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Antioxidant activity of some fresh vegetables and fruits juices

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Abstract

This paper presents the determination of some vegetables and fruits juices antioxidant activity, using the free radical 2,2- diphenyl- 1-picrylhydrazyl (DPPH) method, and correlation of the results with the C vitamin concentration in the sample. It was determined, by the iodometric method, the vitamin C concentration both in the raw materials (apple, beetroot, red cabbage, tomatoes and pink grapefruit) and in the juices obtained from this. It was observed that vitamin C concentration in the juices are higher than in the raw materials, the richest in ascorbic acid being the pink grapefruit juice (81,612 mg/100ml), followed by the beetroot juice (68.014 mg/100ml). The lowest vitamin C concentration was found in the apple juice (20,401 mg/100ml). Analysing the obtained results it was conclude that between antioxidant activity and juices vitamin C concentration exists a direct correlation..

Keywords: vitamin C, antioxidant activity, DPPH, beetroot, red cabbage, apple, tomatoes, pink grapefruit

1. Introduction

Now, in the new millenium, the life conditions determine that the food biologically active substances intake be more important than energy intake. Many diseases specific to the current civilisation, ever-decreasing immunity, have determined the development of a new therapeutic concept, consisting in food-drug use for different diseases prevention or treatment [1-3].

Plants contain high concentrations of numerous redox-active antioxidants, such as polyphenols, carotenoids, tocopherols, glutathione, ascorbic acid and enzymes with antioxidant activity, which fight against hazardous oxidative damage of plant cell components. In animal cells, antioxidant production is much more limited and oxidative damage is involved in the pathogenesis of most chronic degenerative diseases (including cancer and heart diseases) and aging [4-7].

Therefore, plant-sourced food antioxidants like vitamin C, vitamin E, carotenes, phenolic acids, phytates and phytoestrogenes have been recognized as having the potential to reduce disease risk.

Within a juices diet, the amount of potassium inserted is about 6 times more than that in traditional food. Following, the vegetabe juices act favorably on the cardiac muscle whole activity, enhances the labor of heart and have salutary effects in the cardiovasculare diseases. In this regard the fruit juices act better than food without salt [8,9].

Mineral substances, especially the alkali ions, have an alkalising effect, neutralizing the hydrochloric acid from the stomach mucosa. This action, complementary to the pectic substances, makes that fruit juices to be indicate for the hyperacidity control, in the case of digestive diseases.

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First, absence of fat, and the large amount of sugars and vitmanins, on the other hand, make of fruit and vegetable juices valuable adjuvants in liver and gall bladder diseases treatment. Due to the low nitrogenous substances content and of alkalizing action, juices are indicated in acute and chronic kidney diseases, especially in those that begins by retaining albumine, are recommended and for pregnant women [1,9].

Fruit and vegetable juices also prevent the formation of kidney and gall stones, due to their potassium salts richness. Are recommended in acidosis, diabetes, undernutrition, gout, aging tissues. Recent scientific studies have also claimed that the antioxidants found in most fruits and vegetable juices can help lower a person's risks of developing Alzheimer's disease [10].

Vegetable juices, especially those that are low in calories, are affective in obesity treatment, because it attenuates hunger and allows water excess remove, due to the potassium salts [11].

In the infants and children foods, fruit and vegetable juices provides to the body the needs of mineral substances and vitamins [12].

The fruit or vegetable juices with pulp are very important, thase being richer in nutrients, cellulose, pectic substances, than the clear juices.

In the fruit and vegetable juices are found relatively high amounts of isothiocyanate, phenolic antioxidants, indoles and flavones that have a proven anti-cancer action. Epidemiological studies has consistently linked abundant consumption of fruits and vegetables to a reduction of the risk of developing several cancers. The mechanisms responsible for this chemopreventive effect still remain largely unknown but is likely related to the presence of phytochemicals associated with fruits and vegetables [13].

Complex therapeutic effects of fruit and vegetables are correlated with the presence of plant polyphenols, particularly bioflavonoids with different structures and complex biological functions. Thus, of the over 500 known flavonoid compounds, 150 have biological activity, and act as vitamin P, with protective effect in cardiovascular diseases, with antioxidant, antiinflammatory and antitumor action [14].

Many analytical methods have been used for antioxidant monitoring, for instance phenolics in fruit have been monitored by HPLC [15,16] or colorimetrically using the Folin Ciocalteu reagent [17]. The total antioxidant capacity of foods and plant extracts has been assessed by using spectrophotometric methods with DPPH· (2,2-diphenyl-1-picrylhydrazyl) [18-20], ABTS+· (2,2'-azinobis(3-ethylbenzothiazo-line- 6-sulfonic acid)) [18,19] or Fe3+-TPTZ (2,4,6-tripyridyl-*s*-triazine) [21].

The purpose of this paper is to analyze comparatively several types of fruit and vegetable juices, respecting the antioxidante activity (by the DPPH method) and C vitamin content.

.2. Materials and methods

It was used raw materials from the domestic market. Juices were obtained from fresh raw materials undergo operations of scraping, squeezing and filtering. For each samples of raw material and juice it was determinated the vitamin C content and for each juice sample was eavaluated the antioxidant activity using the free radical DPPH method.

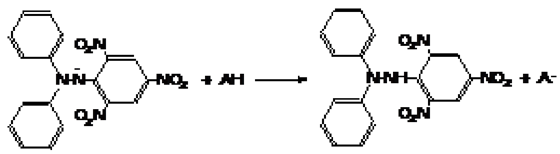
Determination of vitamin C. In order to determine the C vitamin content, it was used the adapted iodometric method. The method principle is color reaction between starch and KI + I solution. Determination of C vitamin is made with the aid of $K_2Cr_2O_7$ in the KI-starch presence. Initially, C vitamin is oxidized and then the following reaction takes place:

$$Cr_2O_7^{2-} + 6\,I^- + 14\,H^+ = 3\,I_2 + 2\,Cr^{3+} + 7\,H_20$$

Thus liberated iodine stains starch blue. Was weighed a sample on analytical balance, then was brought quantitatively into a titration vessel, were added 10 ml of 2n hydrochloric acid, diluted to 50 ml with distilled water, triturated, added 1 ml of 1% starch solution (freshly prepared) and 1 ml 0.1 N potassium iodide, after which the solution was titrated with 0.1 N potassium dichromate until persistent blue color. Quantification of vitamin C content was done according to the relation: 1 ml 0.1 N potassium dichromate is equivalent to 0.008806 g vitamin [22]. All determinations were performed in triplicate, calculating their arithmetic mean.

*Antioxidant activity determination.*DPPH (2,2-diphenyl-1-pycrylhydrazyl) is one of the most stable organic nitrogen radicals and commercially available and has maximum UV-VIS absorbance at 517 nm, giving a bluish color.

In time of reduction process, solution fade from purple to yellow, the DPPH free electron mating with an antiradical antioxidant hidrogen, passing in reduced DPPH-H:



DPPH purple | DPPH-H yellow

Unfolding reaction was monitored spectrophotometrically [23].

It was determined the antioxidant activity of pink grapefruit, apple, beetroot, red cabbage and tomatoes natural juices (clear, without pulp).

Materials used were:

- solution 1mM DPPH (2,2-diphenyl-1-pycrylhydrazyl – free radical, MP Biomedicals, LLC-Germany, in ethanol);
- ethanol 96% (Merck).

For each analyzed juice were taken samples which were diluted with distilled water 1:100, because at lower dilutions happening instantly fading samples. In an stoppered tube were introduced: 0,3 ml diluted sample, 2,6 ml solvent (ethanol) and then 0,3 ml DPPH 1mM solution, after wich this blend was introduced in the spectrophotometer cuvette, for compensation using ethanol or acetone. The absorbance at 517 nm and its variation over time was recorded. The records were made with the Perkin Elmer, Lambda 25 spectrophotometer.

Based on VIS absorption spectra of the different concentration DPPH solutions (figure 1.) was obtained calibration curve: *Absorbance* (517 nm) = *f* (*c*, mM) (figure 2.).

To quantify the antioxidant activity, slope was calculated for solutions of DPPH and reaction rate was determined as the ratio between DPPH concentration derived as a function of time, according to equation:

$$v = \Delta c/\Delta t$$

If the reaction rate is higher, the better antioxidant capacity.

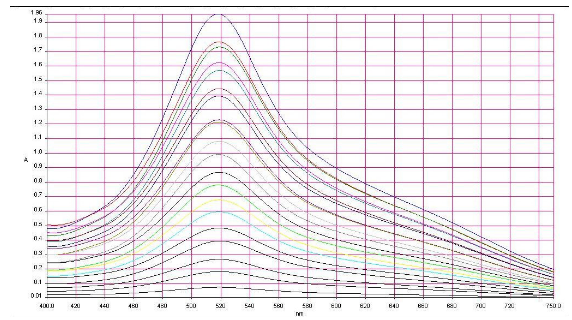


Figure 1. DPPH standard solution VIS spectra

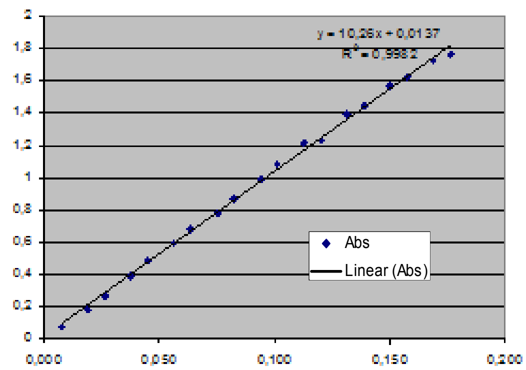


Figure 2. DPPH calibration curve

3. Results and discussions

Vitamin C level. In the table 1. and 2. (figures 3. and 4.) are the results on vitamin C content of juices and raw materials.

Table.1. Vitamin C content of raw materials

Sample	Vitamin C content (mg /100g)
Apple („Golden auriu”)	7,016
Pink grapefruit	35,359
Beetroot	33,840
Red cabbage	23,706
Tomatoes	10,201

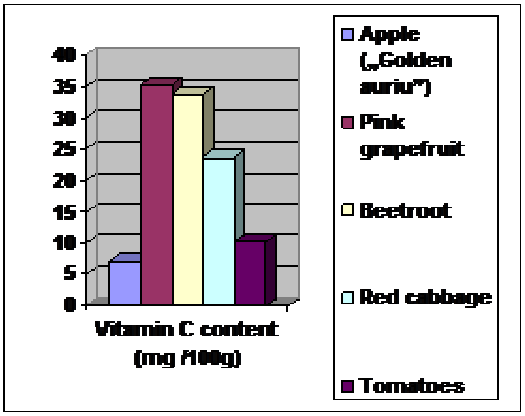


Figure 3. Vitamin C content of raw materials

Between fruits and vegetables examined, the highest vitamin C content was found in the pink grapefruit (35,359 mg/100g), followed by the beetroot (33,840 mg/100g) and red cabage (23,706 mg/100g), the lowest vitamin C content was in apple (7,016 mg/100g). This results are well corelated with the literature data [24-26].

Table 2. Vitamin C content of juices

Sample	Vitamin C (mg/100ml suc)
Apple juice	20,4
Pink grapefruit juice	81,6
Beetroot juice	68,0
Red cabbage juice	60,4
Tomatoes juice	23,3

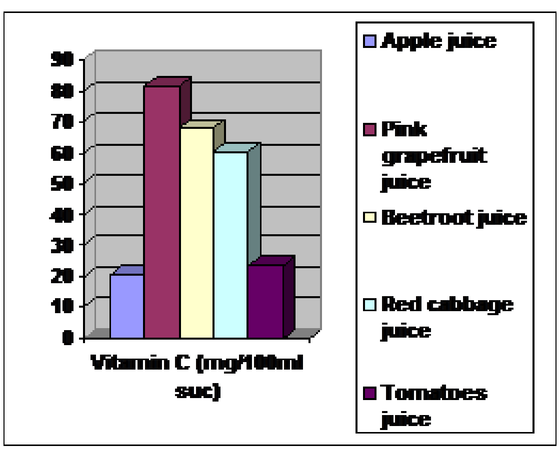


Figure 4.Vitamin C content in juices

There is a higher concentration of vitamin C in juices than in raw materials, the richest in ascorbic acid being pink grapefruit juice (81,612 mg/100ml), folowed by the beetroot juice (68.014 mg/100ml).

The lowest value of the vitamin C concentration was obtained for apple juice (20,401 mg/100ml).

Antioxidant activity. The results concerning antioxidant activity of the samples, expressed by the average reaction speed of DPPH reaction, in μM/s, are presented in the table 3 (figure 5.). Values were calculated using calibration DPPH curve pentru DPPH and Microsoft Excel programme, on basis of the spectrophotometric curves recorded on for each sample (figures 6-10).

Table 3. Average reaction speed of DPPH in the presence of the analized sample

Proba	Diluția	v (μM/s)
Suc sfeclă roșie	1:100	5,087
Suc varză roșie	1:100	5,040
Suc tomate	1:100	3,154
Suc grapefruit	1:100	5,235
Suc mere	1:100	3,133

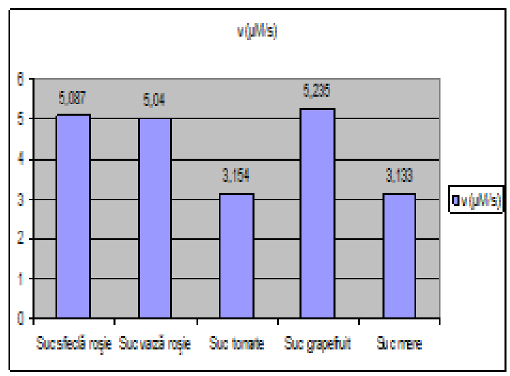


Figure 5. Average reaction speed of DPPH in the presence of the analized sample

The greater reaction speed of DPPH was in the presence of pink grapefruit juice (5,235 μM/s), that can appreciate such as having the strongest antioxidant activity of all samples analyzed. This is followed, in descending order, beetroot juice (v= 5,087 μM/s), red cabbage juice (v=5,040 μM/s), tomatoes juice (v=3,154 μM/s) and apple juice (v=3,133 μM/s).

Correlating results concerning the antioxidant activity with those concerning the vitamin C content, can be seen as the antioxidant activity varies in direct proportion to the vitamin C concentration, for all the analysed samples.

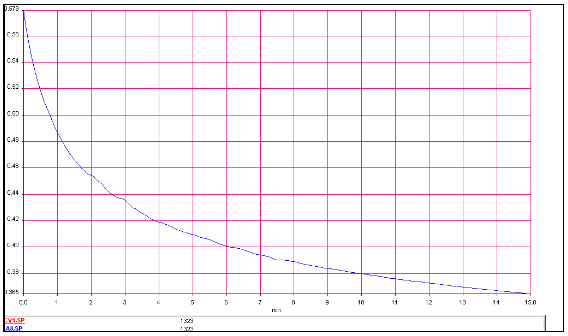


Figure 6. Time variation of the DPPH 1mM solution in the presence of pink grapefruit juice diluted 1:100

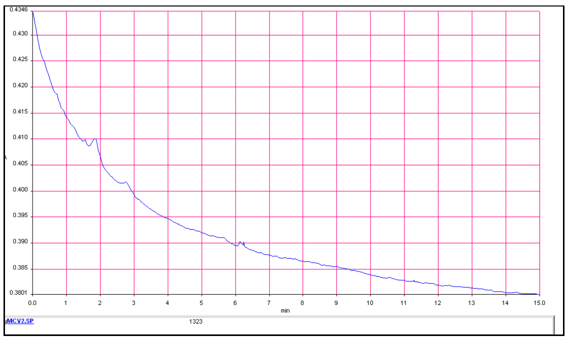


Figure 7. Time variation of the DPPH 1mM solution in the presence of apple juice diluted 1:100

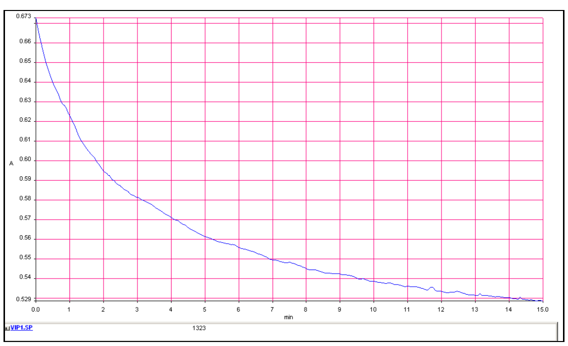


Figure 8. Time variation of the DPPH 1mM solution in the presence of beetroot juice diluted 1:100

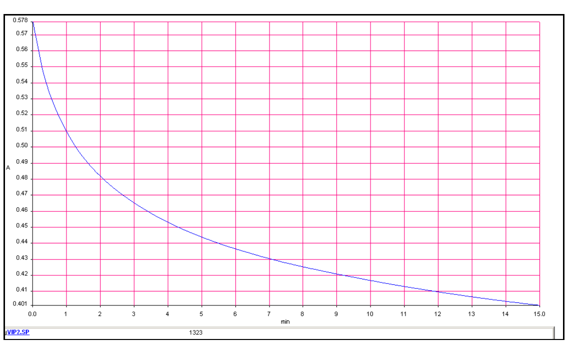


Figure 9. Time variation of the DPPH 1mM solution in the presence of red cabbage juice diluted 1:100

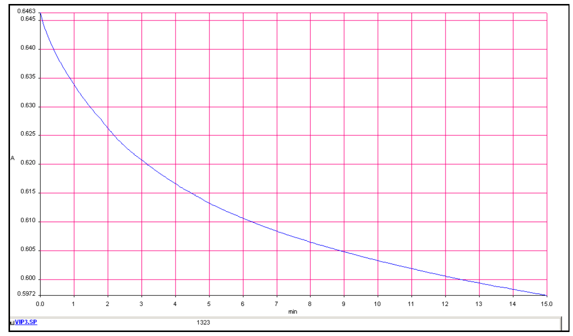


Figure 10. Time variation of the DPPH 1mM solution in the presence of tomatoes juice diluted 1:100

4. Conclusions

This paper aimed to determine the comparative antioxidant activity (using the DPPH free- radical method) of some natural, fresh vegetable and fruit juices (pink grapefruit, apple, beetroot, red cabbage, tomatoes). Knowing that vitamin C is one of the most powerful natural antioxidants we try to establish a correlation between antioxidant activity and vitamin C content of the analysed juices. The results of experiments may be summarized as:

- Between fruits and vegetables examined, the highest content of vitamin C is present in pink grapefruit, followed by the beetroot and red cabbage and the lowest vitamin C content is found in apple.
- Vitamin C concentration is higher in the juices than in the raw materials, the richest in ascorbic acid being pink grapefruit juice, followed by the beetroot juice. The lowest concentration of vitamin C was obtained for apple juice.
- For all the analysed samples the antioxidant activity varies in direct proportion with the vitamin C concentration.

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