APPLES QUALITY INDICATORS VARIATION DURING STORAGE IN CONTROLLED ATMOSPHERE CONDITIONS

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Abstract

The objective of the present study was to determine the quality indicators variation of three organic apple varieties during one year postharvest storage under controlled atmosphere conditions as following: $O_2 - 3\%$, $CO_2 - 5\%$, relative humidity (RH) (%) - 95%, temperature (T) (°C) - 1°C. Apple varieties like 'Golden Delicious', 'Red Prince' and 'Gala' were harvested from an organic orchard in Arad County, Romania. The quality indicators analyses were conducted in triplicates for each apple variety and consisted in determination of dry matter content, total soluble solids, total titratable acidity, ascorbic acid and total anthocyanin content. The obtained results indicated that ascorbic acid and total anthocyanin content storage for all three apple varieties. Indicators like dry matter content and total titratable acidity registered decreases between 19% and 52%.

Key words: *apples*, *organic*, *postharvest*, *quality indicators*.

INTRODUCTION

The consumers are more and more aware of the importance and benefits of consuming fresh and freshly processed fruit (Castell-Perez and Moreira, 2011; Piližota, 2014) in order to maintain human health. The nutritional content of fresh fruits includes vitamins, minerals, fibres, organic acids, and antioxidants, with low quantities of calories, fats and represents a benefit to human health. Because the majority of fruits are harvested seasonally and from different geographical regions, to keep their nutritional content during storage period, is necessary to have effective conservation and storage techniques (Xin et al., 2015).

In Europe, 35% of fruit trees are apple trees, meaning over 450.000 hectares of orchards. The main producer of apples in 2013 was Poland with more than 3,000,000 tons, then Italy and France, while Romania had 503.000 tons of apples. In the last 24 years, the entire European Union area cultivated with apple trees has decreased with 15%, while in Romania decreased by 33%. In the same time, the apple yield increased in Romania by 8% and in Europe by 12% (Badiu et al., 2015).

Due to the role of apples in human nutrition and prevention of diseases it is very important to prolong the storage period with maintaining of quality indicators. Apples consumption can contribute to health protection effects against chronic diseases because of their content in vitamin C, vitamin E and lutein (Lee et al., 2005; Ye and Song, 2008). Also, apples are a rich source of iron and contain high levels of phosphorus, sodium, magnesium, calcium and zinc. The content of potassium and magnesium has been associated with a reduced risk of (Koutsosand cardiovascular disease & Lovegrove, 2015).

The aim of this study was to determine the variation of quality indicators of apple fruits varieties ('Golden Delicious', 'Red Prince' and 'Gala') obtained in an organic orchard and stored for one year under controlled atmosphere conditions

MATERIALS AND METHODS

Samples

Apple varieties like 'Golden Delicious', 'Red Prince' and 'Gala' were harvested in August-September 2017 from an organic orchard in Arad County, Romania and storedfor 12 monthsin controlled atmosphere conditions (O_2 - 3%, CO_2 - 5%, relative humidity (RH) - 95%, temperature (T)°C - 1°C) of Postharvest Technologies Laboratory of Research Center for Studies of Food Quality and Agricultural Products from University of Agronomic Sciences and Veterinary Medicine of Bucharest.

Physical analysis

The analysis of dry matter content was performed with thermobalance Partner MAC 50. The digital refractometer KRÜSS DR301-95 was used for measurements of total soluble solids (TSS) according to Brix reading (Turmanidze et al., 2017). In accordance with Commission Implementing Regulation (UE) No. 974/2014 the obtained results were expressed in percentage (%).

A TitroLine automatic equipment was used for measurements of total titratable acidity (TTA) and consist in titration with 0.1N sodium hydroxide (NaOH) up to pH 8.1 (Skupień, 2006). Results for TTA were expressed in percentages of malic acid (Saad et. al, 2014).

The firmness of fruit was measured with an electronic penetrometer TR equipped with a piston of 11 mm diameter (Rizzolo et al., 2010).

Ascorbic acid identification and quantification

Extraction for ascorbic acid analysis consisted in trituration of 1 g of raw material with 10 mL of metaphosphoric acid (9%, v/v). The extracts obtained were passed through a RC Agilent filter of 0.45 μ m (Turmanidze et al., 2017).

Identification and quantification of ascorbic acid were performed through High Performance Liquid Chromatography (HPLC) using a method adapted afterChanforan et al.,(2012) and the chromatographic equipment Agilent Technologies 1200equipped with UV-DAD detector.

For data processing was used the Agilent ChemStation B.04.03 software (Agilent, USA). The vitamin C chromatographic separation was performed with a ZORBAX XDB-C18 (4.6 x 50 mm, 1.8 μ m i.d.) column and XDB-C18 (4.6 x 12.5 mm, 5 μ m i.d.) (Agilent, USA) guard column. The 2 μ l injection volume, 30°C temperature of the column, 0.5 mL/minflow rate and isocratic elution with 0.05% formic acid in water (v/v) were set during the injection of samples. For the quantitative analysis, a calibration curve was performed through injection of known concentrations of ascorbic acid.

Total anthocyanin content

Extraction for total anthocyanin content consisted in trituration of 0.3 g of raw material with 5 mL of acidified methanol with 1% hydrochloric acid (v/v) for 24h at room temperature in darkness (Giusti & Wrolstad, 2001; Jung et al., 2011). Successive extractions were performed until the residue has become colourless and then was adjusted to 15 ml fixed volume using acidified methanol with 1% hydrochloric acid (v/v).

Specord 210 Plus spectrophotometer was used for measurements of the sample extracts absorbance at wavelength of 530 nm. Results were expressed in mg 100g⁻¹fresh weight and calculated after the below formula:

Total anthocyanin content(mg $100g^{-1}$ FW) = DO530 x F,where DO530 is absorbance at wavelength $\lambda = 530$ nm and factor F = 11.16.

Chlorophyll a, b and total carotenoids content

An adapted method after Lichtenthaler and Wellburn (1983) was used and 1g of fresh sample wasextracted with 80% acetone (v/v). Successive extractions were performed until the residue has become colourless and then was adjusted to 50 ml fixed volume. The absorbance of extracts was measured at 663, 646 and 470 nm against blank (acetone 80%). Formula sused for calculation of chlorophyll a (Ca), b (Cb), and total carotenoids (Cx+c) content were:

Ca μ g/mlextract = 12.21A663-2.81A646 Cb μ g/mlextract = 20.13A646-5.03A663 Cx+c μ g/ml extract = $\frac{1000A470 - 3.27Ca - 104Cb}{229}$

RESULTS AND DISCUSSIONS

Physical analysis

The obtained results are shown in the Table 1. During the storage period in controlled atmosphere conditions it can be observed that firmness decreased for all analysed apple varieties. The 'Red Prince' variety registered the most important loses of firmness, with 30% in comparison with 'Golden Delicious' (aprox. 2%) and 'Gala' (20%). Similar behaviour it was observed by Peck et al. (2006) and mentioned by Mditshwa et al. (2017) which demonstrated that the fruit firmness of 'Galaxy Gala' apples (*Malus* × *domestica* Borkh.) is influenced by production systems and after 6 weeks storage in controlled atmosphere (CA) conditions, the organically grown apples lost 10% of their firmness. Meanwhile, the dry matter content and total titratable acidity registered decreases between 19% and 52%.In the study conducted by Mohammed et al., (2011), noted that the values of the 'Golden Delicious' apples for 100 g raw fruit ranged between 14.53-40.35% for the dry matter content, the humidity 59.65-85.47%, total soluble solids 8.05-10.54%, and total titration acid was 0.47%, of pulp.

Table 1. Variation of apples physic-chemical properties during storage at O₂-3%, CO₂-5%, RH-95%, T-1°C(0 – initial moment after harvesting; 6 – after six months of storage in controlled atmosphere conditions; 12 – after twelve months of storage in controlled atmosphere conditions)

| Sample | Moment of analysis | Dry matter | Firmness | Total soluble solids | Titratable acidity |
|------------|--------------------|------------------|-----------------|----------------------|--------------------|
| | (months) | (%) | (Kg/cm) | (°Brix) | (malic acid %) |
| Golden | 0 | 15.02 ± 0.13 | 8.02 ± 0.60 | 12.24±0.98 | 0.28±0.001 |
| Delicious | 6 | 28.20 ± 0.04 | 8.00 ± 0.70 | 12.20 ± 0.40 | 0.21±0.009 |
| | 12 | 18.42±0.26 | 7.91±0.02 | 13.60±0.25 | 0.15±0.002 |
| Red Prince | 0 | 16.46±0.34 | 6.80±0.60 | 13.24±0.99 | 0.40±0.003 |
| | 6 | 21.88±0.05 | 5.50±0.20 | 13.90±0.60 | 0.27±0.003 |
| | 12 | 13.30±0.42 | 4.90±0.03 | 12.60±0.16 | 0.30 ± 0.005 |
| Gala | 0 | 30.93±0.54 | 6.13±0.50 | 13.38±0.98 | 0.21±0.008 |
| | 6 | 21.70 ± 0.04 | 6.00 ± 0.07 | 13.20 ± 0.00 | 0.23 ± 0.018 |
| | 12 | 14.87 ± 0.07 | 6.04±0.03 | 13.60±0.90 | 0.30 ± 0.005 |

RH – *relative humidity; T- temperature*

Data represent the means of three replicates followed by their standard deviations.

Ascorbic acid identification and quantification and total anthocyanin content

Table 2. Variation of apples ascorbic acid and total anthocyanin content during storage at O_2 -3%. CO_2 -5%. RH%-95%. t °C-1°C (0 – initial moment after harvesting; 6 – after six months of storage in controlled atmosphere conditions; 12 – after twelve months of storage in controlled atmosphere conditions)

| Sample | Moment of analysis (months) | Ascorbic acid content (mg/100g) | Total anthocyanin content (mg/100g) | |
|------------|-----------------------------|------------------------------------|--|--|
| Golden | 0 | 24.51±0.43 | n/a | |
| Delicious | 6 | 0.28 ± 0.02 | n/a | |
| | 12 | 0.38±0.02 | n/a | |
| Red Prince | 0 | 25.36±0.54 | 1.42±0.063 | |
| | 6 | 0.45 ± 0.01 | $0.65 {\pm} 0.005$ | |
| | 12 | 0.4±0.03 | 0.70 ± 0.026 | |
| Gala | 0 | 22.92±0.63 | 2.63±0.114 | |
| | 6 | 0.22±0.01 | 0.94±0.041 | |
| | 12 | 0.15±0.00 | 0.62±0.023 | |

RH – relative humidity; T- temperature; n/a – analysis not applicable for green or yellow fruits as Golden variety. Data represent the means of three replicates followed by their standard deviations.

In general, fruits and vegetables shown gradual decreases in ascorbic acid content as the storage period or temperature increases (Koyuncu and Dilmaçünal. 2010; Stan and Popa, 2015).

Also, in the present work in can be observed that the ascorbic acid and total anthocyanin content registered important decreases during storage period in controlled atmosphere conditions. Decrease of the ascorbic acid content was more than 90% for all apples varieties while for total anthocyanin content it ranged between 50% and 80%.

Chlorophyll a. b and total carotenoids content The variation of chlorophyll a. b and total carotenoids content during storage period in controlled atmosphere conditions is presented below in Table 3.

Table 3. Variation of apples chlorophyll a. b and total carotenoids content during storage at O₂-3%. CO₂-5%. RH%-95%. t °C-1°C (0 – initial moment after harvesting; 6 – after six months of storage in controlled atmosphere conditions; 12 – after twelve months of storage in controlled atmosphere conditions

| Sample | Moment of analysis (months) | Chlorophyll a (µg/ml plant extract) | Chlorophyll b (µg/ml plant extract) | Total chlorophyll (µg/ml plant extract) | Total carotenoids (μg/ml plant extract) | Ratio Chl a/Chl b |
|---------------------|-----------------------------------|---|---|--|---|----------------------|
| Golden Delicious | 0 | 0.46 ± 0.004 | 0.10±0.006 | $0.55 {\pm} 0.010$ | 0.13±0.001 | 4.74±0.281 |
| | 6 | 0.44 ± 0.025 | 0.62±0.012 | 1.06 ± 0.010 | 0.07 ± 0.004 | 0.70 ± 0.014 |
| | 12 | 0.45±0.017 | 0.59 ± 0.042 | 1.04 ± 0.116 | 0.01±0.003 | 0.75 ± 0.034 |

RH - relative humidity; T- temperature;

Data represent the means of three replicates followed by their standard deviations.



Correlation between chlorophyll a and b

Chlorophyll a (µg/ml plant extract)

Figure 1. Correlation between chlorophylla and b obtained results during apples storage in controlled atmosphere conditions

The values of chlorophyll a registered decreases from initial moment of analyse and the chlorophyll b values were higher than initial. Similar behaviours of chlorophyll a and

b were observed also in many studies of various matrix by Tanaka & Tanaka (2011). The explication for these variations is because of chlorophyll a conversion to chlorophyll b

when the chlorophyll a synthesis was stopped under darkness. Chlorophyll a is essential in photochemistry while chlorophyll h is apparently dispensable for their photosynthesis. Instead chlorophyll b is necessary for stabilizing the major light-harvesting chlorophyll binding proteins. Chlorophyll b is synthesized from chlorophyll a and is catabolized after it is reconverted to chlorophyll a. This interconversion system between chlorophyll a and chlorophyll b refers to the chlorophyll cycle (Tanaka & Tanaka, 2011).

The correlation between chlorophyll a and b is inversely proportional and significantly different due to conversion of chlorophyll a to chlorophyll b (Figure 1).

CONCLUSIONS

Ascorbic acid and total anthocyanin content values decreased during one-year postharvest storage for 'Golden Delicious', 'Red Prince' and 'Gala' apple varieties.

Firmness decreased for all analysed apple varieties.

The correlation between chlorophyll a and b is inversely proportional and significantly different due to conversion of chlorophyll a to chlorophyll b.

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