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Effect of roasting on the physico-chemical properties, fatty acids, polyphenols and mineral contents of tobacco (*Nicotiana tabacum* L.) seed and oils

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Abstract

The physico-chemical properties, phytochemicals, mineral contents of tobacco (*Nicotiana tabacum* L.) seeds grown at Samsun province in Turkey were evaluated. The oil contents of tobacco seeds ranged from 20.6% (control) to 29.0% (microwave-roasted). *L*, a** and *b** values of tobacco seeds ranged from 32.38 to 35.61; from 6.32 to 6.78; from 13.72 to 14.27, respectively. Total phenolic contents of tobacco seed extract and oils were reported between 31.02 (oven-roasted) and 34.42 mg GAE/100 g (microwave-roasted) to 4.60 (microwave-roasted) and 6.45 mg GAE/100 g (oven-roasted), respectively. Total flavonoid values of raw and roasted tobacco seed extract and oils were determined between 26.62 (oven) and 67.10 mg/100 g (control) to 21.57 (control) and 44.71 mg/100 g (microwave-roasted), respectively. Gallic acid, 3,4-dihydroxybenzoic acid and catechin are the predominant phenolic components of raw and roasted tobacco seed oils. The amounts of oleic and linoleic acid in raw and roasted tobacco seed oils ranged from 10.23% (oven-roasted) to 12.48% (control) and 73.72% (control) to 76.63% (oven-roasted), respectively. The abundant elements found in seeds were K, P, Ca, Mg, S and Fe. The mineral amounts of the roasted seeds were found higher than that of the control. The highest increase was detected in oven roasted tobacco seeds.

KEYWORDS
bioactive properties, fatty acid, minerals, oil, polyphenols, roasting, tobacco seed

INTRODUCTION

Tobacco (*Nicotiana tabacum* L.; Solanaceae family) is an economically important perennial industrial crop, widely cultivated all over the World. Tobacco, which is the raw material of the cigarette industry and one of the most commercially valuable agricultural crops, is grown in many countries of the World (Kirkova et al., 2016). Tobacco, which is the world's most important non-food agricultural product with an increase in production exceeding four million hectares all over the World (Chen et al., 2012; Poltronieri, 2016). Some previous studies have emphasized the importance of the type and concentration of phenols in tobacco leaves and their antimicrobial and antioxidant activity at different developmental stages (Ru et al., 2012; Torras-Claveria et al., 2012). As a result of the chemical characterization of tobacco seeds, it is important to process the seeds into alternative products, namely oil and pulp, and to determine some usage areas of these products (Deng et al., 1998; Zlatanov et al., 2007). The oil of tobacco seed, which is an important agricultural product for the production of oleo-chemicals, contains significant levels of linoleic acid (Abbas et al., 2008). Therefore, tobacco seed oil falls into the category of semi-drying oil due to its high linoleic acid amount (Ashirov et al., 2020). Since tobacco seed oil is rich in linoleic acid, it can be used in the preparation of long-chain compounds in the formulation of acid protective coatings, plastics, surfactants, dispersants, biolubricants, cosmetics and various synthetics (Awola et al., 2010; Frega et al., 1991).

More than 20% of tobacco plant parts are disposed of as processing waste, which pollutes the environment (Ru et al., 2012). Tobacco seed is a good source of phytochemicals, protein, oil, cellulose, proteins, starch, and minerals and organic acids. Also, tobacco seeds contain contain ∼30%–43% oil (Kirkova et al., 2016; Ranjhan, 1978).Tobacco seed oil contains low concentrations of saturated fat compared with other vegetable oils. Tobacco seed oils contain linoleic (65%–75%), oleic(10%–16%), palmitic (8%–11%) and stearic (2%–3%) acids as dominant fatty acids, and so this nicotine-free oil is comparable to other edible oils (Ashraf-Khorassani et al., 2015; del Piano et al., 2014; Frega et al., 1991; Giannelos et al., 2002; Patel et al., 1998; Stanisavljevic et al., 2007; Stanisavljević et al., 2009; Zdremtan & Zdremtan, 2006). Tobacco seed oil and flour have high antioxidant activity (Ashirov et al., 2020). Therefore, tobacco seed oil is used in the preparation of various salads and sauces. In addition, defatted tobacco seed cakes can also be added to bread (Ashirov et al., 2020). It has been stated that the nutritional value of refined tobacco seed oils used as cooking oil in some European countries is similar to peanut, cottonseed and safflower seed (Talaqani et al., 1986). Recently, studies on alternative uses of tobacco have started and it has gained value especially as an oily plant (Stanisavljević et al., 2009). Tobacco is an industrial crop traditionally used for manufacturing cigarettes. However, due to restriction of European subsidies, an alternative use of tobacco is needed, such as biofuel or biomass (Grisan et al., 2016). The tobacco plant, which is grown in large areas in Turkey, contains a significant amount of seeds with high oil content and the use of these seeds is of great interest to the tobacco manufacturing industry. A limited number of studies have been carried out on the bioactive properties, color values, phenolic components, fatty acid composition and mineral contents of tobacco seeds by applying different roasting techniques. The objective of present research was to investigate the effect of roasting process (oven and microwave system) on the physiochemical properties, fatty acid, polyphenols, element contents of tobaco (*Nicotiana tabacum* L.) seeds.

MATERIALS AND METHODS

Material

Tobacco seeds were provided from tobacco plants harvested in Turkey (Samsun-Bafra province) in 2021. Bafra's GPS coordinates are 41°33′55.5048″ and 35°53′42.8676″. After the seeds are separated from the plant, they are cleaned of foreign materials such as stem, leaves and soil. After the cleaned seeds were taken into cloth bags, they were brought to the laboratory. Seeds were air-dried by laying on a tray with a thickness of ∼1–2 mm in the laboratory. After the seeds were roasted in a laboratory grinder (Sinbo SCM 2934), seeds were ground. Then, they were filled into colored glass jars and analyzed. The ground samples were kept in the refrigerator (4°C) during the analysis.

Methods

Moisture content

Moisture contents of raw and roasted-tobacco seeds were monitored at 100 ± 5°C in an oven until a constant weight was reached.

$$\text{Moisture content}\,(\%) = \frac{\text{Loss in weight}}{\text{Weight of sample}} \times 100.$$

Roasting process

Roasting process of tobacco seed was made in a conventional oven and microwave oven (Arçelik-MD 20 M Vintage) at 160°C for 20 min; and 720 W for 16 min, respectively. Raw and roasted seeds were ground by a grinder (Siemens MC 23200) before analyses.

Color values

A Minolta Chroma meter CR 400 (Konica Minolta, Inc.) was used for determination of color values of tobacco seed samples. The equipment was calibrated against the white surface calibration plate before measurement and the *L* a* b** values which determined according to the CIELab color scale (Pagliarini and Rastelli, 1994). Color measurements consist of three axes: *L**, dark to light, with values ranging from 0 to 100; *a** is green to red with values −128 to +127; and *b** represents values from −128 to +127 from blue to yellow (Fraser et al., 2004).

Protein content

Protein content of the tobacco seeds was determined using AOAC (2005) *Method 945.18-B*. Nitrogen Percentage and protein calculated by the following equations:

Where, Ts = titre volume of the sample (ml), TB = titre volume of blank (ml), 0.014 = M eq. wt. of N_2.

$$\text{Protein}\,\% = \text{Nitrogen} \times 6.25.$$

Oil content

The total oil content of cleaned tobacco seeds was established. Tobacco seeds were ground in a laboratory grinder (Sinbo SCM 2934), through a 0.5 mesh sieve. After weighing 10 g of the ground sample into the Soxhlet cartridge, the cartridge was placed in the Soxhlet apparatus. After the seed sample was extracted with petroleum ether in the Soxhlet Apparatus for 5 h, the solvent in the micella was removed with a rotary vacuum evaporator (Heidolph Laborota 4001, Germany) at 50°C. The remaining oil amount was calculated gravimetrically and the oil content of the seeds was determined (AOAC, 1990).

Extraction of phenolic compounds

Extraction of phenolic compounds from the tobacco seeds were carried out according to the method applied by Jakopic et al. (2011). After 15 ml of methanol were added to ground tobacco seed sample (5 g), it was mixed. After shaking the mixture in an ultrasonic water bath (Bandelin Sonorex, Germany) at 25°C for 30 min, the samples were centrifuged at 6000 rpm for 10 min. The collected methanol phase was evaporated at 40°C. The dried extract was then dissolved in 10 ml of methanol. After adding 12 ml of methanol/water (70:30 vol/vol) to 2 ml of extract, the mixture was thoroughly mixed manually. Then, after the mixture was vortexed for 1 min, the mixture was sonicated in an ultrasonic bath for 10 min. Immediately after sonication, the sample was centrifuged at 6000 rpm for 5 min. *n*-hexane (5 ml) was added to the supernatant, which was removed after centrifugation and mixed using a vortex apparatus. The methanol and *n*-hexane layers were separated in a separator funnel. This step was performed twice with 5 ml of *n*-hexane (Durmaz & Gökmen, 2011).

Total phenolic content

The Folin-Ciocalteau reagent method described by Yoo et al. (2004) were used for the total phenolic contents of tobacco seed extracts. After 1 ml Folin–Ciocalteu reagent and 10 ml Na_2CO_3 were added to tobacco seed extract, mixture was stirred by vortex. After adding deionized water until the sample volume was 25 ml, the samples were kept in the dark for 1 h. The absorbance values of the samples were evaluated at 750 nm in a spectrophotometer (Shimadzu UV mini 1240). A standardization calibration curve was prepared using gallic acid (0–200 mg/ml). Results were described as mg gallic acid equivalent (GAE)/100 g.

Total flavonoid content

For flavonoid analysis of samples, 0.3 ml of $NaNO_2$, 0.3 ml of $AlCl_3$ and 2 ml of NaOH were added to 1 ml of tobacco seed extract, respectively, then mixed well and left in the dark for 15 min. The absorbance of the mixture was determined at 510 nm. The results obtained were determined as mg quercetin equivalent (QE)/100 g (Hogan et al., 2009).

Antioxidant activity

After adding 2 ml of 0.1 mM methanolic 1.1-diphenyl-2-picrylhydrazyl (DPPH) solution to the tobacco seed extract, the mixture was thoroughly mixed with a vortex. Then, after the samples were stored in the dark for 30 min, the absorbance values of the tobacco seed extracts were measured at 517 nm after 30 min of reaction. Results were expressed as mmol trolox equivalent (TE)/kg (Lee et al., 1998).

Determination of phenolic compounds

Chromatographic separation of phenolic compounds of the tobacco seed extracts was performed by HPLC (Shimadzu) equipped with a PDA detector and an Inertsil ODS-3 (5 μm; 4.6 × 250 mm) column. A mixture of 0.05% acetic acid in water (A) and acetonitrile (B) with the flow rate of 1 ml /min at 30°C for mobile phase were prepared. The volume of injection was 20 μl. The peaks were obtained at 280 using a PDA detector, and and 60 min were selected for the total running time for per sample. The elution program was employed: 0–0.10 min 8% B; 0.10–2 min 10% B; 2–27 min 30% B; 27–37 min 56% B; 37–37.10 min 8% B; 37.10–45 min 8% B. The total running time per sample was 60 min. Gallic acid, 3,4-dihydroxybenzoic acid, catechin, caffeic acid, syringic acid, rutin, *p*-coumaric acid, ferulic acid, resveratrol, quercetin, cinnamic acid and kaempferol were used as standards for the determination of phenolic compounds in tobacco seeds. The concentration range for standard curve is 0–200 mg/L.

Fatty acid composition

Fatty acid methyl esters of tobacco seed oil was prepared according to ISO-5509 (1978) method, and fatty acid methyl esters were analyzed by the gas chromatography (Shimadzu GC-2010) equipped with flame-ionization detector (FID) and capillary column (Tecnocroma TR-CN100, 60 m × 0.25 mm, film thickness: 0.20 μm). The temperature for injection block and detector temperature was set as 260°C. Mobile phase

was nitrogen with 1.51 ml/min flow rate. Total flow rate was 80 ml/min and split rate was 1/40. Column temperature was programmed 120°C for 5 min and increased 240°C at 4°C/min and held 25 min at 240°C. Total flow and partitioning rates were set as 80 ml/min and 1/40, respectively.

Determination of mineral composition

Tobacco seeds were dried at 70°C in a drying cabinet till constant weight. Each sample was combusted with 5 ml of 65% HNO_3 and 2 ml of 35% H_2O_2 in a closed microwave system (Cem-MARS Xpress). It was brought to 20 ml with distilled water till the volumes. The amounts of minerals of tobacco seeds were performed by Inductively Coupled Plasma Optical Emission spectroscopy (ICP-OES) (Skujins, 1998).

Statistical analyses

The roasting process was repeated twice, and also the analyses were repeated three times ($n = 6$). The means of significant variation sources were compared using Duncan Multiple Comparison Test with the help of MSTAT-C (MSU) program. The significance level was given as $p < 0.05$ unless stated otherwise.

RESULTS AND DISCUSSION

Physcio-chemical and bioactive properties of tobacco seed and oils

The physico-chemical properties of tobacco seeds roasted in microwave and oven methods are given in Table 1. After tobacco seeds were roasted, their oils were extracted. The total phenolic, flavonoid amounts and antioxidant activities of the oil samples were determined. Results obtained from seed and oils showed some fluctuations depending on roasting types compared with the control. The moisture contents of raw and roasted tobacco seeds ranged from 4.35% (microwave and oven) to 11.22% (control). In addition, the oil yields of seed samples were measured between 20.6% (control) and 29.0% (drybasis) (microwave). This oil ratio varies depending on many factors such as the type of tobacco, growing conditions and cultivation area (Ashirov et al., 2020; del Piano et al., 2014; Giannelos et al., 2002; Hutchens, 1999). The moisture contents of tobacco seeds decreased with the roasting process. In addition, the oil contents of the seeds increased when compared with the control. With the roasting process, the oil globules trapped in the seeds are collected and it is easier to leak out from the seed. The oil amounts of microwave-roasted tobacco seeds increased when compared with the control. Rao (1994) determined 3.0% moisture and 33.7% fat in tobacco seed. The oil contents of tobacco seeds were reported between 24.56% and 41.93% (Abbas et al., 2008; Mohammad & Tahir, 2014). Tobacco seed contains a wide range of oil, up to 36%–41% of the seed weight depending on the type of tobacco, growing conditions and cultivation area (Gofur et al., 1993; Patel et al., 1998). Since tobacco seeds have the highest crude oil content, they can be used as an alternative fuel source in the production of biodiesel (Fornasier et al., 2018). Tobacco seed oil, which has the feature of being used in the production of quick-drying oils, can also be used in oleochemical production, in other fields such as cosmetics and surfactants (Chiririwa et al., 2014).

L^*, a^* and b^* values of tobacco seeds ranged from 32.38 to 35.61; from 6.32 to 6.78; from 13.72 to 14.27, respectively (Table 1). The lowest L^* value was obtained in control sample, and roasting process

TABLE 1 Some chemical and bioactive properties of the oils and tobacco seeds roasted in microwave and conventional oven

Sample	Process	Moisture content (%)	Oil content (%)	L^*	a^*	b^*
Seed	Control	11.22 ± 0.21a**	20.6 ± 1.48c	32.38 ± 0.38b	6.78 ± 0.06a	14.27 ± 0.73a
	Microwave roasting	4.35 ± 0.02b***	29.0 ± 0.84a	35.61 ± 0.45a	6.32 ± 0.18c	14.03 ± 0.20ab
	Conventional roasting	4.35 ± 0.16b	27.1 ± 0.98b	35.51 ± 0.89a	6.41 ± 0.30b	13.72 ± 0.51c

Sample	Process	Total phenolic content (mg GAE/100 g)	Total flavonoid content (mg QE/100 g)	Antioxidant activity (mmol TE/kg)
Seed	Control	32.10 ± 0.37b	67.10 ± 2.43a	0.60 ± 0.01c
	Microwave roasting	34.42 ± 0.75a	34.05 ± 1.20b	0.94 ± 0.02a
	Conventional roasting	31.02 ± 1.05c	26.62 ± 1.37c	0.79 ± 0.02b
Oil	Control	5.79 ± 0.12b	21.57 ± 1.46c	0.69 ± 0.06a
	Microwave roasting	4.60 ± 0.13c	44.71 ± 1.07a	0.66 ± 0.03b
	Conventional roasting	6.45 ± 0.18a	38.71 ± 1.46b	0.60 ± 0.02c

Note: *Color parameters; **standard deviation; ***values within each column followed by different letters are significantly different at $p < 0.05$.

caused a bit increase in L^* values. A significant difference was not observed in both a^* and b^* values by the roasting process. According to our knowledge, there is not a study about L^*, a^* and b^* values of tobacco samples. Contrary to current results, Gunel et al. (2019) reported that L (41.54–55.28) and b (16.45–20.91) values of the carob powder decreased, and also a values (8.88–20.91) increased after roasting process. In a study conducted by Ma et al. (2020), L^* value decreased from 88.62 to 80.99), and a^* increased from 1.21 to 2.55. In addition, b^* values increased from 6.40 to 10.66 in buckwheat flours (Ma et al., 2020). In another study, pan roasting process caused a decrease in L^* value from 83.01 to 72.77, and increase in a^* (from −0.27 to 2.49) and b^* (from 15.49 to 25.25) values of arrowhead (Wani et al., 2016). Also, total phenolic amounts of seed and their oils were reported between 31.02 (oven) and 34.42 mg GAE/100 g (microwave) to 4.60 mg GAE/100 g (microwave) and 6.45 mg GAE/100 g (oven), respectively. While total flavonoid values of raw and roasted tobacco seeds vary between 26.62 (oven) and 67.10 mg/100 g (control), total flavonoid values of tobacco seed oils were reported between 21.57 (control) and 44.71 mg/100 g (microwave). Antioxidant activity values of tobacco seed and oils were measured between 0.60 (oven) and 0.69 mg/kg (control), respectively. The total phenolic amounts of the oven-roasted tobacco seeds was partially decreased when compared with the control. The contents of total phenolic of tobacco seed oils are seen in Table 1, which is the opposite of the processes in the seed. That is, the total phenol values of the oil obtained from the oven-roasted seeds increased when compared with the control. In addition, the total flavonoid amounts of the roasted the seeds decreased when compared with the control, while the total flavonoid values of the oil samples obtained from the roasted tobacco seeds increased when compared with the control. In addition, the antioxidant activity values of the roasted tobacco seeds increased when compared with the control, while the antioxidant activity values of the oils obtained from these seeds were partially decreased. Total phenolic and flavonoid values of tobacco seed oils were significantly decreased when compared with the seeds. The leaves of young and adult plants contained higher amounts of total phenol (14.46–23.05 mg GAE/g) than the other parts (Nasr et al., 2014). Extracts obtained from adult plants petioles dried at 40 and 70°C contained 0.40 and 1.37 mg QE/g flavonoids, respectively (Nasr et al., 2014). These values, which were determined as a result of the analysis at room temperature, were determined as 24.89 ± 0.84 and 32.24 ± 1.52 μg ml/L, respectively (Nasr et al., 2014). Results exhibited some fluctuations compared with results of previous studies. These fluctuations can be probably due to parts used, harvest time, cultural activities, climatic factors and solvents used for extraction.

Phenolic components and their values of raw and roasted-tobacco seed and oils

Phenolic components and their values of raw and roasted-tobacco seed and their oils are illustrated in Table 2. While 3,4-dihydroxybenzoic acid, catechin, rutin and quercetin are found as the major phenolic components in raw and roasted-tobacco seed extracts, the abundant phenolic components of oils obtained from raw and roasted tobacco seeds were gallic acid, 3,4-dihydroxybenzoic acid and catechin. While gallic acid values of raw and roasted tobacco seed extracts vary between 0.12 (control) and 0.83 mg/100 g (microwave), gallic acid values of the oils obtained from raw (control) and roasted tobacco seeds were detected between 2.73 (control) and 4.82 mg/100 g (oven). The 3,4-dihydroxybenzoic acid contents of tobacco seed and oils were determined between 1.19 (oven) and 1.94 mg/100 g (microwave) to 2.73 and 5.66 mg/100 g (microwave), respectively. Catechin values of raw and roasted-tobacco seed extracts were identified between 0.51 (microwave) and 4.25 mg/100 g (control) while catechin contents of the oils provided from raw and roasted tobacco seeds are detected between 1.61 (oven) and 11.98 mg/100 g (microwave). Amounts of rutin in tobacco seed and oils varied between 0.76 mg/100 g (control) 3.01 mg/100 g (microwave) to 0.36 (control) and 1.67 mg/100 g (oven), respectively. Also, while quercetin contents of raw and roasted tobacco seed extracts change between 1.04 (oven) and 1.34 mg/100 g (control), quercetin values of tobacco seed oils were detected between 0.13 (microwave) and 0.77 mg/100 g (oven). Kaempferol amounts of tobacco seed and oil extracts were established between 0.68 (oven) and 1.00 mg/100 g (microwave) to 0.50 (oven) and 1.06 mg/100 g (control), respectively. In general, the contents of phenolic compounds (except for catechin, resveratrol and quercetin) of tobacco seeds and oils roasted in both roasting systems increased compared with the control. While some components increased in microwave roasted seeds and oils extracted from these seeds, some of these components decreased. The amounts of ferulic acid and kaempferol increased in seeds roasted in microwave, but decreased in seeds roasted in oven. In particular, it was observed that the gallic acid, 3,4-dihydroxybenzoic acid and catechin values of tobacco seed oils were significantly higher when compared with the same component amounts detected in the seed. The increase in the content of the phenolic compounds during the roasting process likely resulted in a relative increase in the amounts of the phenolic compounds, possibly as a result of the significant removal of water from the seed by roasting. The characteristics of plants such as growth, yield and biochemical content are affected by climatic conditions (Zandalinas et al., 2018).

TABLE 2 Phenolic compounds of the oils and tobacco seeds roasted in microwave and conventional oven

Phenolic compounds of seed (mg/100 g)	Control	Microwave roasting	Conventional roasting
Gallic acid	0.12 ± 0.06c*	0.83 ± 1.39a	0.36 ± 0.21b
3,4-dihydroxybenzoic acid	1.48 ± 0.42b**	1.94 ± 4.07a	1.19 ± 0.65c
Catechin	4.25 ± 0.36a	0.51 ± 0.11c	1.37 ± 0.51b
Caffeic acid	0.15 ± 0.13c	0.23 ± 0.41a	0.22 ± 0.28b
Syringic acid	0.12 ± 0.14c	1.79 ± 1.58a	1.17 ± 0.27b
Rutin	0.76 ± 1.14c	3.01 ± 3.45a	1.91 ± 0.44b
p-coumaric acid	0.03 ± 0.04c	0.18 ± 0.18a	0.10 ± 0.09b
Ferulic acid	0.11 ± 0.16a	0.12 ± 0.06a	0.07 ± 0.04b
Resveratrol	0.18 ± 0.30a	0.13 ± 0.16b	0.18 ± 0.09a
Quercetin	1.34 ± 0.16a	1.23 ± 0.26b	1.04 ± 0.10c
Cinnamic acid	0.03 ± 0.01b	0.13 ± 0.14a	0.13 ± 0.14a
Kaempferol	0.96 ± 0.10b	1.00 ± 0.58a	0.68 ± 0.10c
Phenolic compounds of oil (mg/100 g)	**Control**	**Microwave roasting**	**Conventional roasting**
Gallic acid	2.73 ± 0.42c*	5.66 ± 0.39a	3.99 ± 0.49b
3,4-dihydroxybenzoic acid	2.07 ± 0.01c**	4.56 ± 1.05ab	4.82 ± 1.95a
Catechin	4.41 ± 0.17b	11.88 ± 0.13a	1.61 ± 0.06c
Caffeic acid	0.11 ± 0.03a	0.08 ± 0.01b	0.06 ± 0.01a
Syringic acid	0.08 ± 0.03c	0.13 ± 0.01a	0.11 ± 0.01b
Rutin	0.36 ± 0.07c	0.89 ± 0.21b	1.67 ± 0.03a
p-coumaric acid	0.04 ± 0.01a	0.05 ± 0.02a	0.04 ± 0.01a
Ferulic acid	0.07 ± 0.01c	0.13 ± 0.04b	0.20 ± 0.01a
Resveratrol	0.16 ± 0.03a	0.07 ± 0.02b	0.08 ± 0.03b
Quercetin	0.27 ± 0.08b	0.13 ± 0.03c	0.77 ± 0.31a
Cinnamic acid	0.19 ± 0.03a	0.09 ± 0.04c	0.17 ± 0.01b
Kaempferol	1.06 ± 0.21a	0.77 ± 0.05b	0.50 ± 0.18c

Note: *standard deviation;**values within each row followed by different letters are significantly different at $p < 0.05$.

TABLE 3 Fatty acid composition of the oils extracted from raw and tobacco seeds roasted in microwave and conventional oven

Fatty acids (%)	Control	Microwave roasting	Conventional roasting
Palmitic	9.40 ± 0.00a*	8.96 ± 0.09b	8.81 ± 0.04bc
Stearic	2.74 ± 0.00a**	2.64 ± 0.00c	2.71 ± 0.01b
Oleic	12.48 ± 0.02a	10.50 ± 0.01b	10.23 ± 0.02c
Linoleic	73.72 ± 0.02bc	76.31 ± 0.04ab	76.63 ± 0.02a
Arachidic	0.20 ± 0.01a	0.19 ± 0.02b	0.19 ± 0.01b
Linolenic	1.27 ± 0.00a	1.19 ± 0.00c	1.20 ± 0.00b
Behenic	0.09 ± 0.01	0.09 ± 0.00	0.09 ± 0.01

Note: *standard deviation;**values within each row followed by different letters are significantly different at $p < 0.05$.

Fatty acid compositions of the oils of raw and roasted-tobacco seed

Fatty acid compositions and their amounts of the oils obtained from raw and tobacco seeds roasted by microwave and oven roasting systems are presented in Table 3. Linoleic, oleic and palmitic acids were the most abundant fatty acids of raw and roasted tobacco seed oils. The predominant fatty acid of tobacco seed oil was linoleic. In the present study, total saturated and unsaturated fatty acid compositions of tobacco seed oil obtained by Soxhlet extraction were determined as ∼12% and 87% in total, respectively. These were followed by stearic and linolenic acids in decreasing order. It has been observed that roasting systems and applied roasting temperature and powers are partially effective on the content of fatty acids in tobacco seeds. While palmitic acid values of raw and roasted tobacco seed oils are detected between 8.81% (oven) and 9.40% (control), stearic acid contents of the oils were

TABLE 4 Mineral (mg/kg) and protein (%) contents of tobacco seeds roasted in microwave and conventional oven

Process	P	K	Ca	Mg	S	Fe	Cu	Mn	B	Zn	Protein (%)
Control	4168.83 ± 106.26c*	9020.53 ± 172.40c	3165.96 ± 114.37c	3466.41 ± 176.06c	2045.74 ± 109.60c	141.73 ± 1.25c	16.44 ± 1.37b	32.63 ± 1.09c	9.64 ± 0.66c	36.40 ± 0.85	24.61 ± 1.10c
Microwave (720 W/16 min)	4238.34 ± 227.55b**	9245.58 ± 189.73b	3385.13 ± 185.84b	3470.01 ± 58.69b	2117.59 ± 155.59b	145.58 ± 2.79b	15.69 ± 0.94c	34.46 ± 0.73b	10.59 ± 0.74b	37.78 ± 1.74	26.01 ± 1.30b
Oven (160°C/20 min)	4643.10 ± 107.73a	11631.91 ± 613.88a	4346.42 ± 102.19a	3951.38 ± 109.66a	2353.06 ± 211.54a	188.60 ± 0.91a	17.63 ± 0.91a	39.83 ± 0.93a	13.61 ± 0.76a	42.91 ± 1.73	26.41 ± 1.18a

Note: *standard deviation;**values within each column followed by different letters are significantly different at $p < 0.05$.

identified between 2.64% (microwave) and 2.74% (control). In addition, oleic and linoleic acid contents of raw and roasted tobacco seed oils were found between 10.23% (oven) and 12.48% (control) to 73.72% (control) and 76.63% (oven), respectively. Linolenic acid contents of oil samples were detected between 1.19% (microwave) and 1.27% (control). The amounts of fatty acid compositions of the oils provided from heat-treated tobacco seeds were reduced compared with the control (except for oleic acid). However, the fatty acids of the oils of the seeds roasted both in the microwave and oven were close to each other in composition. Therefore, tobacco seed oil is included in the oleic-linoleicacid group, which is the largest and most important fatty acid group among oils. Since they do not contain linolenic acid and higher unsaturated fatty acids, they are resistant to serious taste and aroma degradation. There were statistically significant fluctuations among the fatty acids constituent of tobacco oil depending on the different roasting types compared with control ($p < 0.05$). Figure 2 shows a representative fatty acid chromatogram of tobacco oil. The unsaturated fatty acid constituent of tobacco seed oils was found to be close to the amount of poppy and sesame seeds (Kirkova et al., 2015; Kirkova et al., 2016). Tobacco seed oil contained 0.567%–1.233% lauric, 0.7%–1.70% myristic, 0.26% palmitoleic, 8.04%–25.67% palmitic, 3.27%–8.00% stearic, 11.3%–43.8% oleic, 0.67% vaccenic, 14.90%–71.73% linoleic, 0.53%–0.90% alpha-linolenic and 0.33%–0.64% behenic acids (Ashirov et al., 2020; Kirkova et al., 2016; Mohammad & Tahir, 2014; Rao, 1994; Stanisavljević et al., 2009; Zlatanov & Menkov, 2000). Sannino et al. (2017) determined 9.70%–10.03% palmitic, 2.80%–3.00% stearic, 10.9%–11.6% oleic, 74.7%–75.9% linoleic, 0.70%–0.77% linolenic acids in tobacco seed oils. Also, in other studies, the major fatty acids in seed oils were determined linoleic (60%–80%), oleic (10%–20%) and palmitic acids (10%–20%) (Bucciarelli et al., 2013; Srbinoska et al., 2003; Stanisavljević et al., 2009; Zlatanov et al., 2007). The quantitative values of saturated and unsaturated fatty acids determined in this study were very close to those described by Giannelos et al. (2002), Mukhtar et al. (2007), Stanisavljević et al. (2009), Mohammad and Tahir (2014), Kirkova et al., (2016) and Ashirov et al. (2020). Caponio et al. (2003) studied chemical properties of extra virgin olive oil, groundnut oil and sunflower oil heated by conventional electric ovens and microwaves, and they have reported that both unsaturated and polyunsaturated fractions are significantly reduced. This is related to the decrease of unsaturated fatty acids, and the increase in the percentage of the sum of saturated fatty acids (Barreto et al., 2016). Yoshida et al. (2003) studied the effects of microwave treatment on pumpkin seeds, and reported a change in the fatty acid composition of the pumpkin oil through the effect of microwave heating treatment. Also, changes in the fatty acid

composition after microwave pretreatment of oil seeds have been previously reported by Takagi and Yoshida (1999), Abbas et al. (2008). Hassanein et al. (2003) has reported that polyunsaturated fatty acids were reduced as the microwave heating time increases. In addition, Majid et al. (2014) reported that the content of monounsaturated fatty acid varies slightly depending on the heat treatment and the ratio of polyunsaturated fatty acids decreased. In addition, Abbas et al. (2008) have heated corn oil in microwave for different watts and times and the the results obtained showed that the content of linoleic acid in the corn oil was reduced. The slight differences in the fatty acid composition of tobacco seed oil may have resulted from the harvest time of the tobacco used in the study, growing conditions, cultural and climatic factors, or different environmental or geographical and analytical conditions.

Mineral contents and their values of raw and roasted tobacco seeds

Mineral contents of tobacco seeds are given in Table 4. The abundant elements found in seeds were K, P, Ca, Mg, S and Fe. The mineral contents of the roasted seeds increased when compared with the control. The highest increase was detected in oven roasted tobacco seeds. While P contents of raw and roasted-tobacco seeds vary between 4168.83 (control) and 4643.10 mg/kg (oven), K values of seeds ranged from 9020.53 (control) to 1163.91 mg/kg (oven). In addition, Ca and Mg values of raw and roasted tobacco seeds ranged from 3165.96 (control) to 4346.42 mg/kg (oven) and 3466.41 (control) to 3951.38 mg/kg (oven), respectively. S values of seeds changed between 2045.74 (control) and 2353.06 mg/kg (oven). While Fe amounts of tobacco seeds vary between 141.73 (control) and 188.60 mg/kg (oven), Zn amounts of seeds ranged from 36.40 (control) to 42.91 mg/kg (oven). The highest Cu (17.63 mg/kg), Mn (39.83) and B (13.61 mg/kg) were determined in tobacco seeds roasted in oven. Rao (1994) determined 483 P, 178 Ca, 345 Mg, 33 Fe, 7.8 Zn, 3.0 Mn, 0.5 Cu, 0.07 mg/100 g Cr in tobacco seed. The protein contents of raw and roasted tobacco seeds changed depending on roasting types compared with the control. The protein contents of tobacco seeds were reported between 24.61% (control) and 26.41% (oven). There were observed statistically significant fluctuations between treated seeds and control group ($p < 0.05$). Rao (1994) determined 24.7% protein in tobacco seed. Mohammad and Tahir (2014) determined 20.861% to 23.872% protein in tobacco seed. The protein amount of tobacco seed is 23%–28% and most of them are globulins (Deng et al., 1998). Results obtained on mineral and protein were found partially similar compared with literature values. The small difference in the contents of different minerals and protein contents may be due to agricultural and climatic factors, fertilizers, harvest period of tobacco and reagents and solvents used in mineral and protein analysis.

CONCLUSION

The lowest L^* value was obtained in control sample, and roasting process caused a small increase in L^* values. The total phenolic and flavonoids of the oven-roasted seeds was partially decreased when compared with the control. The amounts of phenolic compounds (except for catechin, resveratrol and quercetin) of tobacco seeds and oils roasted in both roasting systems increased compared with the control. The amount of unsaturated fatty acids in tobacco seed oil was higher than the amount of saturated fatty acids. The high content of linoleic acid in tobacco seed oil indicates that tobacco seed oil is a potential source of essential fatty acids. Tobacco seed oil is included in the oleic-linoleic acid group, which is the largest and most important fatty acid group among oils. The mineral contents of the roasted seeds increased when compared with the control. The results indicated that tobacco seed contained large amount of lipid and protein. The increase in mineral and oil content of roasted seeds is likely due to reduced moisture content during roasting. It is considered to be an ideal source of oilseeds as a source of edible oil in terms of its fatty acid composition.

AUTHOR CONTRIBUTIONS

Mehmet Musa Özcan: Investigation; writing. **Nurhan Uslu:** Formal analysis. **Viktar Lemiasheuski:** Data curation. **Duygu Akçay Kulluk:** Mineral analysis. **Nesim Dursun:** Conceptualization.

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CONFLICT OF INTEREST STATEMENT

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

ETHICS STATEMENT

No human or animal subjects were used in the research for this paper.

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