Triacylglycerol and Phospholipid Composition of Hazelnut (*Corylus avellana* L.) Lipid Fraction during Fruit Development

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We analyzed the triacylglycerol and phospholipid contents of hazelnuts from early development to maturity. Both were analyzed by high-performance liquid chromatography coupled to a light scattering detector. Trioleylglycerol, linoleyl-dioleylglycerol, and palmitoyl-dioleylglycerol were the most predominant triacylglycerols throughout development. Triacylglycerols showed small variation during hazelnut development. Phosphatidylcholine, phosphatidylethanolamine, and phosphatidylinositol were the most abundant phospholipids. Traces of phosphatidic acid were also detected. The statistical analysis showed that the positive correlation among the individual phospholipid contents was significant. Phospholipid contents showed a steep decrease during hazelnut development. Triacylgycerols and phosphatidylcholine were isolated by preparative thin-layer chromatography, and their fatty acid profile was determined by gas−liquid chromatography. Triacylglycerols showed a high percentage of monounsaturated fatty acid moieties, whereas phosphatidylcholine had the highest percentage of saturated and monounsaturated fatty acid moieties. The polyunsaturated fatty acid moiety showed low percentages in the triacylglycerol and phospholipid backbone.

Keywords: *Developing hazelnuts; triacylglycerol; phospholipid; composition; fatty acid profile*

INTRODUCTION

At the present, nutritional interest in the lipid composition of vegetable oils is on the increase. Hazelnuts, like other nuts (almonds, walnuts, peanuts, etc.), are valuable ingredients of the Mediterranean diet, which is typical of the countries around the Mediterranean Sea. Spain, the fourth-largest hazelnut-producing country (behind Turkey, Italy, and the United States), produces about 12 000 metric tons year (FAO, 1996), which is 2% of the total world hazelnut production.

Hazelnuts are rich in lipids; about 60% of the hazelnut kernel contains oil (Souci et al., 1994; Paul and Southgate, 1988). Several authors have reported the benefits of nut consumption for the human diet (Sabate, 1993; Sabate et al., 1993; Abbey et al., 1994; Jenkins et al., 1997; Fraser et al., 1992; Prasad, 1997). Vegetable oils are especially valuable because of their fatty acid (Grundy, 1994, 1997; Denke and Grundy, 1992; Grundy et al., 1988), sterol (Gylling et al., 1995; Vanhanen et al., 1993; Miettinen and Vanhanen, 1994; Miettinen and Kesaniemi, 1989; Tilvis and Miettinen, 1986; Vahounoy et al., 1981, 1983), and tocopherol (Jialal and Grundy, 1992) contents. These compounds have been reported to affect plasma lipid and lipoprotein contents in human serum (Denke and Grundy, 1992; Gylling et al., 1995; Vanhanen et al., 1993; Miettinen and Vanhanen, 1994; Miettinen and Kesaniemi, 1989; Tilvis and Miettinen, 1986; Jialal and Grundy, 1992).

More specifically, fatty acids are linked to the triacylglycerol and phospholipid backbone. Triacylglycerols (TAGs), the most important neutral lipid in most vegetable oils included in the human diet, influence total fat and cholesterol absorption in the human lumen (Bracco, 1994; Katan et al., 1994; Kritchevsky, 1994; Nydahl et al., 1994; Berry et al., 1991). Modulating the fatty acid composition of vegetable oils is one of the biggest concerns in the biochemistry of plant lipids (Slabas and Fawcett, 1992; Ray et al., 1993; Martin and Rinne, 1986; Knutzon et al., 1992). Several authors have shown the influence of environmental and varietal factors on the fatty acid composition of oilseeds and nuts (Slack and Roughan, 1978; Slack et al., 1978; Chaiserie and Dimick, 1989; Branch et al., 1990; Lajara et al., 1990).

Hazelnut flower fertilization takes place very late in the season; then, a quick rapid nut and kernel growth leads to maturity in late summer or early fall (Westwood, 1993a; Thompson, 1979). In Spain, hazelnuts grow from late May to late August. During hazelnut development, the oil content rises sharply in early July (Thompson, 1979; Westwood, 1993b). Regardless of other constituents such as sugar and protein compounds, the fatty acid composition of hazelnut kernels changes considerably between the early stage of development and maturity (Neubeller, 1990; Koyunku et al., 1997). When maturity is completed, which usually happens in late summer through midfall, hazelnuts drop and are ready to be picked up. Afterward they are either stored or sold with or without their shells.

We previously reported the influence of environmental factors on the fatty acid, triacylglycerol, and tocopherol contents of hazelnuts harvested in a small area of production in Spain (Parcerisa et al., 1993, 1994, 1995a). Here we show the compositional changes of triacylglycerol and phospholipid contents, including their fatty acid profile, in hazelnuts from an area of production in Catalonia (Spain). Our results provide

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more data on the biosynthesis of lipids in hazelnuts and lay the basis for further research into how to improve both the nutritional quality and the commercial value of hazelnuts produced in Spain.

MATERIALS AND METHODS

Samples. To avoid any variability, hazelnut samples were taken from the same hazel tree every Sunday between the first week of July and the last week of August (1997). On each occasion, 8−10 developing hazelnuts were cut from the tree and stored at 5 °C until analyses were carried out. The samples were labeled with the date when they were collected.

Oil Extraction. The hazelnuts were shelled, and then the kernels were crushed. The sample was poured into a 100-mL glass homogenizing tube followed by 25 mL of a chloroform/methanol mixture (2:1, v/v). The mixture was homogenized at high speed for 2 min using a Polytron homogenizer (Kinematica AG, Littau, Switzerland); afterward 20 mL of a saturated solution of sodium chloride in water was added. The mixture was vigorously shaken for 30 s. After phase separation the lower layer was removed with a Pasteur pipet, dried with sodium sulfate, and filtered off. The chloroform was evaporated in a rotary vacuum pump. Finally the oil content was stored at −17 °C until analyzed.

Analysis of Triacylglycerols (TAGs). The TAGs were analyzed by high-performance liquid chromatography (HPLC). About 200 mg of hazelnut oil was dissolved in 2 mL of acetone. The solution was filtered through a 13-mm diameter and 0.45-μm pore Nylon filter (Lida, Kenosha, WI), using a glass syringe (Sigma, Alcobendas, Madrid, Spain); 10 μL of this solution was injected into the HPLC, a Waters 600E System controller liquid chromatograph (Millipore Corp., Milford, MA) equipped with a Waters 700 Satellite WISP (Millipore) automatic injector (200-μL Rheodyne loop), and connected to a model Sedex 45 light scattering detector (Sedere, Vitry sur Seine, France). The detector conditions were as follows: gain five, temperature of 40 °C, and air pressure of 200 KPa. The mobile phase, which passed through the column at a flow rate of 1 mL min^{-1} at 25 °C, was a mixture of acetone/acetonitrile (64:36, v/v). The stationary phase was a 0.25-m × 4.6-mm i.d. (5-μm pore) Spherisorb ODS 2 column (Teknokroma, Sant Cugat del Vallès, Barcelona, Spain). Data were recorded in a personal computer connected to the HPLC.

The TAGs were identified according to the logarithms of their retention time relative to trioleylglycerol (OOO), based on previous published works (Parcerisa et al., 1994, 1995b). The TAGs were quantified according to their percentage area obtained by integration of the peak as a semiquantitative method.

Analysis of Phospholipids. Phospholipids were resolved and quantified by HPLC following the method proposed by Becard and co-workers (Becard et al., 1990). Approximately 550 mg of hazelnut oil was dissolved in 2 mL of chloroform; then solvent was added to a final volume of 5 mL. The solution was filtered through a 13-mm diameter and 0.45-μm pore Nylon filter (Lida) using a 10-mL glass syringe; 35 μL of this solution was injected into the liquid chromatograph, a Waters 600E System controller (Millipore Corp.), using a Waters 700 Satellite WISP automatic injector (Millipore) (200-μL Rheodine loop), which was connected to a model Sedex 45 light scattering detector (Sedere). The detector conditions were as follows: gain six, temperature of 53 °C, and air pressure of 200 KPa. The solvent system had two liquid phases: phase A, chloroform/methanol/concentrated ammonia (400:97.5:2.5, v/v/v), and phase B, chloroform/methanol/water/concentrated ammonia (300:170:27.5:2.5, v/v/v). The two phases, placed in solvent pumps A and B, passed through the column at 25 °C according to the following gradient system: the percentage of mobile phase A showed a linear decrease from 100% to 0% at min 14, which was mantained until min 23, when it began a linear increase, and reached 100% at min 29, remaining there until min 34. In the same way the percentage of mobile phase B showed a linear increase to 100% at min 14, which was maintained until min 23, when it began a linear decrease, and reached 0% at min 29, remaining there until min 34. The stationary phase was a 0.25-m × 4-mm i.d. (10-μm pore) Spherisorb W column (Teknokroma). Data were recorded in a personal computer connected to the HPLC system.

Phospholipids were identified by comparison of their retention time with pure standard phospholipids: phosphatidylcholine (PC), phosphatidylinositol (PI), phosphatidylethanolamine (PE), and phosphatidic acid (PA). Phospholipids were quantified using phosphatidylcholine as an external standard. The calibration curve was prepared by injecting into the HPLC 35 μL of the following solutions of phosphatidylcholine in chloroform: 0.05, 0.1, 0.2, 0.4, 0.8, and 1.0 mg mL^{-1}. The same response factor was assumed for PI and PE.

Fatty Acid Composition of TAGs and Phospholipids. TAGs and phospholipids (PL) were separated by thin-layer chromatography (TLC) on 0.25-mm film thickness silica gel G plates (0.2 m × 0.2 m x 0.02 m) (Merck, Darmstadt, Germany).

The TAGs were resolved with a solvent system composed of hexane/diethyl ether/concentrated acetic acid (80:20:1, v/v/v) (Kates, 1986). After development, the mobile phase was allowed to evaporate. The TAG band was identified by comparison of its retardation factor value (R_f) with standard trioleylglycerol, which was run on the same TLC plate. The TAG were revealed as yellow-brown spots by exposing the plate to iodine vapor for 5 min; then the silica gel containing the TAG was scraped off and stirred with 5 mL of a mixture of chloroform/methanol (2:1, v/v) for 5 min in darkness.

Phosphatidylcholine (PC) was separated by TLC using a solvent system composed of chloroform/methanol/concentrated ammonia (65:30:4, v/v/v) (Kates, 1986). After development, the plate was dried out. The PC band was identifed by comparison of its R_f value with standard PC, which was run on the same TLC plate. The PC appeared as a yellow-brown spot after exposing the plate to iodine vapor. The silica gel spot containing PC was scraped off and stirred with 5 mL of a mixture of chloroform/methanol (2:1, v/v) for 5 min in darkness. During TLC separation because of the small amounts of other phospholipids found (PI, PA, and PE), they were not analyzed for their fatty acid composition.

The TAG and PC solutions were filtered using a vacuum pump system, transferred to round-bottom flasks, and concentrated under vacuum at 25 °C. Each solution was transferred with a Pasteur pipet to a 10-mL glass screw-capped test tube; 3 mL of a solution of BF_3 methanol complex (20%, wt) was added to each tube; then the mixture was heated for 10 min in a boiling water bath and cooled under cold running water; 1 mL of hexane was then added. The mixture was vigorously shaken for 30 s; then 2 mL of a solution of saturated sodium chloride in water was added. After phase separation, the upper layer was suctioned with a Pasteur pipet. The lower layer was extracted two more times with 1 mL of hexane. The hexane solutions of FAME were mixed together, dried with sodium sulfate, and filtered off. The final solution of FAME was concentrated under a stream of nitrogen.

The FAME were analyzed by gas−liquid chromatography (GLC). The sample, 1−3 μL, was injected into the gas chromatograph (GC), a Hewlett-Packard (HP) 5890 series II (Little Falls, Wilmington, DE), with a flame ionization detector (FID). The GC was equipped with a 60-m fused silica capillary column (0.25-mm i.d. and 0.20-μm film thickness) coated with 90% polybiscyanopropylsiloxane and 10% cyanopropylphenylsiloxane (Supelco, Alcobendas, Madrid, Spain). The oven temperature was programmed as follows: 160 °C for 2 min, raised to 200 °C at 2 °C min^{-1}; this final temperature was maintained for 10 min. The injector and detector temperatures were 200 and 280 °C, respectively. Helium, grade 5.0 (Air Liquide España, Madrid, Spain), was used as carrier gas at a flow rate of 0.97 mL min^{-1} (181.9 KPa). Nitrogen (N_2) was used as a makeup (Air Liquide España). The split ratio was 30:1. Data were processed with the HP 3365 series II Chem station software (version A.03.21) for Windows 3.1 (Microsoft Iberica, Madrid, Spain) using a HP Vectra personal computer attached to the GC/FID.

Table 1. Triacylglycerol Contents (X, Mean, and SD, Standard Deviation) of Developing Lipid Fraction of Hazelnut Samples

	TAG[b]																					
	LLL[c]		LLO[c]		LLP[c]		LLO[c]		PLO[c]		PLP[c]		OOO[c]		POO[c]		POP[c]		PPP[c]		SOO[c]		SOP[c]	
DAF[a]	X	SD	X	SD	X	SD	X	SD	X	SD	X	SD	X	SD	X	SD	X	SD	X	SD	X	SD	X	SD
134	0.6	0.00	3.2	0.28	0.2	0.00	17.9	0.42	2.7	0.42	0.1	0.00	55.7	1.27	14.2	0.35	0.5	0.21	0.1	0.00	4.5	0.28	0.4	0.07
141	0.7	0.07	3.0	0.42	0.3	0.07	9.9	11.11	2.4	0.63	0.1	0.00	65.9	7.00	13.4	1.90	0.3	0.07	0.1	0.07	4.1	0.63	0.1	0.14
148	0.4	0.00	2.1	0.14	0.2	0.07	13.8	1.13	1.8	0.14	0.1	0.00	65.6	0.56	12.4	1.06	0.3	0.07	0.1	0.00	3.2	0.49	0.2	0.07
155	0.4	0.00	1.9	0.14	0.2	0.07	14.3	0.42	2.1	0.35	0.1	0.00	60.9	1.48	14.2	0.98	0.4	0.00	0.0	0.00	5.2	0.35	0.4	0.07
162	0.4	0.00	1.8	0.07	0.2	0.00	13.7	0.14	1.5	0.00	0.1	0.00	65.1	0.21	12.1	0.56	0.6	0.00	0.0	0.00	4.0	0.28	0.5	0.00
169	0.5	0.07	2.3	0.14	0.2	0.00	15.8	0.84	1.8	0.07	0.1	0.07	63.2	1.41	12.3	1.48	0.3	0.14	0.1	0.07	3.4	0.21	0.2	0.07
176	0.5	0.00	2.1	0.14	0.2	0.00	15.7	0.91	1.5	0.07	0.1	0.00	63.7	0.49	11.7	0.98	0.2	0.07	0.1	0.07	4.1	0.35	0.3	0.07

[a] DAF, days after flowering. [b] Triacylglycerol contents expressed as relative percent (%) of total TAG. [c] Trilinoleylglycerol (LLL), dilinoleyl-oleylglycerol (LLO), dilinoleyl-palmitoylglycerol (LLP), linoleyl-dioleylglycerol (LOO), palmitoyl-linoleyl-oleylglycerol (PLO), palmitoyl-linoleyl-palmitoylglycerol (PLP), trioleylglycerol (OOO), palmitoyl-dioleylglycerol (POO), palmitoyl-oleyl-palmitoylglycerol (POP), tripalmitoylglycerol (PPP), stearoyl-dioleylglycerol (SOO), stearoyl-oleyl-palmitoylglycerol (SOP). Data are means of duplicate results.

The FAME were identified by comparing their retention times and equivalent chain length (Hofstetter et al., 1965) with those for standard FAME. The FAME were quantified according to the percentage area obtained by integration of the peak as a semiquantitave method (Parcerisa et al., 1995b).

Statistical Analysis. The statistical analyses were carried out using the Statistical Package for Social Sciences (SPSS) (Hispanoportuguesa SPSS, Madrid, Spain) version 6.1.3 for Windows 95 (Microsoft Iberica).

Reagents and Standards. All the solvents used (diethyl ether, chloroform, methanol, and hexane) were reagent grade and obtained from Panreac (Montcada i Reixac, Barcelona, Spain). The HPLC grade solvents (acetone, acetonitrile, chloroform, and methanol) all were purchased from SDS (Peypin, France). Water for HPLC (Milli-Q) was obtained from a water purification system (Millipore, Milford, MA). The 25% solution of ammonia in water and concentrated acetic acid were analytical grade and obtained from Panreac. Anhydrous sodium sulfate, potassium hydroxide, iodine, and sodium chloride were reagent grade and obtained from Panreac. The BF_3 methanol complex (20%, wt) was purchased from Supelco. Standard FAME were a mixture of caprylic (8%, wt), capric (8%, wt), lauric (8%, wt), myristic (8%, wt), palmitic (11%, wt), palmitoleic (5%, wt), stearic (8%, wt), oleic (5%, wt), linoleic (5%, wt), linolenic (5%, wt), eicosanoic (8%, wt), behenic (8%, wt), erucic (5%, wt), and lingnoceric (5%, wt) fatty acid methyl esters purchased from Sigma. The standard trioleylglycerol was obtained from Sigma. The standard phospholipids (PC, PA, PI, and PE) were purchased from Sigma as part of a kit.

RESULTS AND DISCUSSION

Triacylglycerol Composition. The TAGs were fully separated and identified by HPLC with a light scattering detector. Several authors have used the same technique for TAG analysis of commercial oils (Reske et al., 1997; Pagnucco et al., 1997; Caboni et al., 1992). The composition of TAG is given in Table 1. Twelve compounds were identified and quantified as TAG by HPLC: trilinoleylglycerol (LLL), dilinoleyl-oleylglycerol (LLO), dilinoleyl-palmitoylglycerol (LLP), linoleyl-dioleylglycerol (LOO), palmitoyl-linoleyl-oleylglycerol (PLO), palmitoyl-linoleyl-palmitoylglycerol (PLP), trioleylglycerol (OOO), palmitoyl-dioleylglycerol (POO), palmitoyl-oleyl-palmitoylglycerol (POP), tripalmitoylglycerol (PPP), stearoyl-dioleylglycerol (SOO), and stearoyl-oleyl-palmitoylglycerol (SOP). The predominant TAG was OOO (ranging from 55.7% to 65.9%), followed by LOO (ranging from 9.9% to 17.9%), POO (ranging from 11.7% to 14.2%), and LLO (ranging from 1.8% to 3.2%) (Table 1). Other TAGs had smaller percentages: LLL, LLP, PLO, PLP, POP, PPP, SOO, and SOP (Table 1). These results are consistent with previously published studies (Parcerisa et al., 1994; Shewry et al., 1972; Bazan et al., 1975; Contini et al., 1991a,b). Table 1 reveals an opposite trend between OOO and LOO. This tendency is to be expected from the negative correlation found between OOO and LOO and the positive correlation found between oleic and linolenic fatty acids in previously published works (Parcerisa et al., 1993, 1994). In relation to the most important TAG, the trioleylglycerol proportion shows a small increase during hazelnut development, whereas the linoleyl-dioleylglycerol proportion reveals a slow decrease (Table 1).

Table 2. PL Content (X, Mean, and SD, Standard Deviation) of Developing Lipid Fraction of Hazelnuts

	phospholipids (g/kg)					
	PE[b]		PI[b]		PC[b]	
DAF[a]	X	SD	X	SD	X	SD
134	0.50	0.022	0.86	0.031	3.00	0.044
141	0.09	0.001	0.10	0.002	0.11	0.001
148	0.05	0.000	0.04	0.000	0.13	0.001
155	0.07	0.000	0.08	0.007	0.25	0.003
162	0.06	0.004	0.06	0.003	0.23	0.003
169	0.08	0.001	0.07	0.002	0.24	0.001
176	0.19	0.007	0.15	0.012	0.75	0.011

[a] DAF, days after flowering. [b] Phosphatidylethanolamine (PE), phosphatidylinositol (PI), phosphatidylcholine (PC). Data are means of triplicate results.

Composition of Phospholipids. Phospholipids were fully separated and identified by HPLC (Abidi et al., 1996; Balazs et al., 1996; Picchioni et al., 1996; Melton, 1992; Mounts et al., 1992). Table 2 gives the contents for PI, PE, and PC of the developing hazelnut lipid fraction. Figure 1 shows a hazelnut oil lipid fraction chromatogram in which PI, PE, PC, and PA are indicated. The PC fraction was the most important phospholipid, ranging from 0.11 to 3.0 g/kg of oil, whereas PE and PI were less predominant phospholipids, ranging from 0.05 to 0.5 and from 0.04 to 0.86 g/kg, respectively (Table 2). These contents are consistent with published data on the phospholipid content of other commercial oils (Morrison et al., 1995). Traces of PA were also found in all developing samples. Phospholipid content decreased between the early stage of hazelnut lipid fraction synthesis and the latest stage of maturity; however, a small increase of phospholipid content was observed at the end of maturity when hazelnuts fall off the tree and are ready to be picked up (Table 2). The statistical analysis showed a correlation between PC and PE ($r = 0.9876$, $p < 0.001$), PC and PI ($r = 0.9922$, $p < 0.001$), and PI and PE ($r = 0.9784$, $p < 0.001$) contents. Kates and Marshall (1975) reported a link between phospholipid biosynthesis in plants. At the

Table 3. Fatty Acid Content by GLC of TAGs and PC of Hazelnut Lipid Fraction during Development

	lipid class[a]				fatty acids[b] (%)					
DAF		$C_{12:0}$[c]	$C_{14:00}$	$C_{16:00}$	$C_{18:0}$	$C_{18:1}$	$C_{18:2}$	$C_{18:3}$	SFA[d]	MUFA[d]	PUFA[d]
	PC										
134		20.1	5.5	15.9	5.5	41.2	11.8	0.0	46.9	41.2	11.8
141		4.2	2.5	16.3	6.3	61.3	9.3	0.0	29.3	61.3	9.3
148		9.5	5.5	24.1	5.50	36.9	6.3	12.0	44.6	36.9	18.5
155		6.9	4.9	28.3	21.3	30.0	5.0	3.6	61.4	30.0	8.6
162		4.3	1.7	12.0	4.0	69.5	8.5	0.0	22.0	69.5	8.5
169		12.3	7.5	25.5	6.9	35.7	6.6	5.5	52.2	35.7	12.1
176		7.9	7.5	30.6	12.3	33.1	4.8	3.9	58.2	33.1	8.7
	TAG										
134		0.0	0.0	7.0	3.0	80.2	9.5	0.2	10.0	80.2	9.8
141		0.1	0.1	6.0	2.5	82.0	9.0	0.1	8.8	82.0	9.2
148		1.2	0.2	7.9	2.2	80.6	7.6	0.0	11.7	80.6	7.6
155		0.1	0.1	6.8	3.5	81.6	7.9	0.0	10.4	81.6	8.0
162		0.0	0.0	7.2	4.0	84.1	4.4	0.3	11.2	84.1	4.7
169		0.3	0.0	6.9	2.4	82.4	7.8	0.2	9.6	82.4	8.0
176		0.2	0.1	6.1	2.9	81.8	8.8	0.1	9.3	81.8	8.9

[a] Triacylglycerol, TAG; phosphatidylcholine, PC. [b] Lauric acid, $C_{12:0}$; myristic acid, $C_{14:00}$; palmitic acid, $C_{16:00}$; stearic acid, $C_{18:0}$; oleic acid, $C_{18:1}$; linoleic acid, $C_{18:2}$; linolenic acid, $C_{18:3}$. [c] Fatty acids are defined by the number of carbon atoms followed by the number of double bonds after the colon. [d] SFA, saturated fatty acids ($C_{12:0} + C_{14:00} + C_{16:00} + C_{18:0}$); MUFA, monounsaturated fatty acids ($C_{18:1}$); PUFA, polyunsaturated fatty acids ($C_{18:1} + C_{18:2}$).

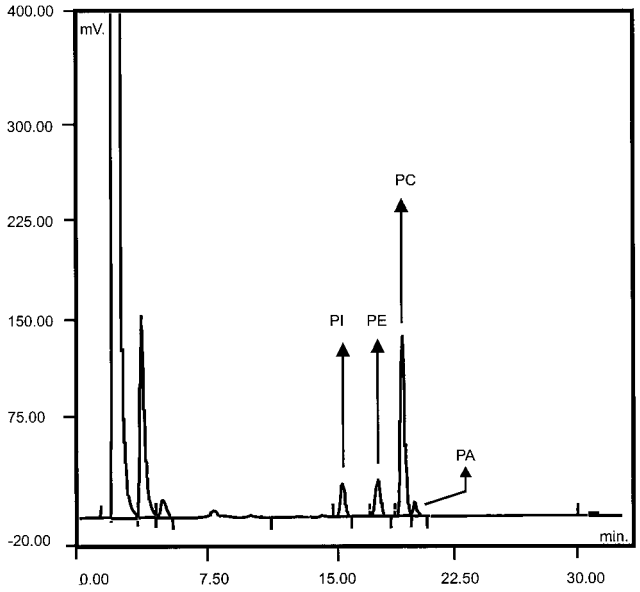

Figure 1. HPLC of hazelnut oil lipid fraction using a light scattering detector (PI, phosphatidylinositol; PE, phosphatidylethanolamine; PC, phosphatidylcholine; PA, phosphatidic acid). HPLC conditions were as follows: mobile phase A, chloroform/methanol/concentrated ammonia (400:97.5:2.5, v/v/v); mobile phase B, chloroform/methanol/water/concentrated ammonia (300:170:27.5:2.5, v/v/v); sationary phase, 0.25 m x 4-mm i.d. (10-mm pore) Spherisorb W column.

beginning of hazelnut development phospholipid contents show their highest value, followed by a steep decrease; however, as hazelnut maturity approaches phospholipid contents reveal a small increase (Table 2).

Fatty Acid Composition of Triacylglycerols and Phosphatidylcholine. We isolated enough samples of TAG and PC by preparative TLC to change their fatty acid moieties into methyl esters. Table 3 shows the fatty acid composition of the TAG fraction and PC. Oleic, linoleic, lauric, myristic, palmitic, stearic, and linolenic were the most abundant fatty acids in TAGs and PC, which confirmed previously published data on hazelnut oil (Parcerisa et al., 1997). Oleic and linoleic were the predominant fatty acids in TAGs, ranging from 80.2% to 84.1% and from 4.4% to 9.5%, respectively. Oleic and palmitic acids were the major fatty acids in phosphatidylcholine, ranging from 30.0% to 69.5% and from 12.0% to 30.6%, respectively (Table 3).

The one-way analysis of variance (one-way ANOVA) showed that SFAs ($C_{12:0} + C_{14:0} + C_{16:0} + C_{18:0}$) (Table 3) were present in different proportions in TAGs and PC (11.7% and 61.4%, respectively) ($p < 0.001$). The PC had the highest percentage in SFA, while TAG had the lowest. The statistical analysis also revealed that MUFA ($C_{18:1}$) were present in different proportions in TAG and PC (84.1% and 69.5%, respectively) ($p < 0.001$). The TAG had the highest percentage in MUFA, whereas PC had the lowest. The one-way ANOVA did not reveal any statistical difference between PC and TAG in their PUFA ($C_{18:2} + C_{18:3}$). Lauric and myristic fatty acids show a low proportion in the TAG, whereas the same fatty acids reveal a higher percentage in PC. The statistical analysis revealed a correlation between MUFA and PUFA in the TAG fraction ($r = -0.7818$, $p < 0.001$). In a similar way the statistical analysis revealed a correlation between SFA and MUFA ($r = -0.9717$, $p < 0.001$) in the PC fraction. In former papers we reported a correlation between monounsaturated and polyunsaturated fatty acids in the overall fatty acid composition of hazelnut lipid fraction (Parcerisa et al., 1993; 1995b). These results are similar to those found by other researchers in oilseeds and nuts (Munshi and Sukhija, 1984; Gurr et al., 1974; Appleby et al., 1974; Ichihara and Noda, 1980). Despite efforts to isolate PE, PI, PA, and PS by TLC, we could not obtain enough samples of these compounds to identify their fatty acid profile.

Hopefully, further research will seek to assess the composition of hazelnut lipid fraction during kernel development for nonsaponifiable compounds (tocopherols and sterols).

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