

Use of sea buckthorn bioactive potential in obtaining of farinaceous functional products

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Abstract

The problem of functional foods with bioactive components, in general, of substances with natural origin, is more than current for food industry. The sea-buckthorn (*Hippophaë rhamnoides L.*), is a rich source of natural bioactive compounds which are playing an important role in the prevention of degenerative diseases. Starting from these considerations, aim of this study was to obtain and characterize some functional flour products such as digestive cookies. Were evaluated the sensory, physic-chemical and antioxidant properties, of digestive cookies with the addition of 10% fresh sea buckthorn (FSB), dried sea buckthorn fruits powder (DSB) and sea buckthorn syrup (SSB). The results showed that the addition of 10% FSB, DSB and SSB in dough led to the improvement of sensory and physic-chemical characteristics of the cookies samples obtained, and consequently to the increase of nutritional value and their antioxidant properties.

Keywords: cookies formulation, cookies quality, bioactive compounds, sea buckthorn, antioxidant activity, functional products

1. Introduction

Together with all other sectors in the food industry, the bakery industry and floury products sector, occupies an important place in the framework of consumer goods manufacturing, mainly due to the fact that bread is a daily consumed, staple food. This is because bakery and floury products, together with all other food products, provide human body a significant part of the necessary substances for the vital activity, maintaining health status and work capacity.

In this context, for the moment, new trends in consumer preferences are oriented to new baked and floury goods to the detriment of classic white bread, man's interest in other specialties other than local or traditional, has increased, and over the last

years functional foods and green products have been intensively promoted. As a result, two main currents raised, depending on market segmentation, namely: one directed to "convenient" bakery products type (small and easy to consume products, most of the times pre-packaged and with a longer shelf-life) characteristic to "fast food" type nutrition, and the other, at the opposite pole, directed to fresh, natural or with benefits to health baked goods, corresponding to a more healthful diet, possibly "slow food" type diet [4, 5].

For this purpose, a series of biochemical, microbiological and nutritional studies, have been carried out, regarding the impact of the diet based on bioactive components functional food, on the quality

of life and in preventing the occurrence of degenerative diseases.

After clinical trials, the specialists have identified a number of benefits for functional food consumption: the body defense by reducing body allergies and activate immune system, prevent illness by preventing hypertension, diabetes, congenital metabolic as well as anti-tumour imbalances, control of certain functions of the body like central nervous system operation, the appetite and natural nutrients absorption, reducing the duration of the convalescence and, ultimately, reducing the effects of aging. So, to achieve functional foods represented at the same time, both a challenge for technologists, famous nutritionists, biochemists and microbiology specialists, and along with the increasing interest for functional foods, new products have been developed and with them, the need for the standards and processes of development, and promotion of these products [3, 4].

Therefore, in the offing to ensure a diet based on bioactive components functional food, the specialists have designed and produced a wide range of assortment functional products. Designing and promoting functional food aim to identify beneficial interactions between a functional component of food and one or more target functions in the body [5, 6].

In functional food category are included all those foods that can be consumed in the normal diet, which contain bioactive compounds, with the potential for the prevention and improvement of the health status. Examples of such foods are those that contain minerals, vitamins, fatty acids, fiber, biologically active substances added food, such as antioxidants and probiotics.

Intensive colored fruits and vegetables (blueberries, blackberries, cherries, kiwi, broccoli, spinach, parsley leaves) have antioxidant effects. Juices obtained from these fruits and vegetables contain polyphenols that induce antioxidant enzyme activity, stimulate proliferation of lymphocytes as well as action of the natural-killer cells [5, 6, 7].

Antioxidants are compounds of endogenous and exogenous origin, through which the body fights

against free radicals. Antioxidants are either antiradical action enzymatic systems (superoxide-dismutase, catalase, glutation-peroxidase) existing in natural fund of the organism, or substances with antiradical action of exogenous origin, from food (vitamin E, vitamin C, carotenoides, glutatione, flavones, polyphenols, selenium).

Foods rich in antioxidants are fresh fruit and vegetables, whole grains, grains [6, 7]. In the great variety of food that is rich in antioxidants, sea buckthorn is one of the most powerful antioxidants that mother offers us each year. These sour fruits contain twice as much vitamin C than hip tree (*Rosa canina*) and up to ten times more vitamin C than oranges or lemons. Nutritionally, the sea buckthorn is a food very rich in A, E, P, K, F, vitamins and trace elements vitamins, in complex B (B1, B2, B6, B9) vitamins and other substances which are important for health. Another interesting aspect on sea buckthorn is that it contains high quantities of β -caroten compared with carrot pulp, so white sea buckthorn is "soaked" in β -caroten [2].

Also, in Romania, sea buckthorn grows naturally and it seems that it has an exceptional content of active principles, perhaps the most valuable in the world. It is to be mentioned that nowhere in the world, the sea buckthorn varieties which are growing naturally, or have been improved do not have a composition at the level of which can be found in the areas of our subcarpathian areas of our country [8].

Starting from these observations, this paper aims the exploitation of the sea buckthorn bioactive potential, in order to obtain some farinaceous functional food products: digestive cookies.

2. Material and methods

2.1. Materials

All raw materials used in these experiments have been purchased from markets of specialized stores.

2.2. Methods

2.2.1. Evaluation of average nutritional value of fresh sea buckthorn of sea buckthorn based products.

In order to evaluate average nutritional value of fresh sea buckthorn and of sea buckthorn based products (FSB), of dried sea buckthorn fruits powder (DSB) and of sea buckthorn syrup (SSB), the following chemical characteristics of these, were carried out:

moisture content, carried out according to A.O.A.C. standard method, 2000 [9]; protein content using Kjeldahl method [10]; lipids content using Soxhlet extraction [11]; carbohydrates content using Lane Eynon method [12].

2.2.2. Technological process for obtaining FSB, DSB and SSB addition digestive cookies. In this study we have obtained 4 types of cookies, as follows: a control sample (C) – without any addition of sea buckthorn, cookies with fresh sea buckthorn (CFSB), cookies with dry sea buckthorn (CDSB) and cookies with syrup of sea buckthorn (CSSB). The technological process for obtaining digestive cookies was the common one, used in sugary cookies manufacturing. The recipe used, was the following one: white wheat flour type 000 - 50%, wholegrain wheat flour type 800 – 25%, oat bran – 25%, brown sugar – 25%, butter – 20%, milk – 10 ÷ 20%, salt – 0.5%, baking soda and baking powder – 1%, eggs – 6% and 10 % FSB, DSB and SSB each. Similarly, a control digestive cookies sample without sea buckthorn or its products addition was performed. After foaming the eggs with sugar, the other ingredients were added. Everything was mixed up for 10 minutes, at 125 rpm, an homogeneous and stable dough being obtained. After that, the dough was let to rest for 40 minutes at 4°C. Then, the dough pieces were laminated (5 mm thickness) and cut, using a rotary moulder (51 mm) and baked at 160°C for 20 minutes. After baking, the cookies were cooled at room temperature, and then packed in PP bags [13].

2.2.3. Sensory and physical-chemical evaluation of FSB, DSB and SSB addition digestive cookies. The digestive cookies samples obtained according to the method described in paragraph 2.2.2., were submitted to sensory and physical-chemical evaluation, aiming: aspect, crumb aspect, color, taste, odor, consistence, diameter, defect cookies, alkalinity, soak capacity, humidity, ash content, according to A.O.A.C. standard method [14,15].

2.2.4. Determination of vitamin C of the samples. **Vitamin C (VC)** was determined by titrimetric method using 2,6-dichlorophenolindophenol sodium salt solution according to official method of analysis described by AOAC [16]. This analysis was performed both for all forms of sea buckthorn

used in recipe (FSB, DSB and SSB) and obtained cookies (C, CFSB, CDSB and CSSB). The results were expressed as mg/100 g.

2.2.5. Determination of total antioxidant capacity (TAC). The analysis of total antioxidant capacity (TAC) was performed on the base of CUPRAC (cupric ion reducing antioxidant capacity) method. CUPRAC method depends on the reduction of cupric neocuproine complex to the cuprous neocuproine complex, at low pH. The neocuproine complex was monitored by measured the absorption at 450 nm. As reference substance, in this study, it was used TROLOX (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid), an antioxidant compound having a structure similar to vitamin E. 1mL 0.01M CuCl₂ solution was mixed with 1mL 7.5·10⁻³M neocuproine (2,9-Dimethyl-1,10-phenanthroline) and 1 mL acetate buffer and than it was added 1.1 mL alcoholic extract obtained from our samples. For control, it was used ethanol 20%. The absorption was read at 450 nm after incubation for 30 min. at 20°C. All determinations were carried out in triplicate. TAC was expressed as μM TROLOX/100g. Correlation coefficient (r) for calibration curve was 0.9784 [17, 18, 19].

3. Results and Discussion

3.1. Average nutritional values of FSB, DSB and SSB. Average nutritional values of FSB, DSB and SSB was evaluated in this study, is given in *Table 1*:

Table 1. Average nutritional values of FSB, DSB and SSB

Components	Experimental values		
	FSB	DSB	SSB
Humidity (g/100g)	82.07	3.25	37.65
Proteins, (g/100g)	1.14	1.19	0.41
Lipids (g/100g)	0.69	0.72	1.55
Carbohydrates (g/100g)	10.18	10.22	64.26

By comparing the values obtained from the analysed samples (FSB, DSB, SSB) with quality standards (STAS 8613/93) [20], it can be observed that these results fit within the limits of admissibility for all samples. Summarizing the data presented in *Table 1*, it can be seen that both the fresh sea buckthorn fruits, and the products based on sea buckthorn, beside the richest sources of vitamins (A, E, C, B1, B2, K and P) [21] and trace elements (K, Mg, Fe, Mn) [22], represent important protein (0.41 ÷ 1.14 g/100g product), lipids (0.69 ÷ 1.55 g/100g product)

and carbohydrates (10.18 ÷ 64.26 g/100g product) sources, too, in comparison with other fruits, nutrients as important to human body in preventing many diseases.

3.2. Sensory evaluation of FSB, DSB and SSB addition digestive cookies. The use of 10% FSB, DSB, SSB to digestive cookies recipe has allowed obtaining of breads with optimal sensory characteristics in accordance with *STAS 1227-3/1990*. Sensory evaluation of the assortments of digestive cookies obtained in the laboratory was performed using the points scale method, which accordingly, they have obtained the following qualifications:

Table 2. Medium scores assigned to digestive cookies samples

Sample	Maximum score	Scores obtained
Control	17.85	good
CFSB	19.71	very good
CDSB	19.07	good
CSSB	19.50	very good

The four assortments of digestive cookies obtained, were sensory evaluated by a number of 20 tasters. According to data shown in *table 2*, it is observed that the cookies samples sensory evaluated fall into the first two quality categories (“very good” and “good”), but from the consumer preference point of view, the digestive cookies with 10% FSB, were the most rated, with a maximum score of 19.71 from the maximum 20 possible points.

Also, the obtained results (*table 2*) have shown that adding 10 % FSB, DSB and SSB, respectively, to digestive cookies samples, led to: a brighter colour (golden yellow) towards to control sample, which shown a brownish colour; form was obtained cookies well defined, undistorted shell surface showed no cracks or other defects; the taste and odour were improved, having a pleasant characteristic aroma in comparison with control sample; digestive cookies samples consistence was tolerable (crispy but not brittle) in comparison with control sample (hard and brittle). Regarding the total quality, it can be observed that the digestive cookies with FSBTF, DSBTFP and SBTS addition, shown higher values (19.07 ÷ 19.71

points) towards control sample (17.85 points). The results obtained in this study fit with those obtained by Ashoush *et al.* [13].



C



CFSB



CDSB



CSSB

3.3. Physical-chemical evaluation of FSB, DSB and SSB addition digestive cookies. After the sensory examination digestive cookies samples were subjected to physico-chemical analysis. The experimental results obtained in this study, are given in *Table 3*:

Table 3. Physical-chemical evaluation of FSB, DSB and SSB addition digestive cookies

Physico-chemical parameters	C	CFSB	CDSB	CSSB
Diameter (mm)	35.81	37.22	37.53	37.90
Thickness (mm)	9.19	10.12	10.24	10.58
Defective cookies (% from weight)	20.60	14.52	15.20	12.82
Water impregnation capacity (g)	5.78	6.39	6.66	13.66
Humidity (%)	7.93	7.55	4.78	6.57
Alkalinity (grade)	2.40	2.90	2.60	2.70
Ash (%)	1.40	2.14	2.42	2.23

The highest dimensions were determined in digestive cookies with the addition of SC (the diameter was 37.90 mm and 10.58 mm thick), while the lowest values of dimensions were recorded at digestive cookies - blank (the diameter was 35.81 mm and 9.19 mm thick) (*Table 3*). The results in this case showed that the addition of FSB in the dough determined an increase of 3.93% in the diameter and 10.11% in thickness of cookies, compared to the blank; the addition of DSB in the dough determined an increase of 4.80% in the diameter and 11.42% in thickness of cookies compared to the blank; the addition of SSB in the dough determined an increase of 5.83% in the diameter and 15.12% in thickness of cookies compared to the blank. According to these results it can be appreciated that the use of three types of additives (FSB, DSB, SSB) in the dough determined an increase of cookies dimensions, compared to the blank.

Regarding of percentage of cookies with defects from the total mass (*Table 3*), it can be seen that the best sensory characteristics it presents cookies with the addition of SSB, where it was 37.76% lower than the blank. But doing a comparison between the three samples of cookies with addition of FSB, DSB and SSB in terms of percentage of defects cookies it can be seen that cookies with the addition of DSB this is the largest (15.20%), and a possible explanation for this result may be the faulty homogenization of sea buckthorn powder in the dough.

From the data presented in *Table 3* it can be seen that this characteristic of cookies, namely soaking capacity increases for all samples of cookies with addition of FSB, DSB and SSB (6.39 ÷ 13.66 g water) compared with the blank (5.78 g water), which proves that these additives promotes the

absorption of large quantities of water in the product.

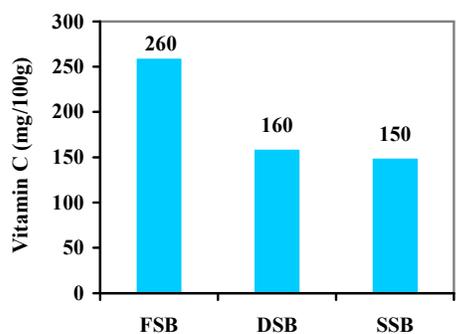
The humidity of samples with addition of FSB, DSB and SSB was without exception lower than the blank (7.93%), ranging from 4.78% in sample with the addition of DSB to 7.55% in sample with the addition of FSB. The big humidity difference found for the samples with the addition of FSB, DSB and SSB is due to different water content of the three types of additives and the low level of hydration of gluten proteins from the dough. Also, the values obtained suggest that the cookies with addition of DSB show greater stability during storage.

Higher alkalinity of biscuits samples with addition of FSB, DSB and SSB (2.60 ÷ 2.90 degrees) than the blank (2.40 degrees) is due to the intake of chemicals, the microbial load or the enzymes that come with the three types of additives in the dough.

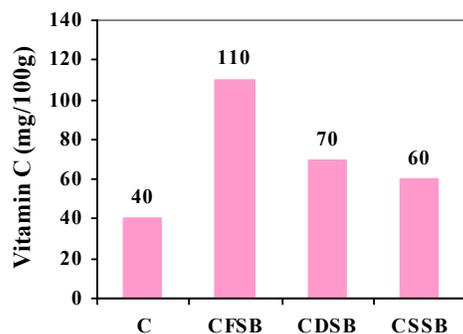
As shown in *Table 3*, it can be seen that the addition of FSB, DSB and SSB in biscuits increases their nutritional value respectively the content of mineral substances which are growing from 1.40% in the blank to 2.42% in the sample with the addition of DSB.

3.4. Vitamin C content in investigated samples. Vitamin C content depicted in *Figure 2* reveals the differences among sea buckhorn products (a) and the changes in response to cookies processing (b). FSB presents the highest content of VC, followed by DSB and SSB. The decreases in the content of VC recorded in the last sea buckhorn products could be explained by destroying of this bioactive compound in response to thermal processing applied for drying of fresh sea buckhorn as well as a result of the fresh sea buckhorn content from SSB (30 g fresh buckhorn/100 g syrup). When we used these forms of sea buckhorn in cookies technology, the recorded values show that the highest content of VC was

displayed by CFSB and the lowest by control sample. The cookies with DSB and SSB presented intermediate values. We can see that the adopted thermal regime (20 min at 160°C) for cookies processing didn't induce a large losses in the VC content. From this point of view, we can recommend the use of sea buckhorn in different form of preservation in the obtaining of various pastries and bakery products. The most important source of VC is FSB, but the main shortcoming derives from its short shelf life. Thus, DSB and SSB represent alternative forms of sea buckhorn which kept this antioxidant potential.



(a)

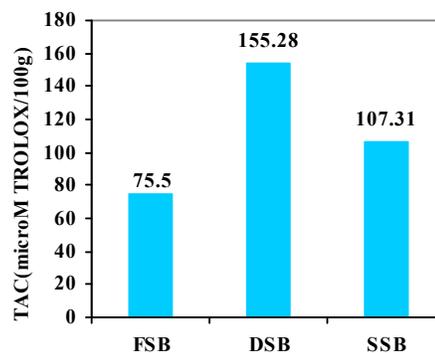


(b)

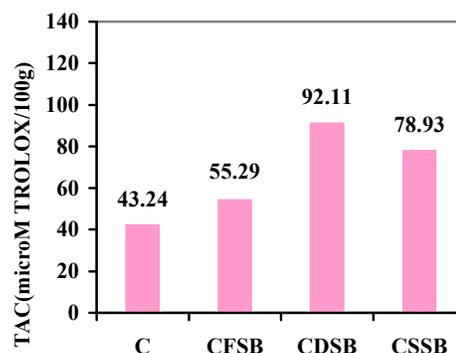
Figure 2. The content of VC in analysed samples (a) sea buckhorn products; (b) cookies

3.5. Antioxidant capacity in investigated samples.

Data presented in *Figure 3* highlight the antioxidant properties recorded for sea buckhorn products and for cookies obtained using these products in technological recipe.



(a)



(b)

Figure 3. TAC values in analysed samples (a) sea buckhorn products; (b) cookies

The results shown in *Figure 3a* reveal that DSB had the highest TAC, followed by SSB and FSB. This fact can be explained by the highest content of bioactive compounds from DSB that are present in addition vitamin C. From literature data, the sea buckhorn contains vitamins B1, B2, folic acid, vitamin C, E, beta-carotene (Pro-vitamin A), and K. Also, it contains carotenoids, flavonoids, phenols and terpenes, these compounds proving antioxidant properties [21,22, 23].

Based on the above mentioned results, these sea buckhorn products can be considered functional food ingredients used further in order to obtain different therapeutic functional food products. A valuable way for exploit this potential is represented by using of these products for processing of different types of cookies.

Depending to the values of TAC found in sea buckthorn products, in Figure 3b can be observed the antioxidant capacity of processed cookies. This parameters varies as follows: CDSB>CSSB>CFSB>C. The best retention of TAC in CDSB strengthen the previously mentioned idea regarding the highest extent of bioactive compounds with antioxidant potential preservation in DSB. Also, SSB is a good way to capitalize the sea buckhorn potential, from this point of view. As regards the use of FSB in thermal processed food products, this product brings along some bioactive compounds and can be considered for this purpose, but the most recommended are DSB and SSB.

4. Conclusions

The results obtained in this study indicate that the addition of 10% FSB, DBS and SSB in dough can be used with success in the obtaining functional cookies technology, thus, by this addition we can see the improvement of both sensory characteristics and physicochemical properties of cookies samples obtained.

On the base ob results obtained after performing this study, we can recommend the use of sea buckhorn in different form of preservation in the obtaining of various pastries and bakery products. The most important source of VC is FSB, but the use of this product is limited due to its short shelf life. The best retention of TAC in CDSB strengthen the previously mentioned idea regarding the highest extent of bioactive compounds with antioxidant potential preservation in DSB. Also, SSB is a good way to capitalize the sea buckhorn potential, from this point of view.

Thus, DSB and SSB represent alternative forms of sea buckthorn which kept this antioxidant potential. Taking into account the proven antioxidant properties of sea buckhorn products as a source of functional food ingredients, the investigated cookies can be considered therapeutic functional food products.

Also, by correlating the results obtained can be appreciated that established recipes from this study can be successfully applied on an industrial scale, thereby achieving valuable products both

nutritionally and in terms of technological requirements quality.

Compliance with Ethics Requirements. Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human / or animal subjects (if exist) respect the specific regulation and standards.

References

1. Bilgicli N., Ibanoglu S., Herken E.N., Effect of dietary fibre addition on the selected nutritional properties of cookies. *J Food Eng.*, **2007**, 78(1), 86–89, [doi:10.1016/j.jfoodeng.2005.09.009](https://doi.org/10.1016/j.jfoodeng.2005.09.009)
2. Bernath J., Foldesi D., Sea buckthorn (*Hippophae rhamnoides* L.) a promising new medicinal and food crop, *J. Herbs Spices Med. Plants*, **1992**, 1(1-2), 27–35, [doi:10.1300/J044v01n01_04](https://doi.org/10.1300/J044v01n01_04)
3. Mozaffarian D., Kumanyika S.K., et al.: Cereal, fruit, and vegetable fiber intake and the risk of cardiovascular disease in elderly individuals. *JAMA*, **2003**, 289(13), 1659 – 1666, [doi:10.1001/jama.289.13.1659](https://doi.org/10.1001/jama.289.13.1659).
4. Thomas D., Frankenberg E. Health, nutrition and prosperity: a microeconomic perspective, *Bull. World Health Organ.* **2002**, 80(2), 106-113, doi.org/10.1590/S0042-9686200200020000_5
5. Anton D. T., Coman A.E., The role of functional foods in promoting child health, *Medical Practice*, **2011**, VI, 2(22), 126-128
6. Szabo Caunii A.R., Food additives-antioxidant. Functional foods bioactive antioxidant components, *Thesis*, **2010**, Iasi,
7. Hurgoiu V., Functional foods, *Romanian Journal of Pediatrics*, **2004**, LIII (1), 18-23.
8. Gutzeit, D., Winterhalter, P., Jerz, G., Nutritional assessment of processing effects on major and trace element content in sea buckthorn juice (*Hippophaë rhamnoides* L. ssp. *rhamnoides*), *Journal of Food Sci.*, **2008**, 73(6), 97-102, [doi: 10.1111/j.1750-3841.2008.00817.x](https://doi.org/10.1111/j.1750-3841.2008.00817.x)
9. AOAC (Association of Official Analytical Chemists) Official Methods of Analysis International. 17th, **2000**, Ed. Washington, DC: AOAC.
10. Kjeldahl J., Determination of protein nitrogen in food products. *Encyc Food Agric.* **1983**, 28, 757–765, [doi:10.1002/jsfa.2740280815](https://doi.org/10.1002/jsfa.2740280815)
11. Yang BR, Kallio HP, Fatty acid composition of lipids in sea buckthorn (*Hippophaë rhamnoides* L.) berries of different origins. *J. Agric. Food Chem.*, **2001**, 49, 1939-1947, [doi: 10.1021/jf001059s](https://doi.org/10.1021/jf001059s)
12. Zhiben ZA., Chemical composition of seabuckthorn fruit., *J Guizhou Agric College China*, **1987**, 3(1), 51-60.

13. Ashoush I.S. Gadallah., M.G.E, Utilization of Mango Peels and Seed Kernels Powders as Sources of Phytochemicals in Biscuit, *World Journal of Dairy & Food Sciences*, **2011**, 6(1), 35-42.
14. A.O.A.C., "Official Methods of Analysis" *Association Official Analytical Chemists of the 16th Ed. International*, 1995, Washington, D.C., U.S.A.
15. STAS 1227/3-1990
16. AOAC, Vitamin C (ascorbic acid) in vitamin preparations and juices. In K. Helrich (Ed.). *Official Methods of Analysis*, 2000, 15th edn. AOAC, Inc., Arlington, VA, pp. 1058.
17. Reşat Apak, Kubilay Güçlü, Birsen Demirata, Mustafa Özyürek, Saliha Esin Çelik, Burcu Bektaşoğlu, K. Işıl Berker, Dilek Özyurt, Comparative Evaluation of Various Total Antioxidant Capacity Assays Applied to Phenolic Compounds with the CUPRAC Assay, *Molecules*, **2007**, 12(7), 1496-1547, [doi:10.3390/12071496](https://doi.org/10.3390/12071496)
18. Apak R, Güçlü K, Özyürek M, Karademir SE. Novel Total Antioxidant Capacity Index for Dietary Polyphenols and Vitamins C and E, Using Their Cupric Ion Reducing Capability in the Presence of Neocuproine: CUPRAC Method. *Journal of Agricultural and Food Chemistry*. **2004**, 52(26), 7970–7981, [doi:10.1021/jf048741x](https://doi.org/10.1021/jf048741x)
19. Apak R, Güçlü K, Özyürek M, Bektas Oğlu B, Bener M., Cupric ion reducing antioxidant capacity assay for food antioxidants: vitamins, polyphenolics, and flavonoids in food extracts, *Methods Mol Biol.*, 2008, 477, 163-193, [doi:10.1007/978-1-60327-517-0_14](https://doi.org/10.1007/978-1-60327-517-0_14)
20. STAS 8613/93
21. Tiitinen, K. M., Hakala, M. A., Kallio, H. P. Quality components of sea buckthorn (*Hippophaë rhamnoides*) varieties. *Journal of Agricultural and Food Chemistry*, **2005**, 53(5), 1692–1699, [doi:10.1021/jf0484125](https://doi.org/10.1021/jf0484125)
22. Venkatesh Meda, S.N. Naik, Santosh Satya, Sea buckthorn berries: A potential source of valuable nutrients for nutraceuticals and cosmeceuticals, *Food Research International*, **2011**, 44(7), 1718–1727, [doi:10.1016/j.foodres.2011.03.002](https://doi.org/10.1016/j.foodres.2011.03.002)
23. Szeto, Y. T., Tomlinson, B., Benzie, I.F.F., Total antioxidant and ascorbic acid content of fresh fruits and vegetables: implication for dietary planning and food preservation. *British Journal of Nutrition.*, **2002**, 87(1), 55-59, [doi: http://dx.doi.org/10.1079/BJN2001483](https://doi.org/http://dx.doi.org/10.1079/BJN2001483)