



RESEARCH ARTICLE

# Phytochemical and nutritional composition of *Elaeagnus macrophylla* Thunb. fruit

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## Abstract

This article examines the morphobiology, ecology and adaptability of *Elaeagnus macrophylla* Thunb. under Uzbekistan's environmental conditions, highlighting its potential in various industries. *E. macrophylla* is a perennial, evergreen shrub reaching heights of 2–8 m tall. Its elongated, silver-colored fruits are covered with small dark orange to red trichomes. In Uzbekistan, flowering occurs later than in its native habitat-blooming in May, with seeds ripening in August-September. The plant's reddish-brown, hairy branches and high antioxidant content make it valuable in the pharmaceutical, food and perfume industries. This study examined the chemical composition and physical properties of *E. macrophylla* grown in Uzbekistan. Key parameters such as fruit moisture, pH, ash content and the mass ratio of pulp to seed were analyzed, along with protein and flavonoid levels. Research was conducted at the Institute of the Chemistry of Plant Substances and the Institute of Bioorganic Chemistry of the Academy of Sciences of Uzbekistan. Advanced techniques such as plasma optical emission spectrometry, gas chromatography, HPLC, Kjeldahl method, standard procedures and a Mettler Toledo pH meter were used. Results showed the fruit's moisture content was 88.92 %, with seed and pulp mass ratios of 57.7 % and 42.3 % respectively. The protein content in raw samples was 1.14 % and 10.27 % on a dry matter basis. The pH value was measured at 3.75, indicating a slightly acidic nature. These findings confirm the plant's adaptability to Uzbekistan's climate and its potential in several industries.

**Keywords:** ash content; *Elaeagnaceae*; *Elaeagnus macrophylla*; flavonoid; fruit; morphology; protein; pH value

## Introduction

In recent years, population growth has led to an increased demand for medicinal and edible plants. Based on botanical research, the identification and introduction into production of medicinal plant populations in various regions serves to meet the population's demand for medicines and food products.

The flowering plant family Elaeagnaceae Juss. includes about 103 species and around 3 genera. These plants are typically evergreen or silver-leaved, fragrant-flowered and consist of small trees or shrubs. Their fruits are natural products used both as food and in medicine. Numerous studies have focused on the ornamental value, chemical composition and nutritional properties of various genera within the Elaeagnaceae Juss. family (1).

Therefore, conducting research on approximately 55 species belonging to 3 genera of the Elaeagnaceae Juss. Family-an important group with significant economic value and distribution across Europe, Asia and North America-is considered essential is valuable for understanding their ecological, nutritional and economic roles (2).

*Elaeagnus macrophylla* Thunb. (family Elaeagnaceae), an evergreen shrub, has a long history of use in traditional medicine. Its fruits are well-known for their beneficial health properties, including antioxidants, anti-inflammatory and anti-diabetic effects. These properties are attributed to the abundance of bioactive compounds found in the fruit, such as polyphenols, flavonoids and vitamins (3).

However, scientific data on the physicochemical composition of the fruit of *E. macrophylla* is still limited. A thorough investigation into its chemical profile is essential for a complete assessment of this plant's potential applications in the food, pharmaceutical and cosmetic industries (4).

The main goal of this study is to determine the physicochemical composition of the *E. macrophylla* fruit, focusing specifically on its content of macro and microelements, fatty acids, sugars and vitamins. The results of this research will provide valuable new information about the nutritional value and bioactive properties of this unique fruit, creating a strong scientific foundation for its future practical use (5).

## Materials and Methods

Under our local conditions, the flowers of *E. macrophylla* bloom in May and the seeds ripen by August–September. The exocarp of ripe fruits consists of a shiny outer skin. The mesocarp is thick and fleshy, rich in organic compounds. The endocarp of the fruit is a hard, woody seed composed of tightly packed mechanical fibers (brachysclereids) (6).

*E. macrophylla* Thunb. is drought-resistant and capable of growing in rocky, sandy and saline soils as well as in sunny or partially shaded areas. It can even tolerate temperatures as low as -15 °C. However, if not properly cared for, *E. macrophylla* Thunb. plants, like many other species, may suffer from chlorosis, which causes the leaves to yellow and dry prematurely. Additionally, it is susceptible to damage from psyllid larvae (Psyllis), which are considered pests. The presence of honeydew on buds and leaves can also promote the growth of mold fungi (7).

Speaking of the beneficial properties of *E. macrophylla* Thunb., its dried fruits have traditionally been used in Chinese medicine to treat coughs, sore throats and other respiratory diseases. The fruit is considered a source of vitamin C and antioxidants. A decoction made from its fruits is used to treat diarrhea, colitis, stomach disorders and respiratory diseases and it is also applied as an anti-inflammatory remedy (8).

As a result of our research, we found that *E. macrophylla* Thunb. was introduced as an ornamental plant for the first time in Uzbekistan in 2022 by the Landscaping Department of Samarkand city (coordinates: 39°39'20" N 66°58'13" E and 39°40'36" N 66°57'59" E). Initially, around 200 saplings were planted along Dahbed Road and Motrid Street and 120–135 of them survived the abnormal cold of winter 2022. Currently, these seedlings are thriving and growing well (Fig. 1).

The chemical composition of *E. macrophylla* Thunb. fruits (pH value, proteins, flavonoids) were studied at the Institute of Plant Chemistry named after S. Y. Yunusov of the Academy of Sciences of the Republic of Uzbekistan and the Institute of Bioorganic Chemistry. Plasma optical emission spectrometry, gas chromatography, UV-Vis spectroscopy, Kjeldahl method, standard methods and pH measurements using a Mettler Toledo (USA) modern pH meter were employed in these studies (9).

*E. macrophylla* Thunb. is drought-resistant and capable of growing in rocky, sandy and saline soils as well as in sunny or partially shaded areas. It can even tolerate temperatures as low as -15 °C. However, if not properly cared for, *E. macrophylla* Thunb. plants, like many other species, may suffer from chlorosis, which causes the leaves to yellow and dry prematurely. The crude protein content was determined according to the standard method. During the procedure, the following equipment and materials were used: analytical balance (accuracy 0.0001 g), filter paper, conical funnel, photo colorimeter (FEK), caustic soda (NaOH), Nessler's reagent, distilled water, concentrated sulfuric acid and hydrogen peroxide (10).

The laboratory sample, after being finely ground in a porcelain mortar, was mixed and spread in a thin layer on a tray. Samples were then taken from various locations in the necessary amounts for each determination. Each sample was placed in pre-dried and weighed beakers, covered with lids and weighed using an analytical balance. The seed samples were dried in a drying cabinet at a temperature of 100 °C -105 °C for 3 hr (11). After the specified time, the beakers were quickly removed from the cabinet, covered and placed in the exicator for 10-15 min to cool. The cooled and weighed beakers were then placed back in the drying cabinet for an additional 30 min, after which they were removed, cooled again and weighed. This process was repeated until a constant mass was achieved (12).

The constant mass is considered to be achieved when the difference between successive weighing does not exceed 0.001 g.

The moisture content of the seeds, in percentage (X), was calculated using the following formula:

$$X = [(P1 - P2) / P1] * 100$$

Here:

P1 is the weight of the sample before drying (in g),

P2 is the weight of the sample after drying (in g),

P is the weight of the powder sample (in g) (18).

To investigate the flavonoid content, the dried vegetative parts of the plant were ground to a particle size of 0.1-1.5 mm. Precisely 1.0000 g of the sample was weighed and extracted with 99 mL of 70 % ethanol at a temperature of 50 °C -60 °C for 2 hr in a flat-bottomed flask equipped with a reflux condenser, under



**Fig. 1.** *Elaeagnus macrophylla* Thunb. A, B - Growth of *Elaeagnus macrophylla* Thunb. under the environmental conditions of Samarkand city.

intensive stirring (13). The resulting solution, along with the flask, was placed in an ultrasonic bath, where extraction was carried out at 35 °C for 15 min, repeated twice with a 10 min interval. The extract was cooled to room temperature. It was first filtered through an ash-free paper filter (blue ribbon) and then 2 mL of the resulting solution was passed through a 0.2 µm membrane filter (14). A 100 µL aliquot of this filtrate was taken and diluted with eluent to a final volume of 1 mL. The analysis was performed using High-Performance Liquid Chromatography (HPLC) under isocratic elution conditions, employing a diode array detector (DAD). Acetonitrile and a buffer solution were used as the mobile phase (15, 16).

To determine the pH of *Elaeagnus macrophylla* fruit, the pulp was first carefully separated from the seed. The extracted pulp was processed under sterile laboratory conditions using a high-speed mixer for 3-4 min until a homogeneous mass was obtained. This homogenized mixture was then filtered through several layers of filter paper to obtain clear juice (17). The pH of the resulting fruit juice was measured using a modern pH meter (Mettler Toledo, USA). Prior to measurement, the pH meter was calibrated with buffer solutions at pH 4.00 and 7.00. The measurement was conducted at room temperature (approximately 22 °C-25 °C). The electrode was fully immersed in the sample and the pH value was recorded after waiting 2-3 min for the reading to stabilize (18).

## Results

The fruit of *Elaeagnus macrophylla* Thunb. growing in China was selected as the research object. The aim of the study was to examine the chemical composition of *E. macrophylla* Thunb. fruits. The analysis was conducted twice in repetition using the

method recommended in the reference. During the process, analytical balances, a drying cabinet, an exicator and glass beaker were used (19).

The moisture content of *E. macrophylla* fruit was determined and the results are presented in Table 1. Two separate samples were analyzed to ensure reliability. The initial weight of the fruit samples was 1.394 g and 2.84 g respectively. After drying, the weights were reduced to 0.155 g and 0.313 g respectively, indicating a significant loss of moisture.

The moisture content for the two samples was calculated to be 88.88 % and 88.97 %, resulting in an average moisture content of 88.92 %. This high moisture content is a characteristic feature of fresh fruits and aligns with the typical composition of many berries. This high water content also suggests that the fruit is highly perishable and may require specific post-harvest handling or processing to extend its shelf life (Table 2). The components of the *Elaeagnus macrophylla* fruit, namely the seed and pulp fractions, were determined. The seeds constituted 57.7 % of the fruit's total mass, while the pulp made up 42.3 %. This result indicates that the edible portion of this fruit is relatively small compared to its seed content. This information is crucial for assessing the fruit's processing potential, as the pulp is the primary part consumed and used for processing. Therefore, separating the pulp from the seeds is essential for producing food products such as juices or jams from *E. macrophylla* fruit. Furthermore, the high proportion of seeds suggests a potential for exploring alternative uses, such as a source for oil extraction, animal feed or bioactive compounds (Table 2).

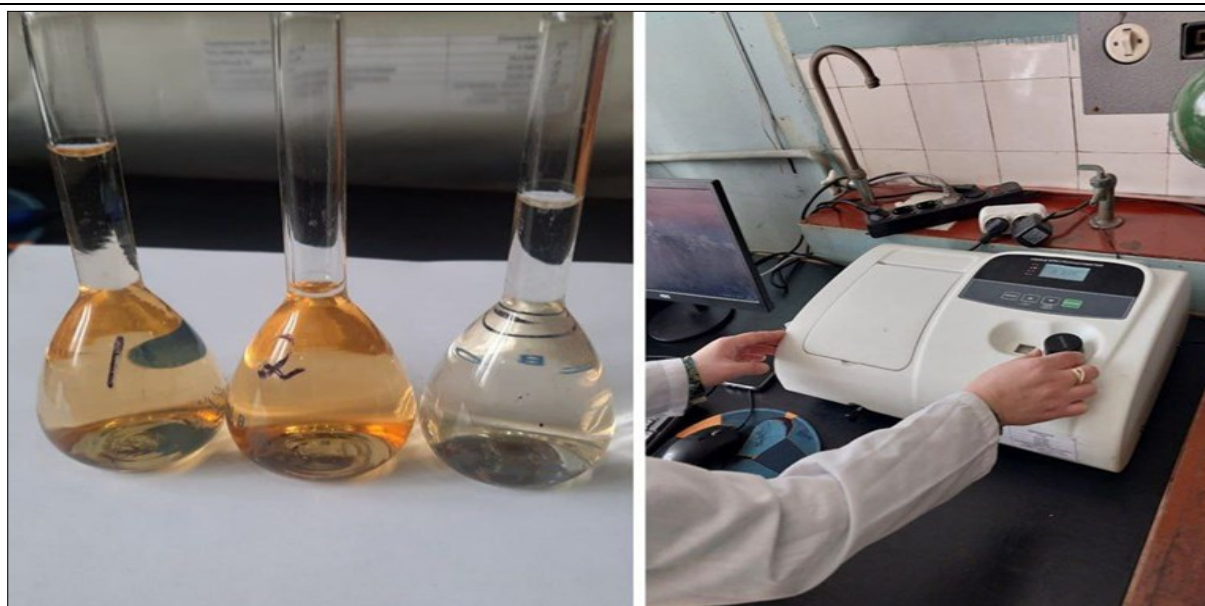
The protein content of the *Elaeagnus macrophylla* fruit was determined using the Kjeldahl method (Fig.2), with the results summarized in Table 3. Two samples were analyzed, with

**Table 1.** Results of moisture content determination in *E. macrophylla* fruit

No.	Sample name	Weight of the crucible (g)	Crucible with initial sample (g)	Crucible with initial sample	Crucible with dried sample (g)	Moisture content (%)	Average value (%)
1	<i>E. macrophylla</i>	14.205	15.599	1.394	14.360	88.88	88.92

**Table 2.** The mass ratio of the pulp and seed of *E. macrophylla* fruit

Sample name	Quantitative composition (mg/100 g)	
	Seed mass fraction (%)	Pulp mass fraction (%)
<i>E. macrophylla</i>	57.7	42.3



**Fig. 2.** Results of protein content determination in *E. macrophylla* fruit.

**Table 3.** Determined protein content in *Elaeagnus macrophylla* fruit

No.	Experiment	Sample weight (g)	Aliquot (mL)	400 nm	Nitrogen	Protein content (%)	Average protein content (%)	Protein content on an absolutely dry matter basis (ADM) (%)
1	Experiment	2.585	0.3	0.174	0.20	1.25	1.14	10.27
2	Experiment	3.156	0.3	0.175	0.16	1.03	1.14	10.27

initial weights of 2.585 g and 3.156 g respectively. The nitrogen content, a key factor in calculating the protein concentration, was measured at 0.20 % for the first sample and 0.16 % for the second.

The raw protein content was calculated to be 1.25 % and 1.03 % for the respective samples, yielding an average protein content of 1.14 % on a fresh weight basis. When converted to an absolutely dry matter basis (ADM), the protein content was found to be 10.27 %. This value, while modest compared to high-protein sources, indicates that *E. macrophylla* fruit contains a notable amount of protein, which contributes to its overall nutritional profile (Fig. 2). This finding is particularly relevant for assessing its potential as a component in food products or dietary supplements (Table 3).

The proximate composition of the *E. macrophylla* fruit was analyzed to determine its key nutritional components. The results, summarized in Table 4, reveal that the fruit has a high moisture content of 88.88 %. This high value indicates that it is a highly perishable fruit, typical of many fresh berries.

The ash content was found to be 3.51 %, which represents the total mineral content of the fruit. This value is relatively high, suggesting that *E. macrophylla* is a good source of essential minerals.

The protein content was also determined, with a protein nitrogen percentage of 0.18 %. The total nitrogen content was 1.14 % and based on these values, the protein content on an absolute dry matter (ADM) basis was calculated to be 10.27 %. This indicates that while the fresh fruit's protein content is low due to high moisture, its dry matter is a notable source of protein, which contributes to its overall nutritional value (Table 4).

Flavonoids are a widely distributed group of phenolic compounds. Several subgroups of flavonoids include flavones, flavanones, flavonols, chalcones, catechins, anthocyanins and others (20). Most of them occur in plants in the form of glycosides or in free form. Flavonoids are present in nearly all plants in various combinations and amounts. Their therapeutic effect results from their combined presence. They exhibit a broad spectrum of pharmacological activity (21). This confirms the importance of identifying flavonoids in medicinal plants. As a result of our research, the content of key flavonoids in the fruits of the species under study-myricetin, hypolaetin, isorhamnetin, gallic acid, hyperoside, resveratrol, apigenin, rutin and quercetin-was determined (Table 5).

The concentrations of rutin and gallic acid flavonoids in *E. macrophylla* fruits were found to be 94.154 mg/g and 13.558 mg/g respectively, indicating a higher concentration compared to the previously studied species. However, the amounts of other flavonoids such as myricetin, hypolaetin, isorhamnetin, gallic acid, hyperoside, resveratrol, apigenin, rutin and quercetin were not detected.

Spectral data were recorded within the wavelength range of 200 to 400 nm (Fig. 3) (Table 6-8).

Signal 1: DAD1 A, Sig=254,4 Ref=360,100

Totals: 672.52941

Signal 2: DAD1 B, Sig=265,4 Ref=360,100

Totals: 603.30030

Signal 3: DAD1 C, Sig=281,4 Ref=360,100

Totals: 655.05500

As a result, the pH value of *Elaeagnus macrophylla* Thunb. fruit was found to be 3.75 (Fig. 4), indicating that the fruit exhibits an acidic environment.

## Discussion

The results of this study confirm the successful acclimatization of *Elaeagnus macrophylla* Thunb. to the environmental conditions of Uzbekistan. The observed shift in phenology-blooming in May and seed ripening in August-September-indicates a flexible adaptation to the local climate (22). These findings are consistent with previous observations by Tani and Sasakawa (2000), who reported that *E. macrophylla* exhibits considerable drought tolerance and can grow in saline soils, although its seed germination decreases significantly at NaCl concentrations above 50 mM.

The measured fruit moisture content of 88.92 % aligns with data reported for other species within the genus. For instance, in a study of *E. angustifolia*, the moisture content of the mesocarp was found to be approximately 82.4 %-83.6 %, demonstrating comparable water retention capacity (23).

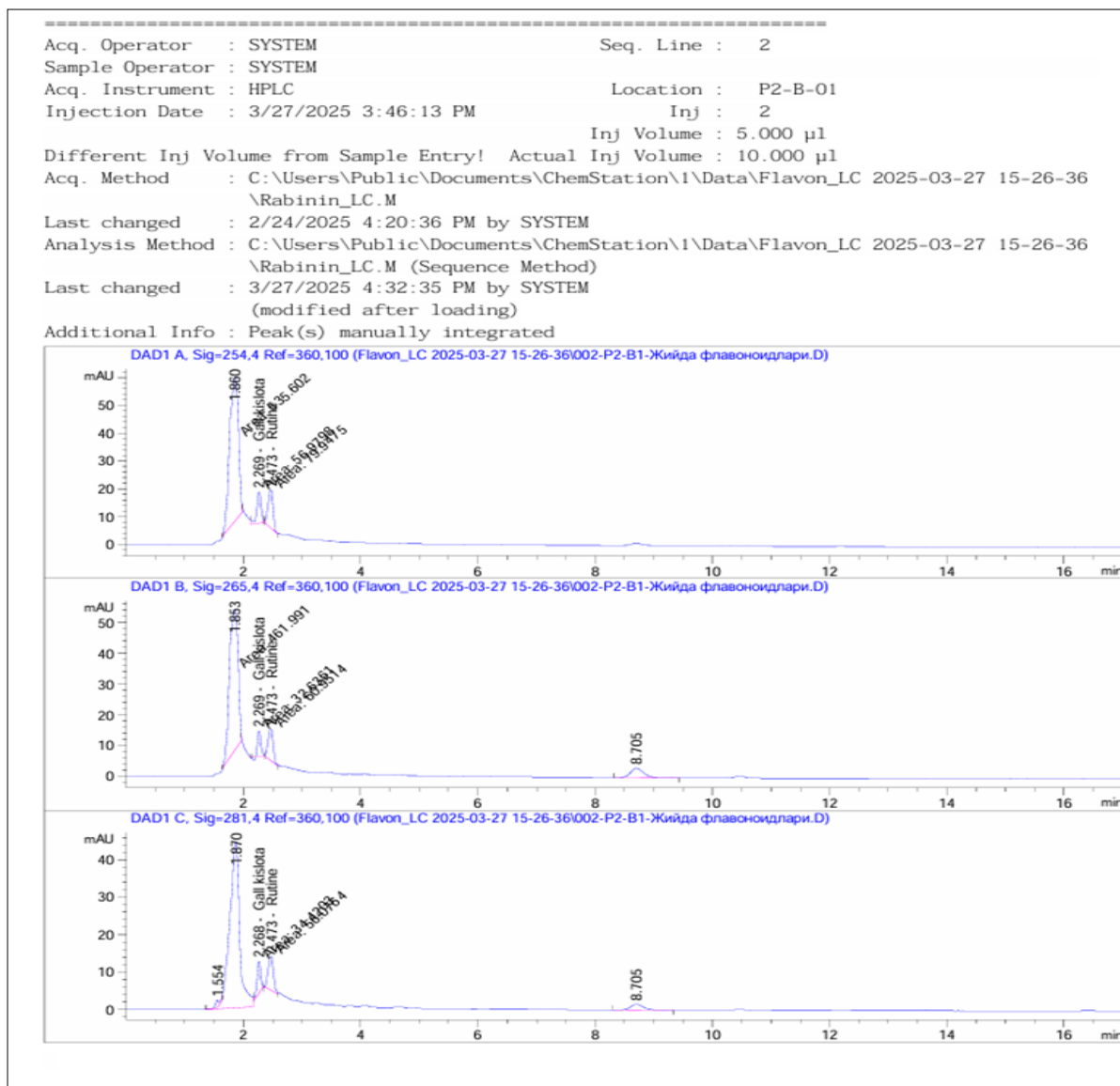
Protein content analysis revealed 1.14 % in the raw fruit sample and 10.27 % on a dry matter basis. These values are comparable to those found in *E. angustifolia*, where protein levels ranged from 11.8 % to 12.8 % in dried fruit samples, further supporting the nutritional potential of *E. macrophylla*.

**Table 4.** Physicochemical analysis results

No.	Name of sample	Moisture content (%)	Ash content (%)	Protein nitrogen (%)	Nitrogen (%)	Protein content in absolute dry matter (ADM) (%)
1	<i>E. macrophylla</i>	88.88	3.51	0.18	1.14	10.27

**Table 5.** Flavonoid content

Name of sample	Flavonoid content (mg/100 g)								
	Myricetin	Hypolaetin	Rutin	Quercetin	Izoramnetin	Gall acid	Giperezid	Apigenin	Resveratrol
<i>E. macrophylla</i>	-	-	94.154	-	-	13.558	-	-	-



**Fig. 3.** Chromatography of flavonoids.

**Table 6.** HPLC analysis results: peak parameters and compound identification based on DAD1-A spectrum at 254 nm

Peak#	Ret time [min]	Type	Width [min]	Area [mAU*s]	Area (%)	Name
1	1.799		0.0000	0.00000	0.0000	Apigenin
2	1.860	MM	0.1728	535.60205	79.6399	
3	2.269	MM	0.0843	56.97982	8.4725	Gall acid
4	2.473	MM	0.0979	79.94753	11.8876	Rutine
5	3.246		0.0000	0.00000	0.0000	
6	3.773		0.0000	0.00000	0.0000	
7	4.482		0.0000	0.00000	0.0000	Gipolaeyetine
8	5.669		0.0000	0.00000	0.0000	
9	6.928		0.0000	0.00000	0.0000	Resveratrol
10	7.356		0.0000	0.00000	0.0000	
11	11.964		0.0000	0.00000	0.0000	Quercetin

**Table 7.** Identification of phenolic compounds in *E. macrophylla* fruit extract by HPLC-DAD at 265 nm

Peak #	Ret time [min]	Type	Area (%)	Area [mAU*s]	Area Name	Name
1	1.853	MM	0.1679	461.99088	76.5773	
2	2.269	MM	0.0656	32.63611	5.4096	Gall acid
3	2.473	MM	0.0980	60.95143	10.1030	Rutine
4	2.608		0.0000	0.00000	0.0000	
5	2.730		0.0000	0.00000	0.0000	Giperazide
6	2.978		0.0000	0.00000	0.0000	
7	3.238		0.0000	0.00000	0.0000	
8	3.698		0.0000	0.00000	0.0000	Izoramnetine
9	4.489		0.0000	0.00000	0.0000	Gipolaeyetine
10	5.111		0.0000	0.00000	0.0000	Miriciten
11	6.929		0.0000	0.00000	0.0000	Resveratrol
12	8.705	BB	0.2365	47.72188	7.9101	

**Table 8.** HPLC analysis results: peak parameters and compound identification based on DAD1-C spectrum at 281 nm

Peak #	Ret time [min]	Type	Area (%)	Area [mAU*s]	Area Name	Name
1	1.554	BV	0.0651	8.36794	1.2774	?
2	1.870	WV	0.1651	532.39056	81.2742	
3	2.268	MM	0.0671	34.42033	5.2546	Gall acid
4	2.473	MM	0.1033	56.07639	8.5606	Rutine
5	2.763		0.0000	0.00000	0.0000	Izoquercetin
6	2.961		0.0000	0.00000	0.0000	
7	3.238		0.0000	0.00000	0.0000	
8	3.706		0.0000	0.00000	0.0000	Izoramnetine
9	4.119		0.0000	0.00000	0.0000	
10	4.473		0.0000	0.00000	0.0000	Gipolaeyetine
11	5.120		0.0000	0.00000	0.0000	Miriciten
12	5.687		0.0000	0.00000	0.0000	
13	6.930		0.0000	0.00000	0.0000	Resveratrol
14	8.705	BB	0.2352	23.79977	3.6332	

**Fig. 4.** pH value of *E. macrophylla* Thunb. fruit.

The flavonoid profile of *E. macrophylla* was characterized by high concentrations of rutin (94.154 mg/g) and gallic acid (13.558 mg/g). The presence of these compounds is pharmacologically significant, as rutin is a powerful antioxidant and anti-inflammatory agent, while gallic acid is known for its strong antioxidant and antimicrobial properties. These findings suggest that the fruit of *E. macrophylla* possesses considerable therapeutic potential (24, 25). These values notably exceed those observed in related species; for example, rutin levels in *E. angustifolia* were reported at 216.5 mg per 100 g of dry weight. The richness in these bioactive compounds underlines the pharmaceutical and antioxidant value of *E. macrophylla* fruits.

The pH value of 3.75 recorded in this study indicates a mildly acidic nature of the fruit pulp, which is similar to pH values reported in other *Elaeagnus* species. This level of acidity is significant as it contributes to the fruit's overall flavor profile and plays a crucial role in its preservation potential. A low pH naturally inhibits the growth of many spoilage-causing microorganisms, thereby extending the fruit's shelf life and making it suitable for processing into products like jams, juices or preserves. This acidity may be favorable for food preservation and product development in the food industry (26).

The results of our experiments showed that proteins and flavonoids are groups of organic compounds essential for the life activities of humans, animals and plants. Overall, *E. macrophylla* has demonstrated robust morpho biological and chemical adaptability under the climatic conditions of Uzbekistan. When

compared to related species, its high bioactive compound content, resilience to environmental stressors and ornamental appeal make it a strong candidate for further application in pharmaceutical, food and horticultural sectors (27).

## Conclusion

It was determined that 1-5 g of the *Elaeagnus macrophylla* Thunb. fruits contain 88.92 % moisture. The seed mass fraction was 57.7 % and the pulp mass fraction was 42.3 %. The protein content in the raw sample was found to be 1.14 % and according to the calculation formula, the protein content on a dry matter basis was 10.27 %. While this study determined the overall composition, future research should focus on isolating and identifying the specific compounds responsible for the observed biological activities (e.g. antioxidant, anti-inflammatory effects). This involves fractionating the fruit extract and testing each fraction to pinpoint the most active components. Our study examined a single sample of *E. macrophylla*. It would be beneficial to analyze the physicochemical composition of fruits from different geographic locations and cultivars. This would help understand how environmental factors (e.g. soil type, climate) affect the concentration of key bioactive compounds.

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## Authors' contributions

LN assisted in writing and submitting the article to the journal. KK conducted the initial review of the manuscript prior to submission. ZU revised and enhanced the manuscript's writing style. BI organized the research work plan. UN participated in laboratory instruction and provided support. BD offered financial support for the project. SA provided practical assistance during the implementation of the study. ZD contributed technical support and assistance with equipment. AS was involved in data analysis, visualization and gave final approval of the manuscript. All authors read and approved the final manuscript.

## Compliance with ethical standards

**Conflict of interest:** The authors declare that they have no competing interests.

**Ethical issues:** None

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