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Evolution of antioxidant activity in sea buckthorn during technological treatments

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Abstract

This paper presents the study of total antioxidant activity evolution during different technological treatments of sea buckthorn. In order to assess the total antioxidant activity, the photocolorimetric method, utilizing DPPH radical, was used. Sea buckthorn was subjected to various technological treatments: freezing, freezing with prior blanching, conservation with sugar, with subsequent refrigeration and pasteurization, with the purpose to determine the stability of the antioxidant activity during these treatments and further storage. From researches it was determined that freezing is the optimal method of conservation, in terms of preserving antioxidant substances. It was also noted that blanching prior to freezing makes the preservation of antioxidant activity more efficient. Freezing juice with pulp proved to be as effective as whole frozen fruit freezing in terms of antioxidant activity's stability immediately after treatment. In the case of sea buckthorn conservation with sugar, total antioxidant activity and sensory properties were better preserved in the chilled product.

Keywords: rutin-fatty acid bioconjugates, cyclodextrins, molecular encapsulation, molecular modeling, docking

docking

1. Introduction

The problems of aging and diseases such as cancer or cardiovascular diseases, are the object of study for scientists worldwide. It was demonstrated that oxidative stress plays an important role in developing these conditions in living organisms. A studied approach to prevent destructive processes is the use of antioxidants, substances with the ability to stop the chain reactions in living cells [8], [9]. Thus, several studies show that antioxidants have a great influence on the human body. Given the destructive processes that occur after culinary treatment, it is beneficial to use products with high content of antioxidants, which are stable to technological treatments.

Antioxidants are also important ingredients of dietary supplements used to maintain health.

Although research suggests that antioxidant supplements are beneficial to health, large clinical trials have not confirmed particular benefits, but have shown instead that an excess of antioxidant supplements may sometimes have negative effects [1], which is why it would be the best for the human body to receive antioxidants from food.

Because foods with the highest content of antioxidants are vegetables and fruits, these are mainly studied. Sea buckthorn was chosen for this research. It is a plant rich in antioxidant compounds like vitamin C, carotenoids, polyphenols etc. It was proven that it can fortify the immune system and prevent senility. More than 10 different drugs were developed from sea buckthorn.

The fruit is used both in therapeutic purposes, to treat hypovitaminosis and avitaminosis, anemia and convalescence, and it can be served as food: juices, syrups, marmelades [2], [3].

Technological treatments have different influence on each antioxidant compound. During the heat treatment, some antioxidants such as vitamin C are inactivated, while others are transformed to become more active or more easily absorbed by the digestive system. There are large losses of vitamin C during boiling and blanching, when water use is no longer used. Vitamin C is unstable in the presence of oxygen [4]. Vitamin A and carotenoids are partially destroyed during thermal processing of food, the destruction being influenced by the presence of oxygen and light [5]. Culinary treatment increases the chemical extractability of carotene, thus increases its bioavailability for the human body [6]. For these reasons many researches were undertaken to determine how different antioxidants transformed during technological processes, which treatments are safer and what methods can be used in order to prevent the degradation of antioxidants.

2. Materials and methods

Initial experiments were carried out using fresh sea buckthorn in state of maturity, fruit with a high content of biologically active substances, many of which have antioxidant properties.

buckthorn subjected Sea was to various technological processes: freezing, freezing with prior blanching, conservation with sugar with subsequent refrigeration and pasteurization, juice freezing, juice pasteurization. In order to assess total antioxidant activity photocolorimetric method using DPPH radical (2,2-diphenyl-1-picrilhidrazil) was used. DPPH method is based on the reduction of DPPH radical absorbance in the presence of antioxidants. DPPH (2, 2-diphenyl-1-picrilhidrazil) is one of the most stable organic radicals which is commercially available [7]. The absorbance was measured in quartz cuvettes (10 cm), using UV-VIS Unicam spectrophotometer at 517 nm.

Sample preparation: To assess the antioxidant activity of the sea buckthorn products alcoholic extracts were used. To prepare the extract 20g of product were weighed, 35 ml of ethanol (96%) were added. Extraction lasted for 7 days.

Analytical determination: 3 ml of sample are placed in the cuvette and 1ml of DPPH solution (1 mM) is added. The blank is prepared by adding 3 ml of sample and 1 ml of ethanol in the cuvette. To prepare the control sample, 3 ml of distilled water and 1 ml DPPH solution are placed in the cuvette and mixed intensely. Distilled water served as

As standard ascorbic acid 10 % solution is used. 3 ml of ascorbic acid solution and 1 ml DPPH solution are placed in the cuvette. To prepare the blank 3 ml of ascorbic acid and 1 ml ethanol are placed in the cuvette and stirred intensely. The absorbance was recorded at 517 nm after t=30 minutes. The solution will fade over time as the DPPH \cdot react with the antioxidant compounds from the sample.

The ability to reduce DPPH· radical (2,2-diphenyl-1-picrilhidrazil) is calculated using the formula:

Capacity of reduction DPPH \cdot (%) = $\binom{A_{\text{control}} - A_{\text{control}}}{A_{\text{control}}}$ \times 100%

 $A_{control}$ absorbance of control sample

Asample absorbance of sample

3. Results and Discussion

After the investigations the data were grouped and a comparative analysis was carried out in order to determine which technological treatment is the best for sea buckthorn preservation in terms of antioxidant activity stability.

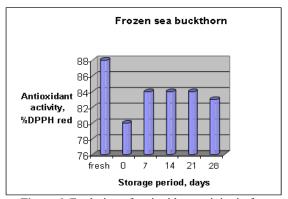


Figure 1. Evolution of antioxidant activity in frozen sea buckthorn

The analysis of frozen sea-buckthorn shows a decrease of antioxidant activity by 9% immediately after treatment. During storage total antioxidant activity remains relatively stable. Organoleptic properties of sea-buckthorn are also very well preserved and not suffer much from thawing.

Compared with the decrease of antioxidant activity in blanched sea-buckthorn, the decrease is sudden. It could be assumed that the effect is due to inhibition of oxidative enzymes during heat treatment of sea buckthorn

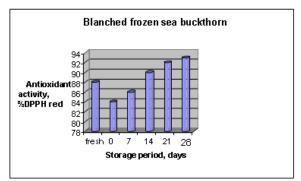


Figure 2. Evolution of antioxidant activity in blanched frozen Figure 4. Evolution of antioxidant activity during sea buckthorn

After tests conducted on following days, a slight increase in antioxidant activity can be noticed, presumably due to diffusion of antioxidant compounds in the cell.

Analysis of frozen sea buckthorn, with a prior 5 minutes blanching in hot water, showed a decrease of approximately 5% immediately after treatment and a slow increase in antioxidant activity in the first 21 days. After 21 days, the analysis showed an increase in total antioxidant activity approximately 5%. This can be explained by the effect exerted by temperature on fruit and vegetable pulp. Following the destruction of tissue the process of blanching causes the extraction of compounds from the cells. The positive effect (elimination of oxygen from plant tissues, enzyme destruction etc) of blanching before freezing, is well known and it was confirmed by the experiments using sea buckthorn.

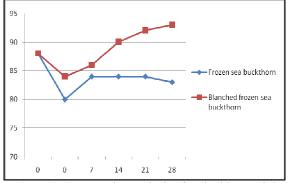
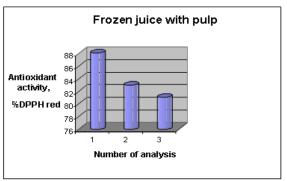


Figure 3. Comparative analysis of antioxidant activity changes in frozen and blanched frozen sea buckthorn

Comparing the two types of freezing of sea buckthorn, a decrease in antioxidant activity immediately after treatment is noted in both cases. In the case of simple freezing a slight increase after 7 days was observed, in the next period the value of total antioxidant activity remained stable.



juice pressing (1), dilution (2) and freezing (3)

The product subjected to a prior blanching showed a lower decrease immediately after treatment and an increase in the following days. Thus, it can be concluded that blanching before freezing had positive effects in terms of retention of antioxidant activity. The analysis of sea buckthorn juice with pulp, diluted twice showed a decrease of approximately 6% of antioxidant activity. After freezing, it had decreased by 2%. In the future it is necessary to observe the evolution of the antioxidant juice during frozen storage. Sensory properties of the juice with pulp are well preserved at freezing-thawing.

In the analysis of sea buckthorn juice with pulp, diluted twice, a decrease of approximately 6% of antioxidant activity was observed. After pasteurization its value had increased by nearly 2%

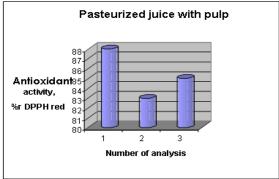


Figure 5. Evolution of antioxidant activity during juice pressing (1), dilution (2) and pasteurization (3)

This effect is presumably explained by the effect of heat treatment on carotenoids [6]. During the heat treatment the extraction of antioxidant compounds in the juice cells takes place. Temperature also has a beneficial effect on the availability of fat soluble antioxidant compounds. However heat treatment had a negative effect on sensory properties of the juice. Data obtained by other researchers also attested unwanted effects caused by thermal treatment. Heating juice for 3 minutes at 90 C caused a clear increase in juice viscosity. Due to coalescence process, oil droplets become larger. If the juice is left 1 or 2 days, it will separate into three phases.

This process is unfavorable from the standpoint of the consumer. To preserve juices HTST treatment at 80-90 C is preferred. The juice is brown in about 6 months of storage at 15-20 C. The color change may be reduced in conditions that prevent oxidation. It is unclear whether this process is due to polyphenoloxydase or Maillard reaction [2].

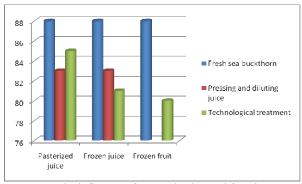


Figure 6. The influence of pasteurization and freezing on sea buckthorn products

The research has shown that the antioxidant activity in frozen juice was greater than the one in the whole frozen fruit and sensory properties were preserved much better than in the pasteurized sea buckthorn juice.

During the analysis of sea-buckthorn chilled with sugar an abrupt decrease in antioxidant activity may be observed immediately after treatment and in the first 14 days, after which it begins to grow. It is assumed that the decrease is due to decomposition of less stable antioxidant compounds, (ex. vitamin C), in the first days and the subsequent increase is due to the diffusion of more stable antioxidant compounds from the solid parts of the fruit in the sugar syrup.

Due to crushing it was expected total antioxidant activity to be higher, but it seems that the alcohol extraction was similar in all the products. Organoleptic properties of this product remained unchanged even after 28 days storage.

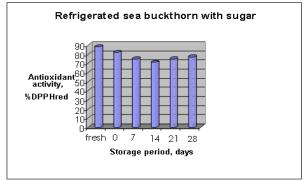


Figure 7. Evolution of antioxidant activity in refrigerated sea buckthorn with sugar

The analysis of pasteurized sea buckthorn with sugar showed that during storage antioxidant activity is slowly decreasing. A decrease of 6 % was noticed immediately after treatment.

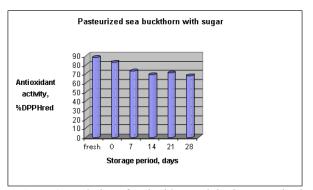


Figure 8. Evolution of antioxidant activity in pasteurized sea buckthorn with sugar

After 14 days a slight increase of antioxidant activity was observed, which can be explained by the extraction and the diffusion of compounds with antioxidant character in the sugar syrup. After 28 days of storage, antioxidant activity in pasteurized sea buckthorn has decreased by 23%. This decrease could be attributed to the action of oxygen that penetrated after opening the jar. It was observed the stability in the evolution of antioxidant activity in the next days.

However sensory properties of pasteurized sea buckthorn with sugar have degraded considerably. A slight browning and the loss of characteristic flavor were noticed.

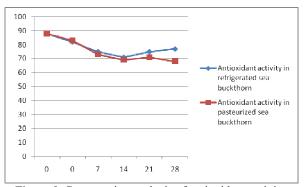


Figure 9. Comparative analysis of antioxidant activity changes in pasteurized and refrigerated sea buckthorn

In comparing the two types of preservation with sugar, one with subsequent pasteurization and another with refrigeration, it was noted that the antioxidant activity decreased immediately after treatment, keeping this trend in the first days. After 21 days an increase in total antioxidant activity in the refrigerated product was noticed, while in the pasteurized product the same downward trend applied. Also, after about a month, in the pasteurized product, adverse changes of sensory properties were noted: mild browning and disappearance of sea buckthorn flavor.

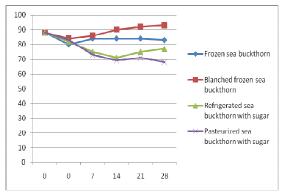


Figure 10. Comparative analysis of the influence of freezing and sugar preservation on antioxidant activity of sea buckthorn

When comparing the four types of conservation, a decrease of total antioxidant activity may be noted in all cases immediately after treatment. In the case of the pasteurized product this trend continued over the following days. The higher stability in the evolution of antioxidant activity was observed in the frozen sea buckthorn, however in the product with the prior blanching the increasing trend continued even after 28 days of storage.

It was also noted that the products preserved with sugar had suffered a greater decrease, presumably due to the addition of sugar in the fruit mass.

4. Conclusion

The research has shown that the most effective method of conservation, in terms of retention of antioxidant activity, is freezing. Blanching before freezing facilitated the retention of total antioxidant activity and improved the process of freezing.

Little difference was observed between the frozen juice and the frozen fruit. Also, sensory properties are well preserved and the juice supports better the freeze-thawing process. In the case of conservation with sugar,

In the case of conservation with sugar, refrigeration was proven to be better than pasteurization, in terms of antioxidant activity stability.

In the future it is necessary to analyze how the storage conditions influence the antioxidants and to reveal the factors which lead to the decrease of antioxidant activity in foods. Also it is necessary to analyze the evolution of each antioxidant, their transformations during treatments and storage.

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