avocado fruit, which is also referred to as "butter fruit", has been eaten in various places, it has gained recognition for its high nutritional value. In particular, the content of lipids in avocado fruit is significantly higher than in other fruits, avocado fruit is used as a high-energy food source. Moreover, avocado oil is one of kinds of oil used all over the world and has been used both as a food and as constituent in cosmetics. The consumption of such highly nutritious avocado fruit in Japan is assumed continue to expanded due to the high level of health consciousness among Japanese; therefore, efforts will be made to provide safe, delicious, domestically produced avocado fruit. It is predicted that the average temperature in Japan will rise due to changes in the global environment, such as the progress of global warming, and the number of areas in Japan where avocado fruit can grow will also increase. There are many reports on the lipids

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contained in avocado fruit¹⁻⁶); however, there has been no study on nutrient components including lipids which are contained at a significantly higher level than in other fruits harvested in Japan. Hence, the lipid and fatty acid compositions in the mesocarp and seed of domestic avocado fruit were analyzed in order to examine the possibility of avocado fruit becoming an agriculture product grown in Japan in the future. The results of the experiments will be described in this report. The same experiments were conducted on commercially available imported avocado fruit, and the results were compared with those of the domestic avocado fruit.

2 EXPERIMENTAL

2.1 Plant material

Experiments were carried out with two avocado cultivars, the Fuerte cultivar and the Bacon cultivar, which were domestically grown in Numazu City, Shizuoka Prefecture. An imported avocado, the Hass cultivar, which was purchased from a supermarket in Tokyo, was subjected to the same experiments for comparison. Samples of these three cultivars were obtained at the fully ripened stage, and the seed and mesocarp were immediately separated. The separated seed and mesocarp were cut into pieces of an appropriate size and subjected to the experimental procedures. Five samples for each cultivar were used to obtain a mean value for each cultivar.

2.2 Isolation of total lipids

Total lipids (TL) were extracted and purified from each avocado fruit according to the method by Folch *et al.* using chloroform/methanol (2:1, v/v)⁷. The obtained total lipids were fractionated into neutral lipids (NL), glycolipids (GL), and phospholipids (PL) by silicic acid column chromatography⁸. The lipid fractions were identified and quantified by thin-layer chromatography using Rf values of standard lipids. Solvent systems were n-hexane:diethylether:acetic acid (80:20:1, v/v) for NL, chloroform:methanol (95:12, v/v)

for GL, and chloroform:methanol:water (65:25:4, v/v) for PL.

2.3 Analysis of fatty acid composition

Fatty acid methyl esters were prepared by transesterification with 15%-BF₃ in methanol. Fatty acid composition was analyzed by detection on a 5890 seriesII GC (Agilent Technologies, Inc., Santa Clarita, CA, U.S.A.) equipped with an FFS capillary column (ULBON-HR-SS-10; 0.25 mm I.D. × 50 m, Shinwa Chemical Industries, Inc., Tokyo, Japan) was used. The FID and injector port were maintained at 250°C. Column temperature was programmed to ramp from 150°C to 210°C at 2°C per minute. The flow rate of helium carrier gas was 0.8 ml/min. Fatty acid methyl esters were identified by comparing retention times between the fatty acid methyl esters and the respective fatty acid methyl ester standards.

3 RESULTS AND DISCUSSION

3.1 Contents and chemical properties of total lipids

The TL content and chemical properties of the mesocarp and seed of each cultivar are shown in Table 1. TL content in the mesocarp of domestic cultivars was $18.7 \pm 0.61\%$ (n=5, average) and $21.8 \pm 0.62\%$ for the Fuerte cultivar and the Bacon cultivar, respectively, while that of the imported Hass cultivar was $18.2 \pm 0.53\%$. As the highest mesocarp TL content was observed in the Bacon cultivar, it is assumed that differences in TL content are attributable to the differences among cultivars. The variation in TL content among the 5 samples of each of the cultivars was not great. The TL content has been reported to increase to 20% or higher after ripening⁵, and the same trend was also observed in the present study for the cultivars grown and harvested in Japan. The TL content in seed was 1.1%, 1.6%, and 1.1% in the Fuerte, Bacon, and Hass cultivars, respectively. TL content was much lower in the seed than in the mesocarp for all the cultivars. The chemical properties (Table 1) of the TL fraction differed between the mesocarp and seed; however, no significant difference in this

Table 1 Chemical Properties of Total Lipids (TL) in Mesocarp and Seed of Avocado Fruits Harvested in Japan.

	mesocarp			seed		
	Fuerte	Bacon	Hass* ¹	Fuerte	Bacon	Hass* ¹
TL content (wt%)	$18.7 \pm 0.61^{*2}$	21.8 ± 0.62	18.2 ± 0.53	1.1 ± 0.16	1.6 ± 0.16	1.1 ± 0.15
Appearance (25°C)	Yellowish green liquid			Yellowish brown liquid		
Acid value	2.3	1.7	1.5	6.9	9.5	5.4
Iodine value	92.4	92.8	94.3	120.9	114.6	118.4
Saponification value	194.2	193.8	195.1	204.2	204.1	203.0

* 1: exported from USA (California)

* 2: average date (n = 5) ± SD

experiment was observed among the domestic cultivars and the imported cultivar.

3.2 Lipid composition

The composition of TL is expressed as a percentage of NL, GL, and PL in Table 2. The percentages of NL in TL were 95% to 97% and 77% to 80% in mesocarp and seed, respectively. While a difference in NL content between the mesocarp and seed was observed, there was no significant difference among the cultivars. GL accounted for around 3% of TL in the mesocarp and 12% to 13% in the seed, and PL accounted for 1% to 2% and 8% to 11%, respectively. Thus, as for the NL content, a difference in distribution between mesocarp and seed was also observed for GL and PL.

3.3 Lipid composition of NL, GL and PL

The lipid composition of NL is shown in Fig. 1. The content of triacylglycerol, which was found to be highest among all the classes in both mesocarp and seed, was 87% to 89% (equivalent to at least 80% of TL) in mesocarp, and 70% to 73% (equivalent to approximately 55% of TL) in seed. A difference was observed between these parts, but no significant difference was observed among the cultivars. In addition to triacylglycerol, other lipids, such as diacylglycerol, free fatty acids, and free sterol, were contained in both the mesocarp and seed, and the contents of these lipids in seed were higher than those in mesocarp.

The lipid composition of GL is shown in Table 3. The content of monogalactosyl-diacylglycerol was found to be highest in both parts, and it was approximately 40% in mesocarp and approximately 57% in seed, respectively. In addition to monogalactosyl-diacylglycerol, other lipids, such as acylsterylglucoside, sterylglucoside, cerebroside, and digalactosyl-diacylglycerol, were also observed. No significant difference was observed among the cultivars.

The lipid composition of PL is shown in Table 4. The contents of phosphatidylethanolamine (39% to 42% in mesocarp and 30% to 32% in seed) and phosphatidylcholine (35% to 37% and 29% to 32%, as well) were high in both parts in all cultivars. Although there was a difference in

the content of these lipids between the parts, these two lipids together accounted for approximately 70% in PL. In addition to these lipids, other lipids, such as phosphatidic acid, phosphatidylglycerol, and phosphatidylinositol, were detected. While there was a difference in the contents of these lipids between the parts, no significant difference was observed among the cultivars.

3.4 Fatty acid composition of TL

The fatty acid composition of TL is shown in Table 5. The content of oleic acid (18:1n9) was highest in mesocarp in all three cultivars. While the content was 58% and 54% in the domestic cultivars Bacon and Fuerte, respectively,

Fig. 1 Lipid Composition (wt%) of Neutral Lipids (NL) in Mesocarp and Seed of Avocado Fruits Harvested in Japan.

Table 2 Lipid Composition (wt%) of TL in Mesocarp and Seed of Avocado Fruits Harvested in Japan.

	mesocarp			seed		
	Fuerte	Bacon	Hass* ¹	Fuerte	Bacon	Hass* ¹
Neutral lipid fraction (NL)	95.2	95.2	96.8	77.1	77.7	80.3
Glycolipid fraction (GL)	3.2	2.7	2.5	12.0	13.2	12.3
Phospholipid fraction (PL)	1.6	2.1	0.7	10.9	9.1	7.4

* 1: exported from USA (California)

* average date (n = 5)

Table 3 Lipid Composition (wt%) of Glycolipids (GL) in Mesocarp and Seed of Avocado Fruits Harvested in Japan.

	mesocarp			seed		
	Fuerte	Bacon	Hass* ¹	Fuerte	Bacon	Hass* ¹
Acylsterylglucoside	31.4	30.4	30.5	18.6	18.0	17.5
Monogalactosyl-diacylglycerol	39.2	42.0	40.0	56.3	57.5	57.7
Sterylglucoside	14.2	13.3	13.4	10.5	10.1	10.8
Cerebroside	11.0	10.7	12.4	10.5	10.7	9.8
Digalactosyl-diacylglycerol	2.7	2.0	2.5	1.7	1.8	2.0
Others	1.5	1.6	1.2	2.4	1.9	2.2

* 1: exported from USA (California)

* average date (n = 5)

Table 4 Lipid Composition (wt%) of Phospholipids (PL) in Mesocarp and Seed of Avocado Fruits Harvested in Japan.

	mesocarp			seed		
	Fuerte	Bacon	Hass* ¹	Fuerte	Bacon	Hass* ¹
Phosphatidic acid	10.4	9.3	11.4	17.6	15.4	14.5
Phosphatidylethanolamine	39.5	41.7	38.6	31.9	30.7	31.3
Phosphatidylglycerol	7.6	6.8	7.0	10.5	13.4	12.6
Phosphatidylcholine	35.9	36.9	34.8	28.9	30.1	31.7
Phosphatidylinositol	1.0	0.8	1.1	4.2	4.0	3.6
Others	5.6	4.5	7.1	6.9	6.4	6.3

* 1: exported from USA (California)

* average date (n = 5)

that was only 46% in the imported Hass cultivar. Such a difference in content is assumed to be due to the difference in cultivar; however, it may also be due to domestic cultivation or damage caused during the process of importing. Further investigation needs to be conducted on this point. Different from other fruits, avocado fruit contains a large amount of fat in the mesocarp and, moreover, it can be eaten readily; therefore, it is expected to be a good source of oleic acid. In addition, the content of linoleic acid was as low as approximately 11%, and the content of monoenoic acids, including oleic acid, was at least 65%. It was also found that, although there was less than 10%, the content of monoenoic acids (16:1n7 and 18:1n7) was high in the imported Hass cultivar. It should be noted that the results of the present study, such as observation of the high oleic acid ratio agreed with previous reports.

The content of linoleic acid was 35 % to 38% in seed, which was the highest, followed by 18:1n9 (22% to 24%) and 16:0 (17% to 19%). The content of α -linolenic acid was found to be 4% to 5%. According to these observations, it was found that the fatty acid composition in seed was

largely different from that in mesocarp. Due to such a difference, the content of total monoenoic acids was at least 65% in mesocarp while it was 29% to 34% in seed, and the content of total polyenoic acids was approximately 12% in mesocarp while it was at least 40% in seed, a significant difference was observed between the parts.

3.5 Fatty acid composition of NL, GL and PL

The differences in the fatty acid composition of main fatty acids among NL, GL, and PL are shown in Fig. 2.

In NL of mesocarp, main fatty acids were 18:1n9, 16:0 and 18:2n6, and the content of 18:1n9 in the imported Hass cultivar was low similarly in the case of TL. In NL of the seed, main fatty acids were 18:2n6, 18:1n9, 16:0 and 18:3n3. As compared to the contents of main fatty acids in TL, some differences (a lower content of 18:2n6 and a higher content of 16:0 of NL) were obtained. Although the proportions in NL were similar to those in TL in mesocarp, a difference between TL and NL was obtained in seed. Such a difference is assumed to be due to the difference in NL content in TL.

Table 5 Fatty Acid Composition (area% in GC) of TL in Mesocarp and Seed of Avocado Fruits Harvested in Japan.

	mesocarp			seed		
	Fuerte	Bacon	Hass* ¹	Fuerte	Bacon	Hass* ¹
12:0	tr* ²	tr	tr	0.6	0.2	0.3
14:0	0.1	tr	0.1	1.0	1.3	0.9
16:0	19.9	18.6	20.0	17.8	17.7	19.0
16:1n7	5.7	4.9	9.2	4.6	4.4	2.4
17:0	tr	tr	tr	0.3	1.1	0.5
18:0	0.7	0.8	0.4	0.9	1.1	1.5
18:1n9	54.4	58.2	45.9	24.0	24.1	22.4
18:1n7	5.3	4.9	9.7	3.6	2.8	2.7
18:2n6	11.6	10.7	12.5	35.3	36.1	38.2
18:3n3	0.8	0.8	1.1	5.3	4.1	5.4
20:0	0.1	0.1	tr	0.7	0.8	0.3
20:1n9	0.4	0.5	0.4	2.0	2.0	1.8
22:0	tr	tr	tr	0.4	0.7	0.5
24:0	tr	tr	tr	0.4	0.7	0.8
Others	1.0	0.5	0.7	3.1	2.9	3.3
Saturated acid	20.8	19.5	20.5	22.1	23.6	23.8
Monoenoic acid	65.8	68.5	65.2	34.2	33.3	29.3
Polyenoic acid	12.4	11.5	13.6	40.6	40.2	43.6

* 1: exported from USA (California)

* 2: trace

In GL in mesocarp and seed, although main fatty acids were similar to those in TL, the contents of main fatty acids differed from each other; a higher content of 18:2n6 and a lower content of 18:1n9 in mesocarp, a lower content of 18:2n6 and a higher content of 16:0 in seed.

In PL of mesocarp, although main fatty acids were similar to those in TL, while the content of 18:3n3 was at least 10% and the content 18:2n6 was high, the contents of 18:1n9 and 16:0 were low. However, since the percentage of PL in TL was low, such properties of PL only slightly affected the fatty acid composition of TL. In PL of seed, the content of 18:1n9 was approximately 45% for PL, and was largely different from that in TL, NL, and GL. This result was similar to the report by Pacetti *et al.* (2007).

4 CONCLUSIONS

The present study examined the lipid and fatty acid compositions of the total lipids of the avocado fruit grown and harvested in Japan located at the northern growth limit of avocado fruit. This study investigated the nutritional value of the avocado fruit, and further, compared the domestic avocado fruit cultivars with an imported commercially available avocado fruit cultivar. The results obtained in the present study showed no significant difference

between the domestic cultivars and the imported cultivars in terms of the chemical properties and the composition of the lipids. However, further investigation should be conducted with more cultivars and in other components. It has been said that there is a difference in taste and also food safety between domestic and imported avocado fruits; thus, it is desirable to increase the amount of domestically produced avocados in the market. If global warming progresses in a way that causes a rise in the average temperature in Japan, avocado fruit may become an agricultural product with the possibility of becoming one of the products in Japan in the future. Hence, it is believed to be necessary to carry out further investigation on avocado fruit, which has a high nutritional value and can be readily eaten, so as to establish a cultivation technique adoptable to Japan through the examination of optimal soil and land features and cultivar selection.

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Fig. 2 Comparison of Main Fatty Acid Composition (%) of TL, NL, GL and PL in Mesocarp and Seed of Avocado Fruits Harvested in Japan.

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