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Fatty acid composition and organoleptic characteristics of *Tilia caucasica* Rupr. fruit oil

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Abstract: The article reports the results of physicochemical, organoleptic characteristics and the fatty acid composition of the oil obtained from the fruits of *Tilia caucasica*. Data on the qualitative composition and quantitative content of fatty acids for oil from *T. caucasica* fruits growing in Azerbaijan, as well as physico-chemical and organoleptic indicators for oil of this species were established for the first time. It was determined that oil yield of the fruits of *T. caucasica* was 12.89%. The color of *T. caucasica* fruits oil was light yellow, with a pleasant taste and had the following characteristics: content of free fatty acids - 1.99 mmol/kg, peroxide value - 3.02, iodine value -128.4, mass fraction of phosphorus-containing substances - 91mmol/kg. The determination of methyl ester level of the oil fatty acids by gas-liquid chromatography revealed the presence of 13 fatty acids: myristic, palmitic, palmitoleic, heptadecanoic, heptadecadienoic, stearic, malvalic, oleic, elaidic, linoleic, linolenic, gandoic, and sterculic. Linoleic acid (46.06%) was the major polyunsaturated fatty acid, among monounsaturated acids - oleic (16.92%), among saturated acids - palmitic (9.36%) and among cyclopropenoid fatty acids sterculic acid (13.35%) presented the highest levels. The evaluation of the physicochemical and organoleptic characteristics, the content of biologically valuable fatty acids in *T. caucasica* fruit oil allow to recommend it for use in food, medicine and cosmetic purposes.

Key Words: *fruit, oil, chromatography*

INTRODUCTION

The genus *Tilia* L. (Malvaceae Juss.) includes about 22 species and 12 subspecies [Alverson et al., 1999; Bayer et al., 1999; Pigott, 2002]. Four species and two natural hybrids grow in Europe and South Asia, four in North America, and the others in East Asia [Liesebach, Sinko, 2008; Mccarthy, 2012]. There are three species of linden wildly growing in Azerbaijan: *T. caucasica* Rupr. (= *Tilia begoniifolia* Stev. = *T. prilipkoana* Grossh. and J. Wagner.), *T. cordata* Mill. (= *Tilia parvifolia* Ehrh. Ex Hoffm.) and *T. platypyllos* Scop. [Askerov, 2006]. The most widespread species in the republic is *T. caucasica* which is most common in mixed forests of the eastern and western parts of the Greater Caucasus (within Azerbaijan), the Quba massive, the northern and central parts of the Lesser Caucasus and also Lenkaran from the lower to upper mountain zones [Flora of Azerbaijan, 1955].

Linden flowers of the species *T. platyphyllos* Scop., and *T. cordata* Mill are useful as a sedative, diuretic, diaphoretic and expectorant in folk medicine. The flowers are also widely used for the treatment of headaches and stomachaches, fainting, diseases of the liver, intestines, and kidneys [Gulnur, 1995]. In Jordan Pharmacopeia, the plant is used to treat diabetes [Otoom et al., 2006]. In addition, the fruits are known as a hemostatic and astringent [Kusmenoglu, Toker, 1998].

Many scientists have studied the chemical composition of different parts of linden species. It was revealed that flowers contain flavonoids, essential oil, tannins, glycosides, saponins, vitamin C, volatile oils, carbohydrates [Ahmadi, Mirza, 1999: Akyuz et al., 2013; Fitsiou et al., 2007; Kowalski et al., 2017; Lupinskaya et al., 2010; Popov, Zarubina, 2013; Toker et al., 2001a, b; Zarubina, Popov, 2012].

There were polysaccharides, tannins, triterpene saponins, 35 bioelements, organic acids (malic, oxalic, succinic), vitamin C, flavonoids, and phenolcarboxylic acids in linden bark [Orlovskaya et al., 2013].

Vitamin C, carotenes, amino acids, tannins, flavonoids were found also in linden leaves [Akyuz et al., 2013; Delnavazi et al., 2015; Zarubina, Popov, 2012]. M.I. Lukanyuk and S.M. Marchishin [2012] studied the fatty acid composition of leaves of five *Tilia* species and reported that palmitic acid among saturated fatty acids, linoleic and linolenic acids among unsaturated were the most abundant in the leaves of all species.

Vegetable fatty oils are the main source providing our body with such essential fatty acids as linoleic, linoleic and arachidic. They are also widely used in the manufacture of medicines [Grigoryeva, Lisishchin, 2002]. One of the important sources of essential fatty

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acids may be products of plant origin, in particular, oils extracted from seeds of wild and cultivated plants. Therefore, identifying new natural sources of essential fatty acids is a priority area for modern science [Zeynalova, Novruzov, 2019].

Despite there are some data contains the composition of fatty acid of oil of various linden species in literature [Bogdanov et al., 2012; Dowd, Ermakov, Panasenko, 2003; Farve, 2013; Karomatov, Abdukhuhidov, 2017; Kusmenoglu, Toker, 1998; Lukanyuk, Marshichin, 2012], but there are no data about species in Azerbaijan. Among three species of the genus *Tilia* growing in Azerbaijan, *T. caucasica* has a large stock and widespread distribution.

Taking into account we set out to study the oil content physicochemical parameters, and the composition of fatty acids, of fruit oil *T. caucasica*, in order to reveal the beneficial properties of this oil.

MATERIAL AND METHODS

Plant material. The plant material (fruits of *T. caucasica*) was collected in September in 2018 in the phase of full ripeness of fruits in the Quba region of Azerbaijan (48°23′24’’E; 41°18′ 40.95’N).

Extraction. The fruits were dried at 105° C for 2 hours on a FAM-100 and then were extracted with n-hexane on a Soxhlet apparatus. The solvent was distilled off in a Rotary Evaporator (ROVA-N2L) and a yield 12.89% of light yellow oil was obtained.

Oil preparation for analysis was carried out according to GOST 31663-2012. In a test tube with a capacity of 20 ml, 1 g oil was weighed out and dissolved in 10 ml of heptane. Then 0.5 ml of a methanolic solution of potassium hydroxide was added to the resulting solution and vigorously shaken for 2 min. After settling, the top layer was selected for chromatographic analysis.

Identification. Fatty acids were converted into their methyl esters according to GOST 31663-2012. HP 6890 gas-liquid chromatograph with flame ionization detector (FID) was used for identification of fatty acids methyl esters (FAMEs). The separation was achieved with capillary column “Agilent 112-88A7” (0.25 mm i.d.×100m×0.20 μm film thickness). The temperature program was initiated at 140° C, held for 5 min and then increased to 240° C at 4° C per min. Carrier gas was H_2, sample injection with flow division. The analysis duration was 45 minutes. Individual FAMEs were identified by comparing to the retention times of standards (Supelco 37 FAME Mix No. 47885-U standard (Sigma Aldrich)). Results are expressed as percentage of total FA [%FA].

Physicochemical and organoleptic indicators were calculated according to standard following methods: GOST R 50457-92; GOST R 51487-99; GOST 18848-73; GOST 5475-69; GOST 5478-2014; GOST R 52676-2006; GOST 5472-50 [GOST R 50457-92; GOST R 51487-99; GOST 18848-73; GOST 5475-69; GOST 5478-2014; GOST R 52676-2006; GOST 5472-50].

RESULTS AND DISCUSSION

Physico-chemical and organoleptic properties. In the literature there are no data on the physicochemical and organoleptic indicators of oil from the fruits of *T. caucasica.* These data are very valuable and important for the use of new vegetable oils for food, medicinal and cosmetic purposes, as well as standardization. The results of physico-chemical and organoleptic studies of the oil extracted from the fruits of *Tilia caucasica* are presented in table.

The oil was light yellow in color, with a pleasant taste and saponification number of 192.8 mg / kg. This indicator is close to sunflower oil [Shah et al., 1984], and to olive oil [Ermakov, Panasenko, 2003], therefore, *T. caucasica* fatty oil can be recommended for food use.

Table. Physicochemical constants and organoleptic properties of fruit oil of *Tilia caucasica* Rupr.

Experiment	Result
Organoleptic parameters	clear liquid
appearance	
colour	light yellow
smell	without smell
taste	a pleasant taste of seeds
Free fatty acids,%	1.99
Peroxide value, mmol O_2 / kg	3.02
Iodine value, IV	128.4
Saponification value, KOH	192.8
Mass fraction of phosphorus-containing substances, mg /kg	91

The amount of free fatty acids for *T. caucasica* is 1.99 mmol/kg, the peroxide value is 3.02, the iodine value is 128.4, and the mass fraction of phosphorus-containing substances is 91 mmol/kg.

Comparison of the data obtained by us on the physicochemical parameters of *T. caucasica* oils with those of oils from *T. platyphyllos* seeds show that they differ greatly. Oils from *T. caucasic*a fruits have iodine number 128.4, and *T. platyphyllos* 59.5, saponification number 192.8 and 240, respectively [Ermakov, Panasenko, 2003].

Fatty acid analysis by gas-chromatography. Gas-liquid chromatography analysis of fatty acid is a very effective method, since separation occurs at a lower temperature and a shorter analysis time. A representative spectrum of gas-liquid chromatography of fatty acids of oil from linden fruits is presented in figure 1. As can be seen from the spectrum, complete separation occurs within 30 minutes.

2. From figure 2, it is evident that linoleic (46.06%), oleic (16.92%), sterculic (13.35%), palmitic (9.36%) and malvalic (8.09%) acid are the most abundant fatty acid, but heptadecanoic (0.11%), gandoic (0.12%) and palmitoleic (0.15%) acids are the less represented fatty acid. The proportion of the other identified fatty acids is totally 5.23 %. The percantage of unidentified fatty acids are 1.13%.

A study of the composition of fatty acids of three linden species (*T. argentina*, *T. platyphyllos*, *T. rubra*) growing in Turkey showed that *T. platyphyllos* and *T. rubra* contain 10 and *T. argentina* 9 fatty acids [Küsmenoğlu, Toker, 1998]. In the oil of all studied species palmitic, amoung saturated fatty acids, linoleic, oleic acids, amoung unsaturated were dominant. Species differ in the total amount of individual fatty acids. The main fatty acid in *T. rubra* oil was linoleic, however, in *T. platyphyllos* - palmitic, in *T. argentina* was oleic acid.

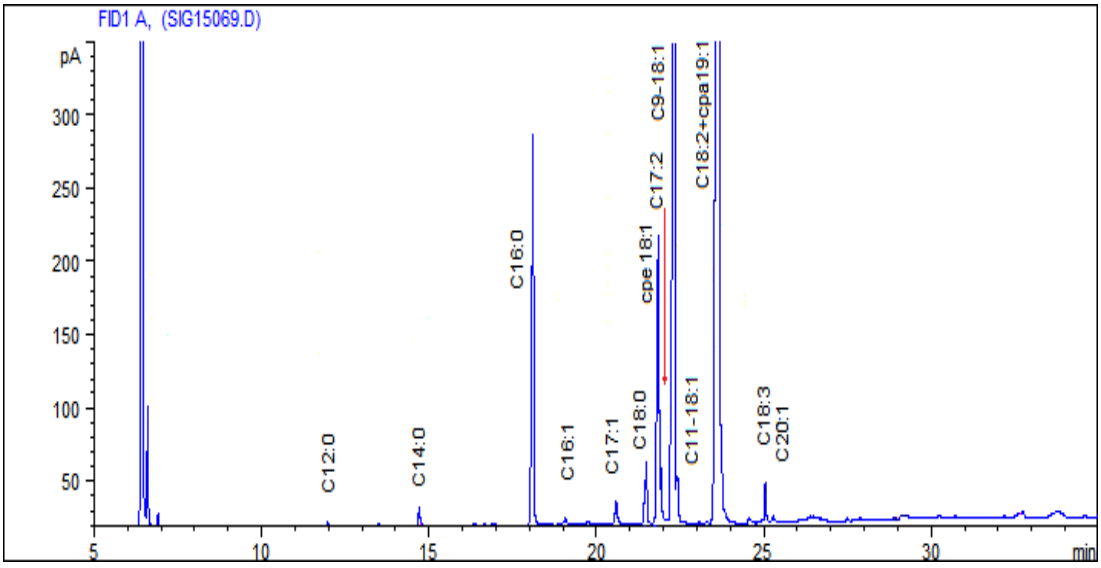

Figure 1. The chromatogram of the fatty acid composition of the oil from the fruits of *Tilia caucasica* Rupr.

The chromatogram shows that 13 fatty acids were identified in the composition of fatty acids of *Tilia caucasica* oil: myristic, palmitic, palmitoleic, heptadecanoic, heptadecadienoic, stearic, malvalic, oleic, elaidic, linoleic, linolenic, gandoic, and sterculic.

It can be seen from the chromatogram that the amount of individual fatty acids respect to the total amount is not the same and varies from 0.11 to 46.06%. The identified fatty acids and the percentange of individual identified acids respect to the total amount are presented in figure

M.K. Dowd and M.C. Farve during the study of the fatty acid composition of oils obtained from seeds of 7 linden species, found that in all samples of *Tilia* spp. seed oils, linoleic acid was the main component of fatty acids; oleic and palmitic acids were present in a smaller amount. This ratio of major and minor fatty acids is characteristic of oils from the seeds of the family Malvaceae. Fatty acids such as myristic, stearic, arachidic, behenic, lignoceric, palmitoleic and cis-vaccenic have been established in the oil of seeds of

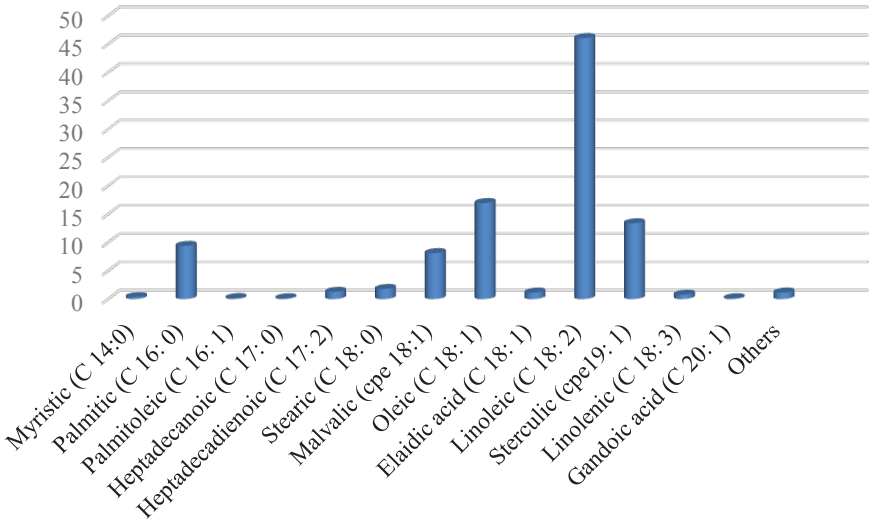

Figure 2. Fatty acid composition of *Tilia caucasica* Rupr. fruit oil.

various linden species. Heptadecanoic, 9-heptadecenoic, 9,10-epoxystearic, and vernolic acids were in small amount. In the seed oil of *T. caucasica* they identified 15 fatty acids. As dominant fatty acids they reported linoleic-52.4%, oleic-21.1%, palmitic-9.76%, and malvalic-6.14% [Dowd, Farve, 2013]. The percantage of these acids in our study are 46.6%, 16.92%, 9.36%, and 8.09%, respectively. As shown they do not differ so much from our results. But among all fatty acids amount of sterculic acid is more significant different. We determined share of this compound 13.35%, however, they presented only 4.30%.

Malvalic and sterculic acids are cyclopropenoid fatty acids. Cyclopropenoid fatty acids were discovered in oil seeds of *T. platyphyllos* by Raju and Resier [1966] for the first time. The total amount of these acids in the oil was 3.2%, while M.K. Dowd and M.C. Farve [2013] reported an higher amounts (9.3%). A comparative analysis of the literature and our data on the content of cyclopropenoid fatty acids (malvalic - 8.09% and steric - 13.35%) shows that the fruit oil of *T. caucasica* growing in the territory of Azerbaijan contains a rather high amount of these acids respect to previous published data. We believe that perhaps these differences are related to the quality of the seed sample (collection time, fruit maturity, soil and climatic conditions), as well as the technique of obtaining derivatives of thiomethyl used to stabilize the acids. Some authors believe that this is due to the decomposition of compounds in the form of methyl esters during chromatography. Perhaps this occurs during the esterification of acids since cyclopropenoid acids are not stable in acid composition [Dowd, Farve, 2013].

The analysis of literature data shows that the content of fatty oil from the fruits of various linden species depends on the species and the place of plant growth. So, the fruits of large-leaved linden growing in Russia contain 10-15% [Bogdanov et al., 2012; Ermakov, Panasenko, 2003] of fatty oil, and in Turkey 8.12%. The yield of oil from the fruits of *T. argentina* and *T. rubra* was 5.6% and 6.16%, respectively [Küsmenoğlu, Toker, 1998]. The results of our studies showed that the fruits of *T. caucasica* differ from other species in their high yield (12.89%) of fatty oil.

Linoleic and linolenic acids are essential nutrients for the body and are not naturally produced in it. Therefore, they must be acquired by the body through food [Grigoryeva, Lisishchin, 2002].

Caucasian linden oil is a very valuable product containing a significant amount of essential fatty acid (C18: 2), which is necessary for human health. Our data allow us to consider linden oil as a semi-drying fatty oil with a high content of essential linoleic acid (46.06%). It should be noted that linoleic acid, which belongs to the fatty acids of the ω-6 family (omega-6), is a structural element of cell membranes, regulates cholesterol metabolism, participates in the formation of tissue hormones - prostaglandins, and is a biochemical precursor of linolenic and arachidonic acids. Under the influence of microelements, enzymes and vitamins in the body, it turns into gamma-linolenic, from which prostaglandin E_1 is synthesized, which, in turn, increases immunity, reduces the risk of cardiovascular diseases, reduces inflammation, regulates the brain and

nervous system, normalizes insulin level, accelerates metabolism [Vles, Gottenbos, 1989].

The high content of linolenic acids in fruit oil from *T. caucasica* makes it suitable for the preparation of cosmetic and dermatological compositions as an antioxidant. The results reported in this work suggests that Caucasian linden oil is a valuable food, medical and cosmetic product and can be used for these purposes.

The results of the investigation of physico-chemical and organoleptic characteristics and fatty acid composition of Caucasian linden seed oil allow to consider the seed oil of this plant as a good source of dietary fatty acid, especially linoleic acid and recommend it for use in food, medicine and cosmetic purposes.

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Tilia caucasica Rupr. meyvələrindən alınan yağın yağ-turşu tərkibi

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Məqalədə *T. caucasica* meyvələrindən əldə edilən yağın yağ turşu tərkibi, fiziki-kimyəvi və orqanoleptik xüsusiyyətləri göstərilmişdir. Azərbaycanda bitən *T. caucasica* növünün meyvələrindən alınmış yağın kəmiyyət və keyfiyyət tərkibi haqda məlumatlar və həmin növün yağının fiziki-kimyəvi və orqanoleptik xüsusiyyətləri ilk dəfə müəyyən olunmuşdur. *T. caucasica* meyvələrindən 12.89% yağ alınmışdır. Yağın fiziki-kimyəvi və orqanoleptik xüsusiyyətlərinin öyrənilməsi göstərdi ki, *T. caucasica* meyvələrindən alınan yağ xoşagələn dadlı, açıq sarı rəngdədir və aşağıdakı xüsusiyyətlərə malikdir: sərbəst yağ turşuları-1.99 mmol / kq, peroksid ədədi -3.02, yod ədədi-128.4, tərkibində fosforun ümumi miqdarı-91 mmol/ kq. Qaz-maye xromatoqrafiya üsulu ilə yağın tərkibində 13 yağ turşusu müəyyən edilmişdir: miristin, palmitin, palmitolein, marqarin, heptadekanin, stearin, malvalin, olein, elaidin, linolein, linoleik, qandoin və stearin. Polidoymamış yağ turşuları arasında linolenin (46.06%), monodoymamış turşular arasında olein (16.92%), doymuş turşular arasında palmitin (9.36%), tsiklopropen turşuları arasında stearin (13.35%) dominantlıq təşkil edir. Qiymətli yağ turşularının olması, o cümlədən yağın fiziki-kimyəvi və orqanoleptik xüsusiyyətləri, *T. caucasica* meyvələrindən əldə edilmiş yağın qida, dərman və kosmetik məqsədlərlə istifadəsini tövsiyə etməyə imkan verir.

Açar sözlər: *meyvə, yağ, xromatoqrafiya*

Жирно-кислотный состав масла плодов *Tilia caucasica* Rupr.

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В данной статье приводятся результаты по изучению жирно-кислотного состава, физико-химических и органолептических характеристик масла, полученного из плодов *T. caucasica.* Данные по качественному составу и количественному содержанию жирных кислот для масла из плодов *T. caucasica,* произрастающего в Азербайджане, а также физико-химические и органолептические показатели для масла данного вида установлены впервые. Было установлено, что выход жирного масла в плодах *T. caucasica* составляет 12.89%. Результаты изучения физико-химических и органолептических характеристик масла показали, что масло из плодов *T. caucasica* светло-желтого цвета, с приятным вкусом и имеет следующие показатели: свободные жирные кислоты составляют 1.99 ммол/кг, перекисное число-3.02, йодное число-128.4, весовая доля фосфор содержащих веществ-91ммол/кг. Изучение метиловых эфиров жирных кислот масла методом газо-жидкостной хроматографии выявило наличие в его составе 13 жирных кислот: миристиновая, пальмитиновая, пальмитолеиновая, маргариновая, гептадеценовая, стеариновая, мальвалиновая, олеиновая, элаидиновая, линолевая, линолеиновая, гандоиновая, стеркуловая. Среди полинасыщенных жирных кислот преобладает линоленовая (46.06%), среди мононенасыщенных кислот - олеиновая (16.92%), среди насыщенных кислот-пальмитиновая (9.36%), а из циклопропеноидных жирных кислот первое место занимает стеркуловая кислота (13.35%). Физико-химические и органолептические показатели, состав и содержание биологически ценных жирных кислот масла плодов *T. caucasica* позволяют рекомендовать его для использования в пищевых, лекарственных и косметических целях.

Ключевые слова: *плод, масло, хроматография*