# 9. LIPID COMPOSITION AND NUTRITIONAL AND PHYSIOLOGICAL ROLES OF PERILLA SEED AND ITS OIL

# HYO-SUN SHIN

Department of Food Science and Technology Dongguk University, Seoul 100-715, Korea

#### INTRODUCTION

The ancient traditional use of Perilla seed and its oil as food or medicine in Southeast Asia is recorded in such old books as "Shen Nong Ben Cao Jing" of China, "Dongeuibogam" of Korea and "Engishikiten" of Japan (Chang, 1995).

In Korea roasted Perilla seed is used widely as flavouring and nutritional sources and mixed with other cereals or vegetables. Perilla oil is used as salad oil and cooking medium because of its distinct flavour. The oil is extracted by mechanical pressing of the roasted seed, then purified by a simple refining process such as filtration. The intact leaves of the plant are used as condiments or flavouring agents for several foods in Korea and Japan, and often deep-fried with batter. Koreans have consumed fresh Perdla leaves with grilled red meat for a long time.

Because Perilla oil is highly unsaturated, it has been used mainly for industrial purposes. Its consumption as a food has been limited to a few countries (Sonntag, 1979). People now recognise the dietary significance of  $\alpha$ -linolenic acid (Holman et *al.*, 1982; Bjerve et *al.*, 1989). Perilla seed and oil are good sources of the  $\alpha$ -linolenic acid and this and other aspects of their dietary value are being researched.

#### LIPID COMPOSITION

Total Lipid Content and Some Physicochemical Characteristics

Perilla seed contains about 38–45% of lipid (Sonntag, 1979; Vaugham, 1970). However, its content varies depending on the variety and growing conditions. Total lipid content of seed of 5 different lines of *Perilla frutescens* Britt. grown in Korea, quantified by the Soxhlet method with ethyl ether, varied from 38.6 to 47.8% (Shin and Kim, 1994). The seed of two varieties of *Perilla* grown in Japan (*P. frutescens* Britt. var. *crispa* Decaisne and *P. frutescens* Britt. var. *acura* Kudo) were extracted with chloroform-methanol (2:1, v/v). The total lipid content was 25.2–25.7% (Tsuyuki eta!, 1978). On the other hand, total lipid content of Perilla seed from India (*P. frutescens*) was very high (51.7%), when determined by the Soxhlet method with ethyl ether (Longvah and Deosthale, 1991). These results show that the total lipid content of Perilla seed varies not only with the variety and environmental factors but also with the nature of the extraction method.

Cultivar	Neutral lipids		Glycolipids		Phospholipids	
	12/I <sup>ds</sup>	0/0 <sup>c</sup>	$wt^b$	%°	wtb	%°
Suwon 8	36.9	93.5	1.5	3.9	1.1	2.7
Suwon 10	40.7	93.9	1.8	4.2	0.8	2.0
Suwon 21	42.7	93.7	2.0	4.4	0.9	2.0
Suwon 24	35.2	91.2	2.2	5.8	1.2	3.0
Yaebsil	44.2	92.5	2.5	5.2	1.1	2.3

 Table 1
 Major lipid classes of Perilla seed'

From Shin and Kim, 1994.

"Percentages of the seed on dry weight basis.

Percentages represent the fraction of a given lipid class with respect to lipid content within a cultivar.

The physicochemical characteristics of lipid extracted from Perilla seeds are as following: 1.4760–1.4784 of refractive index (at 25°C), 192.0–196.3 of iodine value (Wijs), 192.7–197.7 of saponification value and 1.3–1.8% of unsaponifiable matter content (Shin and Kim, 1994). Since Perilla seed oil is much higher in iodine value than other vegetable oils, it has the characteristics of a strong drying oil. Thus, Perilla oil is regarded as an inedible oil and instead is used for manufacturing such industrial products as varnishes, printing ink, linoleum, etc. (Sonntag, 1979; Vaugham, 1970).

### Lipid Classes

As shown in Table 1, the major lipid classes of Perdla seeds are 91.2-93.9% of neutral lipids (NL), 3.9-5.8% of glycolipids (GL) and 2.0-3.0% of phospholipids (PL). This composition is similar to that of other oilseeds having NL as the major component. NL fraction has 88.1-91.0% of triacylglycerols, 4.1-7.5% of sterol esters and hydrocarbons, 1.9-2.7% of free sterols, and a small amount of free fatty acids and partial glycerides (monoglycerides and diglycerides) (Table 2). Esterified steryl glycoside (48.9-54.3%) is the major component in the GL fraction. Other fractions are sterylglycoside (22.1-25.4%), monogalactosyldiacylglycerol (14.7-18.6%) and digalactosyldiacylglycerol (7.9-9.4%). The PL fraction has 50.4-57.1% of phosphatidylethanolamine, phosphatidylcholine (17.6-20.6%), phosphatidic acid (13.6-19.9%), and a small amount of lysophosphatidylcholine (2.9-4.0%), phosphatidylserine and phosphatidylinositol (4.8-6.6%). It was reported that there were no differences in the individual composition pattern comprising NL, GL and PL as the major lipid classes of Perdla total lipids (Shin and Kim, 1994).

Tsuyuki *et al.* (1978) reported that the total lipids of seed of two different Perilla lines were composed of triglycerides (79.79–82.46%), sterol esters (1.74-1.81%), free fatty acids (2.46-2.65%), diglycerides (1.58%), sterol (0.72-0.89%), pigments (3.06-4.18%), monoglycerides (0.59-2.19%), complex lipids (2.37-2.91%) and other (3.19-5.83%). From the seed total lipids they also separated 12 spots of complex lipids by TLC and confirmed 6 kinds of lipids: lecithin, lysolecithin, monogalactosyldiglycerides, cerebrosides, phosphatidylethanolamines and phosphatidylserines.

	- 1		I I		
Lipid class	Suwon 8	Suwon 10	Suwon 21	Suwon 24	Yaebsil
Neutral lipid"					
SE, HĈ	4.1 <sup>b</sup>	6.2	5.7	7.5	5.6
TG	91.0	89.1	88.7	88.1	88.2
FFA	0.3	0.4	0.7	0.3	0.8
FS	1.9	2.5	2.5	2.6	2.7
DG	0.8	0.3	0.9	0.4	1.2
MG	1.8	1.5	1.4	1.0	1.4
Glycolipid <sup>d</sup>					
SG	25.4	23.3	22.1	23.2	24.4
DGDG	7.9	9.2	8.1	8.0	9.4
ESG	50.8	48.9	54.3	53.2	51.4
MGDG	15.8	18.6	15.4	15.6	14.7
Phospholipid'					
LPC	4.0	3.6	3.6	4.0	2.9
PS, PI	4.8	6.1	6.1	6.5	6.6
PC	20.6	17.6	17.9	19.4	20.2
PE	54.2	55.4	57.1	56.6	50.4
PA	16.3	17.4	15.3	13.6	19.9

 Table 2
 Composition of neutral and polar lipids in Perilla seeds"

From Shin and Kim, 1994.

<sup>b</sup>All values are percentages of each lipid class.

<sup>c</sup>SE, sterol esters; HC, hydrocarbons; TG, triacylglycerols; FFA, free fatty acids;

FS, free sterols; DG, diacylglycerols; MG, monoacylglycerols

<sup>d</sup>SG, sterylglycoside; DGDG, digalactosyldiacylglycerol; ESG, esterified steryglycoside; MGDG, monogalactosyldiacylglycerol.

<sup>e</sup>LPC, lysophosphatidylcholine; PS, phosphatidylserine; PI, phosphatidylinositol; PC, phosphatidylcholine; PE, phosphatidylethanolamine; PA, phosphatidic acid,

Park *et al.* (1983) reported the triglyceride composition of total lipids extracted from Perilla seed by using TLC, HPLC, and GLC; the composition of the essential triglyceride of Perilla oil is 68% of  $(C_{18:3}, C_{18:3}, C_{18:3})$ , 6.7% of  $(C_{18:7}, C_{18:3}, C_{18:3})$ , 5.9% of  $(C_{18:1}, C_{18:2}, C_{18:3})$ , 4.3% of  $(C_{16:0}, C_{18:3}, C_{18:3})$ , 3.8% of  $(C_{18:1}, C_{18:2}, C_{18:3})$ , 3.2% of  $(C_{18:1}, C_{18:2}, C_{18:3})$ , 2.0% of  $(C_{16:0}, C_{18:2}, C_{18:3})$ , 1.5% of  $(C_{18:2}, C_{18:2}, C_{18:3})$ , 1.0% of  $(C_{16:0}, C_{18:1}, C_{18:3})$ , etc. The main characteristic is that trilinolenate accounts for 68% of total triglyceride.

### **Fatty Acid Composition**

The major fatty acids of total lipids extracted from Perilla seeds are linolenic  $(C_{18:3})$ , linoleic  $(C_{18:2})$  and oleic  $(C_{18:1})$  acids; palmitic  $(C_{16:0})$  and stearic  $(C_{18:0})$  acids are minor components (Table 3). Perilla oil contains higher unsaturated fatty acids than other vegetable oils, especially high content of linolenic and linoleic acids. Depending on the variety and the growing conditions, the content of the linolenic acid is 54–64%, which

	Shin and Kim	Longvah and Deosthale	<i>Tsuyuki</i> et al.
Fatty acids	(1994)	(199 <b>1)</b>	(1978)
C_16:0	6.3-6.7	8.92	4.04 - 6.64
C <sub>18:0</sub>	1.5 – 1.7	3.77	0.95 – 1.44
C <sub>18:1</sub>	13.2 - 14.9	12.92	13.12 - 18.34
C <sub>18-2</sub>	14.3 - 17.0	17.61	18.19 - 20.03
C <sub>18:3</sub>	61.1 – 64.0	56.76	53.63 - 58.97
Total saturates	7.9 - 8.4	12.69	4.99 - 8.08
Total Unsaturates	88.6 - 95.9	87.29	84.97 – 97.34
Polyunsaturates	75.4 – 81.0	74.37	71.82 - 79.00

 Table 3
 Major fatty acid composition of total lipid in Perilla seed (%)

is not less than that of linseed oil and 6 to 8 times more than that found in mustard and soybean oils. The content of linoleic acid in Perilla oil is similar to that of linseed and mustard oils.

Tsuyuki *et al.* (1978) reported that Perilla seed has small amount (0.08–0.27%, individually) of capric acid ( $C_{10:0}$ ), lauric acid ( $C_{12:0}$ ), myristic acid ( $C_{14:0}$ ), eicosanoic acid ( $C_{20:0}$ ), etc.

The study on the fatty acid composition among major lipid classes of Perilla seed total lipids shows that the fatty acid profile of NL is similar to that of the total lipids; fatty acid profile of GL and PL are high in palmitic, stearic and linoleic acids compared to the neutral lipid fraction. The GL and PL fractions of Perilla seed total lipids contain significant amounts of medium-chain fatty acids, i.e., capric and myristic acids (Table 4).

### **Minor Components**

Sterol and tocopherol were reported to be minor components of Perilla oil. Park *et al.* (1982) analysed the sterol composition of oil extracted from seed (*P. frutescens* Britt. var. *crispa* Decaisne). The results showed that the unsaponifiable matter of Perilla oil was composed of more than 60% of 4-desmethyl sterol fraction and trace amounts of 4,4-

Fatty acids	Neutral lipids	Glycolipids	Phospholipids
C <sub>10:0</sub>	_	1.0 - 14.1	3.2 - 15.7
$C_{14,0}^{10:0}$		1.4 – 15.2	
$C_{14:0}^{10.0}$ $C_{16:0}^{16:0}$ $C_{18:0}^{18:1}$	6.3 – 6.9	0.5 – 13.8	7.7 – 15.9
C <sub>18:0</sub>	1.4 - 1.7	2.7 - 4.4	1.5 - 3.4
C <sub>181</sub>	13.0 - 14.9	13.0 - 14.6	6.9 – 14.1
C <sub>182</sub>	14.4 - 16.0	14.3 - 18.6	15.3 - 29.3
C <sub>18.3</sub>	62.1 - 64.0	40.2 - 56.1	31.6 - 58.2

 Table 4
 Fatty acid composition of major lipid classes Perilla seed <sup>a</sup>

From Shin and Kim, 1994.

Fractions	Campesterol	Stigmasterol	$\beta$ -Sitosterol	$\Delta^5$ -Avenasterol
Total sterol	4.4 - 6.5	0.3 - 0.8	78.0 - 81.7	12.6 - 14.8
Esterified sterol	tr.	tr.	98.9 – 99.6	tr.
Steryl glycoside	3.5 - 7.5	0.2 - 1.2	79.9 - 84.8	11.6 - 13.5
Free sterol	9.4-10.4	3.1 – 10.0	54.7 – 72.6	5.4 - 32.8

 Table 5
 Sterol composition in Perilla oil<sup>a</sup>

'From Park et al., 1982.

dimethyl sterol and 4-monomethyl sterol fractions. Table 5 shows the sterol fraction of Perilla oil; esterified sterol, steryl glycoside, free sterol and sterol composition of total sterol fraction.  $\beta$ -Sitosterol occurs in more than 99% of the esterified sterol fraction and 55–93% in the free sterol fraction. Also they mentioned that the sterol composition of the seed oil is varies with its origin.

The total tocopherol content of total lipids extracted from Perilla seeds was 49.1–67.6 mg/100g oil. The  $\gamma$ -form existed as the major tocopherol (92%) with small amounts of the  $\alpha$ - and  $\delta$ -forms and the  $\beta$ -form absent (Shin and Kim, 1994).

In Perilla seed Park *et al.* (1993) identified physiologically active brassinosteroid, 0.50-0.8ng, as brassonolide, per gram of fresh weight, mainly castasterone with some homodolicholide.

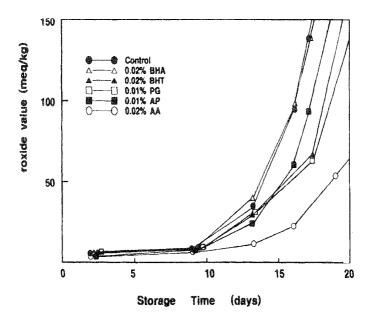
### The Change of Lipid Composition during Maturation and Germination

Min and Kim (1992a, b) studied the change of lipid composition during maturation of Perilla seed. They found that the content of ether-extractable lipids was increased continuously as the seeds matured while the content of triglyceride, the essential component of ether-extractable lipids, increased rapidly at the beginning of maturation and were 61.4–68.2% in matured seed (30 days after flowering). The contents of glycolipids and phospholipids were reduced and the amount of individual component of glyco- and phospholipids varied irregularly.

Kim *et al.* (1994) compared the lipid composition of germinated and non-germinated Perilla seed. They reported that the amount of triacylglycerol among neutral lipids was reduced, and free fatty acids and diacylglycerol were increased during germination. The contents of phophatidylethanolamine among polar lipids increased significantly and the amount of tocopherol, especially  $\gamma$ -form, increased notably.

### THE IMPROVEMENT OF THE OXIDATIVE STABILITY OF PERILLA OIL

The main problem of Perilla oil as edible oil is its high content of linolenic acid, highly unsaturated fatty acid, which is easily oxidized, and thus limitingits use. Thus, the oxidative stability should be improved to minimize the quality deterioration by rancidity.



**Figure 1** Effect of Butylated hydroxyanisole (BHA), Butylated hydroxytoluene (BHT), Propyl gallate (PG), Ascorbyl palmitate (AP) and Ascorbic acid (AA) on The Oxidation of Perilla oil Stored at 60° C (From Yi and Shin, 1989)

Yi and Shin (1989) reported that rancidity was inhibited more significantly when ascorbic acid in reversed micelle form was added to Perilla oil than when the mixed antioxidants (butylated hydroxyanisole, butylated hydroxytoluene, propyl gallate, ascorbyl palmitate) was added (Figure 1). They also mentioned that 8-tocopherol did not have the synergistic effect on ascorbic acid.

Cha and Choi (1990) found that tocopherol did not improve the oxidative stability since Perilla oil contained 400 ppm of tocopherol originally. In the case of organic acids, the antioxidative effect was increased in the following order; L-ascorbic acid > L-ascorbyl stearate > DL-malic acid > tartaric acid > citric acid.

Ahn *et al.* (1991) reported that under the Rancimat AOM test (97.8  $\pm$  0.2°C) the induction period of Perilla oil was longer than that of purified soybean oil, having 16 hours of AOM time when 5% of commercial lecithin was added to Perdla oil having 2 hours of induction period. As more lecithin was added, the induction period increased. The mixture of tocopherol, citric acid and ascorbyl palmitate showed synergistic effect on lecithin.

Kashima*et al.* (1991) reported the antioxidant effect of phospholipids on the oxidative stability of refined Perilla oil (PO), tocopherol-free Perdla oil (POP) and tocopherolenriched Perilla oil (POR). The oxidative stability of PO was increased noticeably by adding phosphatidylethanolamine (PE) and phosphatidylserine (PS). However, the addition of PE and PS did not have the antioxidant effect on POF. The oxidative stability of POR was lower than that of PO even though it had higher tocopherol, and was also increased by adding PE and PS. The reason that phospholipids showed antioxidant effect on Perilla oil was due to its synergistic effect with existing tocopherol.

Kim *et al.* (1994) reported that Perilla oil extracted from germinated Perilla seeds was more stable for oxidation than that extracted from nongerminated Perilla seeds. The oxidative stability was significantly increased in the germinated seeds after being stored more than one year whereas a little increased in the fresh seeds.

# NUTRITIONAL VALUE

The nutritional value of Perilla oil is somewhat different from that of other plant oils. Perilla oil contains especially higher content of linoleic and linolenic acids than other vegetable oils. Therefore, Perilla oil is a good source of essential fatty acids and can be used to optimize fatty acid ratio of n-6 and n-3 by mixing it with other vegetable oils. Essential fatty acids are those that are required by human body for normal growth, maintenance and for normal physiological functions, and are those that are not endogenously synthesized or those that are synthesized only in insufficient quantities. Therefore they must be supplied in the diet. Linoleic acid, a n-6 fatty acid, and  $\alpha$ -linolenic acid, a n-3 fatty acid, are life-saving fatty acids required for phospholipid components of cell membrane, nuclear membrane and mitochondrial membrane. Since the two fatty acids are in a competitive relationship due to their structural similarity, it is harmful unless the two fatty acids are in proper ratios in the diet. Several studies have been published (Bang*et al.*, 1980; Dyerberg, 1986) showing that  $\alpha$ -linolenic acid is closely related to anti-hypertensive effect and anti-thrombosis.

Kim and Kim (1989) reported that as dietary Perilla oil was increased, the concentrations of serum cholesterol and triglyceride was decreased. Other workers (Park *et al.*, 1992; Chung *et al.*, 1986; Nam *et al.*, 1981; Park and Han, 1976) reported similar results. Lee *et al.* (1987) reported that Perilla oil had effects on reducing serum cholesterol. Since below 15% dietary intake level of Perilla oil influences the cellular immune response ability, it is useful for cardiovascular disease or immune response. Therefore, for reducing serum cholesterol and improving immune response, it is recommended to diet with unsaturated fatty acid, such as is available from Perilla oil.

Han et al. (1983) reported that ingested Perilla oil was transformed to eicosapentaenoic acid (EPA) which restrains formation of thromboxane A (TXA<sub>2</sub>) and affects antithrombosis. They reported that when rats were fed ad **libitm** a diet containing 15% of Perilla oil for 15 weeks, the bleeding time was significantly delayed. Also the amount of malondialdehyde was decreased which is an indicator of TXA<sub>2</sub> and increased EPA and decreased arachidonic acid (AA) in the platelet. According to these results, the ingestion of Perilla oil, like fish oil containing high EPA, may result in the delay of bleeding time due to thrombogenesis reduction by increasing the ratio of EPA/AA in the platelet. Therefore, the intake of Perilla oil may be recommended for protecting chronic diseases. But, Suh and Cho (1980) reported that the formation of C<sub>20</sub>-C<sub>22</sub> (n-3) acids was not activated by elongation of C<sub>18:3</sub> (n-3) in Perilla oil in the animal body.

#### HYO-SUN SHIN

	Longvah and	Standall	FAO whole	
Amino acid	Deosthale	et al.	egg protein <sup>a</sup>	
	(1991)	(1985)		
Threonine	181	182	320	
Valine	174	113	428	
Cystine	89	83	152	
Methionine	174	86	210	
Isoleucine	234	100	393	
Leucine	374	363	551	
Tyrosine	244	145	260	
Phenylalanine	311	228	358	
Lysine	240	221	436	
Aspartate	556	513	601	
Serine	443	343	478	
Glutamate	1423	1358	796	
Proline	308	376	260	
Glycine	340	238	207	
Alanine	296	297	370	
Histidine	203	180	152	
Arginine	807	143	381	
Tryptophan	-	78	93	
Essential amino acids	2021	1185	3201	
Total amino acids	6397	5733	6446	

 Table 6
 Amino acid composition (mg/gN) of Perilla frutescens seed protein compared to that of FAO whole egg protein

'From FAO(1978)

On the other hand, when Perilla oil ingestion is roo much, it may result in formation of lipid peroxides in the body causing the depletion of antioxidant materials (Choi et al., 1987). Lee et al. (1976) reported that a deficiency of vitamin E was shown in rats and chicks fed 15% Perilla oil in the diet for 4 weeks. The major symptoms in rats were hair loss around the neck and serious skin lesion and in chicks significant muscle weakness and discoloration of the skin. Kwak and Choi (1992) reported that animals fed Perilla oil showed lower hepatic microsomal lipid peroxidation than animals fed corn oil, and was similar to animals fed tallow even though Perilla oil has a significantly higher polyunsaturated fatty acids/saturated fatty acids (P/S) ratio than corn oil. They suggested that hepatic microsomal lipid peroxidation was influenced not only by the degree of unsaturation of dietary lipids but also by the positions of double bonds in the fatty acids. They also reported that rats fed Perilla oil showed significantly lower prostaglandin E (PGE<sub>2</sub>) and thromboxane B (TXB<sub>2</sub>) production than rats fed corn oil. Therefore, the intake of Perilla oil may decrease tumorigenesis caused by lipid peroxides and eicosanoids. The effects of Perilla oil on formation of peroxides in the body is still in conflict, as mentioned above. Lee and Cho (1988) reported that the addition of vitamin C, vitamin E, ethylenediamine tetraacetate (EDTA) to a Perilla oil diet can slightly decrease the formation of peroxides in the serum and the tissues.

Din	Protein	Food intake	Body weight	t gain PER	NPR
group	(%)	(g/4 weeks)	(g/4 week	ks';)	
Casein	10	268 ± 8.8	77.7 ± 3.2'	2.99 ± 0.88	3.67 ± 0.04
P. frutescens	10	255 ± 11.9	$49.0 \pm 2.2^{\text{b}}$	$2.07 \pm 0.09^{b}$	2.87 10.11"
Protein-free		$103 \pm 6.2$	18.81 3.1		

Table 7Food intake, protein efficiency ratio (PER) and net protein ratio (NPR) in groups of<br/>rats fed casein and Perilla protein (Mean ± SE of six animals in each group)'

Prom Longvah and Deosthale, 1991.

<sup>b</sup>Significantly different at 0.1% level.

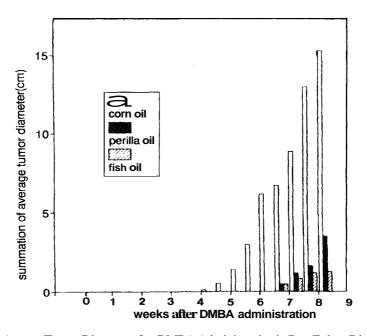
Since the antioxidative effect of Perilla oil is insufficient because of the scanty amount of tocopherol in Perilla oil, supplementation of an appropriate amount of antioxidants, including tocopherol, etc., will enable the reduction in the formation of lipid peroxides. More research should be performed to elucidate the appropriate P/S ratio, the appropriate n-6/n-3 ratio, the formation of lipid peroxides and the prevention of their formation, and the effect on metabolism when Perilla oil is ingested.

In addition to dietary oil, oil seeds generally contain a high content of beneficial protein. Perilla seed contains 18–28% (average 23%) of protein, and the residue left after oil extraction can be used as a protein source for humans and animals. The amino acid composition of Perilla seed protein is shown in Table 6. Amino acid composition varies with the species, the environmental conditions of cultivation, and the method of analysis used. Compared to whole egg, the total essential amino acid content of Perilla protein is very low. Longvah and Deosthale (1991) reported the limiting amino acid of Perilla seed protein was valine; however, Standall et al. (1985) reported it was isoleucine. Table 7 shows the body weight gain, protein efficiency ratio (PER), and net protein ratio (NPR) after animal feeding with Perilla seed protein. According to this result, there was no significant difference in feed intake between animals fed Perilla seed protein and with casein protein. However, body weight gains for animals fed casein protein and Perilla seed protein were 78g/4 weeks and 49g/4 weeks, respectively. PER value of casein protein, 2.99, was significantly higher than that of Perilla seed protein, 2.07. The protein digestibility of these proteins showed similar trends. These results suggested that the quality of Perilla seed protein was poor in terms of absorptiveness and total content of essential amino acids. However, there were no significant quality differences among sesame and other oilseed proteins.

### PHYSIOLOGICAL AND THERAPEUTICAL ROLES

### **Anticarcinogenic Effect**

Many research papers have been published on the physiological roles of eicosapentaenoic acid (EPA) and docosahaxaenoic acid (DHA) which exist abundantly in marine oils (Carrol, 1986; Isoda *et al.*, 1988).  $\alpha$ -Linolenic acid, which exists abundantly in soybean oil, rapeseed oil and Perilla oil, is the same n-3 fatty acid as DHA and EPA. Until recently,



**Figure 2** Average Tumor Diameter after DMBA Administration in Rats Fed on Diets Containing Different Fats (From Yonekura and Sato, 1989)

however, the physiological roles of the  $\alpha$ -linolenic acid had not been extensively studied, and thus, the essentiality of 0C-linolenic acid had not been clear. Among the food components, lipid is the most extensively studied component for the relationship between its consumption and cancer risk. Carroll *et al.* (1981, 1984) reported that there was a significant positive relationship between fat intake and mortality from breast, pancreas, colon, rectum, prostate or ovary cancer. The relationship between the type of fatty acids consumed and the risk of cancer has not been fully understood. However, many researchers have reported that high intake of n-6 type fat (linoleic acid) enhanced the carcinogenecity, and that intake of n-3 type fat such as linolenic acid, EPA or DHA lowered breast and colon cancer risk.

Yonekura and Sato (1989) studied the inhibitory effects of dietary Perilla oil, fish oil and corn oil on the breast cancer in rats. The authors reported that Perilla oil and fish oil showed significant inhibitory effect on the dimethylbenzanthracene (DMBA) induced breast cancer in rats (Figure 2).

Recently, several researchers reported that 0C-linolenic acid had inhibitory effects on breast cancer. Cameron *et al.* (1989) studied the effects of dietary fats on the inhibition of DMBA induced cancer risk in rats by feeding the rats with diets containing 6% either linseed oil ( $\alpha$ -linolenic acid, about 50%), fish oil (EPA, 13%), lard, evening-primrose oil ( $\gamma$ -linolenic acid, 9%), or safflower oil (linoleic acid, 78%). They reported that linseed oil and fish oil significantly suppressed the DMBA induced cancers in rats. It was interesting that inhibitory activity of linseed oil which contained 50% of  $\alpha$ -linolenic acid was

Dietary <sup>b</sup>	Effective no.	No. of rats	No. Of tumors per rat	
group	d rats	with tumors		
sf	26	12 (46%)	$0.6 \pm 0.1^{\circ}$	
SF	25	14 (56%)	$0.8 \pm 0.2$	
PR	26	5 (19%) <sup>d</sup>	$0.2 \pm 0.1^{d}$	
PI.	26	15 (58%)	$0.9 \pm 0.2$	

 Table 8
 Methyl nitrosourea-induced large-bowel tumors in CD-Fischer rats fed with various fat diets<sup>a</sup>

From Narisawa et al., 1990.

"Rats receiving an intrarectal dose of 2mg of methyl nitrosourea 3 rimes a week for 2 weeks were fed with diet containing 5%(sf group) or 12%(SF group) safflower oil, 12% perilla oil (PR group), or 12% palm oil (PL group). The experiment was terminated at week 36.

°Mean ± SEM

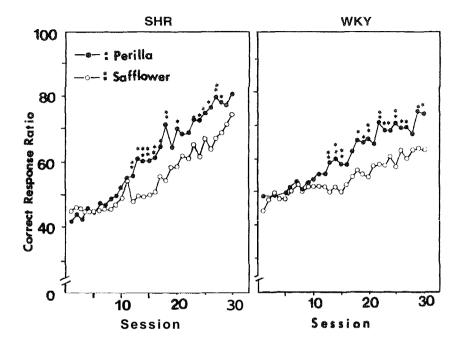
"Significantly different from other groups, p < 0.05 or 0.01 by X<sup>2</sup> test and Student's *t*-test.

significantly higher than that of fish oil which contained EPA and DHA. It was also noted that the cancer development in the groups treated with linseed oil or fish oil along with cancer inducing agent DMBA was significantly lower than that in the control which was treated with lard but no DMBA.

Narisawa *et al.* (1990) examined the methyl nitrosourea (MNU, cancer inducing chemical) treated rats after they had been fed for 35 weeks with diets containing 12% of either Perilla oil, palm oil or safflower oil (Table 8). Perilla oil treatment clearly suppressed large intestine cancer. The number of rats with cancer and the number of cancers per rat in the Perilla oil treated group were about one third and one fourth of other groups, respectively.

Park et *al.* (1993) reported that Perilla oil and fish oil had similar inhibitory effects on the chemically (N-methyl-N'-nitro-N-nitrosoguanidine, MNNG) induced colon cancer in rats. The author explained that Perilla oil and fish oil inhibited the colon cancer because the oil intake affected the content of arachidonic acid, a precursor of TXA<sub>2</sub> and PGE<sub>2</sub>. There are some other published papers on the inhibitory effects of  $\alpha$ -linolenic acid on carcinogenecity (Hori *et al.*, 1987; Fritsche, 1988); these are not detailed here.

It is worthwhile to note that  $\alpha$ -linolenic acid has inhibitory effects on mammary and large intestine cancer risk. It is not an exaggeration to say that in these days humans are constantly exposed to many different kinds of carcinogens derived from the polluted environments. It is thus expected that daily intake of Perilla seed or Perilla oil may reduce the cancer risk. How to use the seed or oil as food ingredient might be an important matter for healthy diet in the future. Cancer death in Japan is reportedly higher than in Korea. Although the reasons for higher cancer death in Japan are not simple, it can be assumed that it might be, at least to some extent, due to the westernized Japanese diets with high intake of linoleic acid. Koreans still relatively well observe traditional diet habits and eat Perilla seed and oil.



**Figure 3** Correct Response Ratio in The Brightness-discrimination Learning Test. (From Yamamoto *et al.*, 1987)

#### **Effects of Brain and Nerve System**

Attention has been drawn to the effects of  $\alpha$ -linolenic acid on the learning ability since it was reported that this acid was an essential fatty acid in the nerve system. Yamamoto et al. (1987) fed SHR rats and WKY rats for 2 generations with linoleic acid rich feed (safflower oil, 5%) or  $\alpha$ -linolenic acid rich feed (Perilla oil, 5%), then carried out a brightness-discriminationlearning test. The authors reported that dietary Perilla oil group in SHR rat and WKY rat showed better correction rate after 10 time tests (1 per day) and it showed the significant difference after 15 time tests (Figure 3). The increased learning ability by the intake of  $\alpha$ -linolenic acid might be due to high concentration of induced DHA in brain from  $\alpha$ -linolenic acid. Bourre *et al.* (1989) also reported that  $\alpha$ -linolenic acid increased learning ability. They fed female Wister rat for 2 generations with feeds containing sunflower oil (no linolenic acid) or soybean oil (1% linolenic acid). Rats from third generation were used for the experiments. The result showed that sunflower oil diet significantly lowered the DHA level in the brain and soybean oil diet significantly increased the DHA level in the brain. The results also showed the positive relationship between the level of DHA in the brain and the  $\alpha$ -linolenic acid contents in diet during brain developing periods. The authors also reported that dietary  $\alpha$ -linolenic acid affected the visual nerve system as a result of a learning ability test in a shuttle box. Low intake of dietary  $\alpha$ -linolenic acid led to abnormal symptoms in electroretinogram. Neuringer *et al.* (1984, 1986) fed prenatal and postnatal infant rhesus monkey with  $\alpha$ -linolenic acid deficient diets. They found that DHA contents in the retina and in the brain of the  $\alpha$ -linolenic acid deficient group were 1/2 and 1/4 of those in the control group, respectively. Thus, the sight of infant monkey with (x-linolenic acid deficiency was failing.

### Effects of Survival Time

Shimokawa *et al.* (1988) reported that  $\alpha$ -linolenic acid rich diets increased life span of rats. They fed hypertensive rats with diets containing 5% of Perilla oil (65% of (x-linolenic acid) or 5% of safflower oil (no linolenic acid) and observed the life span of the rats. The results showed that the mean life spans of the dietary Perilla oil group and the dietary safflower oil group were 59.5 weeks and 50.9 weeks, respectively. That is, the life span of the dietary Perilla oil group was about 17% longer than that of the dietary safflower group.

Renaud *et al.* (1983, 1981) let 25 French farmers, who had been given a saturated fat rich diet, substitute butter with margarine made of canola oil (10% of (x-linolenic acid) for 1 year and determined the serum lipids and platelet aggregation. By substituting the butter with margarine, the percent of  $\alpha$ -linolenic acid in the farmers' diets increased from 1.2% to 3.5%. One year after, the determined EPA content in platelet and serum of the farmers was slightly increased, but the platelet aggregation had decreased significantly. Renaud *et al.* (1983) reported that even though the regular intake of  $\alpha$ -linolenic acid did not greatly increase the serum EPA content, it was important in the prevention of cardiovascular disease and thrombosis. They called it "small is beautiful" and recommended regular intake of  $\alpha$ -linolenic acid.

#### REFERENCES

- Ahn, T.H., Kim, J.S., Park, S.J. and Kim, H.W. (1991) Antioxidative effect of commercial lecithin on the oxidative stability of Perilla oil. *Korean J. Food Sci. Technol.* 23,251–255 (in Korean).
- Bang, B.O., Dyerberg, J. and Sinclair, H.M. (1980) The composition of the Eskimo food in north western Greenland. *Am. J. din. Nutr.*, 33, 2657–2661.
- Bjerve, K.S., Fischer, S., Wammer, F. and Egeland, T. (1989) α-Linolenic acid and long-chain n-3 fatty acid supplementation in three patients with n-3 fatty acid deficiency : effect on lymphocyte function, plasma and red cell lipids, and prostanoid formation. *Am. J. Clin. Nutr.*, **49**, 290–300.
- Bourre, J.M., Francois, M., Youyon, A., Dumont, O., Piciotti, M., Pascal, G. and Durand, G. (1989) The effects of dietary α-linolenic acid on the composition of nerve membranes, enzymatic activity, amplitude of electrophysiological parameters, resistance to poisons and performance of learning tasks in rats. J. *Nutr.*, 119, 1886–1892.
- Cameron, E., Bland, J. and Marcuson, R. (1989) Divergent effects of n-6 and n-3 fatty acids on mammary tumor development in C3H/Heston mice treated with DMBA. *Nutr. Res.*, 9, 383– 393.
- Carrol, K.K. (1984) Role of lipids in tumorigenesis. J. Am. Oil Chem. Soc., 61, 1888--1891.

- Carrol, K.K. (1986) Biological effects of fish oils in relation to chronic disease. *Lipids*, **21**, 731–732.
- Carrol, K.K., Hopkins, G.J., Kennedy, J.G. and Davidson, M.B. (1981) Essential fatty acids in relation to mammary carcinogenesis. *Prop. Lipid* Res., 20, 685-690.
- Cha, G.S. and Choi, C.U. (1990) Determination of oxidative stability of Perilla oil by the Rancimat method. *Korean]. Food Sci. Technol.*, 22, 61–65 (in Korean).
- Chang, G.H. (1995) History of the use of dietary lipids in Korea. Soo Hak Sa, Seoul, pp.143–215 (in Korean).
- Choi, K.W., Park, M.H. Chang, K.S. and Cho, S.H. (1987) Effect of dietary fish oil on lipid peroxidative and antiperoxidative system in rat liver and brain. *J. Korean Soc. Food Nutr.*, 16, 147–155 (in Korean).
- Chung, S.Y., Seo, M.H., Park, P.S., Kanji, J.S. and Kanji, J.O. (1986) Influences of dietary fats and oils on concentration of lipids in serum and liver of rats on hypercholesterolemic diet. *J. Korean Soc. Food Nutr.*, **15**, 75–81 (in Korean).
- Dyerberg, J. (1986) Linolenate-derived polyunsaturated fatty acids and prevention of atherosclerosis. Nutr. Rev. 44, 125-134.
- FAO Nutritional Studies No. 24. (1978) Amino Acid Composition and Biological Data on Proteins. Food and Agricultural Organization of the United Nations, Rome.
- Fritsche, K.L. (1988) Reduced growth and metastasis of a transplantable syngenic mammary tumor by dietary α-linolenic acid. J. Am. Oil Chem. Soc., 65, 509.
- Han, Y.N., Yoon, H.W., Kim, S.H. and Han, B.M. (1987) Effects of Perilla oil intake on bleeding time, thromboxane formation and platelet fatty acid in rats. *Korean J. Pharmacogn.* **18**, *513* (in Korean).
- Holman, R.T., Johnson, S.B. and Hatch, T.F. (1982) A case of human linolenic acid deficiency involving neurological abnormalities. *Am. J. Clin. Nutr.*, **35**, 617–623.
- Hori, T., Moriuchi, A., Okuyama, H., Sohajima, T., Koizumi K., and Kojima, K. (1987) Effect of dietary essential fatty acids on pulmonary metastasis of ascites tumor cells in rat. *Chem. Pharm. Bull.*, 35, 3925–3927.
- Isoda, Y. and Hirano, J. (1988) Cancer and lipids. Hygienic Chem., 34,295-302 (in Japanese).
- Kashima, M., Cha, G.S., Isoda, Y., Hirano, J. and Miyazawa, T. (1991) The antioxidant effects of phospholipids on Perilla oil. J. Am. Oil Chem. Soc., 68, 119–122.
- Kim, C.K., Song, G.S., Kwon, Y.J., Kim, I.S. and Lee, T.K. (1994) The effect of germination of Perilla seed on the oxidative stability of the oil. *Korean* J. Food *Sci. Technol.*, 26, 178–183 (in Korean),
- Kim, W.K. and Kim, S.H. (1989) The effect of sesame oil, Perilla oil and beef tallow on body lipid metabolism and immune response. Korean J. Nutr., 22, 42–53 (in Korean).
- Kwak, C.S. and Choi, H.M. (1992) Effects of intake of Perilla oil or corn oil and 2-acetylaminofluorene treatment on lipid peroxidation, PGE2 and TXB2 production in rats. *KoreanJ. Nutr.*, 25, 351–359 (in Korean).
- Lee, I.S. and Cho, C.S. (1988) Effect of antioxidants added Perilla oil diet on serum and tissue in rats. *Korean Oil Chem. Soc.*, **5**, 29–38 (in Korean).
- Lee, J.M., Kim, W.Y. and Kim, S.H. (1987) A study of Korean dietary lipid sources on lipid metabolism and immune function in rat. *Korean J. Nutr.*, **20**, 350–366 (in Korean).
- Lee, Y.C., Kwak, T.K. and Lee, K.Y. (1976) Relationship between vitamin E and polyunsaturated fat. A comparative animal study emphasizing Perilla seed oil as a fat constituent. *Korean J. Nutr.*, 9,283–291 (in Korean).
- Longvah, T. and Deosthale, YG. (1991) Chemical and nutritional studies on Hanshi (*Perilla frutescens*), a traditional oilseed from northeast India.]. Am. Oil Chem. Soc., 68, 781-784.

- Min, Y.K. and Kim, Z.U. (1992a) Change of glycolipids and phospholipids during maturation of Perilla seed (*Perilla frutescens*). J. Korean Agric. Chem. Soc., 35, 146–151 (in Korean),
- Min, Y.K. and Kim, Z.U. (1992b) Change of lipids during maturation of Perilla seed (*Perilla frutescens*). J. Korean A , Chem. Soc., 35, 139–145 (in Korean).
- Nam, H.K., Sung, H.C. and Chang, I.Y. (1981) Studies on the effect in degree of saturation of fats on serum cholesterol level in the rabbit. J. Korean Soc. Food Nutr., 10, 27–37 (in Korean).
- Narisawa, T., Takahashi., M., Kusaka, N., Yamazaki, Y., Koyama, H., Kotanaga, H., Nishizawa, Y. and Kotsugai, M. (1990) Inhibition of large-bowel carcinogenesis in rats by dietary Perilla oil rich in the n-3 polyunsaturated fatty acid α-linolenic acid. *Ishiyaku*, 153,103–104 (inJapanese).
- Neuringer, M., Connor, WE., Lim, D.S., Barstad, L. and Luck, S. (1986) Biochemical and functional effects of prenatal and postnatal n-3 fatty acid deficiency on retina and brain in rhesus monkeys. *Proc. Natl. Acad. Sci.*, 83, 4021–4025.
- Neuringer, M., Connor, W. E., Petten, C.V. and Barstad, L. (1984) Dietary n-3 fatty acid deficiency and visual loss infant Rhesus monkey. J. Clin. Invest, 73, 272–276.
- Park, H.S. and Lee, S.M. (1992) Effects of dietary n-3 fatty acid and fat unsaturation on plasma lipids and lipoproteins in rats. Korean]. *Nutr.*, 25, 555–568 (in Korean).
- Park, H.S., Kim, J.G. and Cho, M.J. (1982) Chemical compositions of *Perilla frutescens* Britton var. *crispa* Decaisne cultivated in different area of Korea. J. Korean Agric. Chem. Soc., 25, 14-20 (in Korean).
- Park, H.S., Kim, J.G. and Hyun K.H. (1983) Brassinosteroid substances in immature Perilla frutescens seed. J. Korean Agric. Chem. Soc., 36, 197–201 (in Korean).
- Park, H.S., Seo, E.S., Song, J.H. and Choi, C.U (1993) Effects of Perilla oil rich in α-linolenic acid on colon tumor incidence, plasma thromboxane B2 level and fatty acid profile of colonic mucosal lipids in chemical carcinogen-treated rats. *Korean J. Nutr.*, 26, 829–838 (in Korean).
- Park, K.R. and Han, I.K. (1976) Effects of dietary fats and oils on the growth and serum cholesterol content of rats and chicks. *Korean J. Nutr.*, 9, 59–67 (in Korean).
- Park, Y.H., Kim, D.S. and Chun, S.J.(1983) Triglyceride composition of Perilla oil. Korean J. Food Sci. Technol., 15, 164–169 (in Korean).
- Renaud, S. and Nordoy, A (1983) "Small is beautiful" α-linolenic acid and eicosapentaenoic acid in man. The *Lancet*, 21, 1169.
- Renaud, S., Moragain, R., Godsey, F., Domont, E., Symington, I.S., Gillanders, E.M. and Obrine, J. (1981) Platelet functions in relation to diet and serum lipids in British farmers. Br. Heart J., 46, 562–570.
- Shimokawa, T. and Okuyama, H. (1988) Effect of dietary α-linolenate/linoleate balance on mean survival time, incidence of stroke and blood pressure of spontaneously hypertensive rats. Life Science, 43, 2067–2075.
- Shin, H.S. and Kim, S.W. (1994) Lipid composition of Perilla seed.J. Am. Oil Chem. Soc., 71, 619–622.
- Sonntag, N.O.V. (1979) Composition and characteristics of individual fats and oils. In Swern D, (ed.), Bailley's Industrial Oil and Fat Products, John Wiley & Sons, New York, pp. 434–435.
- Standall, B.R., Ako, H. and Standall, G.S.S. (1985) Nutrient content of tribal foods from India : *Flemingia vestita* and *Perilla frustescens. J.* Plant Foods, 61, 1471–1453.
- Suh, M. and Cho, S.M. (1986) Effect of dietary n-3 fatty acids on mitochondria1 respiration and on lipid composition in rat heart. *Korean Biochem. J.*, 19, 160–167 (in Korean).
- Tsuyuki, H., Itoh, S. and Nakatsukasa, Y. (1978) Studies on the lipids in Perilla seed. Research Division in Agriculture, Nihon University, 35, 224–230 (in Japanese).
- Vaugham, J.G. (1970) The Structure and Utilization of Oil Seeds. Chapman and Hall LTD., London, p. 120.
- Yamamoto, N., Saitoh, M., Moriuchi, A., Nomura, M. and Okuyama, H. (1987) Effect of dietary

#### HYO-SUN SHIN

 $\alpha$ -linolenate/linoleate balance on brain lipid compositions and learning ability of rat. J. Lipid Res., 28, 144–151.

- Yi, O.S. and Shin, H.K. (1989) Antioxidative effect of ascorbic acid solubilized via reversed micelle in Perilla oil. *KoreanJ. Food Sci. Technol.*, 21, 706–709.
- Yonekura, I. and Sato, A. (1989) Inhibitory effects of Perilla and fish oil on 7,12dimethylbenz[a]anthracene induced mammary tumorigenesis in Sprague-Dawley rats. *Ishiyaku*, 150,233–234 (inJapanese).