PHYSICO-CHEMICAL PARAMETERS OF APPLE FRUITS AND THE EFFECT OF APPLE JUICE PASTEURIZATION ON VITAMIC C CONTENT

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RESEARCH ARTICLE

Abstract

The objective of this study was to characterize three apple varieties (Idared, Red Delicious and Golden Delicious) grown in Romania, with an emphasis on the physico-chemical characteristics. The influence of pasteurization at 70 C° for 30 minutes on ascorbic acid content was also an important objective. Among the physico-chemical characteristics, the content of Dry matter, Humidity, Total mineral content (ash), Total soluble solids (TSS), Total titratable acidity (TTA) and the pH was determined. Among the 3 apple varieties, Golden Delicious (GD) presented a remarkably higher content of total minerals (0.179 %), TTA (3.66 % malic acid in pasteurized juice and 3.52 % malic acid in unpasteurized juice, respectively) and TSS (14.25 °Brix in unpasteurized juice), compared to the varieties Idared (IR) and Red Delicious (RD). After pasteurization, the content of ascorbic acid decreased significantly, compared to unpasteurized juices, and the highest content was reported by unpasteurized IR juice (4.39 %). However, in the case of the GD variety, the content of TTA increases after pasteurization, compared to unpasteurized juice. The investigated apple varieties are fruits rich in bioactive compounds, and the pasteurization treatment does not lead to drastic decreases in the studied physico-chemical parameters.

Keywords: Apple fruits, Apple juice, Phisyco-chemical parameters, Vitamin C, pasteurized juice.

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INTRODUCTION

Apples are a class of fruit varieties that ripen from late summer to winter and are mostly consumed fresh. The most common domesticated apples are known as *Malus x domestica* (*M. domestica)*. Apples can be stored and consumed in all seasons and attract consumers with their color, shape, size and taste. Quality markers of apples are represented by size, shape, color, taste, smell, strength and firmness (Babojelić et al., 2007; Bordean et al., 2013).

If the fruit is harvested too early, and it does not reach the normal point of maturity, the bioactive compounds present in the fruit do not develop completely, and the organoleptic characteristics will suffer. At the same time, if the apple harvest is delayed, it overripes, which leads to storage risks, making it susceptible to handling and storage diseases (Bogdanescu et al., 2018; Bordean et al., 2013; Cvetković et al., 2012).

Apples are harvested manually, by slightly twisting the fruit, thus preventing the occurrence of physical risks. From the moment of harvest until the moment the fruits reach the storage area, it should not exceed more than 2-3 days, so as not to negatively influence the storage capacity (Bogdanescu et al., 2018; Bordean et al., 2013; Cvetković et al., 2012)..

For long-term storage, fruits belonging to varieties with high economic value should be selected, such as Jonathan, Golden Delicious, Florina, Starking Delicious, Idared, etc (Bassi et al., 2017; Bogdanescu et al., 2018).

The optimal storage temperature may vary depending on the variety, depending on their tolerance to low temperatures, which can lead to physiological disorders of the fruits. Among the cold-resistant varieties with sweet flesh and slightly acidic are the ´Golden Delicious´ and ´Red Delicious´ varieties, which are stored at temperatures between 0° C and 1° C. In the category of acidic varieties and more sensitive to cold, there is also the ´Idared´ variety, which has an optimal storage temperature of about 3-4° C (Bogdanescu et al., 2018).

The ´Red Delicious´ variety originated in Peru, Iowa, USA, in about 1972 and was introduced commercially in 1895. It has become the most important and best studied variety in the world, with a distinctive color and appearance. It is an important variety in the European Union, the USA and many other countries. The fruit is crisp and juicy when

harvested in September and October (Bassi et al., 2017; Doryanizadeh et al., 2016; Ferree David C. & Warrington I., n.d.).

´Golden Delicious´ is the second most popular variety grown after ´Red Delicious´ and was discovered in West Virginia in 1914. The variety is a medium to large pale yellow apple, which is mild and sweet. It is crisp when harvested in September and October (Bassi et al., 2017; Bogdanescu et al., 2018; Ornelas-Paz et al., 2018; Thakur et al., 2024).

´Idared´ is an apple that was scientifically developed in 1942 at the University of Idaho Agricultural Experiment Station and is a cross between the Jonathan and Wegener varieties. Since then, demand for production has increased significantly. ´Idared´ is a medium to large apple with a creamy white flesh that is firm, crisp, and juicy (Bogdanescu et al., 2018; Paunovic et al., 2010; Schechter et al., 1993).

Apples, as well as other fruits such as oranges, grapefruits, and strawberries, are known to possess another layer called the extracellular cuticle. This cuticle protects the apple from physical, chemical, and biological stresses such as wind, temperature, chemicals, and drought when the fruit is in the growing stage, as well as when it is harvested. The literature has shown that the outer layer contributes to these barrier properties. Consequently, apples are exposed to mold infections, physical damage and moisture loss (Arrieta-Baez et al., 2020; Bandici et al., 2022).

The aim of this research was to investigate the influence of juice pasteurization on the vitamin C content, as well as the physicochemical characteristics of three Romanian apple varieties: Idared (IR), Red Delicious (RD) and Golden Delicious (GD).

MATERIAL AND METHOD

Plant Materials

Fresh fruits from three different varieties of apples grown in Romania, namely Idared (IR), Red Delicious (RD) and Golden Delicious (GD), were purchased from the local market and transported to the laboratory of the Faculty of Environmental Protection, University of Oradea, Romania, being then analyzed. Before starting the experiment, the fruits were very well washed and dried, and then used to determine the Sensory analysis, Dry matter, Humidity, Total mineral content (ash), Total soluble solids (TSS), Total titratable acidity (TTA), the pH and the content of Acis ascorbic (Vitamin C).

For the determination of sensory analysis, dry matter, humidity and total mineral content (ash), the fruits as such were used.

For TSS, TTA, pH and vitamin C content determinations, fresh fruits were well homogenized in a blender (WARING), the extract obtained was then filtered, and finally, a clear juice without impurities was obtained. Part of the obtained juice was subjected to the pasteurization process at a temperature of 70° C for 30 min, and part of the juice was kept unpasteurized. The operation was repeated for all three varieties of apple under study, and further the previously mentioned determinations were made, both for unpasteurized apple juice and for pasteurized juice, according to the experimental plan presented in Figure 1.

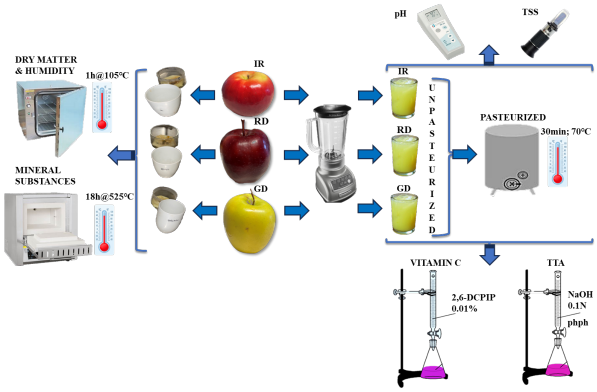

Figure 1. The experimental design for the determination of the physico-chemical parameters of apple fruits and respectively, of apple juices obtained from the varieties IR., RD. and GD.

Sensorial analyze

Ten sensory characteristics (appearance, aroma, taste, texture and aspects related to overall desirability) were evaluated by a team of four men and ten women aged between 19 and 46, students and members of the Department of Food Engineering from the University of Oradea, Romania. Each characteristic was scored between 1 (unacceptable) and 10 (excellent). During the assessment, panel members were given 5 minutes of rest between tests to minimize fatigue and carryover effects. For data collection, sensory evaluation sheets were used and the results define food products quality (Memete et al., 2022).

Determination of dry matter and humidity

Dry matter determination from the conducted analysis was determined through water elimination in the drying oven at 105℃ up to a constant weight value.

The percentual vanished weight is represented by the water content, also referred as humidity (Miteluț et al., 2021; Purcărea, 2015).

Determination of mineral substances

For the determination of mineral substances, the apples were finely chopped, weighed and transferred to ceramic crucibles to withstand the calcination furnace at high temperatures. Mineral substances are inorganic substances that appear after exposure to high temperatures of 525±25℃ in the calcining furnace for 18 hours until the weight of the sample is constant (Harris & Marshall, 2017; Ismail, 2017; Purcărea, 2015).

Determination of total soluble solids (TSS)

Total soluble solids (TSS) results indicate the concentration of sugar in the sample.

Total soluble solids were refractometrically determined from both unpasteurized and pasteurized apple juices. In the first stage, the juice obtained was filtered to obtain a clear juice without impurities. The portable KRÜSS refractometer was turned on, and then brought to 0 with distilled water, after which the Brix readings (in triplicate) of the studied samples were taken at a temperature of 20°C. The results were expressed in °Brix percentage (Memete, Sărac, et al., 2023)..

Determination of total titatable acidity (TTA) and pH

10 ml of clear apple juice, without impurities, was homogenized with 50 ml of distilled water. In an Erlenmayer glass, over the obtained extract, 2.3 drops of phenolphthalein were added, and then titrated with NaOH 0, 1 N until the color turns pink which must persist for 30 seconds. All determinations were performed in triplicate.

Equation (1) was used to interpret the results

$$A = Vx\,10 \qquad (1)$$

Where: A - acidity of the extract expressed in degrees of acidity; V - the volume of the 0.1 N NaOH solution used in the titration, in ml.

For graphical representation, the degrees of acidity were transformed into grams of organic acid that is representative of apple fruits (malic acid).

The pH was determined using a pH meter (Denver Instrument 220) previously calibrated with pH 4.0 and pH 7.0 buffer solutions, the electrode of the pH meter was fully immersed in 100 ml of unpasteurized apple juice and pasteurized, for accuracy of results. All determinations were performed in triplicate at 23°C (Memete et al. 2023; Rosnah et al. 2012).

Determination of vitamin C content

In an Erlenmayer beaker, 10 ml of apple juice and 20 ml of 2% oxalic acid were added, homogenizing together for 5 minutes. After homogenization, 10 ml of sample was transferred into another Erlenmayer beaker and titrated with 0.01% 2,6 dichlorophenol indophenol solution until the appearance of a pink color that persisted for at least 15 seconds. The operation was repeated with standard ascorbic acid solution and the titration volume with 0.01% 2.6 dichlorophenol indophenol solution was noted (Memete, Miere (Groza), et al., 2023; Parveez Zia & Alibas, 2021).

In order to quantify the results, equation (2) was used:

% Vitamin C = (Titration volume of samples / Titration volume of standard acid solution) x 100 (2)

RESULTS AND DISCUSSIONS

Sensory analysis of apple cultivars

The importance of sensory analysis for food products is increasing, especially in terms of smell and taste, because, in addition to the nutritional value and beneficial effects that accumulate in the body through the consumption of certain foods, they attract consumers primarily due to the appearance and their taste. Sensory analysis is also important in the acceptance or rejection of the food product (Khodaei et al., 2021).

The results of the sensory analysis of fresh apples of different varieties (IR, RD and GD) are presented in Figure 2.

The sensory evaluation method of apple fruit considered visual attributes (color), three gustatory attributes (pulp consistency and juiciness of the flesh, taste and ripeness) and harmony of general characteristics. The color of fruits is important in their acceptance or rejection by consumers (Khodaei et al., 2021; Memete et al., 2022).

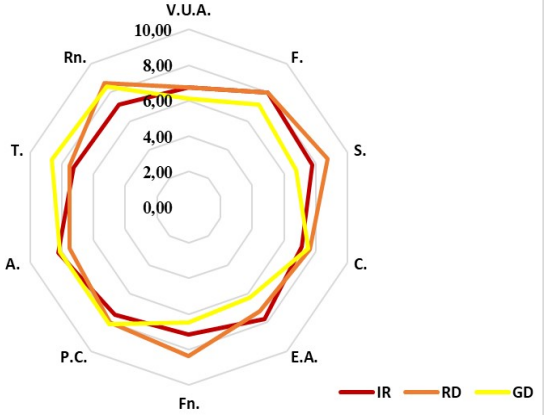

Figure 2. Sensory analysis of three different varieties of apple fruit (IR, RD, GD). V.U.A = Variety Autencity and Uniformity; F.= Form; S. = Size ; C. = Color; E.A. = Exterior Aspect; Fn.= Freshness; P.C. = Pulp Consistency and juiciness of the flesh; A = Aroma; T.= Taste; Rn.= Ripeness.

Regarding the sensory analysis of the three apple varieties (Figure 2), IR reported the highest score for sensory characteristics such as Ripeness (Rn), Size (S) and Freshness (Fn). Analyzing the RD variety, the highest score reported was for Exterior Aspect (E.A.) and Aroma (A). The lowest score was reported for External Appearance (E.A.) compared to IR and GD varieties. Regarding the GD variety, the best score recorded was for Taste (T), followed by Pulp Consistency (P.C) and Color (C).

The content of dry matter and humidity in apple cultivars

As physico-chemical growth and development parameters of the three apple varieties, the percentage of dry matter and humidity was determined for each variety by measuring the weight loss of the fruits after their total dehydration in the oven at 105°C until reaching of a constant weight; The results were presented in Figure 3.

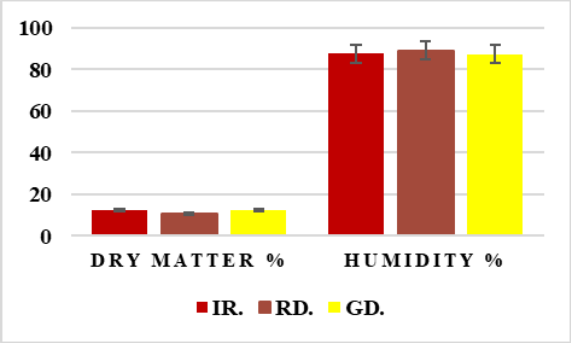

Figure 3. Dry matter and humidity content of IR, ID and GD apple fruits.

Based on the results presented in Figure 3, the percentage of humidity differs according to the variety of fruit studied. The highest humidity percentage was recorded by RD (89.11%), and the lowest by GD (87.48 %). All varieties recorded a high percentage of humidity, with no statistically significant differences reported. Comparing with other studies in the literature, values close to those reported in Figure 3, in terms of dry matter and humidity of the variety IR, were reported in the study of Paunovic et al., 2010 where the dry matter was 15.4 % and the humidity of 84.6 % (Paunovic et al., 2010). Doryanizadeh et al., 2016 reported humidity in the RD variety between 75.76% and 81.5% (Doryanizadeh et al., 2016). Also, Ornelas-Paz et al., 2017 determined the humidity content of GD apples at different ripening stages, and the reported values ranged from 86.0±0.2 % to 86.8±0.1 % (Ornelas-Paz et al., 2018). There are many factors that can influence fruit humidity and dry matter, such as agricultural practices, climatic conditions (high temperatures), fruit maturity level, size, surface-to-volume ratio or external structure. All the listed factors can lead to large differences in humidity, even between the same varieties or varieties of the same species (Bovi et al., 2018; Lufu et al., 2020).

The content of total minerals (ash) in apple cultivars

The content of total minerals was determined for each apple variety, by measuring the weight loss of the fruits after their total dehydration in a calcining oven at 525±25 °C until a light gray ash was obtained; The results were presented in Figure 4.

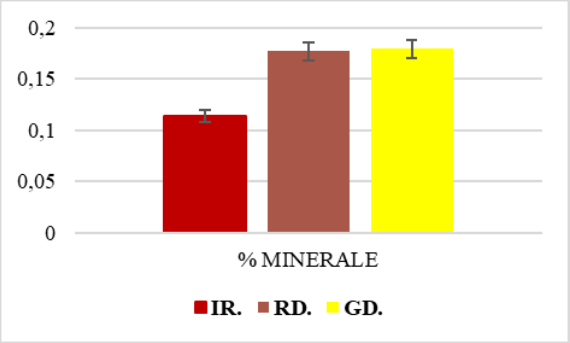

Figure 4. The total mineral content (Ash) of IR, ID and GD apple fruits.

The determination of the ash of food refers to the total minerals and inorganic substances remaining after the food sample has been heated to a very high temperature, removing moisture, volatile and organic substances. The most common minerals and inorganic substances in food are calcium, magnesium, sodium and potassium, but others can also be identified, such as manganese, iron, zinc, in smaller quantities (Harris & Marshall, 2017; Ismail, 2017).

From the results obtained following the determination of the total mineral content and shown in Figure 4, the GD apple varieties followed by the RD reported significantly higher values (0.179% and 0.177, respectively), compared to the IR (0.114%).

The ash content may vary, depending on the degree of processing of the food. Natural foods have lower ash content compared to processed foods (Harris & Marshall, 2017; Ismail, 2017).

Denes, 2023 determined the ash (total mineral content) of a mix of dried Romanian apples from the varieties ´Poinic´, ´Mustos´, ´Dulce´, ´Summer´, ´Winter´, ´Jonathan´ and the reported value was 2.04. Also, in the same study, ash was reported only for the ´Mustos´ variety in different years (2018, 2021), and the values obtained were 1.4 and 1.74, respectively (Denes, 2023).

Physico-chemical parameters determined in unpasteurized and pasteurized apple juices

Physico-chemical determination of apples, such as pH, total titratable acidity (%) and total soluble solids (TSS) expressed in % degree/Brix, were determined from fresh juice, both unpasteurized and pasteurized, obtained for each variety in part.

TTA content in unpasteurized and pasteurized apple juices

The results of total titratable acidity (%) of unpasteurized and pasteurized apple juices are shown in Figure 5.

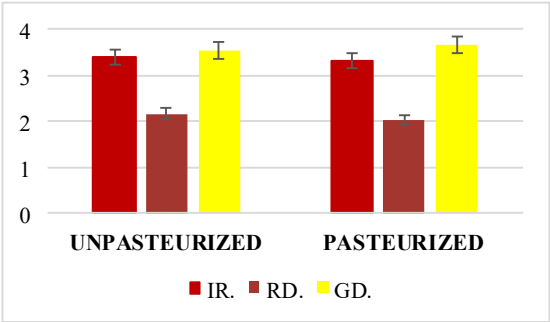

Figure 5. Total titratable acidity of unpasteurized and pasteurized juice obtained from IR, RD and GD apple fruits.

Regarding the total titratable acidity of apple juice (Figure 5), no significant differences were reported between unpasteurized and pasteurized juices for the same variety studied. The highest values were reported for juices obtained from GD apple varieties, both in the case of unpasteurized (3.53±0.01 % malic acid) and pasteurized ones (3.66±0.02 % malic acid). Juices obtained from apples of the RD variety reported the lowest TTA values (2.17±0.03 % malic acid in unpasteurized juice and 2.04±0.01 % malic acid in pasteurized juice respectively); The acidity differences between RD and the other two varieties (IR and GD) are statistically significant in both situations.

Comparing with other literature studies, regarding TTA in GD fruits, Thakur et al., 2024 reported lower values (0.35±0.01 % malic acid) compared to GD fruits in our study (Thakur et al., 2024). Rupasinghe et al., 2010 studied total titratable acidity in 14 red-fleshed apple cultivars, 3 commercial apple cultivars and 3 commercially produced apple juices, and reported values ranged from 0.242 % malic acid (commercial apple cultivar) and 1.28% malic acid (red-fleshed apple) (Rupasinghe et al., 2010).

The pH values in the unpasteurized and pasteurized apple juices

The results of pH of unpasteurized and pasteurized apple juices are shown in Figure 6.

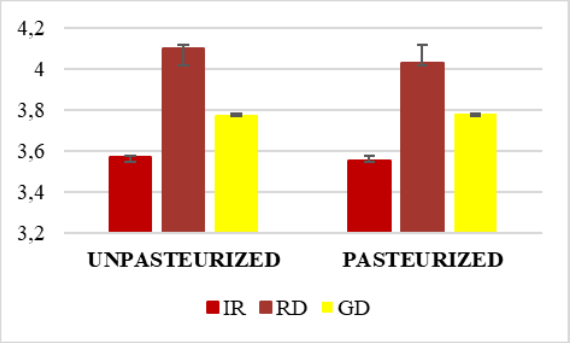

Figure 6. The pH of unpasteurized and pasteurized juice obtained from IR, RD and GD apple fruits.

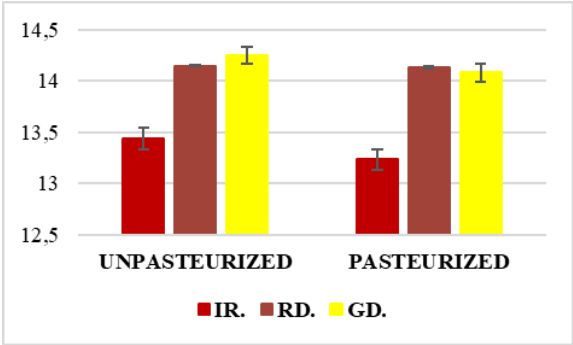

Figure 7. The total soluble solids (TSS) of unpasteurized and pasteurized juice obtained from IR, RD and GD apple fruits.

From Figure 6 it can be seen that the highest pH is reported in the unpasteurized RD juice (4.1), followed by the pasteurized one (4.03), compared to GD and IR. No statistically significant differences were noted between unpasteurized and pasteurized juices of the same apple variety.

Regarding the pH values in the case of the GD variety, similar values were also obtained in the study of Planchon et al., 2004, namely 3.65 (Planchon et al., 2004). Also, a pH of 3.81±0.09 was reported in the study of Thakur et al., 2024 for the variety GD. (Thakur et al., 2024). In the study Bassi et al., 2017, most of the juices obtained from the commercial variety RD had a pH close to 4 (Bassi et al., 2017). In another study, Bogdănescu et al., 2018 determined the pH of eight Romanian apple cultivars, and regarding the ID and GD cultivars the values were similar to those reported in our study (Bogdanescu et al., 2018).

The pH value is closely correlated with the concentration of ascorbic acid in fruit juices. More acidic juices contain higher concentrations of organic acids, especially malic and citric acid, which help protect ascorbic acid from degradation (Bassi et al., 2017; CoSeteng et al., 1989). In addition, the redder the juices, the higher the concentrations of anthocyanins, especially cyanidin-3-O-galactoside which also contributes to the protection of ascorbic acid from oxidation, through its powerful antioxidant capacity (Rupasinghe et al., 2010).

The TSS content in the unpasteurized and pasteurized apple juices

The results of the Total Soluble Solids (TSS) of the juices obtained from the 3 unpasteurized and pasteurized apple varieties (IR, RD and GD) are expressed in % °Brix and presented in Figure 7.

Soluble sugars and organic acids play an important role in determining fruit taste. In apple, fructose is the most abundant soluble sugar, while the predominant organic acid is malic acid which represents up to about 90% of the total organic acids (Monago-Maraña et al., 2021; Zhang & Han, 2021). Apple sweetness is correlated with TSS content, which includes total soluble substances such as organic acids, amino acids, soluble pectins. Also, TSS is an essential parameter in determining the aroma, degree of ripeness, as well as to establish the optimal harvest time of apples (Guan et al., 2015; Monago-Maraña et al., 2021; Zhang & Han, 2021).

TSS analysis indicates the sugar content of juices obtained from 3 apple varieties, unpasteurized and pasteurized. From Figure 7 it can be seen that there were no significant differences between the 3 unpasteurized juices and the 3 pasteurized juices, the highest values being reported by juices obtained from the GD variety in the case of the unpasteurized ones (14.25 °Brix) and RD in the case of the pasteurized ones (14.13 °Brix), and the lowest values were reported by IR in both situations (13.44 °Brix for unpasteurized juice and 13.23 °Brix for pasteurized juice, respectively).

Comparing our results with other studies in the literature, similar results were also reported by Bogdănescu et al., 2018, where the highest TSS values were reported for the GD variety and the lowest were for the IR variety in the case of the GD variety (Bogdanescu et al., 2018). In the study of Planchon et al., 2004 TSS values in apple juice were 13.8 Brix (Planchon et al., 2004). Thakur et al., 2024 also studied the TSS content in GD apples and reported values of 25.48±0.69 °Brix (Thakur et al., 2024). In another study, Monago-Maraña et al., 2021 reported TSS values, determined at 25 °C,

between 15 and 16 % (Monago-Maraña et al., 2021).

The ascorbic acid content in the unpasteurized and pasteurized apple juices

Ascorbic acid (Vitamin C), a powerful water-soluble antioxidant compound, was quantified from the juices obtained from apple fruits and the influence of pasteurization at 70°C on the juice obtained from the three apple varieties: IR, RD and GD was observed. The results obtained were presented in Figure 7.

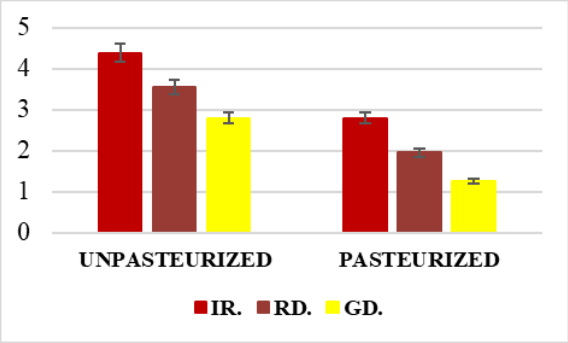

Figure 7. The Ascorbic acid of unpasteurized and pasteurized juice obtained from IR, RD and GD apple fruits.

Apples are among the most consumed fruits in the Western world, either fresh or processed. Therefore, apples are a major source of nutrients and antioxidants. The content of antioxidants, and ascorbic acid in particular, has been extensively studied in other studies, but this content has been shown to be dependent on genotype, geographical position, environmental conditions during the growing and ripening season, agricultural practices, storage conditions as well as processing ones (Farneti et al., 2015; Vegro et al., 2016; Wojdyło et al., 2008). Increased attention to healthy foods has led to a growing interest in identifying varieties with a high ascorbic acid content; However, there are few studies that focus on the factors that influence the content of ascorbic acid in different apple genotypes (Bassi et al., 2017; Kevers et al., 2011; Laslo et al., 2018; Planchon et al., 2004).

Regarding the results of our study, it can be seen in Figure 7 that there is a significant decrease in the content of ascorbic acid in pasteurized apple juice compared to unpasteurized juice. The highest content of ascorbic acid was reported in the juice obtained from apples of the IR variety (4.39 % in unpasteurized juice and 2.80 % in pasteurized juice, respectively), and the lowest content is given by the juice obtained from the GD variety (2.81 % in unpasteurized juice and 1.25 % in pasteurized juice, respectively).

Ascorbic acid in apples degrades both during fruit processing and storage, a fact also confirmed in the study by Kevers et al., 2011, where they observed that ascorbic acid degraded by 80% during three months of cold storage, thus providing a possible explanation for the lower values obtained (Kevers et al., 2011). Also, in the study by Laslo et al., 2018, they investigated the effect of pasteurization on juices obtained from two different varieties of apples grown in Romania (Florina and Liberty). The results demonstrated that the content of ascorbic acid in juices pasteurized at 80° C for 25 minutes, decreased by 73.53% and 76.35% for Florina and Liberty juices, respectively, compared to unpasteurized apple juices (Laslo et al., 2018).

In another study, Planchon et al., 2004 reported ascorbic acid contents ranging from 2.9 to 25.6 mg/100 g FW for whole fruit from 30 old Belgian apple cultivars. Among the 30 varieties studied, we also list the GD variety, for which the reported ascorbic acid content was 11.4 mg/100 g FM (Planchon et al., 2004). Bassi et al., 2017 studied the ascorbic acid content in the pulp of 64 different apple cultivars and reported values between 0.1 mg/100 g and 13.9 mg/100 g. Among the 64 cultivars studied, RD and GD cultivars were also listed which had an ascorbic acid content of less than 1.5 mg/100 g FW in the pulp (Bassi et al., 2017).

CONCLUSIONS

In this paper it is shown that the varieties IR, RD and GD which are widely cultivated in Romania, are fruits with valuable sensory characteristics for human consumption. The pasteurization process of apple juices at 70 °C for 30 minutes increased the level of total titratable acidity in GD juice and significantly decreased the content of Vitamin C, as well as the other studied parameters. Among the apple varieties, GD recorded the highest total mineral content (0.179%) compared to RD (0.177%) and IR (0.114%) varieties. Also, RD was the most appreciative in terms of the sensory characteristics studied and recorded the highest humidity and pH values. From the point of view of vitamin C content, the IR variety stood out with the highest content, both in terms of unpasteurized (4.39 %) and pasteurized (2.80

%) juice. At the same time, the IR variety reported the lowest values of pH, total minerals and TSS, but was the most appreciated for the flavor and external appearance of the fruit. As future perspectives, it is important to study the effect of pasteurization on the evolution of bioactive compounds from apple juices grown in Romania, as well as to establish the best conditions to obtain juices or other food products from apples that maintain a high level of vitamin C, bioactive compounds as well as high antioxidant capacity.

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