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[1]Szent István University, Department of Physics and Control, Budapest, Hungary
[2]Industrial University of Ho Chi Minh City, Institute of Biotechnology and Food Technology, Ho Chi Minh, Vietnam
[3]Szent István University, Department of Refrigeration and Livestock Product Technology, Budapest, Hungary
[4]Szent István University, Department of Postharvest Science and Sensory Evaluation, Budapest, Hungary

Evaluation of precooling temperature and 1-MCP treatment on quality of 'Golden Delicious' apple

László Baranyai[1*], Lien Le Phuong Nguyen[2,3], Mai Sao Dam[2], Tamás Zsom[4], Géza Hitka[4]

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Summary

The presented study simulates commercial practice and focuses on the effect of time gap between harvest and the beginning of cold storage. Apple fruit 'Golden Delicious' was harvested in Hungary and randomly separated into 3 groups for precooling at 1, 4 and 10 °C. After 7 d cold storage, groups were randomly split half to control and the other half was subjected to gaseous 1-methylcyclopropene (1-MCP) treatment for 24 h on their cold storage temperature (1, 4 and 10 °C). All samples were stored for 6 months at 1 °C followed by 7 d shelf-life at ambient temperature. Ethylene production, firmness, total soluble solid content, surface color and disorder incidence were determined. Significant correlation was found between color parameters hue and normalized green with firmness, SSC and ethylene production. Precooling temperature and 1-MCP treatment significantly affected apple quality ($p < 0.01$). Initial storage at 10 °C and application of 1-MCP on this temperature had no clear effect on maintaining fruit quality compared to control after 6 months storage. On the other hand, 1 °C and 4 °C precooling and applied 1-MCP treatment could slow the softening of samples during 6 months storage and in the following shelf-life. Apple quality was observed to change faster for group of 10 °C precooling and slower for groups of 1 °C and 4 °C. According to the results, precooling of apple fruit at 1 °C can be recommended in case the 1-MCP treatment is delayed.

Introduction

Apple (*Malus* × *domestica*) is important fruit with annual production of 83 million tonnes (FAO, 2019). It is the second most consumed fruit after banana. The majority of apple production is used for fresh fruit consumption, therefore postharvest technology applied to maintain its best quality is important. Harvested apple is typically stored for months in cold rooms of 0-5 °C using refrigerated air or controlled atmosphere (DeLong et al., 2004; Matthes and Schmitz-Eiberger, 2009). Postharvest technology subject apples to treatment before long term storage to keep both quality and commercial value. Controlled atmosphere of $\%CO_2$:$\%O_2$ of 1.5:1.5 and 5:3 were found to maintain apple firmness after long term storage and shelf-life the best among other combinations (Jeziorek et al., 2010). The controlled atmosphere of 1.5% O_2 and 0.6-0.9% CO_2 at 3 °C and 5 °C was applied on 'Honyecrisp' apple during 6 months storage (DeLong et al., 2004). Desired low O_2 concentration was obtained by flushing N_2 gas into storage chamber. Controlled atmosphere cold storage of 1.5 °C, 1.2% O_2 and 2.5% CO_2 was also reported to successfully maintain polyphenol content in apple (Matthes and Schmitz-Eiberger, 2009).

Ripening of climacteric fruit can be controlled by 1-methylcyclopropene (1-MCP) successfully (Blankenship and Dole, 2003). The advantage of application of 1-MCP is its effect on respiration and ethylene production. As a result, 1-MCP treatment also affects ethylene induced postharvest disorders. Gago et al. (2015) evaluated effect of harvest date and gaseous 1-MCP treatment on apple quality during and after 6 months storage. It was observed that 1-MCP was able to maintain firmness, reduce electrolyte leakage and slow surface color change. During the experiments, apples from certain orchards developed bitter pit at higher rate as a result of 1-MCP treatment. Those fruit were found to have low Ca and high Mg content and therefore bitter pit is more affected by cultivation technique. Authors also concluded that optimal harvest date is essential, affecting both postharvest disorders and following shelf-life. Milinkovic et al. (2018) investigated 5 apple cultivars and did not find significant difference in their K:Ca ratio, but observed fluctuation of mineral content in consecutive years.

Ericsson and Tahir (1996) evaluated possible factors on bruising of 3 cultivars. Pre cooling with cold air of 3-4 °C was found to make fruit more resistant to bruising, but treatment was less effective for late harvested fruit. Harvest date affected bruising sensitivity significantly. Delay of 10 d could increase damage made by bruising with 20-40% in 'Aroma' and 12-25% in 'Ingrid Marie'. The effect of harvest date was found to depend on cultivar. Timing is also important in application of postharvest treatments. DeLong et al. (2004) delayed cooling treatment with a warming period of 20 °C on 'Honeycrisp' apple in order to decrease soft scald and low temperature breakdown. It was found that delayed cooling of 7 d at 20 °C was beneficial and resulted in less disorder incidence during 4-6 months storage in case of early- and late harvested apple. Positive effect of warming period and delay was confirmed by Watkins et al. (2004). They found that soft scald incidence can be decreased with higher storage temperature for sensitive cultivars. It was also highlighted that low production quantity of such sensitive fruit may not be enough to separate them and fill a storage facility to ensure different storage temperature. Delay before cooling was found to be effective, 1 d at 20 °C was able to prevent soft scald and soggy breakdown disorders during storage at 0 and 0.5 °C.

Common parameters measured to monitor apple quality are fruit firmness, soluble solid content and titratable acidity (Lu and Lu, 2016; Watkins et al., 2004). Nondestructive methods are offered by computer vision in wide range from color imaging of surface defects to hyperspectral imaging of quality based on spatial spectral information. Advanced techniques such as X-ray, MRI (magnetic resonance imaging) and CT (computed tomography) can detect internal structural changes, while diffuse reflectance showed its potential in firmness assessment (Lu and Lu, 2016). Surface color measurement with digital image processing is preferred in postharvest handling due to the high speed and ability to deploy in-line. More sophisticated and time consuming methods are utilized primarily in research. The main goal of the presented research was to simulate the commercial application of 1-MCP. During gradual loading of the storage facility, temperature fluctuates and fruit also have different temperature according to the length of their prior storage. This may affect the efficiency of 1-MCP treatment. Therefore, this study evaluated the

* Corresponding author

effect of precooling temperature of 1, 4 and 10 °C as well as 1-MCP treatment on apple quality during 6 months storage. Another objective of the experiment was to analyze relationship between quality indices and color parameters measured by digital image processing.

Materials and methods

Apple fruit and 1-MCP treatment

Apple fruit (*Malus* × *domestica*) of 'Golden Delicious' cultivar have been collected from orchard in Hungary (at city of Ráckeve in Pest county). Samples were collected during middle of harvest season in September 2018. The experimental design was made to simulate whole harvesting procedure including the time gap between picking and long-term storage. The total amount of 900 fruit was selected for experiment and randomly split into 3 groups. Groups were subjected to 7 d cooling at 1 °C, 4 °C and 10 °C. After this initial precooling, groups were divided into two subgroups. Half was kept as control and another half was subjected to gaseous 1-MCP treatment at the precooling temperature. After 1-MCP treatment, apple fruit were stored for 6 months at 1 °C. Forty fruit of each group was left at ambient temperature for 7 d to test their behavior during shelf-life right after the 1-MCP treatment. After long term storage of 6 months, all groups were withdrawn to ambient temperature for 7 d shelf-life. Experimental plan is introduced on flowchart (Fig. 1).

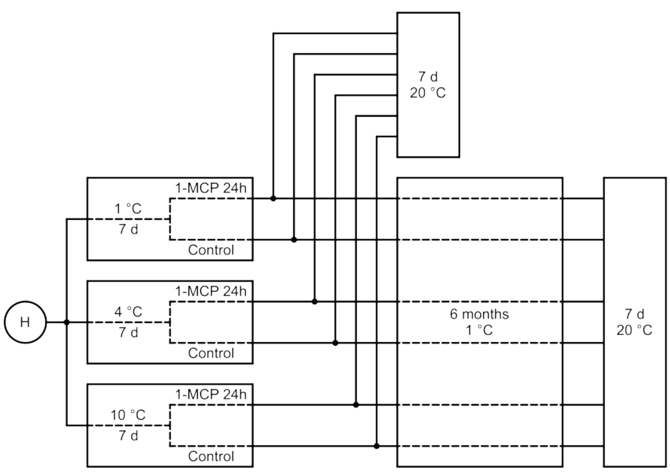

Fig. 1: Experimental plan from harvest (H) with 7 d precooling, 6 months storage and 7 d shelf-life. Steps are simulating: short-term storage in orchard and transportation (7 d, 1-10 °C); long-term storage (6 months, 1 °C); storage in shop and household (7 d, 20 °C).

The gaseous 1-MCP treatment was performed with commercial tablets (0.14% 1-MCP) as an application of the SmartFresh® system, provided by Rohm and Haas Polska Sp.z.o.o. (Warsaw, Poland).

Measurements

Measurements were performed at initial stage 0 d, at 7 d precooling, after 7 d shelf-life following 1-MCP treatment, at the end of 6 months storage and 7 d shelf-life following the 6 months storage. Starch index was measured at 0 d and after 7 d precooling. The ethylene production, firmness, soluble solid content (SSC), disorder incidence, and surface color were measured at 20 °C during the experiment.

Starch index

Starch index was determined using Lugol's solution (aqueous iodine) on half cut fruit and surface pattern was compared to starch index reference guide.

Ethylene production

Ethylene production was determined by an ICA-56 handheld ethylene analyzer (International Controlled Atmosphere Ltd., UK). Apple was placed in a hermetically closed plastic container of 4 L for 1 h before measurement was performed. Measurement was repeated in triplicates. Results were expressed in microliter gas produced per kilogram of fruit in one hour (μL kg^{-1} h^{-1}) calculated on fresh weight basis.

Soluble solid content

Soluble solid content (SSC, %) was determined by a hand-held temperature-compensated ATAGO PAL-1 digital refractometer (Atago Co. Ltd., Tokyo, Japan).

Firmness measurement

Fruit firmness was recorded with a handheld fruit firmness tester (FT 327, Italy). The instrument was mounted on a stand to make measurement more stable. Cylindrical probe of 10 mm diameter was penetrated into the tissue of peeled apple until 10 mm depth. The maximum force was measured at two opposite points on the external circumference. The average was recorded for each fruit. The instrument provided values in kg cm^{-2}, what was transformed to the unit of N.

Disorder incidence

Fruit were visually tested for rot and bitter pit on the skin during storage period. The disorder was determined when a sign of those symptoms occurred. The incidence was calculated as percentage of the total number of fruit.

Surface color

Digital images of HD size (High Definition, 1920 × 1080 pixel) and 24 bit/pixel color depth were recorded with a camera (Samsung WB350F). The optical zoom of 4× was applied. Apples were placed on white paper and this background was used as white color reference during processing. The ROI (region of interest) was selected based on saturation with the threshold of 0.2. Recorded color information was transformed into HSL (hue, saturation, luminosity) space. Green color dominance was calculated as well, with linear normalization (Eq. 1).

$$g_n = G / (R + G + B) \quad (1)$$

where g_n is the normalized green component, R, G and B are red, green, blue intensity values, respectively.

Statistical analysis

All data were subjected to statistical analysis with R version 3.6.0 (R Foundation for Statistical Computing, Austria) using analysis of variance (ANOVA). The effect of the factors of precooling temperature and storage time was evaluated. The ANOVA F value was used to compare effects to the natural variability of collected data. Tukey's method was used as post-hoc test to compare selected groups with $p<0.05$. The results are reported on charts with mean and standard deviation (SD). Figures were created using Microsoft Excel (Microsoft Co., USA).

Results and discussion

Effect of precooling on quality indices

The comparison of initial readings with measurements after 7 d precooling (Tab. 1) showed that apple subjected to 10 °C precooling changed more than that of 1 °C and 4 °C. During the 7 d precool-

Tab. 1: Quality parameters before and after precooling treatment (mean value ± SD)

Parameter	0 d	7 d, 1 °C	7 d, 4 °C	7 d, 10 °C
Starch index	6.51 ± 0.22[a]	7.15 ± 0.24[ab]	7.82 ± 0.21[bc]	8.75 ± 0.27[c]
Soluble solid content, %	12.87 ± 0.22[a]	13.30 ± 0.16[a]	13.20 ± 0.16[a]	13.40 ± 0.16[a]
Firmness, N	72.81 ± 2.52[a]	71.15 ± 2.52[a]	69.32 ± 1.89[a]	66.25 ± 1.89[a]
Hue angle	108.24 ± 0.94[a]	105.12 ± 1.28[ab]	103.45 ± 1.19[bc]	100.25 ± 0.91[bc]
Green dominance, %	44.79 ± 0.73[a]	43.59 ± 0.22[a]	43.71 ± 0.29[a]	44.12 ± 0.48[a]

Different superscript letters in a row show significant difference at $p<0.05$.

ing after harvest, starch index and SSC increased while firmness decreased. On the other hand, no significant difference was observed among fruit according to SSC and firmness. Regarding color attributes, Hue angle value decreased indicating that surface color started to change toward yellow. The green dominance was still similar after precooling, which means that fruit color changed within the green range.

Ethylene production

Ethylene measurements were conducted to evaluate the effect of precooling and 1-MCP application on postharvest life of apple. Precooling temperatures affected the ethylene production. Apples kept at 1 °C and 4 °C showed lower ethylene production during shelf-life both after 1-MCP treatment and after storage than those at 10 °C (Fig 2).

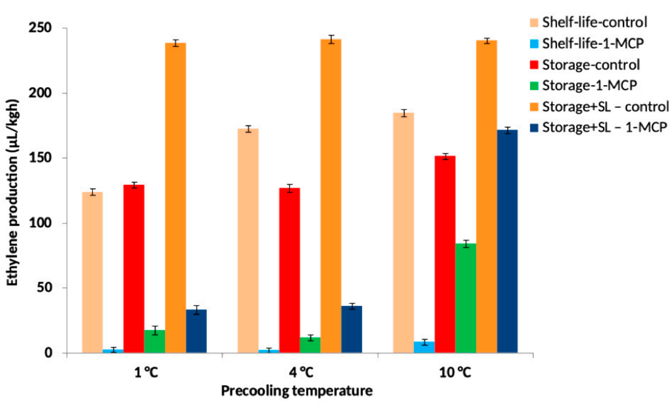

Fig. 2: Ethylene production during experiment (SL: shelf-life). Presented values are mean ± SD.

The 1-MCP inhibited strongly the ethylene production. After 6 months of storage and during the following shelf-life, treated samples showed lower values than control but at different rate. Fruit of 1 °C and 4 °C precooling treated with 1-MCP had lower level in ethylene production than those of 10 °C. At the same time, all control fruit showed high ethylene production, with increasing value from 1 °C precooling temperature to 10 °C. Samples of 1 °C or 4 °C precooling were not significantly different in ethylene production.

Firmness

After 7 d of precooling, firmness decreased slightly for fruit kept at 1 °C and 4 °C, whereas apple from 10 °C had significantly lower values compared to the initial time (Tab. 1). Firmness declined dramatically for all groups throughout shelf-life and storage. The results also showed that 1-MCP reduced the softening of apple for all temperature groups compared to control during the experiment but at different rates (Fig. 3).

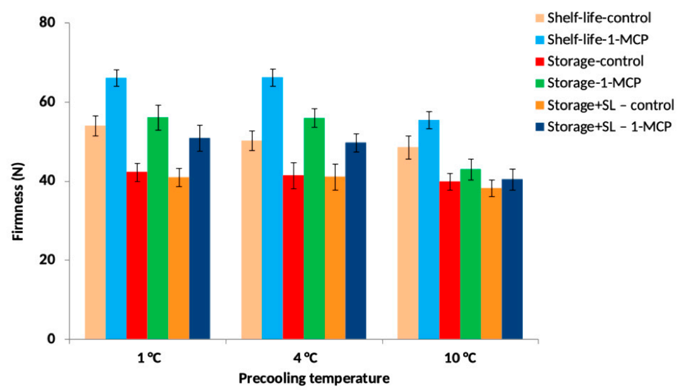

Fig. 3: Firmness during experiment (SL: shelf-life). Presented values are mean ± SD.

Precooling temperature influenced the efficiency of 1-MCP application. 1-MCP strongly affected only the fruit of 1 °C and 4 °C groups. There was no significant difference in firmness between 1 °C and 4 °C precooling either at storage or shelf-life, whereas 1-MCP treated fruit from 10 °C group were much softer. 1-MCP treatment did not show expected effect on apple of 10 °C since no significant difference was detected between control and 1-MCP treated fruit using this precooling temperature.

Soluble solid content

Apple obtained similar SSC values after 7 d precooling in all groups (Fig. 4). However, it increased slightly during shelf-life before storage and after storage.

Fruit subjected to 10 °C precooling had higher values in SSC after 6 months of storage, then declined in the following shelf-life. Nonetheless, there were significant changes and significant differences among groups throughout the experiment.

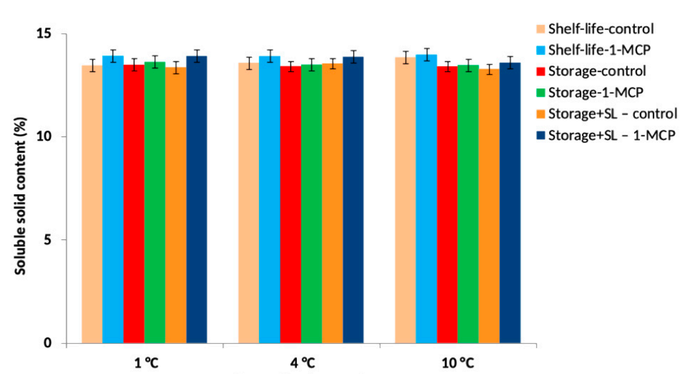

Fig. 4: Soluble solid content during experiment (SL: shelf-life). Presented values are means ± SD.

Bitter pit and rot

The disorder of bitter pit was detected more frequently in 1-MCP treated samples than in control after storage (Tab. 2). After precooling and shelf-life before long term storage, no bitter pit symptom was observed. There were no significant differences in bitter pit occurrence among groups at the end of storage and in the following shelf-life.

The rot infection was detected less frequently for 1-MCP treated fruit than for control in all temperature. The symptom of fungal development only occurred after 6 months of storage. Samples of 1 °C and 4 °C precooling suffered less decay than those of 10 °C (Tab. 2). Fruit subjected to precooling at 1 °C showed the lowest level of bitter pit and rot, while at 10 °C it became very high. No significant difference was observed in bitter pit and rot occurrence between 1-MCP treated and control apples for group of 10 °C.

Surface color

The color development of apple surface was expressed by hue angle values. The surface color of apple continuously changed during the experiment according to the mean value of groups, but no significant difference was found between initial values and that of after 7 d precooling. The hue angle decreased and standard deviation increased after storage for all groups (Fig. 5). The standard deviation was similar for all groups after shelf-life following both precooling and long term storage.

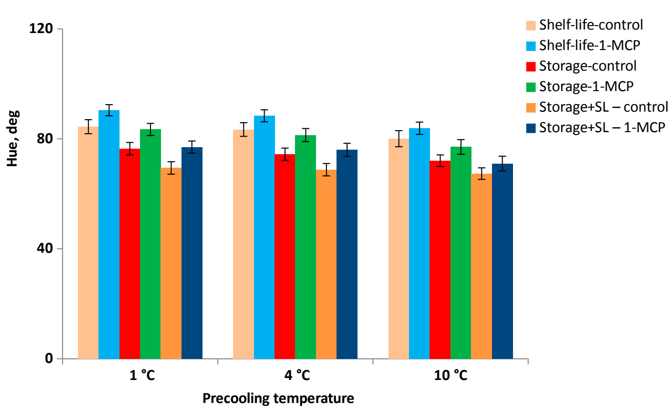

Fig. 5: Hue angle value during experiment (SL: shelf-life). Presented values are means ± SD.

Similar tendency was observed for green dominance. Less difference was found in green dominance before 6 months storage. Green color became less dominant after shelf-life following both precooling and long term storage. Together with decreasing hue angle, they indicate that color changed into the direction of yellow.

Correlation between parameters

Pearson's correlation coefficient was calculated between parameter pairs (Tab. 3). The highest correlation was observed between hue angle and firmness (r = 0.9597). Similarly high correlation was found between firmness and starch index, but starch index is not included in the overall comparison because it was measured until the beginning of long term cold storage. Firmness was highly correlated (r = -0.7715) with ethylene production according to the expectations. Among quality parameters, firmness gained the highest correlation with color, r = 0.9597 for hue angle and r = 0.7721 for green dominance. Ethylene production and SSC obtained lower but still significant correlation values with color.

Tab. 3: Correlation between measured parameters

	Ethylene	SSC	Hue	g_n
Firmness	-0.7715**	-0.3425	0.9597**	0.7721**
Ethylene	1	-0.0256	-0.6733*	-0.4829$^+$
SSC		1	-0.5199*	-0.3294
Hue			1	0.8198**

** significant at $p < 0.001$; * significant at $p < 0.01$; $^+$ significant at $p < 0.05$

Discussion

Maturity stage is the most important factor influencing the efficiency of 1-MCP treatment. The efficacy of 1-MCP is low at advanced maturity stage (Jia et al., 2014; Sozzi and Beaudry, 2007). Our results also indicated that the effectiveness of 1-MCP treatment depends on fruit maturity, in agreement with the previous report (Jia et al., 2014). Application of 1-MCP on apple subjected to 10 °C precooling had a little effect in reducing ethylene production after 6 months storage, while 1-MCP treatment on fruit of 1 °C and 4 °C precooling was effective in suppressing the ethylene production. The reason for this behavior might be that samples of 10 °C precooling for 7 d reached the advanced maturity, therefore 1-MCP did not show the efficacy as strong as other groups. According to this observation, if delay in 1-MCP treatment cannot be avoided due to transportation or optimization of bulk treatment, short-term cold storage at 1 °C is recommended.

The decline in firmness during storage results in soft and mealy fruit decreasing the quality value of fruit (Kader, 1999). In our work, the softening of fruit increased during the precooling period from 1 °C to 10 °C corresponding to the increase of starch index due to advanced ripening (Tab. 1). 1-MCP treatment on fruit of 1 °C and 4 °C precooling delayed the softening of apple during storage and shelf-life, whereas the softening of control samples rose rapidly. Similar behavior was observed for pear using gaseous 1-MCP of 1.0 mL L^{-1} (Gao et al., 2015). Less efficacy of 1-MCP at 10 °C precooling was found perhaps due to incomplete blocking of the ethylene receptors,

Tab. 2: Bitter pit and rot occurrence after 6 months storage and after shelf-life

		After storage		After storage + 7 d shelf-life	
Precooling	Treatment	Bitter pit, %	Rot, %	Bitter pit, %	Rot, %
1 °C	Control	8.45	9.86	13.49	11.32
	1-MCP	10.69	7.33	16.74	9.12
4 °C	Control	8.96	10.45	14.22	12.29
	1-MCP	11.85	7.84	17.48	9.43
10 °C	Control	12.89	12.39	20.83	15.79
	1-MCP	14.97	11.92	21.75	14.61

thus ethylene could exert its action partly in ripening (Blankenship and Dole, 2003; Watkins, 2008). Regarding to delayed 1-MCP treatment, previous reports also indicated that the applied treatments at earlier maturity stage increased the possible storage periods (Kubo et al., 2003; Watkins and Nock, 2005).

Depending on the cultivar and circumstances of treatment, the 1-MCP treated fruit can have higher, lower, or same values of soluble solid content compared to control samples (Gago et al., 2015). Our results showed that 1-MCP had small effect on SSC and SSC of fruit of 10 °C precooling was lower than those at 1 °C and 4 °C at the end of experiment, but not significantly different.

Surface color was found to change gradually from green toward yellow. Hue angle decreased in this direction but green dominance measured by normalized green value did not change significantly. Apple color in control group changed more than groups of 1-MCP treatment. Similar results were found for 'Red chief Delicious' apples (Mir et al., 2001). Ethylene plays an important role in the ripening process. 1-MCP inhibited the ripening by occupying ethylene receptors, so that ethylene is unable to elicit its action (Blankenship and Dole, 2003). Other fruit, including papaya and melon also showed similar results (Bron et al., 2004; Ergun et al., 2005). Color parameters were found to correlate strongly with firmness, followed by ethylene production and SSC.

The percentage of bitter pit was higher in 1-MCP treated fruit than control. In earlier reports, similar results were found for 'Golden Delicious' apple (Gago et al., 2015), 'Granny Smith' apple (Calvo and Candan, 2010). The symptom of bitter pit related to the Ca, K, Mg concentration and cultural technique (Gago et al., 2015). Rot is also one of the main problems during postharvest transport and storage. As shown in Tab. 2, fruit was more susceptible to decay at the end of experiment due to aging and senescence. In addition, ambient temperature during shelf-life also favor the microbial growth (Miccolis and Saltveit, 1995; Yang et al., 2003).

Conclusions

Precooling treatment of 7 d was applied on apple fruit 'Golden Delicious' before gaseous 1-MCP treatment, simulating commercial practice. Results show that 1-MCP treatment successfully delayed fruit ripening, but differences were observed depending on the short-term storage temperature between harvest and treatment. Precooling temperature of 1 °C and 4 °C were more effective, while samples subjected to 10 °C did not respond sensitively to 1-MCP by means of ethylene production, firmness and surface color. The 1-MCP treatment should be applied as soon as possible to prevent progressed aging. In case transportation or optimization of bulk treatment make several days delay, short-term cold storage at 1 °C should be applied to maintain quality and effectiveness of 1-MCP treatment.

Color parameters correlated significantly with fruit firmness, SSC and ethylene production. This observation suggests that surface color can follow changes during ripening. The normalized green color component shown similar behavior to hue angle offering another parameter to use.

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Conflict of interest

No potential conflict of interest was reported by the authors.

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ORCID
László Baranyai https://orcid.org/0000-0001-6177-7364
Lien Phuong Le Nguyen https://orcid.org/0000-0002-5975-9906
Mai Sao Dam https://orcid.org/0000-0002-3170-0785
Géza Hitka https://orcid.org/0000-0003-0942-8102

Adress of the corresponding author:
E-mail: Baranyai.Laszlo@etk.szie.hu