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A RESEARCH ON THE TURBIDITY AND WAX COMPOSITION OF SUNFLOWER AND CORN OILS DURING WINTERIZATION

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Abstract

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The effects of different filter aid materials at different ratios (0.3 or 0.6%, w/w) and different time in winterization on wax content and turbidity of sunflower and corn oils were studied. For both oils, the major part of waxes was removed within 24 h of winterization using 0.6% kieselghur and turbidity was also decreased at the end of the 24 h. Using filter aids is quite effective in removing of waxes so turbidity. The effects of using different filter aid materials and different time of winterization were found statistically important on wax content of both oils ($p<0.001$). Turbidity values showed differences due to the wax content of oils, applied pressure and time.

Key words: Corn oil, filter aids, sunflower oil, turbidity, wax, winterization

Introduction

Winterization is a step in refining some vegetable oils to remove compounds, which crystallize at low temperatures and cause turbidity in oils (Turkulov et al., 1986; Coutinho et al., 2009). The majority of these compounds are waxes. Saturated triglycerides and hydrocarbons along with small amounts of other materials, such as, aldehydes, ketones, sterols, alcohols and oils, which polymerize during refining can be present (Baltanas et al., 1998; O'Brien et al., 2000; Kellens, 2000; Coutinho et al., 2009).

The waxes of vegetable oils are mainly esters of long-chain saturated fatty acids with long-chain aliphatic fatty alcohols having 34 to 50 carbon atoms (Carelli et al., 2002). Waxes have low solubility in oil at low temperature and cause turbidity when they crystallize. Their melting points range from 70 to 80^{o}C (Martini and Anon, 2003; Kanya et al., 2007). The waxes are naturally present in oilseeds but high wax contents are present in their hulls or bran. Depending on the origin of the seeds, the wax content of the hull varies from 1.5 to 3% (Kanya et al., 2003; Baümler et al., 2007). As a result, the content of waxes in oil can change according to the seed variety, its origin, percentage hull removed, temperature and technology used to process the oil (Carelli et al., 2002). While the wax content of crude sunflower oil can vary between 0.02 and 0.35% after refining, this amount decreases to a range of 0-0.06% (Martini and Anon, 2000; Martini and Anon, 2003).

Filter aid materials are added into the oil during winterization to facilitate the filtration of waxes (Raß et al., 2008). The use of filter aid materials provide wide opening holes on the cloth used in plate filters under reduced pressure. Perlite is the most commonly used filter aid in Turkey because of its availability and economic advantages, whereas in Europe and America, kieselghur is used. At the same time, they serve as a promotional material on the filter cloth (Hamm and Hamilton, 2000). The aim of the present study was to determine the effects of using different amounts (0.3% or 0.6%) filter aids (perlite and kieselguhr) for the winterization of sunflower and corn oils at different mixing times (6, 12 or 24 h) on the wax compositions and turbidity values of the oils.

Materials and Methods

Oil samples

Degummed, neutralized and bleached sunflower and corn oils were obtained from commercial vegetable oil refineries in high-density 30-L polyamide tanks.

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Pilot winterization unit

The capacity of the pilot winterization unit was 50 L. The unit was composed of refrigeration and crystallization tanks (1), a digital temperature-control panel (2), a compressor with Freon12 refrigerant (3), holding and balance tanks (4), a plate filter (5), a feed tank (6), a mixer (7), a centrifugal pump (8), junction pipes (9) (Fig. 1). The mixer speed was set at 30 rpm to obtain slow mixing.

Winterization process

Perlite and kieselghur were used as filter aid materials in concentrations of 0.3 and 0.6%. The process was experienced in three different period of time (6, 12 and 24 h). Other parameters were kept constant and the process was carried out in two replications. Also a control winterization was performed without adding any filter aid materials. The winterization process includes three-step cooling, crystallization and filtration. In pre-cooling, the temperature was reduced from ambient temperature to 10°C by mixing in for 2 h with a plate-type heat exchanger. For better crystallization and maturation, the mixture was cooled to 4±1°C. At this temperature the mixture was kept for 6, 12 and 24 h while mixing at 30 rpm. Thus, wax crystals formed during this period matured. After maturation, the mixture was heated to 8-10°C to reduce the viscosity and facilitate filtration. The crystalline phase was removed from the mixture by using filter plates under about 1 bar pressure.

Sampling

After filtering about 3 L oil in 6, 12 and 24 h a 500-mL of sample from each batch was taken for analysis. The sampling design is given in Figure 2.

Determination of Turbidity

Turbidity measurements were done by turbidity meter (VelP Scientifica TB1, Italy). Before each measurement, the instrument was calibrated by its standard solution (0,02; 20; 100 and 800 NTU). Formazine is used as standard solution.

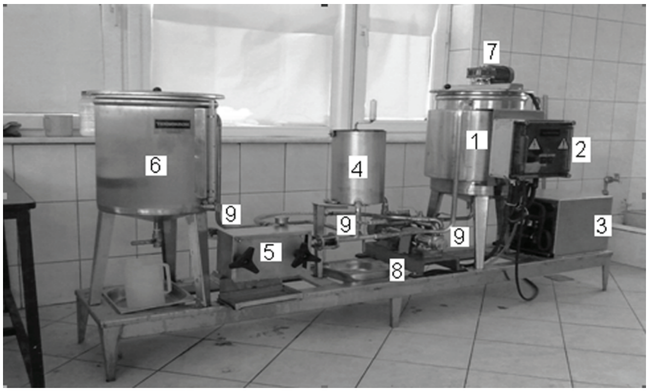

Fig. 1. Laboratory-scale pilot winterization unit

Wax determination

The wax contents of oil samples were determined by using the IOC method (Anon., 2009). The wax component was separated from the samples by column chromatography.

Preparation of samples for wax analysis

A glass column (1.5 mm x 40 cm) was used to separate the waxes. 2 mL water was poured into 100 g of silica gel, which was activated at 350°C for 4 h, and 8 g of that mixture was taken and suspended in n-hexane/diethyl ether (98:2) and then introduced into the column. 200 mg of sample was weighed into a 25-mL flask and then 0.1 mL of internal standard (0.1% lauryl arachidate) and 0.1 mL of 0.1% Sudan-I, which was prepared by n-hexane, were added. The prepared sample was transferred to the chromatography column. n-Hexane was added into the column until the elution was completed. The solvent was evaporated by using rotary evaporator (IKA RV 10 Basic, Germany) from the collected solvent + wax mixtures. The rest of the waxes were dissolved in HPLC-grade n-hexane, then transferred into a test tube and the solvent was evaporated until the 0.5 mL sample remained. 0.8 µL sample was injected directly into the on-column.

Gas chromatography

The instrumentation used for the analyses was as follows: Agilent Technologies GC (model 6890 N) equipped with a

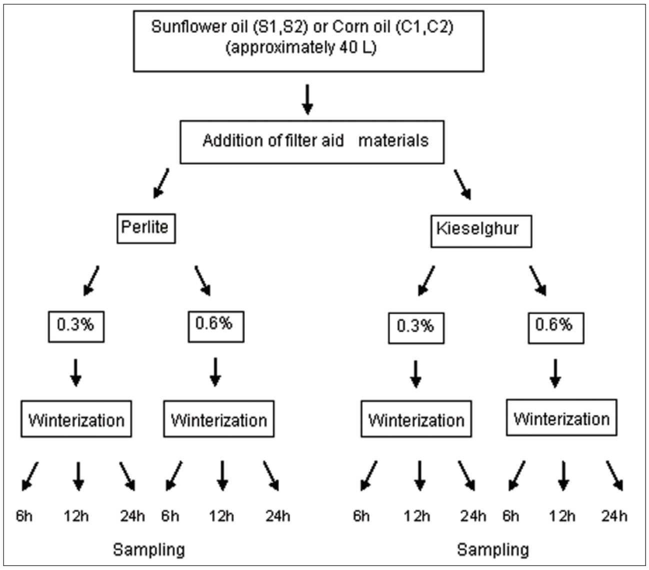

Fig. 2. Factorial experiment plan for winterization sampling process

system for direct on-column injection and a flame ionization detector. HP 5 SE 34 fused silica capillary column (30 m × 0.32 mm i.d., 0.25 mm film thickness; Supelco Inc., Bellefonte, PA, USA) was used for separation of waxes. Helium was used as the carrier gas at 2.1 mL/min flow rate. The detector temperature was 350^{o}C. Oven temperature programming was: initial temperature 80°C, increase at 35°C min^{-1} to 245°C and then held for 15 min. This was increased by 1.40°C min^{-1} to 325°C holding for 25 min, and finally increasing by 35°C min^{-1} to 340°C and holding for 30-40 min.

Identification of peaks and calculation

The wax esters were determined on the basis of retention times of internal standards. The wax chromatogram of olive oil was used for identification of the peaks between C34-C48. Waxes were calculated as esters as the following equation;

$$\text{Ester, mg/kg} = \frac{A_x \text{ x } m_s \text{ x } 1000}{A_s \text{ x } m}$$

$\underline{A_x}$ = area corresponding to the peak fort the individual ester

$\underline{m_s}$ = area corresponding to the internal standard (lauryl arachidate)

A_s = mass of the added internal standard, in mg

m = mass of the sample taken for determination, in g

Statistical Analysis

Data were statistically analyzed by analysis of variance (ANOVA) with the SAS software package. The PROC MIXED procedure was applied following by Fisher's LSD when the differences between applications were significant (p <0.05). Factorial design was used as experimental design. Winterization applications and time were two factors in the experimental design. Filter aids are discussed together with the amounts used and evaluated as a single factor (SAS, 2001).

Results and Discussion

Turbidity values of winterized oils

The effects of using different filter aid materials and different winterization treatments on the turbidity values of sunflower and corn oils are given in Table 1. It can be seen that average turbidity values of sunflower oil samples significantly decreased after winterization. This can be explained that the waxes being removed from high wax content sunflower oil by winterization. Especially, at the end of 24 h of the control winterization, the turbidity decreased up to 1.27 NTU from 221 NTU (Table 1). Turbidity meter is not only used for measuring wax-based turbidity, it is also used for the determination of turbidity caused by filter aid materials, which pass to oil, by the effect of applied pressure, during filtration.

As seen in Table 1, the average turbidity value of control corn oil is quite low (6.60 NTU) when compared to sunflower oil. Generally, at the end of 24 h in all of the winterization applications, the turbidity values of corn oil decreased (Table 1). However, at the end of 12 h of the winterization process in which perlite and kieselghur were used, the turbidity values of corn oil increased because of using high level of filter aid materials about the low wax content.

As a result, in both oil samples, the turbidity values decreased at the end of 24 h of winterization. Some changes were observed due to the applied filtration pressure, filter aid materials and applied time. Industrially when the refining process is completed, oil is filtered by polish filter and become more clear due to the removing all of the facts which cause turbidity.

The effects of using different filter aid materials and different time on the turbidity changes of sunflower oil were found statistically important (p<0.001). There were no differences observed between control winterization and winterization of using 0.6% perlite whereas there is significantly differences between perlite added winterization and kieselghur added winterization. Similarly, there were no differences observed among the samples taken at 6th and 12th hours, whereas there was a decrease was observed at the end of the 24th hour.

The effect of different time of winterization was found statistically important on the turbidity changes of corn oil (p<0.001) whereas using of different filter aid materials did not significantly affect (p>0.05). 24 h of winterization are different from 6 h and 12 h.

Wax content of winterized oils

In both Tables 2 and 3, the effects of using different filter aid materials and different winterization treatments on the wax content and composition of sunflower and corn oils are given. Waxes containing C34 to C48 carbon atoms were detected. C34, C35, C36, C37, C38, C39, C40, C41, C42, C43, C44, C45, C46 and C48 were detected. Total wax content of an oil sample was calculated by the sum of these wax compounds. When the effect of filter aid material and winterization period on the wax content of sunflower oil was taken into account, it can be seen that total wax content of the sample significantly decreased during winterization (Table 2). Initial total wax content of sunflower oil was 1129 mg/kg. 35.9% of total wax content was waxes having odd carbon numbers, whereas 64.1% was waxes having even carbon numbers. Carelli et al. (2002) reported that 41% of total wax content of sunflower oil waxes having odd carbon numbers. According to Baümler et al. (2007), 38% of wax content of winterized sunflower oil was the waxes having odd carbon number. Because of our findings, proportional amount of waxes having

Table 1
Turbidity values of sunflower and corn oil during winterization [a]

Oil type	C.sample*	Control winterization			Perlite						Kieselghur					
					0.3			0.6			0.3			0.6		
	Time	6	12	24	6	12	24	6	12	24	6	12	24	6	12	24
Sunflower oil	221±55.2	2.60±0.2	1.69±0.7	1.27±0.2	4.05±2.3	3.22±1.6	1.92±1.3	5.49±0.04	7.28±0.2	3.34±0.3	5.29±1.5	3.62±1.0	3.34±0.3	5.97±0.9	5.53±0.4	2.87±1.1
Corn oil	6,60±0.2	2.99±1.3	5.00±0.9	2.29±0.9	1.45±1.3	5.34±0.8	2.13±0.5	6.12±2.2	4.51±0.5	2.55±0.1	3.97±1.6	5.44±1.7	1.67±0.9	4.40±0.3	6.70±1.2	2.32±0.6

[a] Data are mean±standard deviation (n=2)
*control sample; degummed, neutralized and bleached oil taken in determined oil companies

Table 2
Wax content and composition of sunflower oil during winterization (mg/kg) [a]

waxes	C.sample*	Control Winterization			Perlite						Kieselghur					
Carbon number					0.3			0.6			0.3			0.6		
	Time	6	12	24	6	12	24	6	12	24	6	12	24	6	12	24
C34	14±1.4	12.5±2.1	9.9±4.5	9.6±3.8	13.3±0.4	9.3±2.0	3.2±1.0	13.1±2.5	6.8±1.1	5.5±0.8	12.2±0.9	7.7±2.5	6.0±0.4	12.8±4.5	8.4±2.1	2.7±1.7
C35	3.4±0.8	3.9±0.3	3.4±0.4	2.6±0.1	3.4±0.2	3.0±0.4	2.3±0.1	3.4±0.8	3.3±1.3	1.9±0.1	3.2±0.4	2.9	2.0±0.1	3.8±0.1	2.5±0.1	2.2±0.4
C36	52.3±2.5	53.3±3.7	39.7±3.0	29.0±1.1	50.2±9.6	36.9±1.2	5.8±0.2	55.3±4.3	20.4±1.9	7.0±1.2	53.2±2.6	33.8±0.8	4.9±0.2	51.6±0.7	15.7±1.1	7.6±0.6
C37	130.2±1.5	115.8±11.8	91.3±3.5	28.7±18.6	81.4±20.4	53.0±13.5	8.6±1.5	102.5±0.4	31.2±10.3	6.2±2.4	113.9±9.0	58.3±1.3	4.6±0.8	115.7±1.3	35.1±6.4	5.1±0.1
C38	22.6±0.8	21.9±1.3	19.5±4.1	12.9±2.1	19.7±3.3	15.7±3.2	7.2±0.4	17±3.7	13.2±1.6	8.4±2.5	19.1±3.6	18.2±0.6	5.8±0.6	23.6±4.0	12.9±3.2	5.0±1.5
C39	37.4±3.0	34.8±6.7	30.6±8.1	21.6±1.7	26.4±5.8	27.6±0.1	8.0±0.9	26.2±2.0	20.6±4.1	4.9±1.3	31.3±1.0	29.1±0.6	5.7±1.5	34.7±0.1	21.0±3.4	5.2±1.5
C40	56.1±3.3	54.4±3.7	49.1±7.2	25.7±2.5	45.9±11.5	41.3±4.7	6.1±0.6	45.9	23.6±5.9	4.4±1.6	52.9±1.8	40.9±7.8	7.4±2.1	56.4±3.7	18.3±3.1	6.7±0.2
C41	161.5±30.2	145.5±28.1	112.2±4.9	36.1±6.4	107.6±18.1	57.3±1.2	12.3±3.1	105.7±5.9	35.8±6.3	9.8±1.7	128.2±19.2	69.6±5.4	7.0±0.4	153.2±7.9	32.7±1.4	8.0±0.8
C42	77.7±36.2	45.7±11.5	35.3±3.2	23.2±1.0	34.7±6.4	30.7±2.0	3.6±0.8	29.4±6.4	25.0±5.6	6.8±1.0	39.9±5.6	32.2±4.4	5.8±0.1	45.6±6.9	13.2±1.7	5.9±0.4
C43	46.6±8.3	38.5±14.1	32.4±9.8	17.3±4.0	32.6±3.4	30.2±4.3	8.7±0.3	22.2±1.4	18.1±3.2	4.4±1.4	33.5±7.4	26.3±6.4	6.2±1.1	42.1±1.7	12.3±1.1	4.8±0.4
C44	163.3±111.1	17.5±2.6	7.6±3.1	6.3±2.8	24.5±10.3	16.5±0.6	4.3±1.6	19.7±10.8	13.4±0.4	2.4±0.4	25.9±7.1	15.4±3.5	5.1±0.9	28.4±1.7	14.1±1.1	3.8±1.6
C45	26.4±6.6	11.3±0.1	9.8±0.6	7.6±0.8	13.5±5.0	8.8±0.2	6.0±0.3	9.6±1.3	7.4±0.4	7.9	12.5±3.3	8.7±0.4	5.4	10.9±0.5	7.4±0.2	4.4±1.7
C46	191.3±100.4	22.2±0.4	20.5±3.0	10.5±4.8	23.2±0.9	17.0±0.4	8.0±1.6	22.0±4.3	12.6±3.7	8.1±0.6	29.3±2.1	22.8±3.7	5.1±0.2	30.6±3.7	17.2±1.6	3.7±4.5
C48	146.1±51.5	43.4±2.6	37.4±4.8	16.4±6.9	38.3±0.1	21.9±0.8	8.5±0.8	35.1±2.3	17.6±0.4	8.3±2.8	48.0±5.1	27.4±4.3	7.3±1.2	52.3±3.5	15.2±2.1	5.3±1.0
Total	1128.6±265.8	620.2±42.0	498.3±2.3	247.1±37.8	514.4±32.8	368.7±19.0	92.1±5.9	506.9±7.7	248.4±25.6	85.9±5.1	602.6±3.7	392.9±12.4	77.8±1.3	661.4±5.9	225.6±17.3	69.8±0.3

[a] Data are mean±standard deviation (n=2)
*control sample; degummed, neutralized and bleached oil taken in determined oil companies

even carbon numbers was higher than the waxes having odd carbon number. The greatest decrease in wax content (69.8 mg/kg) was observed for the sample for which 0.6 % kieselguhr was used and 93.8 % of waxes were removed during winterization. Wax compounds found in sunflower oil at high amounts were; C46 (191.3 mg/kg), C44 (163.3 mg/kg), C48 (146.1 mg/kg), C41 (161.5 mg/kg), C37 (130.2 mg/kg) and C42 (77.7 mg/kg).

As seen in Table 2, total average wax content of sunflower oil decreased to 69.8 mg/kg at the end of 24 h of the winterization process in which kieselguhr (0.6 %) was used. On the other hand, wax content of sunflower oil was 92.1 mg/kg at the end of 24 h of the winterization performed by using 0.3 % perlite as filter aid material

The effects of using different filter aids and winterization periods on the changes in wax contents of the corn oil samples were given in Table 3. Initial average wax content of corn oil (190.8 mg/kg) was decreased to 30.5 mg/kg by using kieselguhr as filter aid material at a concentration of 0.6 % (Table 3). Average total wax content was decreased to 30.5 mg/kg (0.6 % kieselghur) and 34.7 mg/kg (0.6 % perlite) at the end of 24 h by using filter aid materials (Table 3). Because their amounts in oils are low, removal of the waxes from oils is difficult. The amount of waxes having odd carbon number declined to level of 39% from 48% at the end of 24 h by using 0.6% kieselghur. Six h or 12 h are not adequate periods to the remove the waxes from oils.

The average wax contents of the both oils were significantly affected by the winterization process ($p < 0.01$). When the amount of waxes removed from oils was taken into consideration, for both perlite and kieselguhr, 0.6% was optimum compared to 0.3%. However there were no significant differences observed among the concentrations 0.3 and 0.6% ($p > 0.05$).

According to Hamm and Hamilton (2000), wax content should be lowered to 10-100 ppm to obtain cold-resistant oil. Average wax contents of our samples were among these values.

Findings of Brevedan et al. (2000) and Carelli et al. (2002) regarding the wax content of crude sunflower oil are higher than our findings. In addition, our findings related to average wax contents of sunflower and corn oils are similar to the findings of Hamm and Hamilton (2000), Martini and Anon (2005).

Conclusions

By using filter aid materials, C34 and compounds having higher amounts of carbon in their structures could be removed from oil compared to the control sample. Using kieselguhr as filter aid material increased the amounts of waxes removed from oil during process. Optimum winterization period was

Table 3
Wax content and composition of corn oil during winterization (mg/kg) [a]

Waxes carbon number	C.sample*	Control winterization			Perlite						Kieselghur					
					0.3			0.6			0.3			0.6		
	Time	6	12	24	6	12	24	6	12	24	6	12	24	6	12	24
C34	10.5±1.2	9.2±2.3	2.9±2.5	2.4±2.6	6.1±1.9	3.5±0.2	2.2±0.2	9.1±0.3	3.6±1.5	1.9±0.4	7.8±0.1	4.6±1.5	2.7±0.3	9.3±0.6	5.4±5.1	1.8±0.1
C35	5.7±0.5	1.8±0.4	1.4±0.8	1.4±0.8	2.7±0.8	2.4±0.7	2.1±0.4	2.5±0.7	2.4±0.8	1.4±0.7	2.2±0.1	1.8±0.2	2.0	2.5±0.2	2.2±0.1	1.8±0.4
C36	9.8±5.1	8.8±3.2	6.0±4.9	4.4±2.8	5.6±1.0	5.5±0.6	3.3±0.6	5.8±0.1	3.9±0.6	1.4±0.1	4.6±0.4	3.4±1.8	1.8±0.3	6.7±1.1	5.3±2.1	2.2±0.1
C37	11.5±3.6	9.7±0.9	6.9±3.8	5.8±2.8	8.2±1.0	5.9±2.4	3.1±0.1	6.4±2.1	6.2±2.1	1.8±0.2	7.9±2.5	7.4±3.6	2.7±0.2	9.2±2.3	5.5±2.0	2.0±0.4
C38	9.3±2.5	8.7±3.0	4.2±2.3	3.3±2.1	5.9±1.3	5.9±0.9	3.5±0.2	4.9±2.5	4.7±2.6	2.4±0.1	4.1±2.4	3.3±2.0	2.2±0.4	7.5±1.2	6.0±2.0	2.5±0.6
C39	11.4±2.7	6.6±0.6	6.0±0.9	5.4±0.3	8.7±5.4	6.6±2.3	2.4±0.1	6.4±1.3	6.0±1.5	1.8±0.9	6.9±0.6	5.1±1.0	2.5±0.7	6.9±1.3	5.4±1.0	1.8
C40	9.6±0.2	6.8±3.3	6.4±2.8	4.0±0.4	5.3±0.3	5.1±0.9	2.6±0.8	3.5±1.1	2.8±1.2	1.4±0.8	5.9±0.6	4.2±1.4	2.4±0.6	6.1±1.2	6.1±5.2	2.7±0.2
C41	16.6±2.8	9.4±2.5	6.3±3.3	4.9±3.3	5.8±1.3	5.8±1.2	2.1±0.1	6.7±0.6	6.2±1.0	3.2±1.0	7.9±3.0	5.7±0.5	2.5±0.4	9.0±0.7	6.5±1.3	2.6±0.1
C42	16.3±12.6	8.7±4.9	6.6±3.5	4.2±3.1	7.8±0.2	7.0±0.6	1.7±0.4	4.9±4.9	3.7±3.6	1.7±0.9	6.7±2.2	4.1±3.1	2.1±0.1	8.0±3.3	4.2±0.6	2.8±0.1
C43	34.8±4.5	24.3±1.8	19.6±0.3	6.9±0.5	23.8±4.6	6.7±3.5	2.5±0.4	22.5±2.4	19.1±0.8	4.2±0.7	20.8±5.0	11.7±9.3	2.0±1.0	14.5±2.8	9.2±2.8	1.9±0.2
C44	8.3±1.6	7.7±2.0	4.7±3.0	3.4±1.8	7.5±0.7	7.2±1.2	2.8±0.4	7.0±1.1	6.3±1.5	4.1±0.6	7.0±0.6	5.3±0.9	2.3±0.3	5.7±0.7	5.7±0.1	2.3±0.1
C45	12.7±1.9	9.2±4.2	6.5±3.7	3.9±1.2	8.3±1.6	6.4±0.5	2.4±0.7	9.4±1.6	8.3±0.6	3.4±0.1	7.9±1.4	7.3±0.4	2.6±0.6	8.3±0.8	7.8±0.9	2.3±0.4
C46	17.5±3.3	14.3±1.1	4.3±2.1	2.6±1.5	8.5±0.1	7.3±0.3	1.5±0.6	11.9±1.6	8.0±1.7	2.6±0.1	10.3±3.6	7.4±0.8	3.0±0.2	10.0±1.8	6.6±3.7	2.5±0.1
C48	17.3±7.5	13.0±3.1	6.2	3.9±1.1	10.8±1.8	6.6±0.6	2.8±0.4	12.2±5.9	6.2±1.6	3.3±0.3	13.5±3.3	5.2±1.2	2.0±0.1	8.0±0.1	7.7±0.9	1.8±0.1
Total	190.8±8.4	137.8±5.1	89.8±4.7	56.1±0.3	114.9±14.1	81.4±9.8	34.7±0.4	112.8±14.5	87.0±4.2	34.3±1.6	113.0±5.4	76.1±2.3	32.7±1.2	111.5±5.9	83.2±5.5	30.5±1.1

[a] Data are mean±standard deviation (n=2)

*control sample; degummed, neutralized and bleached oil taken in determined oil companies

24 h for adequate removal of waxes. Turbidity, which is the one important physical parameter, decreased generally according to the control sample during winterization. But in some treatments when the filter aid materials pass through filter to the oil, it increased at the end of 12 h of winterization and decreased again in 24 h.

References

Anon., IUPAC, 1991. International Union of Pure and Applied Chemistry. *Determination of triglycerides in vegetable oils in terms of their partition numbers by high performance liquid chromatography*, **63:** 1173-1182.

Anon., AOCS, 1997. Official Methods and recommended practices of the American Oil Chemists Society, *Preparation of Methyl Esters of Fatty Acids*, method Ce 2-66.

Anon., 2009. *Determination of the Content of Waxes, Fatty Acid Methyl Esters and Fatty Acid Ethyl Esters by Capillary Gas Chromatography.* International Olive Council, COI/T.20/Doc. No 28.

Baltanas, M. A., H. Molina and C. Silva, 1998. Rapid methods for predicting the appearance of turbidity in sunflower oil and their comparison with cold tests. *J. Am. Oil. Chem. Soc.,* **75:** 363-370.

Baümler, E. R., G. H. Crapiste and A. A. Carelli, 2007. Sunflower-oil wax reduction by seed solvent washing. *J. Am. Oil. Chem. Soc.,* **84:** 603-608.

Brevedan, M. I. V., A. A. Carelli and G. H. Crapiste, 2000. Changes in composition and quality of sunflower oils during extraction and degumming. *Grasas Aceites,* **51:** 417-423.

Carelli, A. A., L. M. Frizzera, P. R. Forbito and G. H. Crapiste, 2002. Wax composition of sunflower seed oils. *J. Am. Oil. Chem. Soc.,* **79:** 763-768.

Coutinho, C., M.C.C.R.C. Basso, A. P. B. Ribeiro, L. A. G. Gonçalves and L. A. Viotto, 2009. State of art of the application of membrane technology to vegetable oils: A review. *Food Res. Int.,* **42:** 536-550.

Eldin, A. K., 2006. Effects of fatty acids and tocopherols on the oxidative stability of vegetable oils. *Eur. J . Lipid Sci. Tech.,* **58:** 1051-1061.

Ergönül, P. G. and B. Ergönül, 2008. Changes in fatty acid profiles and omega fatty acid contents of selected vegetable oils during refining process. *Electr. J. Environ. Agri. Food Chem.*, **7:** 2655-2660.

Ferrari, R. A., E. Schulte, W. Esteves, L. Brühl and K. D. Mukherjee, 1996. Minor constituents of vegetable oils during industrial processing. *J. Am. Oil Chem. Soc.,* **73:** 587-592.

Huang, A. S., O. A. L. Hsýeh, C. L. Huang and S. S. Chang, 1981. A comparison of the stability of sunflower oil and corn oil. *J. Am. Oil Chem. Soc.,* **58:** 997-1001.

Hamm, W. and R. J. Hamilton, 2000. Edible Oil Processing. *Sheffield Academic Press,* England.

Kanya, T. C. S., K. U. Sankar and M. C. S. Sastry, 2003. Physical behavior of purified and crude wax obtained from Sunflower (*Helianthus annuus*) seed oil refineries and seed hulls. *Plant Food Hum. Nutr.,* **58:** 179-196.

Kanya, T. C. S., L. J. Rao and M. C. S. Sastry, 2007. Characterization of wax esters, free fatty alcohols and free fatty acids of crude wax from sunflower seed oil refineries. *Food Chem.,* **101:** 1552-1557.

Kellens, M., 1997. Le Raffinage des huiles et des graisses, Etat des lieux à la veille de l'an 2000. Traduit de Current Developments in Oil Refining Technology. Extraction De Smet A l'Occasion du Lipidex. De Smet Group Antwerp, Belgium

Lida, H. M. D. N., K. Sundram, W. L. Siew, A. Aminah and S. Mamot, 2002. TAG composition and solid fat content of palm oil, sunflower oil, and palm kernel olein blends before and after chemical interesterification. *J. Am. Oil Chem. Soc.,* **79:** 1137-1144.

Martini, S. and M. C. Anon, 2000. Determination of Wax Concentration in Sunflower Seed Oil. *J. Am. Oil Chem. Soc.,* **77**(10): 1087-1093.

Martini, S. and M. C. Anon, 2003. Crystallization of sunflower oil waxes. *J. Am. Oil Chem. Soc.,* **80:** 525-532.

Martini, S. and M. C. Anon, 2005. Storage of sunflower-seeds:Variation on the wax content of the oil. *Eur. J. Lipid Sci. Tech.,* **107:** 74-79.

Morello, J. R., M. J. Motilva, T. Ramo and M. P. Romero, 2003. Effect of freeze injuries in olive fruit on virgin olive oil composition. *Food Chem.,* **81:** 547-553.

O'Brien, R. D., W. E. Farr and P. J. Wan, 2000. Introduction to Fats and Oils Technology, 2nd edn. AOCS Press, Champaign, IL, USA.

Raß, M., C. Schein and B. Matthaus, 2008. Virgin Sunflower Oil. *Eur. J. Lipid Sci. Tech.,* **110:** 618-624.

Santos, J. C. O., I. M. G. Santos and A. G. Souza, 2005. Effect of heating and cooling on rheological parameters of edible vegetable oils. *J. Food Eng.,* **67:** 401-405.

SAS, 2001. SAS/STAT User's Guide (8.02), SAS Institute Inc., Cary, NC, USA.

Tan, C. P. and Y. B. Man, 2000. Differential scanning calorimetric analysis of edible oils: comparison of thermal properties and chemical composition. *J. Am. Oil Chem. Soc.,* **77:** 143-155.

Tasan, M. and M. Demirci, 2003. Trans FA in sunflower oil at different steps of refining. *J. Am. Oil Chem. Soc.,* **80:** 825-828.

Turkulov, J., E. Dimic, Dj. Karlovic and V. Vuksa, 1986. The effect of temperature and wax content on the appearance of turbidity in sunflowerseed Oil. *J. Am. Oil Chem. Soc.,* **63:** 1360-1363.

Xu, X. Q., V. H. Tran, M. Palmer, K. White and P. Salisbury, 1999. Chemical and physical analyses and sensory evaluation of six deep-frying oils. *J. Am. Oil Chem. Soc.,* **76:** 1091-1099.

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