Effect of natural carotenoids and anthocyanins on properties of healthy food products

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Abstract. The results of studies on the use of concentrated ethanol extracts of hydrophilized natural carotenoid compounds from red carrot root crops and pumpkin fruits, obtained under the developed conditions, consisting in a gradient-step heat treatment of a plant biosystem with subsequent extraction of carotenoids with ethanol and concentration of extracts by removing the solvent are present in work. The concentrates of natural anthocyanins are produced using an acid-free technology for extracting polyphenols from various anthocyanin-containing plant materials (squeezed blueberries, blackcurrants, Isabella grapes, etc.), followed by concentration of the extract by distillation of ethanol under vacuum. The introduction of natural polyphenolic compounds of various plant materials in the form of anthocyanins with high inhibitory properties of oxidative radical processes into the composition of confectionery products (marmalade, caramel, and candy canes) can significantly increase the antioxidant properties of these food products. For example, the addition of concentrates of extracts of natural anthocyanins to jelly marmalade allows increasing its antioxidant properties by 1.5 - 2.5 times. The use of carotenoid and composite carotenoid-anthocyanin antioxidants in the production of alcoholic beverages with low AOA (tinctures, liquors) can increase their antioxidant properties by 1.5 - 3.5 times.

1. Introduction
Among the chemical reactions occurring in human body, the processes with the formation of organic radicals, highly reactive particles with a free electron, are of great importance. Free radicals damage the walls of blood vessels, membranes and other human organs and cause serious diseases. Oxygen in the air catalyzes the formation of radicals due to the intermediate formation of unstable peroxide compounds.

By introducing antioxidants into human diet in the form of food additives or food containing such compounds it is possible to inhibit the formation of radicals [1]. It is advisable to use polyphenolic compounds widely occurring in nature in the form of bioflavonoids (including anthocyanins), as well as polyene carotenoids as antioxidants.

The research is carried out on the effect of natural carotenoid and anthocyanin compounds in food products on their antioxidant activity (AOA).

2. Materials and methods
Carotenoid compounds were obtained as concentrates of ethanol extracts of hydrophilic carotenoids of plant raw materials containing 60-70% of β-carotene and 40-30% of xanthophylls of various
structures by the developed method [2]. Red carrots (Daucus Sativus Roehl) and pumpkin (Cucurbita pepo L.) were used as carotenoid raw materials.

Extraction of natural anthocyanins from anthocyanin-containing raw materials was carried out using the developed acid-free method by extracting polyphenols with ethyl alcohol with 96% of ethanol by volume. (GOST 5962-2013). The obtained extracts were purified from impurities and concentrated by distilling ethanol under vacuum [3]. The extracts of blueberries (Vaccinium myrtillus), black currants (Ribes nigrum) and Isabella grapes (Vitis labrusca V.) were used as anthocyanin-containing raw materials.

To obtain a mixture of carotenoid and anthocyanin compounds, the ethanol extract concentrates of polyphenol and polyene from carotenoid-containing raw materials (red carrots, Daucus Sativus Roehl, or pumpkin, Cucurbita pepo L.) and anthocyanin-containing raw materials (pomace of black currant, Ribes nigrum, or Isabella grapes, V. Vitis labrusca) dried to a moisture content of not more than 10% were used in a weight ratio of 1:2 [4].

The total mass concentration of oxidants in the solution (equivalent to cyanidin-3-glucoside) was determined on the basis of the changes in light absorption with a wavelength of 510 nm upon the change of acidity of solutions with pH from 1 to 4.4 [5], or the standard solution of hydrated cobalt sulfate (GOST 4462-78) in distilled water using photocolorimeter KFK-3 (Russia) according to the technique reported in the literature [6].

Quantitative analysis of carotenoid content (equivalent to β-carotene) was performed using standard solutions of pure β-carotene in ethyl acetate (GOST 22300-76).

Spectral characteristics of solutions of anthocyanin and carotenoid extracts were studied by recording absorption spectra on UV-mini 1240 spectrophotometers (Shimadzu, Japan) in quartz cells with an optical layer thickness of 10 mm.

AOA measurements of extracts of carotenoid and anthocyanin compounds and food products were performed by amperometric method on the Color analyzer Yauza-01-AA (Russia) using technique suggested by A. Ya.Yashin [7]. Rutin was used as a standard substance in AOA studies.

The preparation of confectionery products (marmalade, boiled sweets and jelly candies) was carried out in accordance with the recipe and technology [8]. Anthocyanin compounds were added to the caramel at the stage of sugar mass kneading in the form of alcohol solution with anthocyanin mass fraction of 20%. To make marmalade by traditional technology, a concentrate of blueberry anthocyanin extract was used in the amount of 0.1-1.5 g/kg of ready-made marmalade, 0.25-2 g/kg of black currants and 1-7 g/kg of grapes. Jelly sweets were made using black currant anthocyanins. Anthocyanins were introduced at the stage of marmalade mass tempering in the form of solution with anthocyanin mass fraction of 10%.

Alcoholic beverages (infusions, aperitifs and balsams) were prepared in accordance with the standard recipe and technology [9].

3. Results and discussion

The AOA studies of some food products show that they possess antioxidant properties due to the presence of natural inhibitors of oxidative processes in their composition.

For example, infusions and flavored spirits that are part of alcoholic beverages show antioxidant activity (Table 1).

Table 1. AOA of flavored spirits and liqueurs with some ingredients used in alcoholic beverages

| N  | Ingredient                     | AOA, mg/dm³ |
|----|--------------------------------|-------------|
| 1  | Lemon peel flavored spirit     | 0,56        |
| 2  | Orange peel flavored spirit    | 0,92        |
| 3  | Orange peel infusion           | 27,24       |
| 4  | Tangerine peel infusion        | 32,49       |
| 5  | Cinnamon infusion              | 75,44       |
| 6  | Nutmeg infusion                | 79,03       |
| 7  | Clove infusion                 | 6360        |
Table 1 shows that lemon and orange peel flavored spirits have a lower AOA compared to infusions of cinnamon, nutmeg and clove due to the fact that the infusions are produced by more effective distillation method.

Ethanol concentrates of natural anthocyanin and carotenoid compounds obtained for research possessed characteristics shown in Table 2 and 3.

**Table 2.** Main organoleptic and physico-chemical properties of ethanol concentrates of natural anthocyanin extracts

| N | Parameter                  | Description                      |
|---|----------------------------|----------------------------------|
| 1 | Physical form and color    | Thick syrup-like liquid of dark red color |
| 2 | Odour                      | Faint, distinctive               |
| 3 | Density at 20 °C, kg/m³    | 1500                             |
| 4 | Dry solids weight ratio, % | 60                               |
| 5 | Anthocyanin content in g/kg, min | 100                         |
| 6 | Solubility in water        | Complete                         |

Anthocyanin extracts from various raw materials contain different amounts of anthocyanin compounds: blueberry extract – 160 g/kg; black currant extract – 80 g/kg; grape extract – 15 g/kg.

**Table 3.** Main organoleptic and physico-chemical properties of ethanol concentrates of carotenoid extracts

| N | Parameter                                      | Description                                      |
|---|-----------------------------------------------|--------------------------------------------------|
| 1 | Physical form and color                        | Transparent yellow-orange ethanol extract        |
| 2 | Odour                                          | Characteristic to raw materials used             |
| 3 | Density at 20°C, kg/m³                         | 820                                             |
| 4 | Extractives content, %                         | 8,0 (max)                                       |
| 5 | Carotenoid content equivalent to β-carotene, mg/dm³ | 1000 (min)                                    |
| 6 | Solubility in water                            | Soluble in ethanol solutions with ethanol content of at least 10 % by volume |

When anthocyanins and carotenoids are simultaneously extracted with ethyl alcohol containing 96% of ethanol by volume, these compounds pass into the solvent both in free form, and as a result of the formation of associates of electron-donating carotenoids with electron-acceptor anthocyanins.

The study of the antioxidant properties of jelly marmalade (containing sucrose, molasses, citrus pectin, citric acid, sodium lactate and flavoring) shows a low AOA of this food product (table. 4), associated with the weak antioxidant properties of its components. For example, the AOA of granulated sugar is 0.5 mg / 100g, molasses is 1.8 mg/100g, and citric acid is 4.3 mg/100g.

However, the introduction of natural polyphenolic anthocyanins from various plant raw materials with high reducing properties into the composition of marmalade, allows not coloring and fortifying the product, but also increasing significantly its antioxidant activity. (Table. 4).
Table 4. Effect of natural anthocyanin compounds of various anthocyanin-containing raw materials on the AOA of jelly marmalade during product storage

| Extract                  | Dosage , g/kg | AOA, mg/100 g | Relative change in AOA % |
|--------------------------|--------------|---------------|--------------------------|
|                          |              | 1  | 30   |                        |
| Control sample           | -            | 2,78 | 2,59 | 6,8                      |
| Blueberry                | 0,25         | 4,28 | 3,15 | 26,4                     |
| Blackcurrant             | 1,00         | 6,86 | 5,89 | 14,1                     |
| Blackcurrant             | 0,50         | 4,37 | 3,34 | 23,6                     |
| Grape                    | 1,00         | 4,62 | 3,64 | 21,2                     |
| Grape                    | 0,015        | 2,72 | 1,73 | 36,4                     |

The study results (Table 4) indicate that the AOA of marmalade increases with the concentration of natural anthocyanins in the original extract and, consequently, in the food product. The more natural polyphenols the marmalade contain, the less its AOA decreases during storage. The use of natural anthocyanin concentrates from fruit and berry pomace in the formulation of jelly marmalade can increase its AOA by 1.5-2.5 times.

Table 5. Antioxidant properties (AOA, mg/100 g) of some confectionery products in the presence of natural anthocyanins

| Confectionary item     | Content of blackcurrant extract g/kg |
|------------------------|-------------------------------------|
|                        | 0        | 0,25    | 0,50 | 0,75 | 1,0 | 1,5 |
| Hard candy             | 3,2      | 4,6     | 4,6  | 6,6  | 7,2 | 12,5 |
| Pulled hard boilings   | 4,7      | 5,5     | 5,5  | 6,3  | 10,0| 12,9 |
| Jelly sweets           | 1,7      | –       | 2,1  | 2,3  | 3,5 | 5,0 |

The dependence of AOA of candies and jelly sweets on the concentration of anthocyanins in their composition is almost linear. The results of research show that the antioxidant properties of pulled hard boilings are slightly higher compared to hard candies due to the differences in production technologies.

Table 6. Antioxidant properties of some alcoholic beverages depending on the preparation recipe and shelf life

| Drinks                  | Type of antioxidant                          | AOA, mg/100 g | Relative change in AOA, % |
|-------------------------|---------------------------------------------|---------------|--------------------------|
|                         | without AO                                  | 1  | 30   |                        |
| Dessert infusion        | Carotenoids                                 | 0,79 | 2,21 | 1,68 | -24,00 |
| Bitter infusion         | Carotenoids and anthocyanins                | 1,00 | 3,40 | 2,70 | -20,60 |
| Dessert liqueur         | Carotenoids and anthocyanins                | 3,00 | 5,50 | 4,50 | -18,20 |
| Balsam                  | Carotenoids and anthocyanins                | 51,50 | 68,00 | – | – |

The use of alcoholic beverages with high AOA in the diet will also allow prolonging human life and enhancing protection against various diseases. Hydrophylic carotenoids and a mixture of...
carotenoids and anthocyanins from various plant raw materials were analyzed as natural antioxidants in the composition of alcoholic beverages. The obtained beverages are enriched with vitamins and have an increased AOA (Table. 6).

Carotenoids, in comparison with anthocyanins, possess reduced antioxidant properties because of different chemical structure of these compounds, affecting the AOA of alcoholic beverages. The increased AOA of the balsam Vostochnaya Skazka is due to the presence of infusions and fruit drinks which are made from natural raw materials containing polyphenols with high antioxidant properties in its composition.

4. Conclusion
The results of these studies show that the introduction of concentrates of natural anthocyanin and carotenoid extracts in food can increase their antioxidant properties and increase the content of A and P-vitamins.

The introduction of anthocyanins in the formulation of jelly marmalade, candies and jelly sweets helps to increases the antioxidant properties of the manufactured products by 1.5-3 times. The use of carotenoid and composite carotenoid-anthocyanin antioxidants in the production of alcoholic beverages such as infusions and liqueurs with low antioxidant activity also increases the antioxidant properties by 1.5-3 times.

References
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