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BULGARIAN TRADITIONAL FOODS – SOURCES OF ANTIOXIDANTS

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Many scientific studies are focused on antioxidant activities in prevention, on their beneficial role for health status and age retardation. The main source of antioxidants for humans is the diet.

The aim of the study was to present data for antioxidant compounds in traditional Bulgarian foods, typical of Bulgarian diet.

The traditional foods selected for this study were: onion, pepper, tomatoes, green beans, apples, and wild fruit varieties – blueberries, blackberries, raspberries. Antioxidant vitamins E and C were determined by RP-HPLC/fluorescence and UV detection. Total phenolics were determined by the Folin–Ciocalteu assay. Total flavonoids were measured by the aluminum chloride assay. Total carotenoids were determined by RP-HPLC/UV-Vis.

The results showed that the highest amount of vitamin E was found in red peppers (2.94 mg/100g), and in raspberries (1.47 mg/100g), comparable with the level in sesame (2.4 mg/100g). Red peppers were evidenced to be the richest source of vitamin C (102 mg/100g). Total phenolics and total flavonoids as a marker of food antioxidant potential had the highest level in blueberries (670 mgGAE/100g, 190.3 mgCE/100g) and blackberries (355mgGAE/100g, 55.5 mgCE/100g). Total carotenoids are mostly detected in tomatoes (7.70 mg/100g), followed by spring onion (3.50 mg/100g).

The modern analytical techniques enable the assessment of antioxidant constituents in some foods typical of the Bulgarian diet, and furthermore, on the basis of the results the assumption could be made that the Bulgarian longevity is associated not only with yogurt consumption, but with the abundance of antioxidants in the traditional diet as well. *Acta Medica Medianae 2013;52(1):5-8.*

Key words: *antioxidants, phenolics, flavonoids, carotenoids, vitamins E and C, traditional foods*

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Introduction

One of the current trends in nutritional science outlines the defense of traditional foods in response to the massive invasion of other cuisines, not typical of the diet of the respective population. Many researchers present evidence for the role of the traditional diet in health protection and in achieving longevity (1-4). Nutrigenomics also contributes and provides explanations in this aspect as well (5).

The defense of traditional foods could be realized only by comprehensive knowledge of their nutrient composition and content of bioactive components. The current achievements of analytical techniques provide this opportunity (spectrometry, gas and liquid chromatography with various detections). They have expanded significantly the spectrum of compounds that can be determined precisely in foods, aiming to assess their nutritional and biological value (6-9).

One hypothesis explaining the importance of food as preventive factor and factor for health protection and for retarding ageing processes is the "antioxidant hypothesis" confirmed by the studies of numerous researchers. A number of compounds having antioxidant activity were discovered and identified that, when included in the diet support the antioxidant protection of the organism. Some of them are compounds of the carotenoid, flavonoid, terpens, vitamins and other groups (10-14).

In the past, the longevity of the Bulgarian population was mainly associated with Bulgarian yogurt. Nowadays, the new scientific evidence enables us to state also the role of antioxidants provided by traditional Bulgarian foods.

The aim of the study was to present data for antioxidant compounds in traditional Bulgarian foods, typical of Bulgarian diet.

Materials and methods

The traditional foods selected for this study were: onion, pepper, tomatoes, green beans, apples, and wild fruit varieties – blueberries, blackberries, and raspberries, characteristic for mountain regions of the country.

Antioxidant vitamin E (α-Tocopherol) was determined by RP-HPLC with fluorescence detection (Ex=285 nm, Em=345 nm). Isocratic elution with methanol, flow rate 1.0 ml/min and temperature set at 400C was applied. Sample treatment included saponification prior to extraction with KOH and ethanol. The limit of detection (LOD) of the method was 0.006 mg%, whereas the limit of quantitation (LOQ) was 0.02 mg%. The recovery rates was between 88-95%, and relative standard deviation RSD=10%.

Vitamin C was determined by RP-HPLC/UV detection at 254 nm and 35 0C by using ion pair chromatography with hexadecyl-trimethylammonium bromide as mobile phase modifier, and methanol/ phosphate buffer with pH=5.5 elution (35:65 v/v). The analytical parameters of the method were as follows: LOD=0.02 mg%, and LOQ=0.05 mg%. The analytical recovery was 92%, and RSD=1.9%.

The total phenolic content of fruits and vegetables were determined by using the Folin–Ciocalteu assay. The determined analytical parameters of the total phenolics method were as follows: LOD - 0.4 mg GAE/100 g fresh weight; LOQ 1.2 mg GAE/100 g fresh weight; recovery 97% and repro-ducibility (RSD) 2.7 %.

The total flavonoid content was measured by the aluminum chloride colorimetric assay. The LOD was 0.6 mg CE/100 g fresh weight; LOQ - 1.8 mg CE/100 g fresh weight; recovery - 96% and repro-ducibility (RSD) - 3.7 % (15).

The total carotenoids content was measured as a sum of lutein, zeaxanthin, lycopene, β- crypto-xanthin, α-carotene, and β-carotene with RP-HPLC method using mobile phase of acetonitrile: methanol (95:5) (eluent A) and acetonitrile: methanol: ethyl acetate (60:20:20) (eluent B) with gradient elution: 100 % A for 5 min, followed by linear gradient to 100 % B at 13 min. The detection was carried out with UV-Vis detector at 450 nm. Carotenoids were quantified using external calibration method. The constructed six pointed calibration curves (n=3) were linear (correlation coefficients >0.99) in the working range: 0.04–5 μg/ml. The LOD and LOQ of the method for individual carotenoids varied within the range 0.45-0.86 μg%, and 1.35-2.57 μg%. The recoveries were very high – 95-103 %, and RSD very small – 2.5-4.7%.

Results and discussion

The results of the present study are shown in Table 1. All data refer only to edible portion and are expressed in mg/100 g of fresh weight of the sample. The results for total phenolics are expressed in mg gallic acid equivalent for 100 g, and for total flavo-noids in mg (+)-catechin equivalent for 100 g food.

The results show the highest amount of vitamin E is in red peppers (2.94 mg%), followed by raspberries (1.47 mg%). In comparison with other dietary sources of α-Tocopherol, these levels are logically lower than those in plant oils, like sunflower-seed refined oil (44.9 mg%) or olive oil (11.9 mg%) for instance, but they are comparable with the vitamin E level in some seeds, like sesame (2.4 mg%). The quality of the eaten food determining the level of α-Tocopherol in the organism is also important.

Vitamin C, besides its own antioxidant properties in aqueous media, can regenerate the oxidized forms of vitamin E. Red peppers again, as for vitamin E, are the richest source of vitamin C (102 mg%). This is the highest amount of ascorbic acid found in natural food, according to the Bulgarian food composition table. Greater amounts of vitamin C are found only in fortified food products. The high consumption of this vegetable in summer in Bulgaria makes it the most significant dietary source of vitamin C, since the Bulgarian traditional diet does not imply high consumption of citrus fruit, like the Mediterra-nean one. The results show that green peppers and spring onion have also great amounts of ascorbic acid – 102 mg% and 45.6 mg%, respectively. These data enable the statement that in the traditional Bulgarian diet vegetables provide a greater share of vitamin C intake, since in fruits its level varies between 3.5 mg% in blueberries and 26.3 mg% in raspberries.

Total phenolics and total flavonoids are a very popular characteristic of food antioxidant potential. Their highest level was found in blueberries (670 mgGAE/100g, 190.3 mgCE/100g) and blackberries (355mgGAE/100g, 55.5 mgCE/100g). It can be outlined that phenolics content in green peppers (246.7 mgGAE/100g) is higher than in red ones (173.2 mgGAE/100g), and significantly greater than that in tomatoes (76.9 mg GAE/100g). Tomatoes and peppers are characteristics for the diet of lowland regions of Bulgaria, and wild fruits varieties are

Table 1. Content of antioxidant vitamins E and C, total phenolics, flavonoids, and carotenoids in selected traditional Bulgarian foods

Food	Vitamin E mg/100g	Vitamin C mg/100g	Total Phenolics mg GAE/100g	Total Flavonoids mg CE/100g	Total Carotenoids mg/100g
Onion	0.43	11.7		-	-
Onion, spring	-	45.6	120.0	16.0	3.50
Pepper, red	2.94	143	173.2	13.7	1.20
Pepper, green	0.70	102	246.7	27.4	0.66
Tomatoes	0.34	17	76.9	12.8	7.70
Green beans	0.02	16.6	35.5	4.1	0.54
Apple	0.61	5.76	112.6	41.7	0.76
Blueberry	-	3.5	670.9	190.3	0.29
Blackberry	0.61	14.23	355.3	55.5	0.44
Raspberry	1.47	26.3	178.6	26.6	0.37

(-) – not analyzed

more typical for the mountain regions of the country. According to the results, the antioxidant load of the traditional Bulgarian diet is divided– vegetable sources for lowland regions and seasonal fruit sources for mountain regions.

The carotenids are plant pigments with nutritive (α-Carotene and β-Carotene) and non-nutritive (Lutein, Zeaxanthin, Lycopene, and β-cryptoxanthin) properties. However, all representatives of this group have strong antioxidant potential. The total carotenoids are higher in tomatoes (7.70 mg/100g), followed by spring onion (3.50 mg/100g), and red peppers (1.20 mg/100g).

A wide spectrum of antioxidants dietary intake involves more effective defense against the free radicals and reactive oxygen spices, with all the mechanisms of bioavailability and bio-efficacy in place. However, the first stage of the assessment of this process is the recognition of the qualitative and quantitative composition of the dietary antioxidants in foods. On the basis of such detailed data, preventive diets and healthy nutrition regimes can be established.

Conclusion

The modern analytical techniques allow us to make the assessment of antioxidant constituents in some foods typical of the Bulgarian diet, and furthermore, on the basis of the results, the assumption could be made that the Bulgarian longevity is associated not only with yogurt consumption, but with the abundance of antioxidants in the traditional diet, too. Furthermore, the presented data can be used for establishment of a data base for antioxidants in foods characteristic of the Balkan region as Bulgarian traditional foods are an integral part of the Balkan diet.

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TRADICIONALNE BUGARSKE NAMIRNICE – IZVORI ANTIOKSIDANATA

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Mnoge naučne studije bave se preventivnom ulogom antioksidanata i njihovim značajem za ljudsko zdravlje i usporavanje procesa starenja. Glavni izvor antioksidanata za ljude jeste ishrana.

Cilj ovog istraživanja bio je da predstavi rezultate koji se odnose na sastav antioksidanata u tradicionalnim bugarskim namirnicama karakterističnim za bugarsku ishranu. Ispitivane su sledeće namirnice: crni luk, paprika, paradajz, boranija, jabuke, različite vrste divljeg voća poput borovnica, kupina, malina. Nivo vitamina E i C određen je RP-HPLC fluorescencijom i UV detekcijom. Ukupan nivo fenola meren je Folin-Cicoalteu analizom, a nivo flavonida pomoću aluminijum hlorid analize. Ukupan nivo karotenoida određivan je pomoću RP-HPLC/UV-Vis.

Rezultati su pokazali da se najveća količina vitamina E nalazi u crvenoj paprici (2.94 mg/100gr) i u malinama (47 mg/100g), u odnosu na količinu ovog vitamina u susamu (2.4 mg/100g). Najbogatiji izvor vitamina C jeste crvena paprika (102 mg/100g). Najviši nivo fenola i flavonida kao markera antioksidanta u hrani prisutan je u borovnicama (670 mgGAE/100g, 190.3 mgCE/100g) i kupinama (355mgGAE/100g, 55.5 mgCE/100g). Karotenoidi su najviše prisutni u paradajzu (7.70 mg/100g), a potom u mladom luku (3.50 mg/100g).

Savremene analitičke tehnike omogućile su procenu antioksidanata u određenim namirnicama karakterističnim za bugarsku ishranu. Štaviše, na osnovu ovih rezultata može se pretpostaviti da je dugovečnost stanovnika Bugarske povezana ne samo sa unosom jogurta, već i sa bogatim izvorima antioksidanata u tradicionalnoj ishrani. *Acta Medica Medianae 2012;52(1):5-8.*

Ključne reči: *fenoli, flavonidi, karotenoidi, vitamini E i C, tradicionalne namirnice*