Evaluation of antioxidant activity, polyphenols and vitamin C content of some exotic fruits

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Abstract
The aim of this work was to determine the antioxidant activity (by CUPRAC method), polyphenol content (Folin-Ciocâlteu method) and ascorbic acid (iodometric method) of some exotic fruits: pineapple, kiwi and pomegranate, purchased on the local market. The highest antioxidant capacity had the pomegranate sample (0.290 mM Trolox/l) - even diluted 1:2, compared to other samples, followed by kiwi sample (0.115 mM Trolox/l). The lowest antioxidant activity was recorded in the pineapple sample (0.109 mM Trolox/l). In terms of total polyphenols content, the sample with the highest concentration was pomegranate (0.129 mg gallic acid/l) - diluted 1:2, followed by pineapple (0.072 mg gallic aci /l). The lowest polyphenol content was found in kiwi fruit. The highest content of vitamin C was found in kiwi (91.20 mg/100g), followed by pineapple (45.30 mg/100g).

Keywords: pineapple, kiwi, pomegranate, antioxidant acticity, CUPRAC method, vitamin C

1. Introduction
Exotic fruits are that which are not native and that are cultivated outside, available at their place of origin. Among the exotic fruit in Romania, they are part and pineapple (Ananas comosus L.), kiwi (Actinidia chinensis L.) and pomegranate (Punica granatum L.). It is known that these fruits are rich in antioxidants and other important phytonutrients for human health [1-3]. In recent decades, growing more and more interest to researchers on the role of antioxidants on human health, also appeared a lot of studies related to the type and effects of antioxidants from fruits and vegetables [4-6].

We know that antioxidants are the substances able to prevent or inhibit oxidation processes in the human body and also in food products. Almost all edible plant products contain antioxidants. Among antioxidants, polyphenols are the most numerous group and they are present in fruits and vegetables, in their products, leguminous plants, grains, teas, herbs, spices and wines [6-10]. The antioxidant properties of polyphenols are mainly due to their redox properties, which allow them to act as reducing agents, hydrogen donors and singlet oxygen quenchers [11]. As antioxidants, polyphenols prevent oxidation of vitamin C and adrenaline from enzymes containing copper while enhancing effect of vitamin C [7].

Vitamin C (ascorbic acid) is one of the most important vitamin that exist in fruits and vegetables [12]. Vitamin C may protect cell membranes against lipoperoxidation through two mechanisms: (1) directly by intercepting free radicals formed in the aqueous cytosol and (2) indirectly through participation in the regeneration of vitamin E [7].
The aim of this scientific paper was to evaluate the antioxidant activity (using CUPRAC method) and also polyphenol (Folin- Ciocâlteu method) and vitamin C content in some exotic fruits (kiwi, pineapple, pomegranate) from the Romanian market.

2. Materials and methods

Fruits (kiwi, pineapple, pomegranate) were purchased on the domestic market being analyzed fresh. To determine the antioxidant activity and polyphenols content, from each type of fruit were weighed 2 g and then was comminuted, subsequently subjecting it to extraction with 20 ml 20% ethanol for 2 hours. The filtered samples were subsequently subjected to analysis.

2.1. Determination of antioxidant activity

To determine antioxidant activity of samples was used CUPRAC method that uses as reference substance reagent Trolox - an antioxidant that mimics the structure of vitamin E, but is both fat-soluble and water-soluble [13].

The reagents used in the analysis by this method were: 0.01 M CuCl₂ solution, neocuproine alcoholic solution of 7.5 \times 10^{-3} M, ammonium acetate buffer solution.

Was mixed 1 ml cuprous solution with 1 ml neocuproine alcoholic solution and 1 ml of acetate buffer. Over this solution was added 1.1 mL solution containing standard or sample and stirred well. After 30 minutes, was determined the absorbance at 450 nm, using ethanol as reference substance. The molar absorption coefficient for Trolox in the method CUPRAC is: \( \varepsilon = 1.67 \times 10^4 \) l·mol⁻¹·cm⁻¹.

The results were expressed in nmol Trolox/g dry matter.

2.2. Determination of polyphenols content

To determine the total polyphenol content was used Folin-Ciocâlteu method based on the reducing properties of the phenols to hexavalent molybdenum from the poly-phosphomolybdate [14,15].

The reagents used were as follows: Folin-Ciocâlteu reagent (FC) 2M diluted 1:10 and 7.5% sodium carbonate solution.

Preparation of the calibration curve and the samples was done by mixing 2.5 ml reagent FC diluted 1:10 with 0.5 ml of sample or standard solution of concentration: 0.4- 0.6- 0.8- 0.2- 1.0- 1.2 μM/ml gallic acid. After 10 minutes (time needed to completion of the reduction reaction) were added 2 ml of 7.5% sodium carbonate solution for neutralization and alkalinisation of the reaction medium and the formation of reduced polyphosphomolybdates, colored blue. After approx. 2 hours the absorbance was read at 750 nm. The concentration of polyphenols it was expressed as gallic acid.

2.3. Determination of vitamin C content

The ascorbic acid content of the samples was determined by iodometric method. The principle of this method is the color reaction between starch and I₂ + KI solution. Vitamin C concentration is done with K₂Cr₂O₇ in the presence of KI-starch. Liberated iodine color the starch in blue.

It was weighed at analytical balance a sample of 0.05-0.1 g, was made quantitatively into a titration flask, there were added 10 ml of 2N HCl, diluted to 50 ml with distilled water and triturated. Then was added 1 ml of 1% starch solution (freshly prepared) and 1 ml of 0.1 N KI and then titrated with aqueous 0.1 N K₂Cr₂O₇ to a persistent blue color. The dosage of the vitamin C content was made according to:

\[ 1 \text{ mL } K₂Cr₂O₇ \times 0.1N \text{ corresponds to } 0.008806 \text{ g Vitamin C} \ [16]. \]

3. Results and discussions

3.1. Antioxidant activity level

Using Trolox standard solutions was obtained the calibration curve on the basis of which have been made suitable calculations for the antioxidant activity of exotic fruits analyzed samples.

Pineapple and kiwi samples could be analyzed as such (not necessary for other dilutions) and pomegranate sample had to be diluted 1: 2, being more concentrated in antioxidants.

The results on the antioxidant capacity of a sample of pineapple, pomegranate and kiwi taken in analysis are shown in Figure 1.
The values we have obtained experimentally showed that the highest antioxidant capacity presents pomegranate sample (0.290 mM Trolox/l) - even diluted 1:2, compared to other samples, followed by kiwi sample (0.115 mM Trolox/l). The reduced antioxidant activity showed the pineapple sample (0.109 mM Trolox/l).

3.2. Total polyphenols content

With the aid of the calibration curve were calculated concentration of polyphenols in samples of exotic fruits. The results of the total polyphenol content of the tested samples are shown in Figure 2.

The results show that of the three types of exotic fruits analyzed, the highest content of vitamin C was found in kiwi (91.20 mg/100g), followed by pineapple (45.30 mg/100g). High content of ascorbic acid in kiwi (almost 2 times higher than in pineapple), makes the antioxidant activity of this fruit to be higher than for pineapple, although it has a total polyphenol content slightly lower than this. Regarding pomegranate notice that has the lowest levels of vitamin C (23.15 mg/100g) of the three analyzed fruits, but due to its very high concentrations of polyphenols (more than 14 times higher than in kiwi and almost 2 times higher than in pineapple), this fruit shows the highest antioxidant activity of fruits analyzed.

4. Conclusions

1. Pomegranate fruit showed the highest antioxidant activity (0.290 mM Trolox/l) followed by kiwi fruit (0.115 mM Trolox/l) and pineapple (0.109 mM Trolox/l).

2. The highest concentration of polyphenols was found also in the pomegranate (0.129 mg gallic acid/l), followed by pineapple (0.072 mg gallic acid/l).

3. Of the three types of exotic fruits analyzed, the highest content of vitamin C was found in kiwi (91.20 mg/100g), followed by pineapple (45.30 mg/100g).

4. Although pomegranate has the lowest levels of vitamin C (23.15 mg/100g) of three fruits analyzed, due to very high concentrations of polyphenols, this fruit shows the highest antioxidant activity of fruits analyzed.

Compliance with Ethics Requirements. Authors declare that they respect the journal’s ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human / or animal subjects (if exist) respect the specific regulation and standards.
References


