COMPARISON OF TOTAL POLYPHENOL CONTENT AND ANTIOXIDANT CAPACITIES OF FRUIT AND VEGETABLE JUICE CONCENTRATES MEASURED BY DIFFERENT METHODS

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Abstract

In our research, the total polyphenol content and antioxidant capacity of 6 types of 65% fruit juice concentrates (rosehip, sour cherry, sea buckthorn, beetroot, jerusalem artichoke, papaya) were measured and compared with TEAC and FRAP methods. Despite the fact that the highest TPC value was measured for sour cherries and the second highest for rosehips, the highest antioxidant capacity was measured not for sour cherries but for rosehips, using both (FRAP and TEAC) antioxidant capacity measurement methods. The antioxidant capacity of sour cherries ranks second after rosehips in the case of the TEAC measurement method, while in the FRAP method, it ranks only third in the order of the 6 samples examined. The third highest TPC value was measured in sea buckthorn, whose antioxidant capacity measured by the TEAC method - similarly - is also in third place, but measured by the FRAP method, ahead of cherries, it ranks second. The values of beetroot, Jerusalem artichoke and papaya juice concentrates were in exactly the same (4-5-6th) position in all three measurements (TPC, TEAC, FRAP). Based on our results, it can be stated that the results measured with different antioxidant capacity methods are not comparable with each other, only the values measured by the same method can be compared. Rather, we can interpret the results obtained in different methods as complementary, which point out the different nutritional properties of each plant and their unique complexity.

Introduction

More and more publications worldwide confirm the health-damaging effects of free radicals and the health-protecting effects of antioxidants, therefore there is an increasing demand for more and more accurate determination of the antioxidant capacity of various foods in order to consume the required amount of antioxidants [1] regularly. Several studies have confirmed the excellent health-protecting effects of rosehip [2], sour cherry[3], sea buckthorn [4], beetroot [5], jerusalem artichoke [6], papaya [7], which are due to their outstanding but significantly different nutritional values.

Antioxidant capacity can be defined as the combined effect of all antioxidant compounds in the examined sample. Over a hundred methods have been developed in recent decades to measure antioxidant capacity [8], but every examination method has advantages and disadvantages. Neither method is suitable for accurately modelling the biochemical processes taking place in the body on its own, so it is of paramount importance to formulate a conclusion about the sample based on the combined results of several test methods.

The measurement methods developed so far can be divided into 2 main groups [9]: hydrogen atom transition (HAT: Hydrogen Atom Transfer) and electron transition (ET: Electron Transfer). HAT methods are primarily based on reaction kinetics: they measure how effective the sample is against a given free radical and determine its free radical scavenging capacity [10]. In the case of ET methods, antioxidant capacity can be inferred from the degree of colour change during the reaction [11]. The antioxidant capacity results determined by the HAT and ET methods are not necessarily correlated since the reducing capacity of a sample is not necessarily related to its radical scavenging capacity [11]. The most commonly used methods worldwide mainly belong to the ET group due to their simplicity, speed and low cost. The TPC, FRAP, and TEAC methods used in our present studies also belong to the ET group.

Materials

In our experiment, 4 types of fruit juice concentrate with a dry matter content of 65% (belonging to botanically different families) were used: rosehip (*Rosa canina),* sour cherry (*Prunus cerasus),* sea buckthorn (*Hippophae rhamnoides),* Papaya *(Carica papaya)* and 2 types of root-vegetable juice concentrates beetroot (*Beta vulgaris*) and jerusalem artichoke (*Helianthus tuberosus).* All juice concentrates were in accordance with food safety rules, strictly controlled, produced under the HACCP quality assurance system, stored and distributed in an aseptic manner (distributed by Intercooperation Ltd.). The chemicals used were purchased from Sigma-Aldrich.

Analytical methods

Determination of total polyphenol content (TPC) by Folin-Ciocalteu method: The Folin-Ciocalteu spectrophotometric method by Singleton and Rossi [12], at 760 nm is an electron transfer based on 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 µM 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 juice concentrates was carried out based on Benzie and Strain's procedure [13], 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: The total antioxidant capacity was measured with Trolox-equivalent antioxidant capacity (TEAC) method described by Miller et al. at 734 nm [14]. The method is based on ABTS+ free radical scavenging by antioxidants measured with a spectrophotometer. For the calibration Trolox (the hydrophilic analogue of vitamin E) was used, and results were expressed in µMtrolox equivalent/g DM.

Results and discussion

I.) Results of total polyphenol content (TPC) by Folin-Ciocalteu method

The results of the measurements of the 6 types of concentrated juice showed that the total polyphenolic content of sour cherry juice concentrate was the highest (76,2 µMGS/g), and papaya was the lowest (14,9 µMGS/g). Sour cherries are followed by rosehips (69 µMGS/g), sea buckthorn (58.5 µMGS/g) and beetroot (57.3 µMGS/g), the latter two with only a slight difference. It should be noted that the first 4 TPC values are in similar ranges, with a maximum difference of only 18.9 µMGS/g. But Jerusalem artichoke (20,9 µMGS/g) and papaya (14,9 µMGS/g), which have a much lower TPC value, are significantly separated from the first 4 types of samples (Fig. 1).

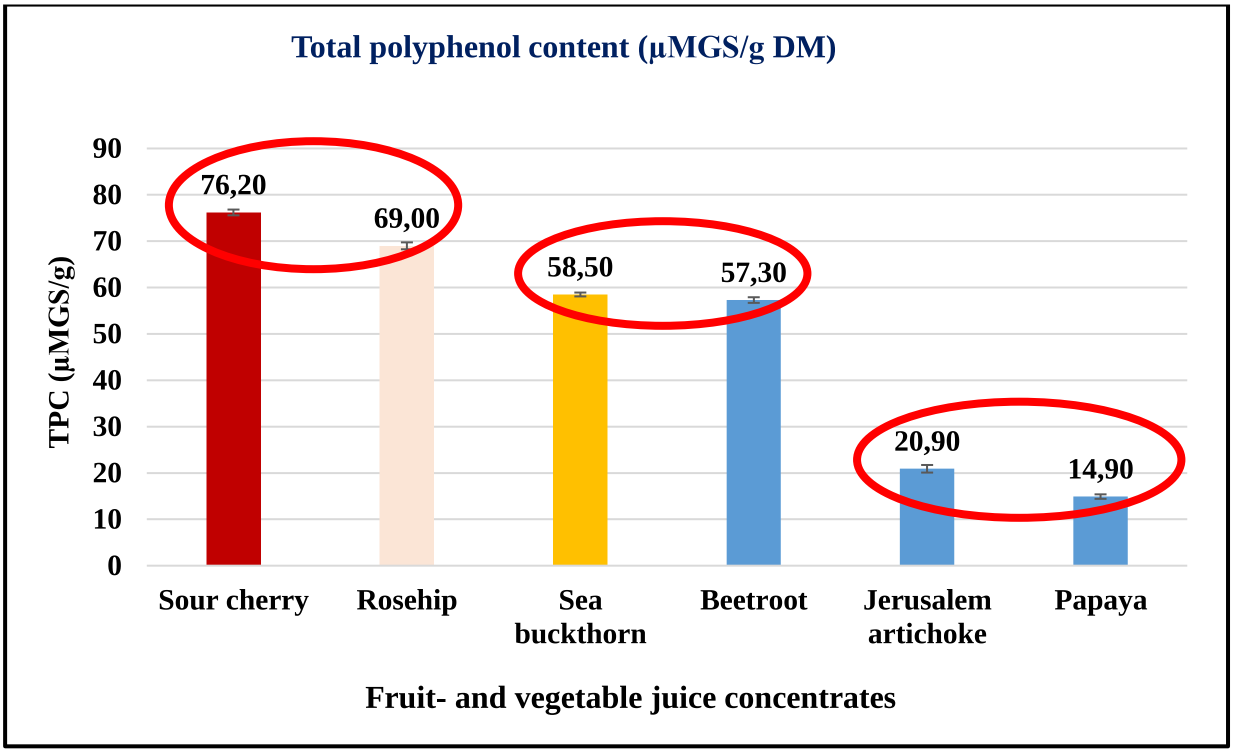

Fig. 1. Total phenolic content of different juice concentrates (μMGA/g DM)

II.) Antioxidant capacity measurement results using TEAC and FRAP methods

In our research, we investigated the antioxidant capacity of 6 types of 65% juice concentrates using the TEAC method based on free radical scavenging capacity (Fig.2.) and the FRAP method based on iron-reducing ability (Fig. 3). Our results were as follows:

1.) For both measurement methods (FRAP, TEAC), outstanding antioxidant capacity values were measured in the rosehip juice concentrate. The antioxidant capacity of the other 5 types of juice concentrate was so significantly separated from rosehip in both methods that a separate (smaller) figure had to be made within the large figure for better illustration (Fig.2-3).
2.) With the TEAC measurement method, the sour cherry (188.59 μMtrolox/g) is in second place, followed by the sea buckthorn (160.56 μMtrolox/g) in third place by a small margin.
3.) At the same time, in the FRAP measurement method, the order of the same 2 concentrates is reversed, and it should be emphasized that here they are not in the same range (as in the case of TEAC), but the sea buckthorn (481.78 μMAS/g) shows a significant difference, more than 10 times higher FRAP value than sour cherries (43.37 μMAS/g).
4.) The order of beetroot, Jerusalem artichoke, and papaya is the same for TPC, TEAC and FRAP measurements, so these have been marked with the same (blue) colour.
5.) In TEAC measurement, Jerusalem artichoke (20.57 μMtrolox/g) and papaya (19.53 μMtrolox/g) show almost similar values, from which beetroot is separated by a significant margin, more than 5 times higher (108.94 μMtrolox/g).
6.) In contrast, in the FRAP measurement method, beetroot (37.8 μMAS/g) and sour cherries (43.37 μMAS/g) show similar antioxidant capacity values, compared to which Jerusalem artichoke (10.17 μMAS/g) and papaya (4.85 μMAS/g) are almost 4 times lower.

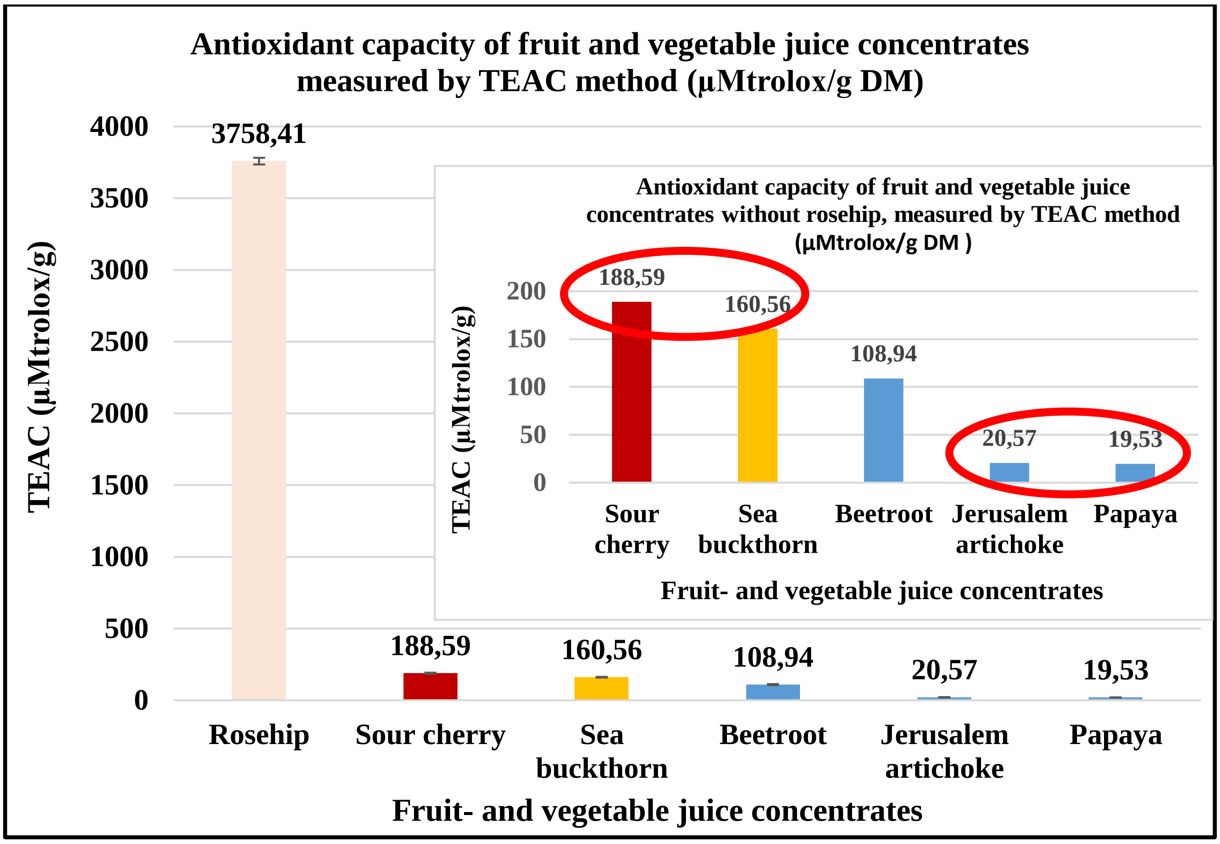

Figure 2. Antioxidant capacity of 6 types of fruit and vegetable juice concentrates measured by TEAC method (µMtrolox/g DM)

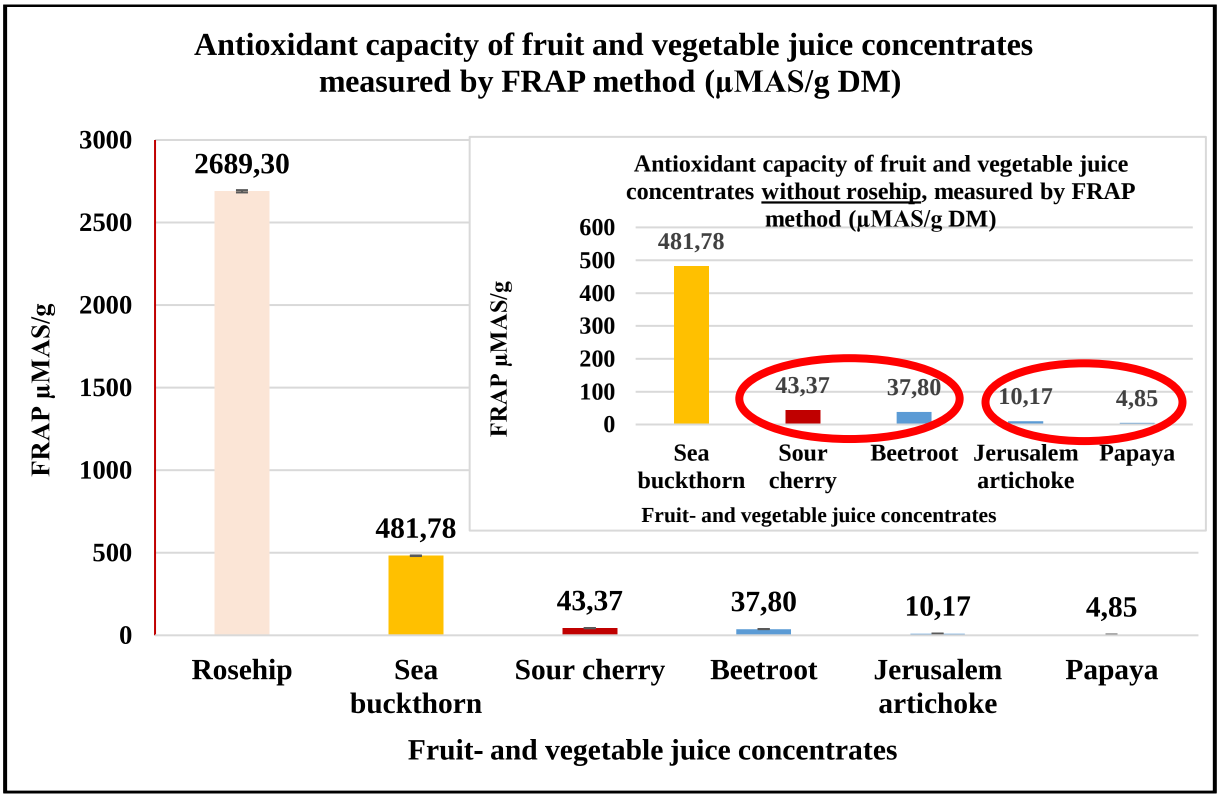

Figure-3. Antioxidant capacity of fruit and vegetable juice concentrates measured by FRAP method (µMAS/g DM)

III.) Sequences due to differences in methods and relationship between total polyphenol content and antioxidant capacity values measured by 2 different methods (FRAP, TEAC)

Despite the fact that the highest TPC value was measured for sour cherries and the second highest for rosehips, the highest antioxidant capacity was measured not for sour cherries but for rosehips using both (FRAP and TEAC) antioxidant capacity measurement methods. Sour cherries came second with TEAC measurement, while FRAP only came third (Table 1). The TPC value of sea buckthorn came in third, yet a higher antioxidant capacity was measured with the FRAP method than with cherries with the best TPC value.

Table-1. The order of the 6 types of concentrated juice in the results of TPC, TEAC, FRAP tests

	Sequence					
Methods	**1**	**2**	**3**	**4**	**5**	**6**
TPC	**Sour cherry**	**Rosehip**	**Sea buckthorn**	**Beetroot**	**Jerusalem artichoke**	**papaya**
FRAP	**Rosehip**	**Sea buckthorn**	**Sour cherry**	**Beetroot**	**Jerusalem artichoke**	**papaya**
TEAC	**Rosehip**	**Sour cherry**	**Sea buckthorn**	**Beetroot**	**Jerusalem artichoke**	**papaya**

Conclusion

Our results draw attention to the fact that there may be very significant differences between different antioxidant capacity measurement methods; therefore, it is not possible to draw far-reaching conclusions based on the test results of individual vegetables, fruits and their juice concentrates using only a few measurement methods.

In the case of 3 concentrates (beetroot, jerusalem artichoke, papaya), we saw a correlation between TPC and antioxidant capacity measured by 2 methods. At the same time, the antioxidant capacity values of the 2 fruits with the highest total polyphenol content (sour cherry and rosehip) were not correlated with their TPC values and their order.

These results suggest that, in addition to TPC, several other biologically active components may play a significant role in the antioxidant capacity of fruits and vegetables (such as the significant vitamin C content in rosehip), which requires further research.

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