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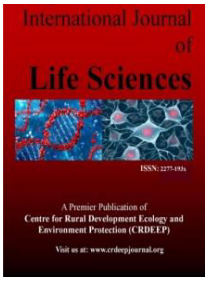

Full Length Research Paper

Prospects for the use of Physiologically Active Compounds of *Elaeagnus umbellata*

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ABSTRACT

One of the priority tasks, facing science in modern ecology terms, is the enrichment of the human ration with raw plant materials, on the basis of which effective therapeutic, preventive or functional products will be created. In this regard, various species of the family Elaeagnus L., which are distinguished by a fairly high content of biologically active compounds, are of great importance. The most common species of the family Elaeagnus L. is Elaeagnus umbellata. The aim of the research has been to study the quantitative content of biologically active compounds,in particular, phenolic compounds - common phenols, flavonoids, catechins,found in fruits and leaves of plants, collected in different regions of Georgia, as well as to determine their antioxidant activity, using modern physicochemical and instrumental methods. The object of the study has been the fruits and leaves of the Elaeagnus umbellata plant, collected in 4 different regions of Georgia (Senaki - 28 meters above sea level, Ozurgeti - 80 meters above sea level, Khelvachauri - 80 meters, Keda - 256 meters above sea level). Studies have shown that the total amount of phenols in fruits is 117,98 mg / 100 g - 989,42mg / 100 g, in leaves - 1128,69 mg / 100 g - 2546,78 mg / 100 g. Flavonoids in fruits - 57,81 mg / 100 g - 781,57 mg / 100 g, in leaves - 580,65 mg / 100 g - 1238,65 mg / 100 g, catechins in fruits - 36,12 mg / 100 g - 147,92 mg / 100 g, in leaves - 418,39 mg / 100 g - 1147,36mg / 100 g, Antioxidants in fruits - 5.94 (52.04%) - 12.99 (40.3%), in leaves 0.47 (53.02%) - 2.44 (41.02%). The samples collected in Senaki and Ozurgeti were distinguished by high rates, what is due to a similar geographical location. Based on the data obtained from research, Elaeagnus umbellata can be used as a new raw material for the production of products used for preventive purposes.

Introduction

In modern environmental conditions, taking into account the ongoing modern research of mankind, it is important to find promising plant raw materials, which can be used as a prophylactic, preventive and therapeutic agent. World civilization is closely related to the introduction of wild plants into culture and their subsequent selection according to the quantity and quality of the crop. Plants included in the human diet differ not only according to nutritional value, but as well as therapeutic and prophylactic one.

One of the priority tasks facing science is enrichment of the human diet with fresh plant materials, on the basis of which effective therapeutic, preventive and functional products can be created. The use of the same functional products is not limited in different age groups. Medicinal plants are considered a vital part of human civilization, since even in the modern era, about 80% of the world's population relies on the use of medicinal plants to treat complications [13].In this respect, various species of the family Elaeagnus L. are important[11].. Their research is being conducted in many countries of the world in order to expand the scope of their application [1,4,24]. The most common species in Georgia is *Elaeagnus umbellata*. There is no information on the distribution of other species in Georgia, as well as there is practically no scientific literature on the chemical and biochemical composition and use of this plant, with the exception of traditional medicine, where it is used for gastrointestinal diseases as an anti-inflammatory, binding or anthelminticagent;as vitamin remedy; as expectorant for bronchitis[1,2, 11].. Fruit tincture has a hypotensive and mild analgesic effect. It is also effective in relieving secondary complications associated with diabetes [24].A concentrate of tannin colloidal substances can be obtainedfrom the fruits of *Elaeagnus umbellata*. It is also used in Traditional Chinese Medicine. The leaf extract can

be used to treat asthma and chronic bronchitis. The flowers of *Elaeagnus umbellata* are used to treat tetanus [6,21]. Seed extract and oil are used as a cough stimulant and a cure for lung diseases [16, 19].

According to modern scientific research, the plant has a wide range of pharmacological and therapeutic effects[11,12], including antimicrobial, insecticidal, antioxidant, healing, cardioprotective, hypolipidemic, antibacterial, antimutagenic, anti-cancer and gastroprotective [4, 21] and anti-tumor action effects [24]. In China, Japan and Korea, the fruits of the plant are used in the diet as a healthy raw material [12].The healing effect of the plant is due to the content of biologically active compounds, such as phenolic compounds, flavonoids, alkaloids, carotenoids, glycosides, terpenoids, steroids, minerals, vitamins, organic acids and others [8,12, 14, 15]. It is also an excellent source of vitamins A, C and E. [11]. Research has widely been conducted in Turkey, the Black Sea and the Balkans, Europe and Asia, the Middle East, Pakistan, China, Korea, India [11,19,24,], Taiwan and Iran [15].

The study of the plant is important in the sense that the raw material can be found in large quantities both in western (Guria, Adjara, Samegrelo) and in eastern Georgia. The plant is widespread both in the wild and in culture [21,25]. **Elaeagnaceae** is one of the species of the family Elaeagnus L. *Elaeagnus umbellata* as a plant that grows easily in various environmental conditions, is cultivated from the territories of North Asia to the Himalayas and Europe[3, 4,20]. *Elaeagnus* has about 80 species. It is widespread in the subtropical and temperate regions of East and Southeast Asia, rarely found in the rest of Asia and in Central Europe.

Elaeagnus umbellata grows in humid areas, but can withstand severe drought; it grows along river banks, swamps, forests and ravines, along roadsides, fences and irrigation canals; it can usually be found in the lower part of the mountains, sometimes in the middle zone; it is bred in gardens and parks; it often goes wild; easily propagated by seeds, root cuttings and root seedlings; it grows mainly in wetlands, it is also found on dry steep slopes, it tolerates temporary flooding with water, it also tolerates soil salinity, It is also resistant to high and low temperatures [12], its roots do not absorb salts, therefore, salt accumulation in the upper parts of the earth is not observed [8,9]. In Italy, this species is mainly used for ornamental purposes, as it can easily grow more than 2 m and does not require specific plant care [12, 13].

The dry fleshy pulp of the fruit in its wild forms makes up 52% of the weight of the whole fruit and contains more than 40% sugars. According to the literature, the fruits of *Elaeagnus umbellata* contain glucose, fructose and sucrose, of which fructose is the predominant sugar (20-35% and glucose 15-25%) [24]. Also found in organic acids are malic acid, citric acid, quinic acid, of which malic acid is predominant. In addition, the fruit contains potassium and phosphorus salts, as well as 10% more protein [9,20].

Various photochemicals have been identified and exhibited from flowers, such as: palmitic acid (16.9%), eugenol (11.1%), methyl-palmitate (10.5%), 4-methyl anisole (33-42.7%) and 4-methyl-phenol (10.9-13.3%) and others [19]. The fruits of *Elaeagnus umbellata* are eaten raw, and bread flour is also enriched with its raw materials. Gumphis of Elaeagnus umbellate can be used in the textile industry, for the manufacture of adhesives and varnishes. The leaves and bark of the plant are used to tan leather and dye it black or brown. Bacteria living in cocoons on the roots of *Elaeagnus umbellata* fix free nitrogen in the atmosphere; therefore, *Elaeagnus umbellata* enriches the soil with nitrogen [5, 21].

The object of the study was the fruits and leaves of the *Elaeagnus umbellata* plant grown in 4 different areas of Georgia (Senaki - 28 m above sea level, Ozurgeti - 80 m above sea level, Khelvachauri - 80 m, Keda - 256 m above sea level). The aim of our research has been to study the quantitative content of biologically active compounds,in particular, phenolic compounds - common phenols, flavonoids, catechins,contained in the fruits and leaves of Elaeagnus Umbellate plants collected on the territory of 4 different regions of Georgia - Samegrelo, Guria, the coast of Adjara and the upper mountainous Adjara (Senaki, Ozurgeti, Khelvachauri, Keda), as well as their antioxidant activity, using modern physicochemical and instrumental methods.

Materials and methods

Sample preparation: At the first stage of the study, 50% and 96% extract of the extractant, ethyl alcohol, were taken for each sample. For analysis, 5 grams of an untreated pulp sample was taken, extracted three times at 70-750 ° C, and the sample was fixed. We then defined the connections using the above methods.

Determination of total phenolic content.

Total phenolic content of the extracts was determined using the Folin-Ciocalteau reagent. Folin-Ciocalteau reagent was diluted 10 times with distilled water. The *Elaeagnus umbellata* extract solution (50 µL) was mixed with 1 ml diluted Folin-Ciocalteau reagent, 1 ml sodium bicarbonate solution (7.5%), and 2 ml distilled water. The mixture was incubated at room temperature for 15 min. The absorbance of the solution was determined at 730 nm using a spectrophotometer and compared with gallic acid equivalents (GAE) calibration curve. The total phenolic content was expressed as mg gallic acid equivalents of 30 gram fresh E. umbellata leaves and fruit [25,26].

Determination of total flavonoid content

Flavonoid content was determined according to the method described by Bonvehi et al. (2001) with some modifications. An appropriate dilution (50 µL) of the extract wasmixedwith1mlof 2% AlCl3 in methanol solution (5% acetic acid in methanol). The

mixture was allowed to react for 10 min and the absorbance was read at 430 nm against a blank sample without reactants. Rutine was used as the standard for the calibration curve. Total flavonoids content of the extracts was expressed as mg rutine equivalents of 5 gram fresh *Elaeagnus umbellata* [25,26].

Determination of catechins - by spectral method –

The sample was extracted with 80% ethyl alcohol at a temperature of 70-75^{0}C. To 1 ml taken from the total volume of the extract was added 3 ml of vanillin reagent and after 3 minutes, the optical density of the red-colored sample was determined at 500 nm. 1 ml of the appropriate extract and and 3 ml of vanillin reagent are controlled. The data obtained from the determination are recalculated on the (+) catechin calibration curve. The content of catechin is calculated by the formula: X = (D K V F) * 1000 / m

Where, X - content of catechins, in mg / kg; D - optical density; K - 35.0 (+) per catechin (conversion ratio); F - dilution factor; V - total volume of the extract, ml; m - mass of raw material taken for extraction, g.

Determination of antioxidant activity with DPPH method –

Antioxidant activity against stable radicals was determined by inhibiting 50% of free radical -DPPH (2,2-Diphenyl-1-picrylhydrazil). DPPH - ALDRICH firm, LOT # STBD4147V (product of Germany) [26]. Inhibition of free radical scavenging (DPPH) activity is calculated by the following formula: $\mathbf{In\% = A_C - A_S/A_C * 100,}$ where the absorption of A_C - DPPH alcohol solution and the absorption of A_S - analytical extract [4, 9, 25].

Results

There has been determined and studied the quantitative content of biologically active compounds,in particular, phenolic compounds - common phenols, flavonoids and catechins in the fruits and leaves of the plant Elaeagnus Umbellata, collected in 4 different regions of Georgia (Senaki, Ozurgeti, Khelvachauri, Keda), as well as their antioxidant activity. The study found that the maximum amount of total phenols, flavonoids, catechins was observed in 96% of the fruit and 50% of the alcohol in the leaves (Table N 1, 2, 3, 4).

Table 1. Quantitative content of total phenols, flavonols and catechins of Elaeagnus umbellata fruits and leaves taken in Samegrelo (Senaki)

№	Sample name	Common phenols mg/100 g	Flavonoids mg/100 g	Catechins mg/100 g	Antioxidant activity is 50% inhibited by the mg sample A
1	Pulp50% C_2H_5OH	550,85	437,39	83,42	5.94
2	Pulp96% C_2H_5OH	989,42	781,57	147,92	10.01
3	Leaf 50% C_2H_5OH	1588,358	747,35	693,16	0.48
4	Leaf 96% C_2H_5OH	1349,956	682,42	590,53	1.13

Table 2. Quantitative content of total phenols, flavonols and catechins of Elaeagnus umbellata fruits and leaves taken in Guria (Ozurgeti)

№	Sample name	Common phenols mg/100 g	Flavonoids mg/100 g	Catechins mg/100 g	Antioxidant activity is 50% inhibited by the mg sample A
1	Pulp50% C_2H_5OH	257,85	173,87	59,34	9.38
2	Pulp96% C_2H_5OH	309,11	201,01	76,54	10.60
3	Leaf 50% C_2H_5OH	2546,78	1238,65	1147.36	0.47
4	Leaf 96% C_2H_5OH	2366,94	1145,67	1012,36	1.78

Table 3. Quantitative content of total phenols, flavonols and catechins of Elaeagnus umbellata fruits and leaves taken in Adjara seaside (Khelvachauri)

№	Sample name	Common phenols mg/100 g	Flavonoids mg/100 g	Catechins mg/100 g	Antioxidant activity is 50% inhibited by the mg sample A
1	Pulp50% C_2H_5OH	568,20	429,61	65,36	7.70
2	Pulp96% C_2H_5OH	687,68	538,62	108,36	7.90
3	Leaf 50% C_2H_5OH	1389,661	735,43	584,80	0.48
4	Leaf 96% C_2H_5OH	1238,07	680.54	481,60	1.69

Table 4. Quantitative content of total phenols, flavonols and catechins of Elaeagnus umbellata fruits and leaves taken in upper Adjara (Keda)

№	Sample name	Common phenols mg/100 g	Flavonoids mg/100 g	Catechins mg/100 g	Antioxidant activity is 50% inhibited by the mg sample A
1	Pulp50% C_2H_5OH	117,98	57,81	36,12	12.04
2	Pulp96% C_2H_5OH	203,41	96,99	53,86	12.99
3	Leaf 50% C_2H_5OH	1219,59	675,64	465,40	0.89
4	Leaf 96% C_2H_5OH	1128,69	580,65	418,39	2.44

The content of total phenols in *Elaeagnus umbellata* fruits varies from 117,98 mg/100 g to 989,42 mg/100g. The highest rate was observed in the sample taken in Senaki area at 989,42 mg/100 g, and the lowest in the sample Keda of 117,98 mg/100g. The content of total phenols in *Elaeagnus umbellata* leaves varies from 1128,69 mg/100g – 2546,78 mg/100g. The highest rate was observed in the sample taken in Ozurgeti area at 2546,78 mg/100g, and the lowest in sample Keda at 1128,69 mg/100g. The content of total flavonoids in *Elaeagnus umbellata* fruits varies from 57,81 mg/100g to 781,57 mg/100g. The highest rate was observed in the sample taken in Senaki area at 781,57 mg/100g, while the lowest rate was observed in sample Keda at 57,81 mg/100g. The content of total flavonoids in *Elaeagnus umbellata* leaves varies from 580,65 mg/100g – 1238,65 mg/100g. The highest rate was observed in the sample taken in Ozurgeti area at 1238,65 mg/100g, and the lowest in the sample Keda at 580,65 mg/100g. The content of catechins in *Elaeagnus umbellata* fruits varies from 36,12 mg/100g to 147,92 mg/100g. The highest rate was observed in the sample taken in Senaki area 147,92 mg/100g and the lowest in the sample Keda of 36,12 mg/100g. The content of catechins in *Elaeagnus umbellata* leaves varies from 418,39 mg/100g to 1147,36 mg/100g. The highest rate was observed in the sample taken in Ozurgeti area 1147,36 mg/100g, and the lowest in the sample Keda 418,39 mg/100g. We also determined their antioxidant activity. Determination of all extractants taken for extraction showed that 50% extract, where the total amount of phenols was present, the maximum antioxidant was also high. Antioxidant rate of 50% inhibitors in *Elaeagnus umbellata* fruits fluctuates between 5.94 (52.04%) - 12.99 (40.3%) of the mg sample. A good rate was observed in the sample taken in Senaki area 5.94 (52.04%) and in the low sample taken in keda 12.99 (40.3%). The antioxidant rate of 50% in the leaves of *Elaeagnus umbellata* in the mg sample ranges from 0.47 (53.02%) to 2.44 (41.02%). The high rate was observed in the sample taken in Ozurgeti area 0.47 (53.02%) and low in the sample taken in Keda 2.44 (41.02%).

As can be seen from the survey results, the data differ depending on geographic location and climate. The lower the distribution of plants from sea level is, the higher the amount of studied elements in the plant is. Also, the content of these compounds in the leaves of the plant is almost 2-10 times higher, depending on the subclasses of compounds and the concentration of the alcohol solution during extraction.

Conclusion

Using modern physicochemical and instrumental methods, we have studied the quantitative content of biologically active compounds, in particular, common phenols, flavonoids, catechins, found in the fruits and leaves of the *Elaeagnus umbellata* plant, collected on the territory of 4 regions of Georgia - Samegrelo, Guria, coastal Adjara and high-mountainous Adjara (Senaki, Ozurgeti, Khelvachauri, Keda), as well determined their antioxidant activity. The samples, taken in Samegrelo and Guria, have a particularly high data rate due to their similar geographic location. The correlation has also beendetermined. The content of total phenols is directly proportional to the antioxidant content. Compared with the literature data, the results obtained show that plants widespread in Georgia are characterized by a high content of physiologically active compounds, what makes it possible to obtain about 5-20 g of biologically

active compounds from one kilogram of fruit, and 20-50 g - from one kilogram of leaves. Based on the data obtained as a result of research, in the future *Elaeagnus umbellata* can be used as a new raw material for a preventive, functional product or dietary supplement.

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