DETERMINATION OF ANTIOXIDANT CAPACITY IN SOME FRUIT CONCENTRATES AND POWDERS BY DIFFERENT METHODS

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Abstract

There is a great demand on consumption of natural and healthy foods, which includes also fresh fruits and food products made by different technological processes. In our experiment, fruit concentrates and powders of elderberry (*Sambucus nigra* L.) and blackcurrant (*Ribes nigrum* L.) were studied. For evaluating their health protective effect, the antioxidant/ reducing properties of both fruits were obtained. The total phenolic content (TPC) and the antioxidant capacity determined by the FRAP, TEAC, and DPPH methods were analysed. Results are expressed on a dry matter basis for better comparison. TPC and antioxidant activity detected by different methods were higher in elderberry (*Sambucus nigra* L.) for both concentrates and powders than in blackcurrant (*Ribes nigrum* L.). Dry powders made from fruit concentrates showed in almost all cases a lower antioxidant activity than the concentrates. The results clearly show that it is recommended to characterize the antioxidant properties in as many ways as possible in order to evaluate the beneficial effects of fruits and their processed goods on the human organism.

Introduction

One way to increase your fruit and vegetable consumption is to produce a food that is easy to prepare, does not require a lengthy preparation, is of good quality and is available at all times of the year. Vacuum drying is one of the methods developed to meet these needs to the fullest. The low drying temperature and the short drying time used in the technology enable the production of long-lasting goods with favourable nutritional and organoleptic properties.

By choosing the right diet, we can prevent certain diseases. Eating vegetables and fruits rich in vitamins and antioxidants can reduce the risk of their occurrence because free radical reactions responsible for the onset of diseases can be delayed or inhibited by antioxidants [1,2,3,4].

The antioxidant or reducing capacity is the combined effect of all the antioxidant compounds in a system. Several methods have been developed for evaluation of antioxidant power, the number of them exceeds one hundred [5].

Methods for measuring antioxidant capacity are commonly classified into two groups, as hydrogen atom transfer (HAT)- and electron transfer (ET)-based assays [6]. Of course, all methods have their strengths and weaknesses, the processes in the body cannot be exactly tracked, only an approximate characterization and comparison of the tested samples is possible. For example, the FRAP and TPC assays work on a non-physiological pH (pH 3.6 and pH 10, respectively) [7], and the DPPH and TEAC methods use free radicals that do not occur in the body [8,9].

Experimental

All chemicals for the experiments were purchased from Sigma Aldrich.

Sample preparation

Samples were taken from filtered frozen (-18°C) concentrates of elderberry (*Sambucus nigra* L.) and blackcurrant (*Ribes nigrum* L.).

The powders of both fruits were produced in a tray-type LMIM LP-405 vacuum oven with three parallel tray drying (temperature detection per tray) at a pressure range of 20 mbar to atmospheric pressure, at 10 °C to 60 °C for 240 minutes after a short warm-up time.

The dry material was crushed using a coffee grinder (Delonghi KG49), and samples were stored in sealed polyethylene bags at -32 °C until measurement.

Both fruit concentrates and lyophilized samples (diluted with distilled water) were analysed for analytical measurements. The diluted aqueous solutions were placed in a cooled ultrasonic water bath for 30 minutes. The samples were then centrifuged at 13,500 rpm at 10 ° C for 15 minutes. In all cases, the analytical tests were carried out on the supernatants.

Electron transfer methods detect the reducing capacity of solutions. The reactions are followed by colour changes, which can be monitored spectrophotometrically (Thermo Scientific –Evolution 300 UV-VIS spectrophotometer).

Analytical methods

Determination of total phenolic contents (TPC) by Folin-Ciocalteu method: The Folin-Ciocalteu spectrophotometric method by Singleton and Rossi [10], at 760 nm is an electron transfer based assay and shows the reducing capacity, which is expressed as phenolic content. Gallic acid (GA) was used to prepare the standard curve. The results were expressed as mM GA/g of dry matter (DM).

Determination of antioxidant capacities by FRAP (Ferric Reducing Antioxidant Power) method: Measurement of ferric reducing antioxidant power of the fruit extracts was carried out based on the procedure of Benzie and Strain [11], at 593 nm. Ascorbic acid (AA) was used as a standard to prepare the calibration solutions. Results were expressed as μMAA/g DM.

Determination of antioxidant capacities by TEAC (Trolox Equivalent Antioxidant Capacity) method: Miller and coworkers [12] described the Trolox equivalent antioxidant capacity (TEAC) method. The assay is based on formation of the ABTS•+ cation [2,2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid)] and its scavenging by antioxidant sample constituents measured by spectrophotometry at 743 nm (decay of green/blue chromophore absorbance is inversely associated with antioxidant sample content). Trolox, a hydrophilic vitamin E analog, was used as a standard and values were expressed as mM TE (Trolox equivalent)/g DM.

Determination of antioxidant capacities by DPPH (2,2-Diphenyl-1-pikrylhydrazyl) radical-scavenging activity method: The spectrophotometric method (517 nm) of DPPH radical-scavenging activity was carried out according to the method of Blois [13] and modifications by Hatano and others [14]. The results were expressed as mM of TE (Trolox equivalent)/g DM.

In the last two methods, artificial radicals were used that do not occur in the living organism [6], and in the case of the DPPH method the anthocyanins interfere in the measurement [7].

Results and discussion

Figure 1.A represents data for the total polyphenolic content of elderberry and blackcurrant samples. It can be clearly seen that the TPC of the elderberry concentrate was extremely high (708.0 mM GA/g DM). For the blackcurrant concentrate, 415.7 mM GA/g DM was measured. Comparing the obtained results with literature data [9,15] it can be said that they are almost identical. Vacuum drying resulted in a decrease in the TPC, the loss of phenolics was 7.5% for elderberry and 18.0% for blackcurrant.

The antioxidant capacity measured by the FRAP method (Fig. 1.B) showed similar correlations. The elderberry concentrate had an outstanding antioxidant power (522.8 µMAA/ g DM), followed by blackcurrant concentrate (369.6 µMAA/ g DM). The decrease on antioxidant capacity after vacuum drying was 9.7% in the case of elderberry, while very high in blackcurrant (32.4%).

The antioxidant activity determined by the TEAC method (Fig. 1.C), was significant higher for elderberry concentrate and powder than for blackcurrant samples. Vacuum drying resulted in an increase in radical scavenging properties for both product types, which may be explained by a possible change in the endogenous components.

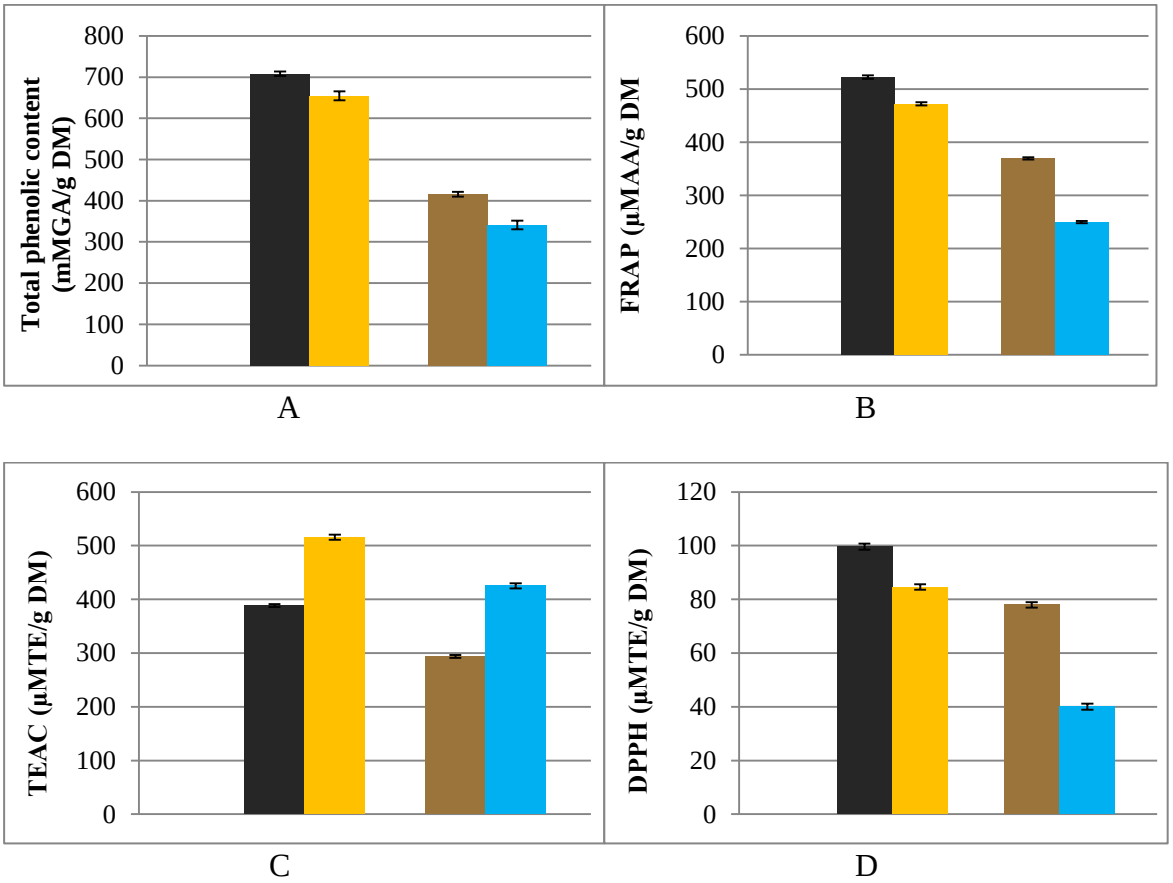

Figure 1. The antioxidant capacity measured by different methods
A) total phenol content, B) FRAP, C) TEAC and D) DPPH

legend:
elderberry concentrate elderberry powder black currant concentrate
black currant powder

In the case of DPPH method (Fig. 1.D), the elderberry concentrate showed the highest antioxidant capacity (99.6 µM TE/ g DM), followed by blackcurrant concentrate (78.0 µM

TE/ g DM). Vacuum drying caused a significant decrease of the antioxidant power, a decline of 15.1% was found for elderberry and 48.6% for blackcurrant.

Conclusion

Total phenolic content and antioxidant capacity determined by different methods (FRAP, TEAC, DPPH) were higher in elderberry (*Sambucus nigra* L.) than in blackcurrant (*Ribes nigrum* L.). Comparing fruit concentrates to powders made from them, significant differences could be found. The concentrates had a higher TPC and antioxidant activity measured by FRAP and DPPH methods than the powders, however, using the TEAC assay just the opposite was obtained for both fruits. These results clearly show that more methods are needed to determinate the antioxidant status of the fruit samples, and to characterize the health protective effect of different food products.

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