Small scale frying of potatoes in sunflower oil: thermoxidative alteration of the fat content in the fried product

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Laboruntersuchungen zum Fritieren von Kartoffeln in Sonnenblumenöl: Thermooxidative Alterung des Fettanteils in dem Fritiergut

Summary: Sets of 500 g of potatoes were fried discontinuously 15 times in 3 l of sunflower oil without addition of unused oil. The fat content of fried potatoes increased with the number of fryings. The quality of the extracted oil from the fried potatoes was evaluated by a combination of column and high performance size exclusion chromatography, and compared with their respective fryer oils. Total polar content (mg 100 mg oil⁻¹) increased after 15 fryings from 6.2 ± 0.35 to 18.7 ± 0.81 in the oil in the frier and to 21.2 ± 0.92 in the fries. Triglyceride polymers, triglyceride dimers and oxidized triglycerides (mg 100 mg oil⁻¹) increased progressively and significantly with the number of fryings either in the fryer oil or in the extracted oil, being the content of triglyceride dimers significantly higher in the extracted oil than in the oil in the fryer. Potatoes from the 15th frying contained more total alteration-oil-products, triglyceride polymers, triglyceride dimers and oxidized triglycerides (g 100 g fresh matter⁻¹) than potatoes from the 8th frying (all p < 0.01). This fact may be of special relevance for some people who usually eat large quantities of potatoes fried in oils with a null or low turnover of fresh oil.

Zusammenfassung: In 3 l Sonnenblumenöl wurden 15 mal aufeinanderfolgend je 500 g Kartoffeln, ohne zusätzliche Beigabe von ungebrauchtem Öl, fritiert. Der Ölgehalt der fritierten Kartoffeln nahm bei jedem Fritieren zu. Die Qualität des aus den fritierten Kartoffeln extrahierten Öls wurde durch eine Kombination von Säulenchromatographie und high performance size exclusion chromatography bestimmt, und mit den betreffenden Fritierölen verglichen. Der Gehalt polarer Fettbestandteile (mg/100 mg Öl) stieg nach 15 Fritierungen von 6.2 ± 0.35 auf 18.7 ± 0.8 in dem Fritieröl und auf 21.2 ± 0.92 in dem Fritiergut. Polymere, dimere und oxydierte Triglyceride (mg/100 mg Öl) nahmen stufenweise und signifikant mit der Anzahl von Fritierungen zu, sowohl in dem Öl als auch in dem aus den Kartoffeln extrahierten Fritieröl. Der Inhalt an Triglyceriddimeren war bedeutend höher in dem extrahierten Öl als in dem Fritieröl. Kartoffeln, die zum 15. Mal fritiert wurden, wiesen eine höhere Konzentration an gesamten Zersetzungsprodukten, Polymeren, Dimeren und oxydierten Triglyceriden (mg/100 mg frische Materie) auf als Kartoffeln, die zum 8. Mal fritiert wurden (alles p<0.01). Diese Tatsache kann vor allem für die Leute wichtig sein, die normalerweise große Mengen von fritierten Kartoffeln essen, die in einem Öl gebraten wurden, zu dem keine oder nur kleine Mengen frisches Öl zugegeben worden waren.

Key words: Column chromatography – size-exclusion chromatography – polar fraction – deepfat frying – fat composition – fried potatoes – sunflower oil Schlüsselwörter: Säulenchromatographie – high performance size exclusion chromatography – Polar-Teilung – Ölbad-Fritierung – Fettbestandteile – fritierte Kartoffeln – Sonnenblumenöl

Introduction

When we study frying, we do not simply study fats and oils, foods, or even processing equipment and conditions, but rather a composite of these elements taken together as a system (2).

Most of the work on heated fats has been carried out in the laboratory on oils heated without food fried in it. However, less studies have been performed on oil with food fried in it and on the lipid exchange between the bath oil (fryer) and the food during the frying operation (10, 14, 17, 22, 23).

Recently in Spain, a marked increase in the consumption and use for frying purpose of sunflower oil and a decrease in the use of olive oil have been described (15). Economical reasons are involved in the use for frying of sunflower oil instead of olive oil. The second one, however, is shown to be more adequate for frying purposes because of its thermal stability.

Analysis of decomposition products from the thermal and oxidative treatment of fats and oils has been widely studied (12). However, systematic studies concerning deep-fat frying are far from being completed, because the number and variety of products formed in the frying process are great, and the nutritional and toxicological consequences of their consumption are largely unknown. Consequently, there is a need to define the alteration level at which the fats or oils must be discarded. It is claimed that a fat or oil must be discarded when its polar fraction is more than 25 % (2, 4). However, such critical alteration level has not been established for lipid content of foodstuff, such as fried potatoes.

Guillaumin (11) and Sebedio et al. (25) found similar lipid composition (polar components), for the french fries and the oils used for frying operation. However, Pokorny (20) indicated that fat retained in the substrate was more oxidized than fat in the pan.

Although many analytical methods have been used for the determination of monomers, dimers and higher polymers in oxidized fat and oils, the technique of high-performance size-exclusion chromatography (HPSEC) may be considered one of the most promising (3).

The aims of this work are: 1) to compare the content of polar material in sunflower oil-fried potatoes with that of the oil used for frying, 2) to evaluate by using the technique of HPSEC the polymers and oxidized materials of both the oil and the fried potatoes, and 3) to analyze the possible preferential absorption by the potatoes of the altered components formed throughout the fryings.

Materials and methods

Materials and reagents

Refined sunflower oil (Córdoba, Spain) and potatoes (Valencia, Spain) were purchased at a local store. Chloroform, methanol, hexane, diethyl ether, petroleum ether, acetic acid, silica gel 60F 250 plates (20 x 20 cm glass) for thin-layer chromatography and silicagel 60 (0.063-0.200 nm) for column chromatography were obtained from E. Merck AG, Darmstadt, FRG. Columns for high performance size exclusion

chromatography (HPSEC) were obtained from Hewlett Packard (Palo Alto, CA). Oleic, linoleic and linolenic acids, diolein, dilinolein, dilinolenin, triolein, trilinolein and trilinolenin were obtained from Sigma Co., St. Louis, Missouri.

Methods

Performance of fryings: Domestic deep-fat fryers with a 3 l aluminum vessel were used for frying. The potatoes were chopped into slices ca. 2 mm thick. The proportion of food to frying oil in the repeated frying was kept at 500 g to 3 l by eliminating one fryer after each four fryings and emptying its contents to make up the volume of the other fryers to 3 l because so much oil is removed along with the fried potatoes. A total of 15 fryings were carried out. Time required to reach and maintain the bath oil at 180° C before introduction of potatoes was 20 min. Potato slices were then fried for 8 min. After the end of each frying, the oil was again heated to 180° C before beginning a new frying (time required 10 min). A set of two fryings was successively carried out, then the oil was allowed to cool until the next day. Every day the same frying procedure was repeated. The fryings took place over 4 consecutive days. On the last day only three fryings were performed. The overall time the oil was heated throughout the whole experiment can be estimated to be 5 h and 50 min. More details of the frying method have previously been described (22, 24).

Aliquots of 50 ml from the unused oil, and from the fourth, eighth, 12th and 15th fryings were taken for analyses. The fried potatoes from the fourth, eighth, 12th, and 15th fryings were kept at -20° C under nitrogen atmosphere until analyses were made.

Lipid extractions: The lipid of the fried potatoes was extracted four times with diethyl ether using a 1:50 (w/v) ratio in each extraction operation.

Separation of polar and nonpolar components: The quantity of polar components in the oils and in the lipid extracts was determined by the silicagel column chromatography method of Waltking and Wessell (31) slightly modified by Dobarganes et al. (7). An accurately weighed sample of 1 ± 0.01 g was dissolved in 20 ml petroleum-ether/diethyl ether 87:13 (v/v) when unused oil was analyzed, and 90:10 (v/v) when used oils and when extracted oils from potatoes were analyzed. A final elution of the column with chloroform/methanol 1:1 (v/v) was performed to improve the recovery of the sample (8). Total recovery could be estimated higher than 99 %.

The purity of each column-chromatographed-fraction was then checked by thinlayer chromatography using a mixture of hexane/diethyl ether/acetic acid (80:20:1) as solvent system (1).

High-performance size-exclusion chromatography (HPSEC): Polar fractions previously obtained by column chromatography, as described above, were analyzed by HPSEC following the Dobarganes et al. method (8). The isolated polar fractions were analyzed in a Konic 500A chromatograph (Barcelona, Spain) with a 10 μl sample loop. A Hewlett Packard 1037A refractive index detector and two 300 mm x 7.5 mm i.d. (5 μm particle size) 0.01 μm and 0.05 μm PL gel (polystyrene-divinylbenzene) columns (Hewlett-Packard) connected in series were operated at 40° C. HPLC-grade tetrahydrofuran served as the mobile phase at 1 ml min⁻¹.

Sample concentration was 10 to 15 mg ml $^{-1}$ in tetrahydrofuran. All eluents as well as samples were precleaned by passing them through a filter ($^2\,\mu m$). To evaluate the hydrolytic and thermoxidative products, pure fatty acids, diglycerides, triglycerides and total polar fractions at different concentrations were studied. Correlations obtained between the detector response and the weight of different compound groups injected were linear (r>0.99). The response factors for fatty acids, diglycerides, triglycerides and total polar components were similar. To this aim, a mix of oleic, linoleic and palmitic acids, mix of diolein, dilinolein and dilinolenin, and mix of triolein, trilinolein and trilinolenin were studied at different concentrations. Because standards for triglyceride dimers and triglyceride polymers are not available, regression lines relating retention times and molecules weight (MW) of compounds such as free fatty acids, monoglycerides, diglycerides and triglycerides were drawn. Then, the retention times of the sample chromatogram peaks were extrapolated on an extension of these regression lines. An MW of \sim 1800 and an MW of \sim 3000 correspondend to MW of triglyceride dimers and triglyceride trimers, respectively.

Statistical analyses: Knewman-Keuls multiple comparisons test, and the unpaired-Student-t test (27) were used.

Results and discussion

In studies on frying conditions, Varela (28, 29) found that the temperature of the fat at which the food was fried, had relatively little bearing on the thermal damage done to the food, provided that the food had a high water content. Because of water evaporation, the temperature inside the food does not rise above 100° C. In addition, the fat does not begin to penetrate the food until a substantial part of the water it contains has evaporated. As a result, the hot fat acts on the food for a very short time.

The efficiency of the sunflower oil used in repeated fryings of potatoes was evaluated measuring the oil loss of the fryers and studying both the weight and the fat content variations of the potatoes. In this report the decrease of sunflower oil in the fryers was of 63.3 ml per frying and fryer. In other research, done under the same frying conditions, our group (cited in (24)) found a decrease of olive oil in the fryers of 56.3 ml per frying. Varela (28, 30) indicated that olive oil forms a crust that protects the food against absorption of oils, whereas other fats do not form such a defined crust and the food contains more fat after frying. This would explain the greater oil loss (19.5 %) in the fryer when sunflower oil is used instead of olive oil.

The fat content of the fried potatoes increased significantly from $26.7 \pm 0.88 \%$ (w/w) (mean \pm SD of the four first fryings) to $29.5 \pm 1.21 \%$ (w/w) (mean \pm SD of the three last fryings). The higher polar content found when the number of fryings with sunflower oil increases (Table 1) would influence this higher fat content, because according to Blumenthal (2) the more altered the culinary fat is, the higher the mass transfer and surfactant production are, increasing therefore the fried food fat content. However, Sebedio et al. (25) described no significant differences in the lipid content of potatoes between the 1st and the 30th frying operations in either peanut or soybean oil.

Moreover, many other factors have been reported to affect oil uptake (26). The oil uptake criterion, U_R, expressing the weight ratio between oil uptake and moisture

Number of fryings	n	Fryer oil	Fried potatoes
0	3	6.2 ± 0.35^{a}	
4	3	10.7 ± 0.64^{b}	12.4 ± 1.58^{bc}
8	3	14.4 ± 0.81^{c}	16.5 ± 0.60^{d}
12	2	16.4 ± 0.81^{d}	18.3 ± 0.32^{e}
15	2	18.7 ± 0.81^{e}	21.2 ± 0.92^{f}

Table 1. Polar compounds (mg 100 mg oil⁻¹) in the fried potatoes and the fryer oil as a function of number of frying operations effected^a

loss introduced by Pinthus et al. (19), ($U_R = \text{oil uptake (g)/water removed (g)}$) seems to be more accurate than the oil uptake without considering the effect of water.

 U_R of fried potatoes shows a significant increase with the number of fryings, 0.10 ± 0.005 (mean \pm SD of the 3 last fryings) vs 0.07 ± 0.007 (mean \pm SD of the four first fryings).

Arroyo et al. (1) and Sebedio et al. (25), indicated that many reactions take place during frying, such as oxidation, polymerization, hydrolysis, cyclization and isomerization, which led to the formation of a complex mixture of products.

The alteration of the fryer oil and the oil extracted from fried potatoes was followed by the amount of polar component (Table 1) and the concentration of triglyceride polymers, triglyceride dimers and oxidized triglycerides (Table 2).

The amount of polar compounds (mg 100 mg oil-1) was 6.2 in the starting sunflower oil (Table 1). This value increased to 18.7 after 15 fryings. The amount of polar component in the fries (mg 100 mg extracted oil-1) was 21.2 after the 15th frying. It should be noted that the level of polar content in the fries from the eighth, 12th and 15th fryings was significantly higher than in the frying medium. Thus, the critical level of 25 % polar content is achieved more rapidly in the fries than in the frying oil. In consequence, we would also need a reference value for fried product reinjection. Guillaumin (11) and Sebedio et al. (25) after 30 frying operations, found similar lipid composition (polar components, polymers, fatty acid isomers) for the french fries and the oils used for the frying operations. Pokorni (20) studying the effect of different substrates on deterioration of the frying oil, indicated that fat retained in the substrate was more oxidized than fat in the pan, probably due at least in part to sorption of oxidation products in the surface layers of the fried substrate.

As has been indicated above, the polar fraction was analyzed by HPSEC. This procedure allows the concentration and determination of degradation compounds such as triglyceride polymers, triglyceride dimers, oxidized triglycerides, diglycerides and free fatty acids (1, 6, 8). The HPSEC chromatograms of the polar fraction from unused and used sunflower oil and of the polar fraction from the fries after the 15th frying are presented in Fig. 1.

^a Values (mean ± SD) bearing a common letter are not significantly (Knewman-Keuls multiple comparison test) different. n = number of samples analyzed.

Table 2. Thermal oxidative and hydrolytic compounds in fried potatoes and fryer oil as a function of number of frying operations effected^a

	Number o 0 (n = 3)	Fryer oil Number of frying operations $8 (n = 3)$	15 (n = 2)	Fried potatoes Number of frying operations 8 (n = 3)	ations 15 (n = 2)
Thermoxidative alteration (T) - Triglyceride polymers (% on polar fraction) (mg 100 mg oil ⁻¹)	(T) 1.3 ± 0.15^{a} 0.1 ± 0.01^{a}	9.7 ± 0.73^{b} 1.4 ± 0.12^{b}	$12.8 \pm 0.52^{\circ}$ $2.4 \pm 0.09^{\circ}$	9.9 ± 0.63^{b} 1.6 ± 0.14^{b}	$14.3 \pm 0.66^{\circ}$ $3.0 \pm 0.3^{\circ}$
 Triglyceride dimers (% on polar fraction) (mg 100 mg oil ¹) 	16.0 ± 0.64^{a} 0.9 ± 0.09^{a}	36.3 ± 2.42^{b} 5.2 ± 0.35^{b}	$35.8 \pm 0.81^{\text{b}} \\ 6.7 \pm 0.27^{\text{c}}$	36.5 ± 0.61^{b} 6.0 ± 0.32^{bc}	38.9 ± 0.31^{b} 8.2 ± 0.33^{d}
 Oxidized triglycerides (% on polar fraction) (mg 100 mg oil⁻¹) 	55.1 ± 0.48^{a} 3.4 ± 0.22^{a}	38.9 ± 2.35^{b} 5.6 ± 0.29^{b}	40.5 ± 1.34^{b} 7.6 ± 0.34^{cd}	41.8 ± 0.75^{b} 6.9 ± 0.22^{c}	38.1 ± 0.11^{b} 8.1 ± 0.38^{d}
Hydrolytic alteration (H) - Diglycerides (% on polar fraction) (mg 100 mg oil ⁻¹)	22.1 ± 0.50^{a} 0.4 ± 0.08^{a}	$10.4 \pm 0.90^{\rm b}$ $1.5 \pm 0.06^{\rm a}$	7.5 ± 0.28^{c} 1.4 ± 0.01^{a}	8.9 ± 0.32^{d} 1.5 ± 0.02^{a}	6.9 ± 0.19^{c} 1.5 ± 0.01^{a}
 Free fatty acids (% on polar fraction) (mg 100 mg oil⁻¹) 	6.7 ± 0.05^{a} 0.4 ± 0.02^{a}	5.5 ± 1.22^{b} 0.8 ± 0.10^{b}	$2.3 \pm 0.26^{cd} \\ 0.6 \pm 0.03^{c}$	$2.9 \pm 0.13^{\circ} \\ 0.5 \pm 0.01^{ac}$	1.9 ± 0.13^{d} 0.4 ± 0.01^{a}
T/H ratio	2.5	5.3	8.4	7.3	10.2

^a Values are mean ± SD. Values in the same row bearing a common letter are not significantly (Knewman-Keuls multiple comparisons test) different. n = number of samples analyzed.

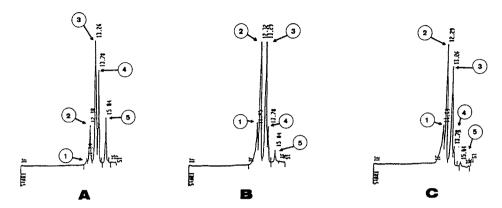


Fig. 1. High-performance size-exclusion chromatograms of the polar components of unused (A), and used oil sample from the 15th frying (B), and of extracted oil of fried potatoes from the 15th frying (C). Peaks 1, 2, 3, 4, and 5 are triglyceride polymers, triglyceride dimers, oxidized triglycerides, diglycerides and free fatty acids, respectively. Conditions: Column-series connected 0.01 µm and 0.05 µm PL gel (polystyrene-divinylbenzene), 300 mm x 7.5 mm i. d., 5 µm particle size; eluant: tetrahydrofuran at 1 ml min⁻¹, 10 µl injection volume, refractive index detection.

Frying operations significantly increased the percentage (% on polar fraction) of polymers and dimers of triglycerides, and decreased the percentage of oxidized triglycerides, diglycerides and free fatty acids either in the fryer oils or in the extracted oils after 8 or 15 fryings (Table 2). Thermoxidative alteration products did not show any difference in those percentages between the frying oil and the extracted oil. However, fryer oils contained higher percentages of hydrolytic alteration products than their respective extracted oils (Table 2).

When the results of the different compounds are given in mg 100 mg oil⁻¹, triglyceride polymers in the fryer oil increased 14- and 24-times after the eighth and 15th fryings, respectively. Triglyceride dimers in the oil also increased 5.6- and 6.7-times after the eighth and 15th fryings, respectively (Table 2). Gere (9) indicated that heating or frying causes a decrease in the nutritive value of fats (e.g., sunflower oil, rapeseed oil and lard) as a result of decomposition of polyunsaturated essential fatty acids bringing about the formation of alteration products, such as polymeric triglycerides. The amount of oxidized triglycerides also increased significantly 1.5- and 2.2-times after the eighth and 15th fryings, respectively.

Alteration products also increased in the fries, because of the amount of trigly-ceride polymers, triglyceride dimers and oxidized triglycerides in fries from the 15th frying vs the unused oil increased 30.0-, 8.2-, and 2.4-times, respectively (Table 2). Thus, the thermoxidative alteration seems to be more intensive in the fries than in the fryer oil. However, only the amount of triglyceride dimers after the 15th frying were significantly higher in the fries than in the oil (Table 2). Pérez-Camino et al. (18) found that polymers (polymers plus dimers) and oxidized triglycerides tended to be higher in lipids from potatoes than in frying oils when sunflower was used in the final preparation of prefried potatoes.

The amount of diglycerides in the oil and in the fries was similar and it remained quite stable throughout the fryings. Free fatty acids increased in the fryer oil but not in the fries (Table 2).

It was already described (22) that the amount of free fatty acids did not correlate with the amount of either polymers or polar content. Arroyo et al. (1) found a high correlation between polar component and triglyceride polymers, triglyceride dimers, oxidized triglycerides and diglycerides, while free fatty acids did not correlate significantly with the amount of polar content. Then, the measurement of free fatty acids, perhaps, could not be the best criterion to test the state of degradation of an oil.

From Table 2 it can be deduced that either in the fryer oil or in the fries thermal oxidative compounds contribute more than hydrolytic compounds to oil alteration. The ratio of thermal oxidative to hydrolytic alteration compounds (T/H ratio) also increased with the number of fryings in the bath oil and in the fries. T/H ratio was higher in fried potatoes than in the bath oil (Table 2). Thermal oxidative products have been related to the adverse physiological effects seen after ingestion of altered fat (4, 5).

Blumenthal (2) has stated (sic) "If the oil is only 75 % pure and 10 % has been picked up on the food, the consumer eats 2.5 % total polar materials by weight of fried food. The polar fraction contains all the degradation chemicals in an oil which may be toxic and which account for the cooking and eating qualities of an oil".

Table 3. Altered lipid content (g 100 g fresh matte	or ¹) of potatoes from the eighth and 15th fryings ^a
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	8th frying (n=3)	15th frying (n=2)	8th vs 15th fryings
Total alteration oil products	3.5 ± 0.13	5.1 ± 0.21	p < 0.01
Triglyceride polymers	0.3 ± 0.02	0.7 ± 0.07	p < 0.01
Triglyceride dimers	1.3 ± 0.02	2.0 ± 0.07	p < 0.01
Oxidized triglycerides	1.5 ± 0.09	1.9 ± 0.10	p < 0.01
Diglycerides	0.3 ± 0.03	0.4 ± 0.05	NS
Free fatty acids	0.1 ± 0.04	0.1 ± 0.05	NS

^a Values are means \pm standard deviations of the indicated (n = number of) samples. NS = no significant variation (unpaired Student's *t*-test).

Table 3 shows that potatoes from the 15th frying contained more total oil alteration products, triglyceride polymers, triglyceride dimers and oxidized triglycerides (g 100 g fresh matter⁻¹) than potatoes from the eighth frying. That fact may be of special relevance for some people who usually eat large quantities of potatoes fried in oils with a null or low turnover of unused oil, as normally is done at home, in bars or in restaurants, because thermoxidative components have been associated with fat toxicity. Combe et al. (4) have shown an absorption rate of 4 % for polymers, 53 % for oxidized monomeric acids and 96 % for cyclic monomers of thermal origin. Nolen et al. (16) have found that both monomeric and dimeric materials were more rapidly absorbed and were therefore more toxic than longer chain polymers. However, Márquez-Ruiz (13) in studies with ¹⁴C suggests that, prior to its absorption,

structural changes in the oxidized polymers would decrease their molecular weight and would account for the higher digestibility of the polymers when compared to the dimers of thermal origin.

In conclusion, discontinuous and successive fryings of potatoes in sunflower oil increased the level of total polar compounds in the oil and in the oil extracted from fries. Thermoxidative alterations took place preferentially to hydrolytic process, containing the oil extracted from fried potatoes slightly but significantly higher triglyceride dimers than the respective frying oils.

According to this data, research could be addressed to establish the maximum level of oxidized triglycerides, dimers and polymers of triglycerides (potential toxics) that should be accepted in fried products.

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