Fatty Acid Evolution During the Storage of Ground, Roasted Coffees

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ABSTRACT: During storage, the quality of coffee decreases, partially because coffee lipids degrade by lipolysis and oxidation. The aim of this study was to compare the evolution of the fatty acid (FA) profile in two ground, roasted coffees—Brazilian Arabica 100% (A100), and a blend of Brazilian Arabica 80% and Indian Cherry Robusta 20% (A80/R20)—throughout 180 d of storage. Linoleic acid (40.1% in A100, 40.2% in A80/R20) and palmitic acid (36.4% in both samples) were the main total FA. No significant change in the FA profile was observed throughout the storage period. A significantly higher total free fatty acid (FFA) initial concentration appeared in A80/R20 coffee (1108.5 mg/100 g fat) compared with A100 coffee (730.2 mg/100 g fat). Except for the first week of storage, similar FA oxidation patterns were found for both coffees, but FFA generation was faster in the A80/R20 blend than in A100.

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KEY WORDS: Coffee, fatty acids, ground and roasted coffee, lipids, storage period.

Espresso coffee consumption is increasing on a domestic level throughout the world. Likewise, ground, roasted coffee is one of the most widely consumed products from a commercial point of view. However, just after roasting, the aging process begins for most coffees. During storage, ground, roasted coffee loses its aroma and fresh flavor because of lipid oxidation and the degradation of some aroma compounds (1).

The amount of lipids in roasted coffee is around 17% on a dry weight basis in arabicas and around 11% in robustas (2). The majority of compounds are triacylglycerols (TAG) (75%), whereas free fatty acids (FFA) make up only 1% of the total lipid content (3). Coffee lipid degradation can take place by two different simultaneous mechanisms: acylglycerol hydrolysis and oxidation. Lipolysis releases FFA, which are more prone to oxidation than esterified fatty acids (FA), particularly long-chain unsaturated FA (4). Even coffees stored under vacuum and low oxygen pressure show lipid oxidation because of the initial presence of free radicals, whose formation is promoted by pyrolysis reactions during the roasting process (5,6). The autooxidation of lipids is a complex process, but model studies have revealed that their rate of autooxidation is affected by the FA composition, the degree of unsaturation in the FA moieties, the presence and activity of pro- and antioxidants in the food matrix, and the partial pressure of the oxygen (4). Baesso *et al*. (5) showed that the most important factor in coffee deterioration is the specific surface area of coffee particles in contact with oxygen from the air. Hence, the coffee-grinding process is critical for the preservation of coffee quality because of the increase in specific surface area.

The FA composition of green and roasted coffees has been described by some authors (3,7–9). The influences of several factors (temperature, oxygen, and water activity) on lipid oxidation during roasted coffee storage have been studied (10–16). However, no studies concerning the evolution of the FA profile in ground, roasted coffee during storage have been reported.

The aim of this research was to study and compare the FA profiles of two ground, roasted coffees (Arabica and an Arabica/Robusta blend) throughout their storage period as influenced by the grinding grade (fine and coarse).

EXPERIMENTAL PROCEDURES

Materials. Brazilian Arabica and Indian Cherry Robusta green coffees were provided by a local factory.

Pure reference standards of lauric, myristic, palmitic, palmitelaidic, palmitoleic, heptadecanoic, stearic, elaidic, oleic, linolelaidic, linoleic, linolenic, arachidic, arachidonic, eicosapentaenoic, behenic, brassidic, erucic, and docosahexaenoic acid methyl esters, and heptadecanoic acid were purchased from Sigma (Steinheim, Germany).

Sample preparation. Two types of green coffee, Brazilian Arabica and Indian Cherry Robusta, were roasted separately using a precision coffee roaster (Hearthware™) at 260°C for 5 min and 20 s. Roasted coffees were maintained in stainless-steel containers for 24 h to release CO_2. The roasted coffees were then blended in commercial percentages, and two roasted coffee samples were prepared for analysis: Brazilian Arabica 100% (A100) and a blend of Brazilian Arabica 80% and Indian Cherry Robusta 20% (A80/R20).

(i) Grinding grade selection. Coffee beans were ground by means of an automatic M01 Azkoyen grinder. The grinder had 19 grinding levels, 1 for the coarsest point level and 19 for the finest. To select the grinding grade, espresso coffees were brewed from each sample with an experimental prototype espresso coffeemaker at the same conditions applied by Andueza *et al*. (17). A volume of 40 ± 2 mL and a percolation time between 18 and 24 s were used. Levels 3 and 11 were selected as coarse and fine grinds, respectively.

(ii) Packaging. Ground, roasted coffee samples were packaged in 250-g trilaminated, waterproof, opaque bags under

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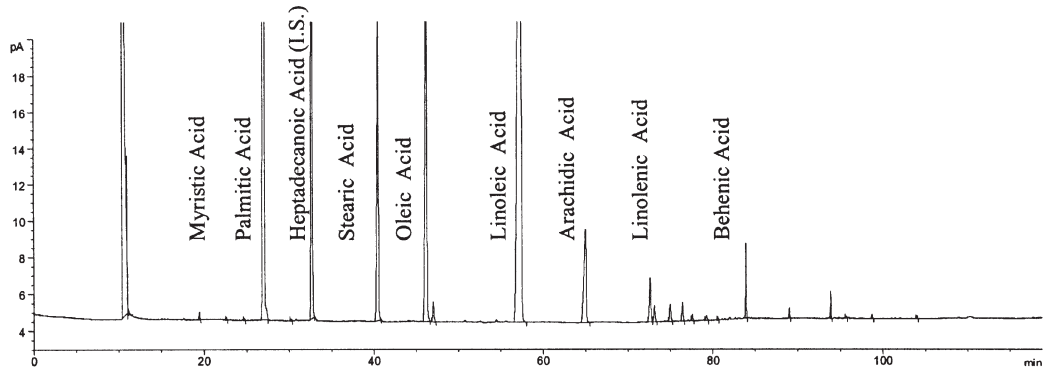

FIG. 1. Chromatogram of the FA in a ground, roasted Brazilian Arabica 100% coffee sample (A100). I.S., internal standard.

vacuum using a manual packer (Ramon Series VP Model 450) and were stored at 25°C for 180 d.

Methods. (i) Moisture. Moisture was determined by oven-drying 5 g of coffee to a constant weight (14 h, 103 ± 2°C) using the standard method (18). Each sample was analyzed in triplicate.

(ii) Total fat. Total fat was extracted with petroleum ether from previously dried samples by the Soxhlet method (19). Each sample was analyzed in triplicate.

(iii) Total FA analysis. Total lipids were extracted using trichloromethane and methanol (2:1, vol/vol) according to the method of Bligh and Dyer (20). The FA analysis was performed by GC after previous methylation of the total lipids with BF_3/methanol (21). A 0.5-μL quantity of the final solution was injected into an SP-2560 capillary column (100 m × 0.25 mm × 0.2 μm; Supelco, Bellefonte, PA) in a gas chromatograph (Agilent 6890). The injector temperature was 250°C, and the carrier gas was helium (89.3 mL/min linear speed). The oven temperature was held at 165°C for 70 min, then raised at 4°C/min to 220°C and held there for 35 min. The FID detector temperature was maintained at 250°C. Peaks were identified by comparison of their retention times with those of standard compounds (Fig. 1). Individual FA were quantified using heptadecanoic acid methyl ester as an internal standard. Each sample was analyzed in triplicate.

(iv) FFA analysis. Total lipids were extracted using trichloromethane and methanol (2:1, vol/vol) according to the method of Bligh and Dyer (20). The extraction of FFA from total lipids was carried out using an activated ion-exchange resin (Amberlite A-26) following the method of Needs *et al.* (22). FFA were then methylated directly. A 1-μL quantity of the final solution was injected into the gas chromatograph, and the resultant peaks were identified by comparison of their retention times with those of standard compounds. Individual FFA were quantified as mg FFA/100 g fat using heptadecanoic acid as the internal standard. Each sample was analyzed in triplicate.

TABLE 1
Moisture (g/100 g) and Fat (g/100 g d.m.) Evolution in Finely and Coarsely Ground, Roasted Coffee Samples During Storage Under Vacuum at 25°C[a]

Time (d)	0	30	90	180
A100				
Moisture				
Fine	2.8 ± 0.1[b]	1.8 ± 0.1[a]	1.8 ± 0.1[a]	1.6 ± 0.0[a]
Coarse	2.8 ± 0.1[b]	1.9 ± 0.0[a]	1.8 ± 0.1[a]	1.9 ± 0.0[a]
Fat				
Fine	14.4 ± 0.1[a]	14.2 ± 0.0[a]	14.3 ± 0.1[a]	14.8 ± 0.1[a]
Coarse	12.4 ± 0.1[a]	12.2 ± 0.1[a]	12.4 ± 0.1[a]	12.8 ± 0.1[a]
A80/R20				
Moisture				
Fine	2.6 ± 0.1[b]	1.8 ± 0.0[a]	1.8 ± 0.0[a]	1.9 ± 0.1[a]
Coarse	2.6 ± 0.1[c]	1.7 ± 0.0[a]	1.8 ± 0.0[a,b]	1.9 ± 0.0[b]
Fat				
Fine	13.3 ± 0.1[a]	13.0 ± 0.1[a]	13.2 ± 0.0[a]	13.4 ± 0.2[a]
Coarse	11.0 ± 0.1[a]	10.9 ± 0.2[a]	11.1 ± 0.1[a]	11.1 ± 0.1[a]

[a]All values are shown as means ± SD ($n = 3$). In each row, different roman superscript letters indicate significant differences ($P < 0.05$) among different analysis times in each kind of sample. A100, Brazilian Arabica 100% coffee; A80/R20, a blend of Brazilian Arabica 80% and Indian Cherry Robusta 20% coffees; d.m., dry matter.

TABLE 2
Total FA in Ground, Roasted Coffee Samples[a]

	A100	A80/R20	LS
Lauric (12:0)	ND	ND	—
Myristic (14:0)	ND	ND	—
Palmitic (16:0)	36.4 ± 1.7	36.4 ± 1.0	NS
Palmitelaidic (16:1t)	ND	ND	—
Palmitoleic (16:1)	ND	ND	—
Stearic (18:0)	8.8 ± 0.3	9.0 ± 0.6	NS
Elaidic (18:1t)	ND	ND	—
Oleic (18:1)	9.0 ± 0.2	9.2 ± 0.1	NS
Linolelaidic (18:2t,t)	ND	ND	—
Linoleic (18:2)	40.1 ± 0.5	40.2 ± 0.3	NS
Linolenic (18:3)	1.1 ± 0.1	1.1 ± 0.0	NS
Arachidic (20:0)	3.2 ± 0.2	3.2 ± 0.1	NS
Arachidonic (20:4)	ND	ND	—
Eicosapentaenoic (20:5)	ND	ND	—
Behenic (22:0)	0.8 ± 0.1	0.7 ± 0.1	NS
Brassidic (22:1t)	ND	ND	—
Erucic (22:1)	ND	ND	—
Docosahexaenoic (22:6)	ND	ND	—
∑SFA	49.4 ± 0.8	49.3 ± 0.4	NS
∑MUFA	9.0 ± 0.2	9.2 ± 0.1	NS
∑PUFA	41.4 ± 0.6	41.4 ± 0.3	NS
∑PUFA/∑SFA	0.8	0.8	NS
∑MUFA/∑SFA	0.2	0.2	NS

[a]All values are shown as means ± SD ($n = 3$). Results are expressed as the percentage of total methyl esters identified. ND, not detected; SFA, saturated FA; MUFA, monounsaturated FA; PUFA, polyunsaturated FA; LS, level of significance; NS, not significant ($P > 0.05$); for other abbreviations see Table 1.

(v) Data analysis. A one-way ANOVA and the T-Tukey b *a posteriori* test, with a level of significance of 95%, were applied for each coffee sample along the time continuum. Student's *t*-test was applied each time between coffee samples or grinding grades. The software package SPSS v. 9.0 (SPSS Inc., Chicago, IL) was used.

RESULTS AND DISCUSSION

The moisture and fat contents for the coffees stored under vacuum are shown in Table 1. A significant decrease in moisture was observed in the first 30 d, but after that, it remained constant up to the end of the study (180 d). The fat content remained at

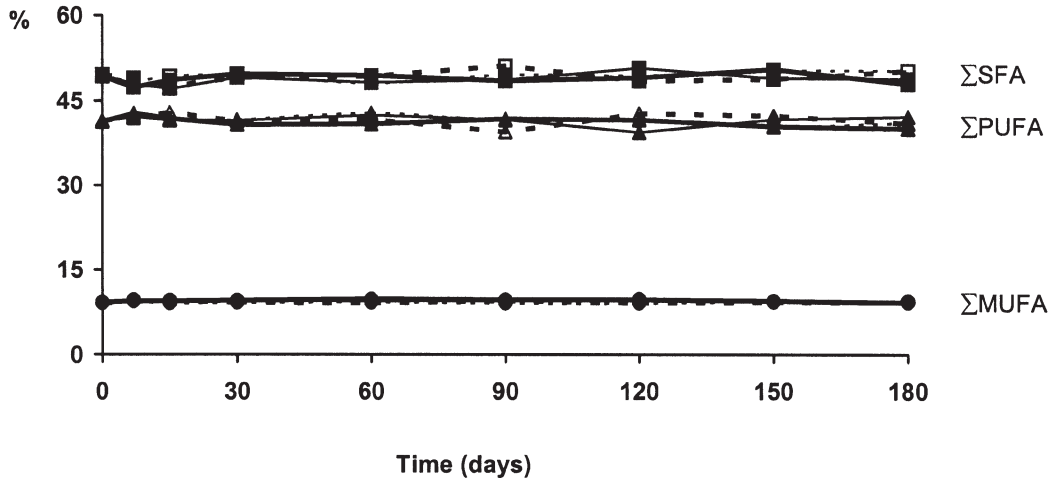

FIG. 2. Evolution of the total FA in ground, roasted coffee samples under vacuum during storage at 25°C. Finely ground A100 (light dashed line, open symbols), coarsely ground A100 (heavy dashed line, open symbols), finely ground A80/R20 (light solid line, filled symbols), coarsely ground A80/R20 (heavy solid line, filled symbols), saturated FA (SFA) (■), monounsaturated FA (MUFA) (●), and polyunsaturated FA (PUFA) (▲). A80/R20, a blend of Brazilian Arabica 80% and Indian Cherry Robusta 20% coffees; for other abbreviation see Figure 1.

TABLE 3
FFA (mg/100 g fat) Evolution in A100 Finely and Coarsely Ground, Roasted Coffee During Storage Under Vacuum at 25°C[a]

Time (d)	0	7	15	30	60	90	120	150	180
Myristic (14:0)									
Fine	Trace	0.4 ± 0.0	Trace	0.2 ± 0.0	0.2 ± 0.0	0.6 ± 0.0	Trace	0.8 ± 0.1	1.2 ± 0.1
Coarse	Trace	Trace	Trace	0.2 ± 0.0	Trace	Trace	Trace	1.8 ± 0.2	1.1 ± 0.1
LS	—	—	—	NS	—	—	—	***	NS
Palmitic (16:0)									
Fine	298.8 ± 13.2^{a}	537.9 ± 5.7^{c}	530.5 ± 24.9^{c}	558.9 ± 10.0^{c,d}	522.4 ± 29.4^{c}	473.9 ± 16.7^{b}	466.9 ± 10.2^{b}	595.8 ± 3.0^{d,e}	605.2 ± 21.3^{e}
Coarse	298.8 ± 13.2^{a}	479.1 ± 6.1^{c}	401.3 ± 16.4^{b}	498.0 ± 16.3^{c}	413.3 ± 13.9^{b}	428.7 ± 10.7^{b}	426.2 ± 6.9^{b}	668.0 ± 12.7^{d}	652.5 ± 1.6^{d}
LS	NS	***	**	**	**	*	**	***	NS
Stearic (18:0)									
Fine	53.0 ± 1.5^{a}	105.5 ± 3.9^{d,e}	112.1 ± 3.6^{e}	110.7 ± 3.0^{e}	96.5 ± 4.5^{c}	96.1 ± 2.8^{c}	78.0 ± 0.8^{b}	107.1 ± 1.7^{e}	99.5 ± 3.5^{c,d}
Coarse	53.0 ± 1.5^{a}	101.1 ± 2.1^{e}	79.7 ± 0.3^{b,c}	86.6 ± 3.2^{d}	78.0 ± 2.9^{b}	81.1 ± 1.0^{b,c}	84.0 ± 2.0^{c,d}	99.8 ± 1.6^{e}	109.1 ± 0.3^{f}
LS	NS	NS	**	***	**	***	**	**	*
Oleic (18:1)									
Fine	62.3 ± 2.7^{a}	109.9 ± 0.8^{d}	108.7 ± 2.1^{d}	111.4 ± 2.6^{d,e}	102.4 ± 5.4^{c}	97.9 ± 1.6^{c}	91.0 ± 1.0^{b}	118.8 ± 0.2^{b}	116.3 ± 2.3^{e,f}
Coarse	62.3 ± 2.7^{a}	99.5 ± 1.7^{c}	84.5 ± 3.6^{b}	98.5 ± 2.1^{c}	81.6 ± 2.3^{b}	84.3 ± 1.7^{b}	87.0 ± 1.3^{b}	123.7 ± 1.5^{d}	123.5 ± 0.7^{d}
LS	NS	***	***	**	**	***	*	**	*
Linoleic (18:2)									
Fine	276.3 ± 14.3^{a}	431.9 ± 10.6^{d}	408.2 ± 9.0^{c,d}	427.2 ± 17.2^{c,d}	412.4 ± 24.0^{c,d}	371.2 ± 7.6^{b}	396.2 ± 7.1^{b,c}	486.1 ± 2.9^{e}	510.5 ± 4.5^{e}
Coarse	276.3 ± 14.3^{a}	377.0 ± 4.7^{d}	297.1 ± 18.4^{a,b}	426.0 ± 6.7^{e}	325.1 ± 7.5^{b,c}	336.9 ± 10.9^{c}	340.5 ± 5.5^{c}	576.1 ± 22.2^{g}	527.4 ± 7.6^{f}
LS	NS	***	***	NS	**	*	***	*	*
Linolenic (18:3)									
Fine	7.6 ± 0.3^{b}	11.0 ± 1.1^{c}	10.0 ± 0.7^{c}	10.0 ± 0.6^{c}	10.3 ± 1.0^{c}	5.4 ± 0.5^{a}	10.7 ± 0.2^{c}	13.0 ± 0.4^{d}	14.2 ± 0.9^{d}
Coarse	7.6 ± 0.3^{a,b}	9.0 ± 0.3^{b}	6.9 ± 0.5^{a}	11.4 ± 0.2^{c}	8.2 ± 0.8^{a,b}	8.2 ± 0.2^{a,b}	8.5 ± 0.4^{a,b}	15.7 ± 1.6^{e}	13.9 ± 0.3^{d}
LS	NS	*	**	*	NS	***	***	*	NS
Arachidic (20:0)									
Fine	25.5 ± 0.9^{a}	52.9 ± 3.0^{d,e}	58.7 ± 0.8^{f}	56.9 ± 2.8^{e,f}	45.3 ± 2.9^{c}	48.5 ± 1.5^{c,d}	35.1 ± 1.5^{b}	51.8 ± 2.1^{d,e}	43.9 ± 2.0^{c}
Coarse	25.5 ± 0.9^{a}	54.0 ± 0.9^{e}	38.5 ± 0.6^{b,c}	40.2 ± 1.9^{b,c}	36.7 ± 4.3^{b}	38.7 ± 0.7^{b,c}	42.1 ± 2.5^{c}	39.9 ± 1.1^{b,c}	48.3 ± 0.3^{d}
LS	NS	NS	***	***	*	***	*	***	**
Behenic (22:0)									
Fine	6.6 ± 0.5^{a}	15.1 ± 0.8^{d,e}	16.2 ± 0.8^{e}	16.2 ± 0.9^{e}	11.5 ± 0.9^{c}	14.5 ± 1.0^{d,e}	8.7 ± 0.7^{b}	14.1 ± 0.4^{d}	11.0 ± 0.3^{c}
Coarse	6.6 ± 0.5^{a}	16.2 ± 0.3^{e}	9.8 ± 0.7^{b,c,d}	10.3 ± 0.7^{b,c,d}	9.6 ± 1.1^{b,c}	9.3 ± 0.6^{b}	11.4 ± 1.2^{c,d}	9.3 ± 0.6^{b}	11.5 ± 0.2^{d}
LS	NS	NS	***	***	NS	***	*	***	NS
Total FFA									
Fine	730.2 ± 31.7^{a}	1264.8 ± 0.8^{c,d}	1244.3 ± 36.4^{c,d}	1291.7 ± 36.8^{d}	1201.2 ± 67.5^{c}	1108.0 ± 23.3^{b}	1086.7 ± 15.4^{b}	1387.6 ± 0.9^{e}	1402.0 ± 34.5^{e}
Coarse	730.2 ± 31.7^{a}	1135.5 ± 13.2^{d}	918.7 ± 38.0^{b}	1171.3 ± 30.2^{d}	952.6 ± 7.1^{b,c}	987.2 ± 23.6^{c}	999.6 ± 7.9^{c}	1534.4 ± 31.9^{e}	1488.3 ± 7.5^{e}
LS	NS	**	***	*	*	**	***	***	**

[a]All values are shown as means ± SD (n = 3). In each row, different roman superscript letters indicate significant differences ($P < 0.05$) among different times. For abbreviations see Tables 1 and 2. *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

TABLE 4
FFA (mg/100 g fat) Evolution in A80/R20 Finely and Coarsely Ground, Roasted Coffee During Storage Under Vacuum at 25°C[a]

Time (d)	0	7	15	30	60	90	120	150	180
Myristic (14:0)									
Fine	Trace	0.4 ± 0.1	0.4 ± 0.0	0.2 ± 0.0	Trace	Trace	0.7 ± 0.1	0.2 ± 0.0	1.4 ± 0.0
Coarse	Trace	Trace	Trace	0.3 ± 0.0	Trace	1.1 ± 0.2	1.0 ± 0.1	0.6 ± 0.0	0.7 ± 0.1
LS	—	—	—	***	—	—	*	***	**
Palmitic (16:0)									
Fine	464.2 ± 6.0^{b}	473.8 ± 12.4^{b}	495.1 ± 16.0^{b}	481.0 ± 4.4^{b}	425.6 ± 21.0^{a}	573.2 ± 24.1^{c,d}	598.2 ± 5.2^{d}	602.3 ± 8.4^{d}	554.8 ± 4.3^{c}
Coarse	464.2 ± 6.0^{c}	424.0 ± 4.4^{b}	378.2 ± 8.7^{a}	483.6 ± 31.9^{c,d}	401.4 ± 11.7^{a,b}	366.9 ± 4.9^{a}	566.0 ± 24.0^{e}	521.1 ± 12.5^{d}	516.6 ± 11.9^{d}
LS	NS	**	***	NS	NS	***	NS	***	**
Stearic (18:0)									
Fine	87.9 ± 1.2^{a}	87.8 ± 2.9^{a}	84.2 ± 7.3^{a}	82.7 ± 1.1^{a}	83.8 ± 3.3^{a}	102.2 ± 2.0^{b}	110.6 ± 0.5^{c}	128.4 ± 1.4^{d}	109.9 ± 1.1^{c}
Coarse	87.9 ± 1.2^{c}	83.2 ± 1.2^{c}	71.0 ± 3.7^{a}	87.3 ± 2.5^{c}	77.6 ± 2.6^{b}	71.9 ± 1.2^{a}	96.2 ± 3.2^{d}	84.5 ± 1.2^{c}	101.1 ± 3.2^{d}
LS	NS	NS	*	*	NS	***	**	***	*
Oleic (18:1)									
Fine	100.4 ± 1.5^{b}	98.0 ± 2.1^{b}	101.1 ± 2.6^{b}	98.1 ± 0.8^{b}	90.6 ± 2.8^{a}	119.3 ± 0.8^{c}	123.9 ± 1.7^{c}	132.2 ± 2.0^{d}	124.2 ± 2.3^{c}
Coarse	100.4 ± 1.5^{c}	92.0 ± 2.0^{b}	78.2 ± 2.5^{a}	104.9 ± 4.2^{c,d}	84.8 ± 5.1^{a}	81.5 ± 0.4^{a}	124.2 ± 3.5^{f}	108.1 ± 1.5^{d,e}	114.1 ± 0.9^{e}
LS	NS	*	***	*	NS	***	NS	***	**
Linoleic (18:2)									
Fine	391.8 ± 4.6^{b,c}	377.7 ± 6.7^{b}	408.2 ± 25.8^{c}	413.9 ± 5.22^{c}	339.0 ± 11.4^{a}	473.0 ± 1.4^{d,e}	463.6 ± 3.8^{d,e}	481.5 ± 13.0^{e}	450.2 ± 7.2^{d}
Coarse	391.8 ± 4.6^{c}	343.2 ± 5.1^{b}	290.1 ± 5.4^{a}	428.5 ± 19.5^{d}	317.5 ± 24.7^{a,b}	305.6 ± 3.9^{a}	537.4 ± 21.0^{f}	470.2 ± 9.7^{e}	443.1 ± 8.1^{d,e}
LS	NS	**	*	NS	NS	***	**	NS	NS
Linolenic (18:3)									
Fine	8.8 ± 0.6^{a,b}	9.0 ± 0.4^{a,b}	10.3 ± 1.4^{b,c}	10.1 ± 0.2^{b,c}	8.2 ± 0.6^{a}	11.8 ± 0.1^{c}	10.7 ± 0.4^{c}	11.5 ± 0.8^{c}	11.8 ± 0.0^{c}
Coarse	8.8 ± 0.6^{c}	7.7 ± 1.5^{b,c}	6.0 ± 0.2^{a}	10.4 ± 0.3^{d}	7.4 ± 0.6^{b}	7.2 ± 0.2^{a,b}	14.1 ± 0.7^{e}	11.6 ± 0.7^{d}	10.6 ± 0.9^{d}
LS	NS	**	*	NS	NS	***	**	NS	NS
Arachidic (20:0)									
Fine	42.0 ± 4.1^{a,b}	42.6 ± 1.0^{a,b}	36.2 ± 3.0^{a}	38.0 ± 1.7^{a}	42.6 ± 5.6^{a,b}	49.4 ± 1.5^{b,c}	53.6 ± 1.7^{c,d}	71.1 ± 3.5^{e}	59.6 ± 2.0^{d}
Coarse	42.0 ± 4.1^{b,c}	39.4 ± 1.1^{a,b,c}	33.6 ± 3.6^{a}	40.6 ± 2.8^{a,b,c}	39.2 ± 2.7^{a,b,c}	36.6 ± 0.7^{a,b}	44.6 ± 3.1^{c}	36.1 ± 1.0^{a,b}	52.3 ± 3.8^{d}
LS	NS	*	NS	NS	NS	***	*	***	*
Behenic (22:0)									
Fine	12.5 ± 1.4^{c,d}	11.0 ± 0.6^{b,c}	8.3 ± 0.8^{a}	10.2 ± 0.4^{a,b}	13.0 ± 0.3^{c,d}	13.8 ± 0.9^{d,e}	14.1 ± 0.5^{d,e}	21.3 ± 1.7^{f}	15.7 ± 0.1^{e}
Coarse	12.5 ± 1.4^{c}	9.0 ± 0.2^{a,b}	9.7 ± 1.2^{a,b,c}	11.3 ± 1.2^{b,c}	10.9 ± 1.2^{a,b,c}	10.1 ± 0.4^{a,b,c}	11.6 ± 1.2^{b,c}	8.2 ± 0.6^{a}	15.8 ± 1.2^{d}
LS	NS	**	NS	NS	NS	**	*	***	NS
Total FFA									
Fine	1108.5 ± 11.3^{b}	1101.3 ± 24.7^{b}	1144.8 ± 2.4^{c}	1133.0 ± 13.3^{b,c}	1002.7 ± 5.2^{a}	1343.5 ± 20.1^{d,e}	1375.5 ± 8.4^{e}	1448.7 ± 12.1^{f}	1327.8 ± 16.2^{d}
Coarse	1108.5 ± 11.3^{c}	1008.0 ± 12.8^{b}	867.6 ± 8.2^{a}	1167.3 ± 53.8^{c,d}	938.8 ± 38.9^{a,b}	880.2 ± 11.3^{a}	1426.1 ± 67.1^{f}	1240.5 ± 23.5^{d,e}	1254.5 ± 15.1^{e}
LS	NS	**	***	NS	NS	***	NS	***	**

[a]All values are shown as means ± SD (n = 3). In each row, different roman superscript letters indicate significant differences ($P < 0.05$) among different times. For abbreviations see Tables 1 and 2. *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

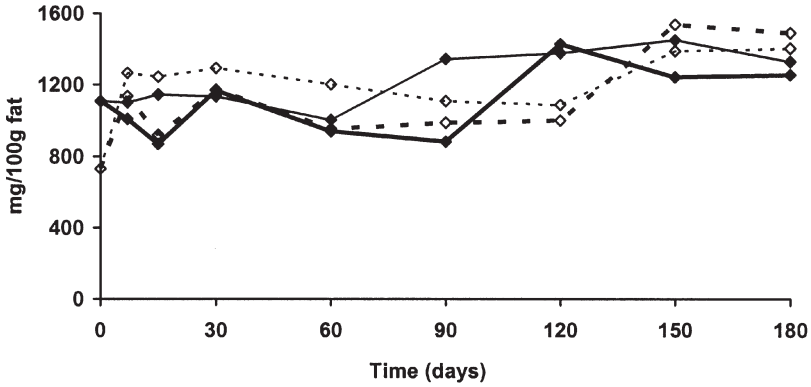

FIG. 3. Evolution of total FFA in the ground, roasted samples under vacuum during storage at 25°C. Finely ground A100 (light dashed line, open symbols), coarsely ground A100 (heavy dashed line, open symbols), finely ground A80/R20 (light solid line, filled symbols), and coarsely ground A80/R20 (heavy solid line, filled symbols). For abbreviations see Figures 1 and 2.

the same level throughout the storage period. Furthermore, a higher fat content was observed in A100 than in A80/R20, in agreement with the findings of Illy and Viani (2). Likewise, in accordance with the findings of Folstar (23), the fat content was higher in the finely ground samples. This result was due to an easier fat release from the interior of the cell to the surface.

FA profiles in the A100 and A80/R20 ground, roasted coffees are shown in Table 2. There were no significant differences in the FA percentages between A100 and A80/R20. Linoleic acid (40.1% in A100, 40.2% in A80/R20) and palmitic acid (36.4% in both samples) were found to be the main FA. Moderate and almost equal percentages of stearic acid (8.8–9.0%) and oleic acid (9.0–9.2%) were also found, whereas linolenic, arachidic, and behenic acids were present in minor amounts. These findings agreed with those reported by Martín *et al*. (8) and Speer and Kölling-Speer (9).

The evolution of FA in the ground, roasted coffee samples during storage at 25°C is shown in Figure 2. No significant changes in the FA percentages were observed throughout the storage period. Furthermore, significant differences were not observed in the FA profiles for either coffee variety or grinding grade.

Tables 3 and 4 show the FFA concentration of the coffee samples (A100 and A80/R20). Initially, among the seven detected FFA, palmitic acid (40.9–41.9%) and linoleic acid (35.3–37.8%) predominated. As previously written, linoleic acid was the predominant FA in the FA profile, which included the esterified and FFA. Hence, linoleic acid seemed to remain more esterified than palmitic acid. Moderate amounts of free stearic acid (7.2–7.9%) and free oleic acid (8.5–9.0%) were found. A significantly higher initial concentration of total FFA was found in the A80/R20 coffee (1108.5 mg/100 g fat in A80/R20 vs. 730.2 mg/100 g fat in A100). Similar results were reported by Speer and Kölling-Speer (9) for green coffee. Consequently, A80/R20 ground, roasted coffee was more prone to oxidation than A100.

In Figures 3, 4, and 5, the total FFA, saturated FA (SFA), monounsaturated FA (MUFA), and polyunsaturated FA (PUFA) evolution in the coffee samples is shown throughout the storage period. During the first week, a significant increase in all FFA,

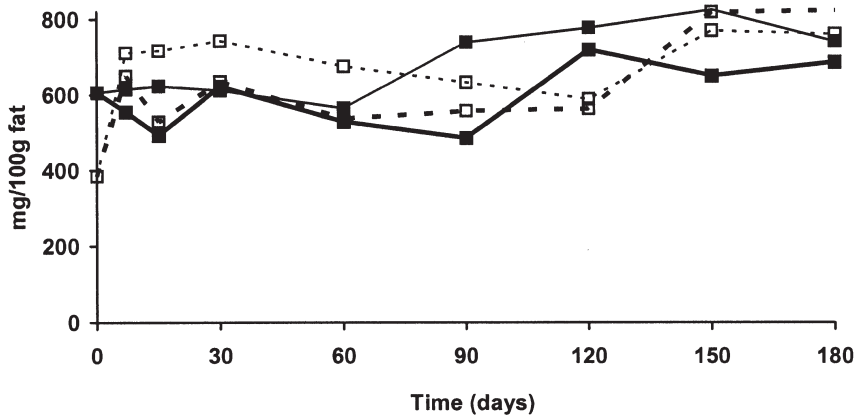

FIG. 4. Evolution of saturated FFA in the ground, roasted samples under vacuum during storage at 25°C. Finely ground A100 (light dashed line, open symbols), coarsely ground A100 (heavy dashed line, open symbols), finely ground A80/R20 (light solid line, filled symbols), and coarsely ground A80/R20 (heavy solid line, filled symbols). For abbreviations see Figures 1 and 2.

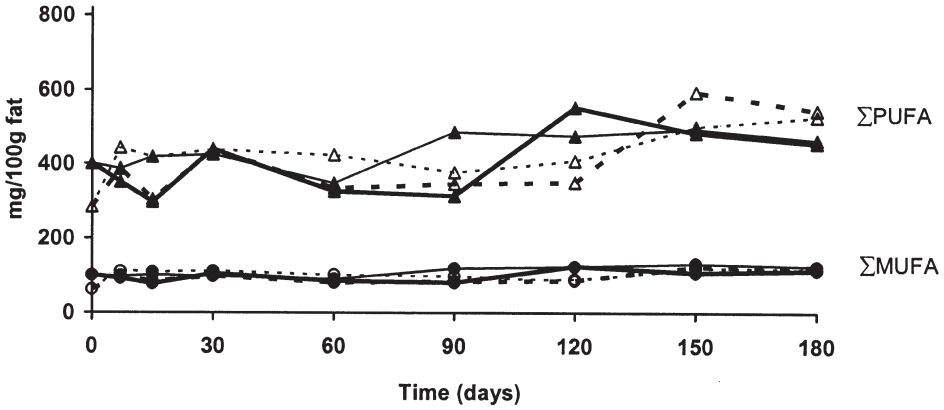

FIG. 5. Evolution of unsaturated FFA in the ground, roasted samples under vacuum during storage at 25°C. Finely ground A100 (light dashed line, open symbols), coarsely ground A100 (heavy dashed line, open symbols), finely ground A80/R20 (light solid line, filled symbols), and coarsely ground A80/R20 (heavy solid line, filled symbols). MUFA (●), and PUFA (▲). For abbreviations see Figures 1 and 2.

mainly in SFA, was observed for A100. For acylglycerols, the primary HO-groups in positions 1 and 3 of the glycerol molecule are preferentially esterified with SFA (4,23), implying that SFA would be released faster. Between 7 and 15 d, there was a significant decrease of FFA in the coarsely ground coffee. After that, a significant increase in FFA was found up to 30 d, probably owing to the onset of lipolysis reactions. However, in the finely ground coffees, significant FFA changes were not found between 7 and 30 d.

From 30 d, a significant FFA decrease occurred in all samples, showing the predominance of oxidation up to 60 d for finely ground A80/R20 and up to 90 d for coarsely ground A80/R20 and both coarsely and finely ground A100. In A100 samples, no significant differences were found between 90 and 120 d, in agreement with the peroxide value (PV) plateau reported by Ortolá *et al*. (10). In Figure 5, the highest maximum points could be observed for PUFA generation in coarsely ground samples occurring at 120 d in A80/R20 and at 150 d in A100. For acylglycerols, the 2-position of glycerol is preferentially esterified with PUFA, mainly linoleic acid (23). Hence, these late PUFA increases could be explained by PUFA liberation from the more protected 2-position owing to the preferential hydrolysis at the 1- and 3-positions in previous times. For finely ground coffees, the FFA increase was lower, possibly because of lipolysis during the first month.

As shown in Table 5 and Figure 6, the amounts of FFA increased in coffee samples at the end of the storage period. On the other hand, in finely ground A80/R20 samples, higher in-

TABLE 5
Changes in the FFA Composition (mg/100 g fat) of Finely and Coarsely Ground, Roasted Coffee During Storage Under Vacuum at 25°C[a]

	A100			A80/R20		
	0 d	180 d	%Δ	0 d	180 d	%Δ
ΣSFA						
Fine	384.0 ± 14.5	759.6 ± 26.9	97.8	606.5 ± 5.2	740.1 ± 6.8	22.0
Coarse	384.0 ± 14.5	821.2 ± 2.0	113.8	606.5 ± 5.2	685.8 ± 18.2	13.1
ΣMUFA						
Fine	62.3 ± 2.7	116.4 ± 2.3	86.8	100.8 ± 1.8	124.2 ± 2.3	23.7
Coarse	62.3 ± 2.7	123.5 ± 0.7	98.2	100.8 ± 1.8	114.2 ± 0.9	13.3
Σ PUFA						
Fine	283.8 ± 14.6	524.8 ± 5.5	84.9	400.6 ± 5.0	462.1 ± 7.1	15.3
Coarse	283.8 ± 14.6	542.6 ± 9.0	91.2	400.6 ± 5.0	453.8 ± 7.4	13.3
ΣPUFA/ΣSFA						
Fine	0.7	0.7	—	0.7	0.6	—
Coarse	0.7	0.7	—	0.7	0.7	—
ΣMUFA/ΣSFA						
Fine	0.2	0.2	—	0.2	0.2	—
Coarse	0.2	0.2	—	0.2	0.2	—

[a]Values are shown as means ± SD ($n = 3$). %Δ, increase in the percentage of FFA from the initial point; for other abbreviations see Tables 1 and 2.

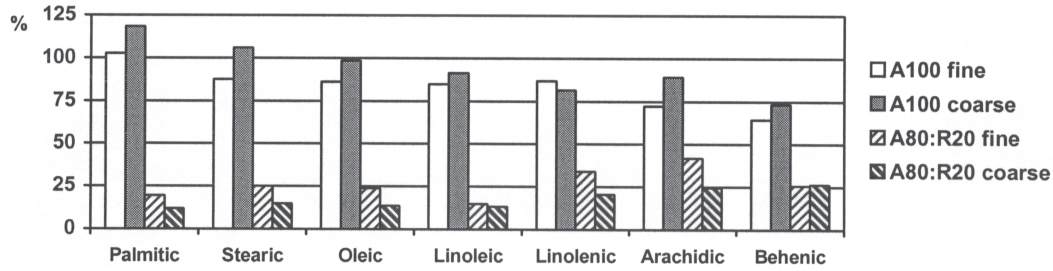

FIG. 6. Percentage FFA increases after 180 d of storage under vacuum at 25°C. For abbreviations see Figures 1 and 2.

creases were observed for every FFA, whereas these increases were higher in coarsely ground A100 samples for every FFA, except linolenic acid.

The initial FFA concentration was higher in A80/R20 coffee samples. However, throughout the storage period, FFA increases were due to TAG hydrolysis, and FFA decreases were due to oxidation reactions. Except for the first week, similar patterns were shown for both coffees, but FFA oxidation seemed to occur faster in the A80/R20 blend. Eventually, at the end of storage (180 d), the FFA concentration increased in all coffee samples.

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REFERENCES

1. Clarke, R.J., Coffee, in *Handbook of Food and Beverage Stability: Chemical, Biochemical, Microbiological, and Nutritional Aspects*, edited by G. Charalambous, Academic Press, London, 1986.
2. Illy, A., and R. Viani, Roasting, in *Espresso Coffee: The Chemistry of Quality*, edited by A. Illy and R. Viani, Academic Press, London, 1995, pp. 88–120.
3. Nikolova-Damyanova, B., R. Velikova, and G.N. Jham, Lipid Classes, Fatty Acid Composition and Triacylglycerol Molecular Species in Crude Coffee Beans Harvested in Brazil, *Food Res. Int. 31*:479–486 (1998).
4. Belitz, H.D., W. Grosch, and P. Schieberle, Lipids, in *Food Chemistry,* edited by H.D. Belitz, W. Grosch, and P. Schieberle, Springer-Verlag, Heidelberg, 2004, pp. 157–242.
5. Baesso, M.L., E. Correa Da Silva, H. Vargas, J.G. Cortez, and J. Pelzl, Use of Electron Spin Resonance for the Determination of Staling of Roast Coffee in Polyethylene Bag Packs, *Z. Lebensm. Untersuch. Forsch. 191*:24–27 (1990).
6. Morrice, A.E., N. Deighton, S.M. Glidwell, and B.A. Goodman, Free Radical Scavenging Reactions in Coffee, in *Proceedings of the 15th International Colloquium on the Chemistry of Coffee* (Montpellier, France), ASIC, Paris, 1993, pp. 644–649.
7. Lercker, G., E. Turchetto, A. Lucci, M.F. Carboni, R. Bortolomeazzi, G.T. Bertacco, N. Frega, and F. Bocci, Coffee Lipids. Note II: Some Parameters of Determination, *Indust. Aliment. 35*:1186–1193 (1996).
8. Martín, M.J., F. Pablos, A.G. González, M.S. Valdenebro, and M. León-Camacho, Fatty Acid Profiles as Discriminant Parameters for Coffee Varieties Differentiation, *Talanta 54*:291–297 (2001).
9. Speer, K., and I. Kölling-Speer, Chemistry I: Non-volatile Compounds. 1C: Lipids, in *Coffee: Recent Developments*, edited by R.J. Clarke, and O.G. Vitzthum, Blackwell Science, Oxford, 2001, pp. 33–49.
10. Ortolá, M.D., C.L. Gutiérrez, A. Chiralt, and P. Fito, Lipids Oxidation in Roasted Coffee, *Alimentaria* (Enero–Febrero):49–53 (1997).
11. Ortolá, M.D., L. Londoño, C.L. Gutiérrez, and A. Chiralt, Influence of Roasting Temperature on Physicochemical Properties of Different Coffees, *Food Sci. Technol Int. 4*:59–66 (1998).
12. Cardelli, C., and T.P. Labuza, Application of Weibull Hazard Analysis to the Determination of the Shelf Life of Roasted and Ground Coffee, *Lebensm.-Wiss. Technol. 34*:273–278 (2001).
13. Labuza, T.P., C. Cardelli, B. Anderson, and E. Shimoni, Physical Chemistry of Roasted and Ground Coffee: Shelf Life Improvement for Flexible Packaging, in *Proceedings of the 19th International Colloquium on the Chemistry of Coffee* (Trieste, Italy), ASIC, Paris, 2001.
14. Koelsch, C.M., T.W. Downes, and T.P. Labuza, Hexanal Formation *via* Lipid Oxidation as a Function of Oxygen Concentration: Measurement and Kinetics, *J. Food Sci. 56*:816–834 (1991).
15. Nicoli, M.C., N. Innocente, P. Pittia, and C.R. Lerici, Staling of Roasted Coffee. Volatile Release and Oxidation Reactions During Storage, in *Proceedings of the 15th International Colloquium on the Chemistry of Coffee* (Montpellier, France), ASIC, Paris, 1993, pp. 557–565.
16. Huynh-Ba, T., M.C. Courtet-Compondu, R. Fumeaux, and P. Pollien, Early Lipid Oxidation in Roasted and Ground Coffee, in *Proceedings of the 19th International Colloquium on the Chemistry of Coffee* (Trieste, Italy), ASIC, Paris, 2001.
17. Andueza, S., M.P. de Peña, and C. Cid, Chemical and Sensorial Characteristics of Espresso Coffee as Affected by Grinding and Torrefacto Roast, *J. Agr. Food Chem. 51*:7034–7039 (2003).
18. International Organization for Standardization, Roasted Coffee: Determination of Moisture Content. Method by Determination of Loss in Mass at 103°C, ISO/DIS 11294-1993, International Organization for Standardization, Geneva, 1993.
19. Association of Official Analytical Chemists (AOAC), Petroleum Ether Extract of Roasted Coffee, 920.97, in *Official Methods of Analysis*, 17th edn., edited by W. Horwitz, AOAC, Gaithersburg, MD, 2002.
20. Bligh, E.G., and W.S. Dyer, A Rapid Method of Total Lipid Extraction and Purification, *Can. J. Biochem. Physiol. 37*:911–917 (1959).
21. Association of Official Analytical Chemists (AOAC), Preparation of Methyl Ester, 969.33, in *Official Methods of Analysis,* 17th edn., edited by W. Horwitz, AOAC, Gaithersburg, MD, 2002.
22. Needs, E.C., D. Graema, G.D. Ford, A.J. Owen, B. Tuckey, and M.A. Anderson, Method for the Quantitative Determination of Individual Free Fatty Acids in Milk by Ion Exchange Resin Adsorption and Gas–Liquid Chromatography, *J. Dairy Res. 50*:321–329 (1983).
23. Folstar, P., Lipids, in *Coffee, Vol. 1: Chemistry*, edited by R.J. Clarke and R. Macrae, Elsevier Science Publishers, New York, 1985, pp. 203–222.

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