Quality Assurance and Safety of Crops & Foods, 2019; 11 (7): 631-638
SPECIAL ISSUE: *Scientific trends of global food laws and regulations*

Postharvest quality changes of organic strawberry Regina cultivar during controlled atmosphere storage

M.E. Popa[1*], A. Stan[2], V. Popa[2], E.E. Tanase[1], A.C. Mitelut[1] and L. Badulescu[2]

[1]University of Agronomic Sciences and Veterinary Medicine of Bucharest, Faculty of Biotechnologies, 59 Marasti Blvd., District 1, Bucharest, Romania; [2]University of Agronomic Sciences and Veterinary Medicine of Bucharest, Research Center for the Study for Food and Agricultural Products Quality, 59 Marasti Blvd., District 1, Bucharest, Romania; pandry2002@yahoo.com

Received: 30 November 2018 / Accepted: 19 September 2019

RESEARCH ARTICLE

Abstract

Fresh produce remains metabolically active even after harvest and continues to respire and lose water. This fact represents a current challenge for establishing the optimal conditions of controlled atmosphere for prolonging the shelf life of fresh vegetal products. In this study, the shelf life and the best storage practices for high quality organic strawberries were determined. Organic strawberries were stored in four different conditions: air (control); 5% O_2, 10% CO_2, 85% N_2, 75% relative humidity (CA I); 5% O_2, 15% CO_2, 80% N_2, 75% relative humidity (CA II) and 5% O_2, 10% CO_2, 85% N_2, 95% relative humidity (CA III). It was established that the best storage conditions in controlled atmosphere of the tested organic strawberries were 5% O_2, 15% CO_2 and 80% N_2 with 75% relative humidity at 3 °C, from physical-chemical, microbiological and nutritional points of view.

Keywords: organic strawberries, shelf life prolongation, controlled atmosphere

1. Introduction

Increasing consumer demand for safe food products and environmentally friendly food technologies makes organic food production one of the fastest-growing market segments over the last few years. Consumers expect an increased quality of organic products compared to conventional ones, while protecting nature and reducing environmental pollution. Berries, especially strawberries, are very attractive for consumers due to their unique aroma, texture and colour, both fresh and a large variety of food products and snacks (Tylewicz *et al.*, 2017).

Strawberries are popular fruits grown around the world, with an annual production in Romania of about 23,000 tons in 2016 (http://www.fao.org/faostat/en/#data/QC). They are rich in vitamin C, fibre, polyphenols, potassium and present various benefits for human health (Aaby *et al.*, 2018; Pan *et al.*, 2014).

Strawberries have a very short post-harvest life, mainly due to sensitivity and susceptibility to mechanical damage, rapid dehydration, and infections caused by several microorganisms (Aday *et al.*, 2013; Pan *et al.*, 2014; Siedliska *et al.*, 2018; Tylewicz *et al.*, 2017). Their shelf life depends on the cultivar, degree of ripeness, harvesting conditions, handling and storage. The limiting factors regarding the shelf life of strawberries are a combination of appearance, taste, texture and microbial growth (Nielsen and Leufven, 2008).

Controlled atmosphere (CA), with low O_2 and/or high CO_2, together with temperature contributes to the maintenance of storage quality, freshness and the extension of shelf life of perishable fruits and vegetables (Deng *et al.*, 2007; Zhao *et al.*, 2019). The packaging method using modified atmosphere conditions represents one of the most important ways to increase the shelf life of strawberry (Gholami *et al.*, 2017). The maintenance of fruit in contact with the controlled atmosphere leads to a decrease in the rate of chemical and biochemical reactions, as well as the reduction of growth of microorganisms (Barikloo *et al.*, 2018).

Thus, modified atmosphere packaging is a technique used to modify the atmosphere of gas inside the packaging using perforated or non-perforated polymeric films, thereby reducing product quality loss and improving the shelf life of packaged products by lowering moisture and reducing metabolic and microbial activity. Regarding strawberries, they are not sensitive to refrigeration temperatures, the recommended conditions for modified atmosphere being 0-5 °C, 5-10% O_2 and 15-20% CO_2 (Xanthopoulos *et al.*, 2012).

The objective of this study was to establish the shelf life and determine the best storage practices for high quality organic strawberries.

2. Materials and methods

Raw material

Organic strawberries cultivar Regina were purchased from a local producer (Giurgiu, Romania). Prior to packaging, the strawberries were sorted to have the same ripening stage, uniform size (diameter (mm): 31±0.94), absence of any physical damage and fungal infection.

Experimental design

Approximately 200 g of organic strawberries were placed in a single layer in polyethylene terephthalate perforated trays. The storage conditions were: air (control); 5% O_2, 10% CO_2, 85% N_2, 75% relative humidity (CA I); 5% O_2, 15% CO_2, 80% N_2, 75% relative humidity (CA II) and 5% O_2, 10% CO_2, 85% N_2, 95% relative humidity (CA III). Three controlled atmospheric (CA) cells were connected and controlled individually through Besseling Group B.V. BatNetWin (Oosterblokker, the Netherlands) and parameters like relative humidity, temperature, injection or absorption of O_2, CO_2 and N_2 were setup before effectively storing the strawberries. All experiments were performed at 3 °C. The samples were analysed on day 0, 7, 14, 25 and 32 during storage. Before analysis, samples were homogenised in sterile bags using a bag mixer (Interscience™ Bagmixer, Saint Nom, France). All determinations were done in duplicate.

pH

pH was determined using a pH meter WTW INOLAB 720 series type with automatic temperature compensator (Xylem Analytics GmbH, Weilheim, Germany).

Sugar

The level of sugars was measured as Brix° by a Krüss Refractometer (Krüss Optronic GmbH, Hamburg, Germany) and correlated with the amount of soluble solids (expressed as sucrose concentration) using the conversion table or read directly on the refractometer's scale.

Firmness

Organic strawberry fruit firmness was measured using a digital penetrometer Turoni FR 5120 (T.R. Turoni srl, Forli, Italy) fitted with an 8 mm diameter flat probe and the results were expressed as kgf/cm^2.

Respiration and transpiration rate

The respiration rate was determined by measuring the amount of CO_2 produced during fruits respiration. For this purpose, a known volume and mass of strawberries was tested with a CO_2 analyser to determine how much CO_2 was produced in a known volume of air in 10 min. Results were expressed in mg CO_2/kg/h. The transpiration rate measurement was accomplished by gravimetric method, by repeated weighing of the samples at 10 min intervals. Results were expressed as ml water/100 g product/h.

Colour

Colour assessment was conducted at room temperature using a HunterLab colorimeter, Miniscan XE Plus (Hunter Associates Laboratory, Inc., Reston, VA, USA). Fruit colour was described using L*, a*, and b* CIE coordinates. L* axis represents the lightness with values ranging from 0 (black) to 100 (white), the a* axis is red-green with negative values for green and positive values for red, and b* axis is blue-yellow with negative values for blue and positive values for yellow (Palonen and Weber, 2019).

Microbiological analysis

Total aerobic count (TAC), and yeast and moulds were determined for the analysed samples. Briefly, 10 g of homogenised sample were mixed with 90 ml of sterile distilled water and properly diluted (using the decimal dilution method). Plate count agar culture media was used for TAC determination and malt extract agar culture media was used for yeast and moulds determination, both purchased from Scharlu. After inoculation, the Petri dishes were incubated as follows: 24 h at 35 °C for TAC, 72 h at 25 °C for yeast and 5 days at 25 °C for moulds determination. Results were expressed as colony forming unit (cfu)/g.

Antioxidant activity

The effect of antioxidant activity on 1,1-dipheny-l-2-picrylhydrazyl (DPPH) was estimated according to the procedure described by Villaño *et al.* (2007), with some modification presented further. To obtain DPPH solution (60 μM), 2.36 mg DPPH were diluted in 100 ml ethanol. For samples preparation, 5 g of fruit were macerated in 25 ml ethanol (75%) for 48 h in the dark, at room temperature, then extract solutions were filtered through filter paper. For each measurement, 0.05 ml filtered extract solution was added to 1.95 ml DPPH ethanolic solution. These solutions were vortexed thoroughly, and incubated in dark at room temperature for 30 min (Gülçin, 2010). After 30 min, sample absorbance was measured at 515 nm (t=30 min) against DPPH ethanolic solution alone (t=0 min). Results were expressed as quercetin equivalents. All measurements were performed in triplicate.

Ascorbic acid content

The content of ascorbic acid from strawberry samples was determined by extracting 10 g of fruit pulp in 100 ml of 1% oxalic acid in a homogeniser for 1 min. The extract was filtered and 2 ml from the extract solution, 1 ml oxalic acid 1%, 5 ml tampon solution, 2 ml indophenol (2, 6-dichlorophenol indophenol) and 20 ml xylene, were placed in a centrifuge tube and centrifuged for 20 min at 40 °C and 9,000 rpm. After measuring the absorbance at 500 nm, ascorbic acid content was expressed as mg/100 g product.

Total polyphenols content

Total content of polyphenols was determined using the Folin-Ciocalteu method (Arnous *et al.*, 2002; Rover and Brown, 2002). Briefly, for each measurement, 1.58 ml distilled water, 20 µl filtered extract solution (5 g of fruit macerated in 25 ml ethanol (75%) for 48 h in the dark at room temperature, then filtered through filter paper), and 100 µl Folin-Ciocalteu reagent were mixed and after exactly 1 min 300 µl Na_2CO_3 (20%) was added. The solutions were mixed and stored in the dark at room temperature for 2 h. After 2 h, sample absorbance was measured at 765 nm against ethanol solution (75%). Total polyphenol concentration was expressed as mg/l gallic acid equivalents (GAE). All measurements were performed in triplicate.

3. Results and discussion

In this experiment, compared with air-stored organic strawberries, storage in controlled atmosphere conditions decreased the losses of strawberry firmness, sugar content and pH. The results of the physical-chemical analyses are presented in Table 1.

The pH of the tested samples slightly increased during the storage period, for all storage conditions. Similar results were obtained by Van de Velde *et al.* (2019) in their study, where pH values of strawberries increased during CA storage (for both 10% CO_2 and 20% CO_2 conditions). The initial sugar concentration of the organic strawberries was 6.6%. During storage period, the sugar levels decreased slightly for the samples stored in CA II and CA III conditions; most likely the sugars were consumed through the respiration process. On the contrary, for the control sample and CA I conditions, the level of sugars slightly increased, most probably due to dehydration of the tested samples. Similar results were obtained by Kahramanoğlu (2019), sugar concentration of strawberries showing a

Table 1. Physical-chemical analysis results on strawberries.

Storage conditions at 3 °C	Time of analysis (days)	pH	Brix (°Bx)	Firmness
Raw material before storage[1]	0	3.5±0.01	6.6±0.06	0.38±0.12
Control (air)	7	3.78±0.01	6.2±0.06	0.39±0.13
	14	3.79±0.03	6.6±0.06	0.49±0.13
	25	The samples presented visible mould on the surface.		
	32	The samples presented visible mould on the surface.		
CA I – 10% CO_2 75% RH	7	3.77±0.01	6.7±0.06	0.39±0.13
	14	3.9±0.06	6.7±0.06	0.51±0.17
	25	3.83±0.03	6.8±0.06	0.44±0.16
	32	The samples presented visible mould on the surface.		
CA II – 15% CO_2 75% RH	7	3.79±0.01	6.8±0.06	0.43±0.11
	14	3.87±0.01	6.5±0.06	0.43±0.19
	25	3.85±0.01	6.4±0.06	0.44±0.14
	32	3.95±0.01	5.9±0.06	0.31±0.1
CA III – 10% CO_2 95% RH	7	3.8±0.01	7.7±0.06	0.59±0.13
	14	3.86±0.04	6.8±0.06	0.53±0.14
	25	3.89±0.01	6.8±0.1	0.34±0.16
	32	The samples presented visible mould on the surface.		

[1] CA = controlled atmosphere; RH = relative humidity.

declining trend in the first days of storage, and then being increased. The firmness of the strawberries tended to increase during the storage period.

Regarding the physical – chemical analysis, the best results were obtained for strawberries stored under controlled atmosphere conditions CA II (5% O_2, 15% CO_2 and 80% N_2 with 75% relative humidity, stored at 3 °C). Under CA II storage parameters strawberries were kept for 32 days without mould appearing on the surface, while the other samples (stored under CA I and CA III) could not be analysed on day 32 due to mould development, and the control developed visible moulds starting with 25th day.

Respiration and transpiration (Figure 1 and 2) rate measurements were conducted along 14 days for control sample, 25 days for samples stored in CA I and CA II, and 32 days for samples stored CA III conditions, the last one representing the best sample from visual appearance and storage life time point of view, due to high CO_2 concentration (15%) which has a role in reducing the intensity of respiration process.

As shown in Table 2, the values of lightness (L*), redness (a*) and yellowness (b*) parameters showed an increasing trend when strawberries samples were stored under CA I conditions and Control sample, while for the samples stored in CA II and CA II conditions the values decreased.

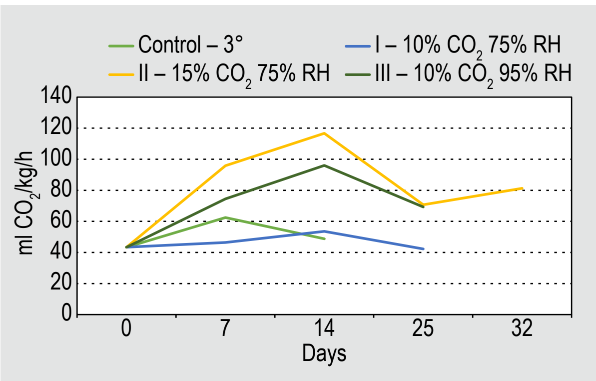

Figure 1. Graphical representation of strawberry respiration rate during storage.

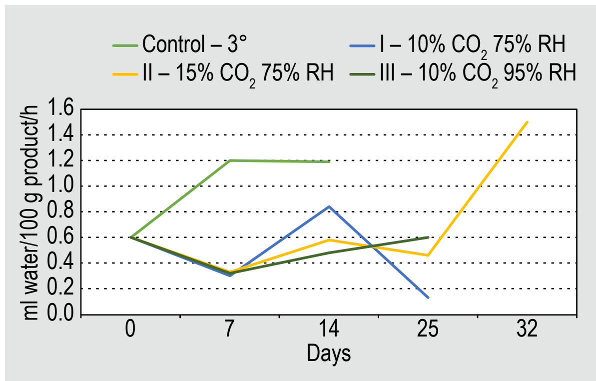

Figure 2. Graphical representation of strawberry transpiration rate during storage.

Table 2. Colour changes during storage of organic strawberries.

Storage conditions at 3 °C	Time of analysis (days)	L*	a*	b*
Raw material before storage[1]	0	24.86±0.66	36.71±0.19	26.57±0.46
Control (air)	7	23.57±0.28	35.01±0.26	23.64±0.49
	14	28.06±0.25	37.08±0.27	26.34±0.37
	25	The samples presented visible mould on the surface.		
	32	The samples presented visible mould on the surface.		
CA I – 10% CO_2 75% RH	7	25.90±0.15	35.51±0.15	23.85±0.30
	14	27.27±0.15	37.09±0.20	25.30±0.38
	25	26.66±0.12	33.49±0.12	20.93±0.28
	32	The samples presented visible mould on the surface.		
CA II – 15% CO_2 75% RH	7	26.13±0.20	35.59±0.22	24.26±0.47
	14	24.95±0.30	34.19±0.61	20.86±0.45
	25	25.96±0.47	33.58±0.21	20.45±0.32
	32	22.23±0.14	32.00±0.14	19.19±0.23
CA III – 10% CO_2 95% RH	7	26.43±0.14	35.92±0.12	24.12±0.19
	14	26.14±0.29	34.23±0.21	20.98±0.34
	25	25.46±0.18	32.36±0.17	19.43±0.18
	32	The samples presented visible mould on the surface.		

[1] CA = controlled atmosphere; RH = relative humidity.

The results of microbiological analyses (Table 3) show that both the total number of mesophilic aerobic germs and the number of yeasts increased during storage in a controlled atmosphere, and moulds were present throughout the storage period for all samples. However, visible on the surface only after 25 and 32 days of storage, under the conditions of the Control sample, CA I and CA III (Figure 3), compared with the samples stored in CA II on which no mould colony was identified. This fact could be explained by the higher concentration of CO_2, which inhibited the development of mould on the surface of the strawberries.

Regarding the ascorbic acid content of strawberry, a slight decrease can be observed for all analysed samples, regardless of the storage conditions (Figure 4).

Figure 5 shows the evolution of antioxidant activity during the 32 days of storage under controlled atmosphere conditions of strawberry samples, which was determined by the DPPH method. Clear differences between samples can be observed. Thus, the samples stored 25 days in 15% CO_2 and 75% relative humidity presented a concentration of 1,276.59±54.60 µM equivalents of quercetin.

As shown in Figure 6, the total phenolic content of strawberries decreased during the storage period for all tested samples, the highest value being 361.47±21.78 mg/l GAE for the fresh strawberry sample. However, according to Table 4, the samples stored in CA II presented the lowest percentage of decrease in polyphenol content.

Figure 3. Strawberries appearance at the end of storage period.

During the storage period, it was observed content reducing of the analysed nutritional parameters for all samples but at the same time, controlled atmosphere could ensure a clear extension of the shelf life in comparison with control stored in air at the same temperature. The obtained results are presented in Table 4.

Table 3. Results of microbiological analysis of strawberries.

Storage conditions (3 °C)[1]	Time of analysis (days)	Total plate count (cfu/g)	Yeasts (cfu/g)	Moulds[2]
Raw material before storage	0	0.04×10^5	0	+
Control (air)	7	0.225×10^5	0.935×10^5	+
	14	1.90×10^5	1.65×10^5	+
	25	The samples presented visible mould on the surface.		
	32	The samples presented visible mould on the surface.		
CA I – 10% CO_2 75% RH	7	0.082×10^5	0.09×10^5	+
	14	0.185×10^5	0.0745×10^5	+
	25	2.0×10^5	2.65×10^5	+
	32	The samples presented visible mould on the surface.		
CA II – 15% CO_2 75% RH	7	3.4×10^3	6.1×10^3	+
	14	5.1×10^3	2.0×10^3	+
	25	1.25×10^4	5.0×10^4	+
	32	1.0×10^4	6.21×10^4	+
CA III – 10% CO_2 95% RH	7	1.4×10^3	1.7×10^3	+
	14	1.8×10^4	1.25×10^4	+
	25	8.5×10^4	2.2×10^4	+
	32	The samples presented visible mould on the surface.		

[1] CA = controlled atmosphere; RH = relative humidity.
[2] + indicates the presence of mould during microbiological analysis.

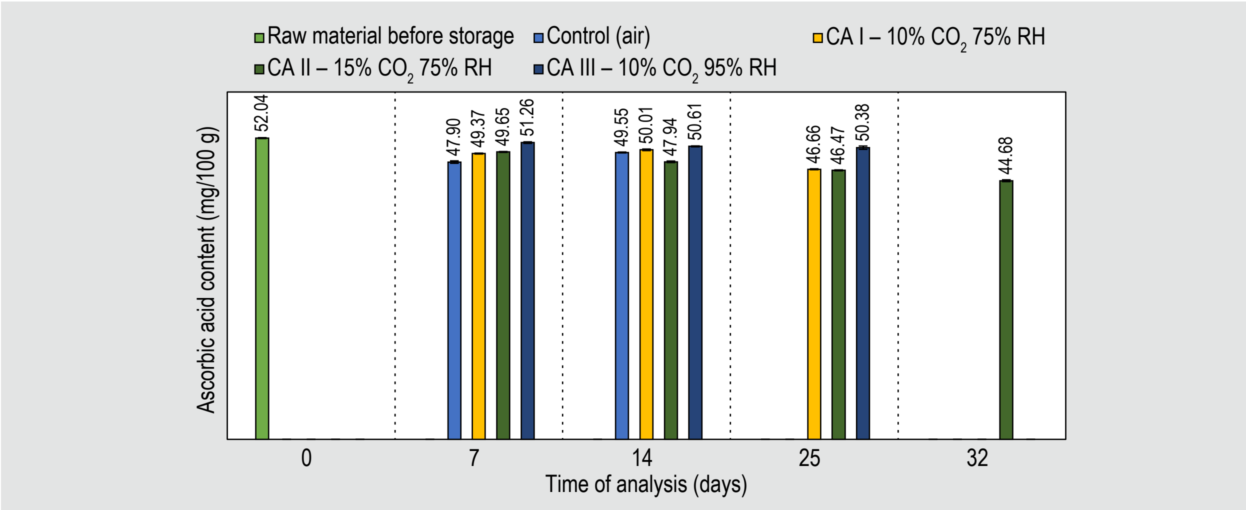

Figure 4. Graphical representation of ascorbic acid content evolution during storage.

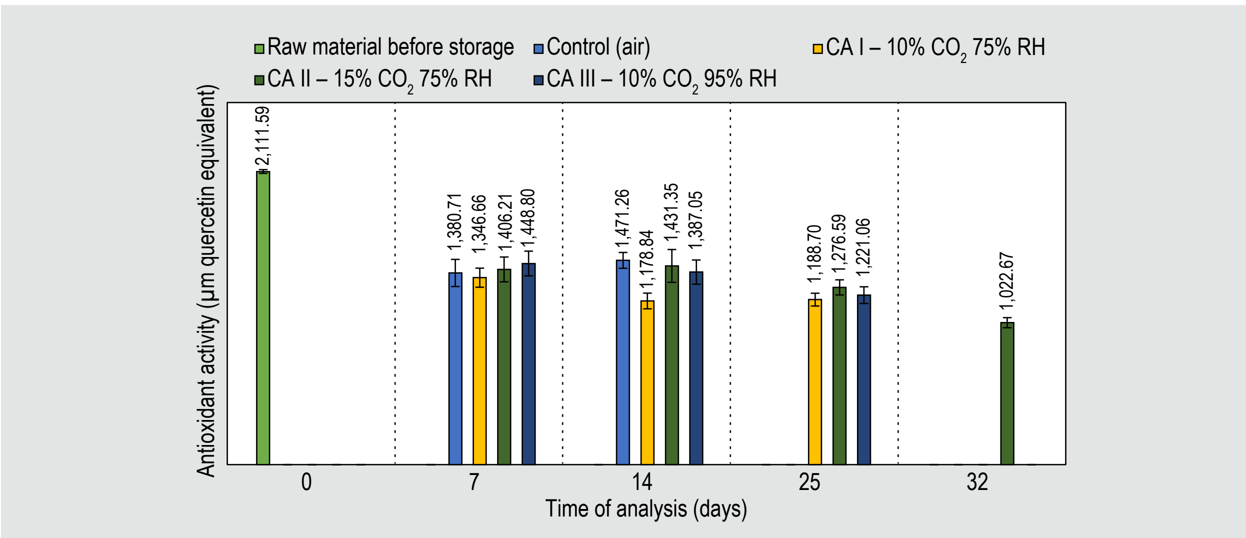

Figure 5. Graphical representation of antioxidant activity evolution during storage.

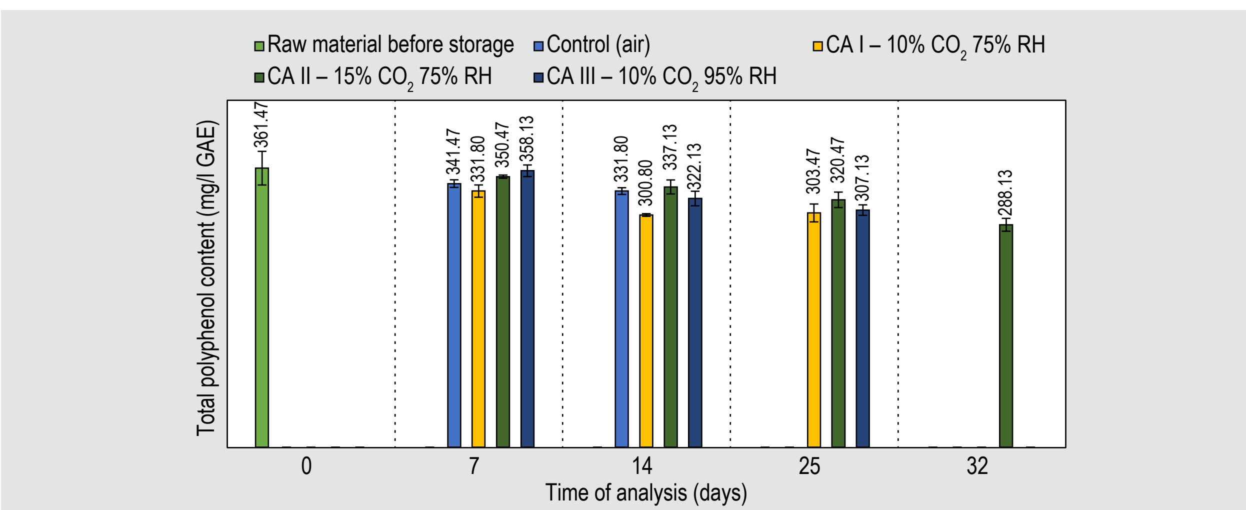

Figure 6. Graphical representation of total polyphenol content evolution during storage.

Table 4. Nutritional parameters at the end of shelf life in different storage conditions.

Strawberries (sample)[1]	Storage temperature (°C)	Shelf life (days)	Extension of shelf life (%)	Ascorbic acid content reducing (%)	Antioxidant capacity reducing (%)	Total polyphenols content reducing (%)
Control (air)	3	14	0	5	30	8
CA I – 10% CO_2 75% RH	3	25	78.5	11	44	16
CA II – 15% CO_2 75% RH	3	32	128.5	11	40	11
CA III – 10% CO_2 95% RH	3	25	78.5	4	42	15

[1] CA = controlled atmosphere; RH = relative humidity.

Conclusions

Strawberries kept in modified atmosphere maintained their quality parameters and appearance better than those stored in air (control). Although there are some losses of valuable compounds such as ascorbic acid and polyphenols the benefits obtained regarding the shelf life extension are evident. This advantage is very important taking into consideration seasonal characteristic of this fruits.

It was established that the best storage conditions in controlled atmosphere of the tested organic strawberries were 5% O_2, 15% CO_2 and 80% N_2 with 75% relative humidity at 3 °C.

Acknowledgement

Financial support for this project was provided by funding bodies within the CORE Organic Plus FP7-ERA-NET-618107 Ensuring quality and safety of organic food along the processing chain and with cofounds from the European Commission UEFISCDI – ERA NET for Romanian Partner USAMVB – 'Innovative and eco-sustainable processing and packaging for safe and high-quality organic berry products with enhanced nutritional value' EcoBerries.

References

Aaby, K., Grimsbo, I.H., Hovda, M.B. and Rode, T.M., 2018. Effect of high pressure and thermal processing on shelf life and quality of strawberry purée and juice. Food Chemistry 260: 115-123.

Aday, M.S., Temizkan, R., Büyükcan, M.B. and Caner, C., 2013. An innovative technique for extending shelf life of strawberry: ultrasound. LWT – Food Science and Technology 52: 93-101.

Arnous, A., Petrakis, C., Makris, D.P. and Kefalas P., 2002. A peroxy-oxalate chemiluminescence-based assay for the evaluation of hydrogen peroxide scavenging activity employing 9,10-diphenyl-anthracene as the fluorophore. Journal of Pharmacological and Toxicological Methods 48(3): 171-177.

Barikloo, H. and Ahmadi, E., 2018. Shelf life extension of strawberry by temperatures conditioning, chitosan coating, modified atmosphere, and clay and silica nanocomposite packaging, Scientia Horticulturae 240: 496-508.

Deng, Y., Ying, W. and Yunfei, L., 2007. Effects of high CO_2 and low O_2 atmospheres on the berry drop of Kyoho grapes. Food Chemistry 100: 768-773.

Gholami, R., Ahmadi, E. and Farris, S., 2017. Shelf life extension of white mushrooms (*Agaricus bisporus*) by low temperatures conditioning, modified atmosphere, and nanocomposite packaging material. Food Packaging and Shelf Life 14, Part B: 88-95.

Gülçin, I., 2010. Antioxidant properties of resveratrol: a structure-activity insight. Innovative Food Science and Emerging Technologies 11: 210-218.

Kahramanoğlu, I., 2019. Effects of lemongrass oil application and modified atmosphere packaging on the postharvest life and quality of strawberry fruits. Scientia Horticulturae 256: 108527.

Nielsen, T. and Leufven, A., 2008. The effect of modified atmosphere packaging on the quality of Honeoye and Korona strawberries, Food Chemistry 107: 1053-1063.

Palonen, P. and Weber, C., 2019. Fruit color stability, anthocyanin content, and shelf life were not correlated with ethylene production rate in five primocane raspberry genotypes, Scientia Horticulturae 247: 9-16.

Pan, L., Zhang, W., Zhu, N., Mao, S. and Tu, K., 2014. Early detection and classification of pathogenic fungal disease in post-harvest strawberry fruit by electronic nose and gas chromatography-mass spectrometry. Food Research International 62: 162-168.

Rover, M.R. and Brown, R.C., 2013. Quantification of total phenols in bio-oil using the Folin–Ciocalteu method. Journal of Analytical and Applied Pyrolysis 104: 366-371.

Siedliska, A., Baranowski, P., Zubik, M., Mazurek, W. and Sosnowska, B., 2018. Detection of fungal infections in strawberry fruit by VNIR/SWIR hyperspectral imaging. Postharvest Biology and Technology 139: 115-126.

Tylewicz, U., Tappi, S., Mannozzi, C., Romani, S., Dellarosa, N., Laghi, L., Ragni, L., Rocculi, P. and Dalla Rosa, M., 2017. Effect of pulsed electric field (PEF) pre-treatment coupled with osmotic dehydration on physico-chemical characteristics of organic strawberries. Journal of Food Engineering 213: 2-9.

Van de Velde, F., Méndez-Galarraga, M.P., Grace, M.H., Fenoglio, C., Lila, M.A. and Pirovani, M.E., 2019. Changes due to high oxygen and high carbon dioxide atmospheres on the general quality and the polyphenolic profile of strawberries. Postharvest Biology and Technology 148: 49-57.

Villaño, D., Fernández-Pachón, M.S., Moyá, M.L., Troncoso, A.M. and García-Parrilla, M.C., 2007. Radical scavenging ability of polyphenolic compounds towards DPPH free radical. Talanta 71: 230-235.

Xanthopoulos, G., Koronaki, E.D. and Boudouvis, A.G., 2012. Mass transport analysis in perforation-mediated modified atmosphere packaging of strawberries. Journal of Food Engineering 111: 326-335.

Zhao, X., Xia, M., Wei, X., Xu, C., Luo, Z. and Mao, L., 2019. Consolidated cold and modified atmosphere package system for fresh strawberry supply chains. LWT – Food Science and Technology 109: 2017-2015.