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Comparison of 1-MCP treatment on four melon cultivars using different temperatures

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Summary

The aim of this work was to evaluate the effect of 1-methylcyclopropene (1-MCP) treatment on musk melons (Cucumis melo L. var. reticulates Naudin) using different temperatures during the treatments. Three application temperatures (5, 10 and 20 °C) and four melon cultivars ('Centro', 'Lillo', 'Donatello' and 'Celestial') were investigated. Three groups of each cultivar were treated with 625-650 nL L⁻¹ gaseous 1-MCP for 24 h at 5, 10 and 20 °C. During the 24 h long gaseous 1-MCP treatment, the control group was kept at 5 °C. After treatment, all samples were stored at ambient temperature (20 °C) for 10 d. The results showed that 1-MCP treated melons released less ethylene and CO2 compared to controls. Moreover, 1-MCP application could slow the softening as well as the color change of melon throughout shelf-life in comparison to controls. No significant differences were observed among 1-MCP treatment temperatures for four melon cultivars. 1-MCP did not reduce disease severity of treated melons.

Keywords: 1-methylcyclopropene, cantaloupe, storage, shelf-life, *Cucumis melo*

Introduction

Cantaloupe (*Cucumis melo* L. var. *reticulates* Naudin) is one of the most common melon varieties due to its attractive orange flesh, unique flavor and nutritional value (AGUAYO et al., 2007; SILVEIRA et al., 2008). However, short-term shelf-life due to microbial growth and deterioration in appearance and nutritional value result in losing market value (FALLIK et al., 2000). Thus, the time for transport, sale and consumption of melon is limited and this produce is used primarily for fresh-market consumption (ABADIAS et al., 2014).

1-Methylcyclopropene (1-MCP) is an ethylene action inhibitor (SISLER, 2006). 1-MCP successfully controls the ripening of fruit and vegetable during storage and transport by preventing negative ethylene effects (SISLER, 2006). Recently, many studies were published about the effects of 1-MCP in delaying ripening and maintaining texture, firmness, taste and appearance of fruit (ZHAO et al., 2018; CHENG et al., 2019). Responses of melon to 1-MCP were also reported. 1-MCP had strong effect on 'Galia' melon at intermediate to advanced stages of ripening (ERGUN et al., 2005). Those authors found that the change of surface color was dramatically delayed by 1-MCP. 1-MCP retained the firmness of intact and fresh-cut melon compared to control (GAL et al., 2006; ERGUN et al., 2007). In addition, the symptom of water soaking in fresh-cut melon was also postponed by 1-MCP (ERGUN et al., 2007). Fresh-cut 'Charentais' melon treated with 1-MCP accumulated higher values of aroma volatiles than untreated samples (AMARO et al., 2012). The benefit of 1-MCP in extending the shelflife of muskmelon was to retain firmness and soluble solid content, suppress the overripe flavor development (AGEHARA et al., 2018). In literature, most of the studies focused on the effect of 1-MCP concentration (ALVES et al., 2005; ERGUN et al., 2005; DE MELO et al., 2008), different treatment duration (GAL et al., 2006) or application form including gaseous 1-MCP and aqueous 1-MCP (AMARO et al., 2012; SHI et al., 2014; BAI et al., 2014; AGEHARA et al., 2018).

In addition, 1-MCP treatment on melon was mainly carried out at ambient temperature within a day after harvest (ALVES et al., 2005; ERGUN et al., 2005; ERGUN et al., 2007; GAL et al., 2006; SHI et al., 2014; AGEHARA et al., 2018). The 1-MCP treatment is commonly performed at 20-25 °C for 12-24 h in experiments to reach desired effects on crops (BLANKENSHIP and DOLE, 2003). 1-MCP may reach ethylene receptors more easily at higher temperature (WATKINS, 2016). For example, 1-MCP treatment affected broccoli (*Brassica oleracea*) at both 5 and 20 °C, however, it showed better effectiveness at 20 °C (KU and WILLS, 1999). In case of penstemon (*Penstemon hartwegii*), 1-MCP treatment at 2 °C was not effective whereas application at 20 °C completely controlled ripening (SEREK et al., 1995). For coriander (*Coriandrum sativum*), 1-MCP did not yield benefit at low application temperature (5-10 °C) because of low affinity to the ethylene receptors (JIANG et al., 2002).

Nevertheless, in commercial practice, it is not easy to carry out the 1-MCP treatment at the harvest day due to transport or occasional lack of the air tight storage room (WATKINS and NOCK, 2005). In commercial facilities, fruit are amassed in storage rooms at cold temperature after harvest. The 1-MCP treatment is carried out when rooms are filled to desired level. Therefore, commercial utilization of 1-MCP is usually conducted at cold temperature (WATKINS and NOCK, 2005). Moreover, melon has a high rate of respiration and senescence when storage temperature is above 5 °C after harvest (EDWARDS and BLENNERHASSETT, 1994). Therefore, precooling followed by 1-MCP application at cold temperature is the most suitable. Although there are several studies published about 1-MCP application on melon, information about response to different treatment temperature is still limited.

The aim of this study was to investigate the effect of 1-MCP treatment temperature (using 5, 10 and 20 °C) on four melon cultivars ('Lillo', 'Centro', 'Donatello' and 'Celestial') during shelf-life at 20 °C. The results of this work provide useful information for commercial application of 1-MCP.

Materials and methods

Materials

Fruit of four cultivars ('Lillo', 'Centro', 'Celestial' and 'Donatello') of the muskmelon variant (*Cucumis melo* L. var. *reticulates* Naudin) were harvested at the half slip stage in July 2017 and 2018. Cultivars were selected according to their popularity and high production quantity in Hungary. Fruit were selected for uniformity of size, shape and absence of external damage. Small and large diameters of melons were 12.0 ± 0.3 cm and 14.0 ± 0.2 cm, respectively. Harvested

fruit were transported to the laboratory (Budapest, Hungary) and treatments were applied within 24 h of harvest. Fruit temperature was 20 ± 1 °C during transportation. For each cultivar, melons were randomly divided into four groups (15 fruit per group). Fruit of three groups were subjected to 1-MCP treatment at different temperatures and the others were stored as controls. The shelf-life was monitored and quality measurements were performed at 20 °C.

The 1-MCP (0.14 % 1-MCP tablet, AgroFresh, USA) as an application of the SmartFresh[®] system was provided by Rohm and Haas Polska Sp.z.o.o. (Warsaw, Poland).

1-MCP application

Melons were kept at 5, 10 and 20 °C, respectively, for 24 h before treatment. Three groups were treated with 625-650 nL L⁻¹ gaseous 1-MCP in an air-tight plastic box of 500 L at 5, 10 and 20 °C, respectively, on the 1st day after harvest for 24 h. The control group (untreated) was kept at 5 °C during this period. After treatment, the samples of the four groups were stored at room temperature (20 °C) and at relative humidity (RH) of 55%.

Measurements

Measurements of stiffness, ethylene production, respiration rate, rind color, chlorophyll fluorescence and disease severity were performed according to NGUYEN et al. (2019). All parameters except disease severity were measured at the initial time of 0 d, followed by 5 d and 10 d. Disease severity was assessed at 4 d, 7 d and 10 d. During the experiment, non-destructive tests were performed with 15 randomly selected fruit each interval. Respiration was evaluated in triplicates due to limitation of resources.

Stiffness

Fruit firmness was estimated using the acoustic vibration method (CHEN and DE BAERDEMAEKER, 1993) and expressed as the parameter Stiffness (S, $g^{2/3} s^{-2}$). Stiffness of the samples was determined at 2 points on the exterior circumference of each fruit, using an AFS DTF V0.0.0.105 acoustic firmness instrument (AWETA, Nootdorp, The Netherlands).

Ethylene production

Ethylene production was determined by an ICA-56 handheld ethylene analyzer (International Controlled Atmosphere Ltd., Tonbridge, UK). One melon 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 fresh mass in one hour (μ L kg⁻¹ h⁻¹).

Respiration rate

Carbon dioxide production was measured in a closed respiratory system for 1 h. The system was built with hermetically closed acrylic sheet containers equipped with FY A600-CO2H carbon dioxide sensors (Ahlborn Mess-und Regelungstechnik GmbH, Holzkirchen, Germany). Measurement was repeated in triplicates. Changes in CO₂ concentration was recorded with an Almemo 3290-8 data logger (Ahlborn Mess-und Regelungstechnik GmbH, Holzkirchen, Germany) and results were expressed as mL kg⁻¹ h⁻¹.

Surface color

Melon rind color was measured with a portable Minolta Chroma Meter CR-400 (Minolta Corporation, Osaka, Japan). Standard CIE L*, a* and b* color characteristics were determined at three points on the external circumference of each fruit. The hue angle value was calculated as arctangent of $b*/a^*$.

Chlorophyll fluorescence

Chlorophyll fluorescence was determined at three equidistant points on the equator of each fruit by a PAM WinControl-3 controlled MONI-PAM multi-channel chlorophyll fluorometer (Heinz Walz GmbH, Effeltrich, Germany). From the recorded minimal and maximal chlorophyll fluorescence signals (F_0 , F_m) the potential maximum quantum yield of the photosystem II (F_v/F_m) was calculated.

Disease severity

Melons were examined for mold growth on the rind or stem during storage. Infection was expressed on the scale of 1-3, where 1 = good, fruit without decay; 2 = fair, fruit with moderate decay; 3 = bad, fruit with severe decay. Decay was visually assessed according to the total area of infection on rind or stem. Disease severity was calculated as an average score of all melon within a group (YANG et al., 2003).

Statistical analysis

All data were subjected to statistical analysis with IBM SPSS version 22 (IBM Corp, New York, USA) using analysis of variance (ANOVA). The effects of cultivar, treatment temperature and storage time were evaluated. The ANOVA F value was used to compare effects to natural variability of readings. Tukey's method was used as post-hoc test to compare groups with p<0.05. On figures, results are reported as means and standard deviations.

Results

Ethylene and CO₂ production

The initial ethylene concentrations largely varied among fruit of the four cultivars, ranging from 36 μ L kg⁻¹ h⁻¹ to 46 μ L kg⁻¹ h⁻¹ (Fig. 1). Fruit of 'Lillo' had the lowest ethylene production followed by those of 'Donatello', 'Celestial' and 'Centro'. Ethylene production of both untreated and treated fruit decreased during storage, but at different rates.

Rates of ethylene release of controls gradually declined during storage (Fig. 1), while that of treated fruit pronouncedly decreased after treatment, obviously reflecting a direct response to 1-MCP. Observed differences in rates of ethylene release and its production kinetics between fruit of the four cultivars were significant (p<0.001; Tab. 1). According to the observed drop in ethylene production, fruit of cultivars 'Centro' and 'Donatello' responded more sensitively to 1-MCP than the others.

There were no significant differences between the effects of treatment temperature on ethylene production of fruit of the four cultivars. The initial carbon dioxide release rates (approximately 28 mL kg⁻¹ h⁻¹) were almost the same for fruit of the four melon cultivars (Fig. 2). However, during the experiment, the carbon dioxide production of 'Lillo' fruit decreased more sharply than that of other cultivars (Fig. 2).

According to ANOVA, cultivar but not treatment temperature affected respiration significantly (p<0.001; Tab. 2).

Again, 1-MCP treatment but not temperature affected CO_2 production of treated fruit compared to that of controls. In this context, sensitivity of ethylene production to 1-MCP treatment was 4.4 times higher than that of CO_2 production. Ethylene and CO_2 production followed similar tendency, their values strongly correlated during the experiment (r = 0.920).

Stiffness

The stiffness of all melons declined throughout storage. However, softening of 1-MCP treated fruit was pronouncedly lower than that of untreated samples (Fig. 3). This clearly indicated that 1-MCP suppressed softening of fruit.



Fig. 1: Ethylene production of fruit of four melon cultivars during shelf-life at 20 °C. Data are shown as means ± SD.



Fig. 2: Carbon dioxide production of fruit of four melon cultivars during shelf-life at 20 °C. Data are shown as means ± SD.

Tab. 1: Statistical evaluation of ethylene production (ANOVA)

Factors for Ethylene	Main effect	Interaction effect	
,		× Temperature	× Cultivar
Time	***	_	***
Treatment	***	-	***
Temperature	-	-	_
Cultivar	***	-	-

Tab. 2: Statistical evaluation of CO₂ respiration (ANOVA)

Factors for CO ₂	Main effect	Interaction effect	
		× Temperature	× Cultivar
Time	***	_	***
Treatment	***	-	**
Temperature	_	-	_
Cultivar	***	-	-

*** p<0.001, ** p<0.01, * p<0.05, N = 144

*** p<0.001, ** p<0.01, * p<0.05, N = 144



Fig. 3: Stiffness of melon during shelf-life at 20 °C. Data are shown as means ± SD

Tab. 3: Statistical evaluation of Stiffness (ANOVA)

Factors for Stiffness	Main effect	Interaction effect	
		× Temperature	× Cultivar
Time	***	**	***
Treatment	***	-	**
Temperature	*	_	_
Cultivar	***	-	-

*** p<0.001, ** p<0.01, * p<0.05, N = 720

According to the ANOVA (Tab. 3), effects on fruit stiffness of 1-MCP treatment and cultivar but not of temperature were significant (p<0.001). Only fruit of 'Donatello' responded significantly to changes in temperature (p<0.05) but this was less than 1% of the effect of 1-MCP treatment (ANOVA F=3.3 << 370.1). Stiffness values correlated strongly with maximal chlorophyll fluorescence (F_m , r = 0.976) and efficiency of Photosystem II (F_v/F_m , r = 0.938).

Chlorophyll fluorescence parameters

During storage, treated melons retained higher F_v/F_m than controls (Fig. 4). The results showed that 1-MCP treatment clearly affected F_v/F_m (p<0.001). Additionally, cultivar was found to have only weak interaction effect with 1-MCP treatment (ANOVA F = 5.3, p<0.01). According to the statistical analysis, primarily storage time and 1-MCP treatment contributed to changes in chlorophyll fluorescence (p<0.001; Tab. 4). Besides the strong relationship with stiffness, F_v/F_m obtained significant correlation with hue angle (r = 0.871; p<0.001). This agreement between surface color and green pigment measurements was expected.

At the end of the experiment, controls of all cultivars had the lowest F_0 and F_m (data not shown). Obviously, chlorophylls of untreated samples were more rapidly degraded than that of treated fruit, indicating the retardation of ripeness and/or senescence.

Tab 4: Statistical evaluation of fluorescence F_V/F_M (ANOVA)

Factors for F _V /F _M	Main effect	Interaction effect	
		× Temperature	× Cultivar
Time	***	_	_
Treatment	***	-	**
Temperature	_	-	-
Cultivar	-	-	-

*** p<0.001, ** p<0.01, * p<0.05, N = 720

Hue angle value

The hue angles of all samples decreased during shelf-life at 20 °C (Fig. 5). The color of the rind of controls turned to yellow more rapidly than that of treated samples. Thus, this color change is often used as an indicator of ripening (DONG et al., 2002). This is also supported by our results as hue angle significantly correlated with chlorophyll fluorescence and stiffness (r = 0.898; p<0.001). Both 1-MCP treatment and cultivar but not temperature significantly affected (p<0.001) hue angles (Tab. 5). Temperature had only minor interactive effect with cultivar (ANOVA F = 2.4, p<0.05).

Disease severity

Ripe fruit can be more sensitive to microbial infection due to softer texture and changing compounds, therefore 1-MCP treatment might affect disease severity, too. In the presented experiment, 1-MCP treatment showed smaller effect on disease severity (ANOVA F = 7.2, p < 0.01) compared to storage time and cultivar (Tab. 6).

Severity of disease of all fruit increased with increasing treatment temperature during prolonged storage (c.f. days 7 and 10; Tab. 7). Severity of disease was always highest in the controls of all cultivars and difference among groups were not significant. Nevertheless, the susceptibility of melons depended on the cultivar. 'Lillo' and 'Donatello' fruit better retained appearance than those of 'Celestial'



Fig. 4: F_v/F_m of fruit of the four melon cultivars during shelf-life at 20 °C. Data are shown as means \pm SD



Fig. 5: Hue angle value of four melon cultivars during shelf-life at 20 °C. Data are shown as means ± SD

Tab. 5: Statistical evaluation of hue angle (ANOVA)

Factors for Hue angle	Main effect	Interaction effect	
		× Temperature	× Cultivar
Time	***	_	***
Treatment	***	-	*
Temperature	_	-	*
Cultivar	***	-	-

Tab. 6: Statistical evaluation of disease severity (ANOVA)

Factors for disease severity	Main effect	Interaction effect	
		× Temperature	× Cultivar
Time	***	_	_
Treatment	**	-	_
Temperature	_	-	-
Cultivar	***	-	-

*** p<0.001, ** p<0.01, * p<0.05, N = 720

*** p<0.001, ** p<0.01, * p<0.05, N = 720

and 'Centro' after 10 d of shelf-life.

At the end of the experiment, disease severity was high for all samples. It might be the result of initial infection from soil and lack of surface cleaning. Microorganisms present on rind surface can develop rapidly during transport and storage (BASTOS et al., 2005), especially when temperature promotes microbial growth (YANG et al., 2003).

Tab. 7: Disease severity of melon cultivars during shelf-life at 20 °C

Cultivars	Sample	4 th day	7 th day	10 th day
Lillo	1-MCP _{5°C}	1.0	1.33 a	2.27 a
	1-MCP _{10°C}	1.0	1.40 a	2.33 a
	1-MCP _{20°C}	1.0	1.53 a	2.47 a
	Untreated	1.0	1.60 a	2.53 a
Donatello	1-MCP _{5°C}	1.0	1.33 a	2.33 a
	1-MCP _{10°C}	1.0	1.47 a	2.33 a
	1-MCP _{20°C}	1.0	1.47 a	2.40 a
	Untreated	1.0	1.53 a	2.47 a
Centro	1-MCP _{5°C}	1.0	1.53 a	2.53 a
	1-MCP _{10°C}	1.0	1.60 a	2.60 a
	1-MCP _{20°C}	1.0	1.67 a	2.60 a
	Untreated	1.0	1.90 a	2.70 a
Celestial	1-MCP _{5°C}	1.0	1.53 a	2.53 a
	1-MCP _{10°C}	1.0	1.67 a	2.47 a
	1-MCP _{20°C}	1.0	1.67 a	2.60 a
	Untreated	1.0	1.80 a	2.70 a

Subscripts indicate 1-MCP treatment temperature. Means followed by the same letters in columns are not significantly different, p<0.05.

Discussion

Our results showed that 1-MCP reduced the respiration rate, ethylene production and pulp softening. It was coincident with earlier report for 'Galia' melon (DE LIMA et al., 2004; GAL et al., 2006). Those authors found that fruit treated with 1-MCP for 12 h at 25 °C was firmer and lower in carbon dioxide production compared to control. However, mass loss, total titratable acidity, pH, soluble solids content, total soluble sugars content were not affected by 1-MCP. Similar results were observed in our study. At the end of measurement, the mass loss of melon was about 6% (data not shown), and the soluble solid content around 7.5% (data not shown) for all samples. Nevertheless, the effect of 1-MCP on mass loss, soluble solid content also depends on cultivar. For example, there was a difference in mass loss between treated and untreated fruit for 'Orange Flesh' and 'Charantais' melon, but not for 'Galia' melon (ALVES et al., 2005; DE MELO et al., 2008). Similarly, 1-MCP treatment decreased the soluble solid content for 'Orange Flesh' melon, while 1-MCP had no effect on soluble solid content for 'Galia' and 'Charantais' melon (ALVES et al., 2005).

In our work, there was no difference between three different treatment temperatures for all cultivars. Studies of 1-MCP application on melon were usually carried out at ambient temperature (ALVES et al., 2005; DU CHATENET et al., 2000; ERGUN et al., 2007; GAL et al., 2006; SHI et al., 2014). Earlier publications reported that 1-MCP treatment at cold temperature had less effect than at warm temperature. The reason was found to be the lower affinity of ethylene receptor to 1-MCP at cold temperature (BLANKENSHIP and DOLE, 2003). 1-MCP delays the ripening by occupying ethylene receptors, so that ethylene is unable to elicit its action. The application time of 12-24 h is generally considered long enough to reach full response (BLANKENSHIP and DOLE, 2003). Application of gaseous 1-MCP at high concentration was also reported to induce desired effect, such as 1 mL L⁻¹ for pear (GAO et al., 2015). The effect of 1-MCP also depends on factors such as cultivar (WEI et al., 2019), stage of maturity, concentration, temperature and treatment time (WATKINS, 2006). In case of apple, DEELL et al. (2002) reported a relationship between temperature and time. The effective exposure time of apple at 3 °C was 9 h, while the equivalent time required to reach the same effect at 23 °C was only 6 h. Similarly, a 6 h 1-MCP treatment of apple at 20 °C had the same effectiveness as a 24 h treatment at 0 °C and 5 °C (DAUNY and JOYCE, 2002). However, WATKINS and NOCK (2005) did not find difference between 24 h 1-MCP treatment of warm and of cold fruit of four apple cultivars.

In the presented study, 1-MCP treatments markedly reduced the ethylene and carbon dioxide production in fruit of all four melon cultivars. This observation is in agreement with a prior study on melons (ALVES et al., 2005). Similar results were also reported for other muskmelon varieties, and 'Galia' (ERGUN et al., 2007; GAL et al., 2006) and 'Charentais' melons (DU CHATENET et al., 2000). In addition, application of 1-MCP maintained the firmness of melon. The efficiency of 1-MCP in retaining the firmness was found for other fruits including tomatoes (WILLS and KU, 2002), pears (KUBO et al., 2003; GAO et al., 2015), apples (MILINKOVIC et al., 2018), apricots and plums (DONG et al., 2002). Pulp softening, respiration and ethylene production of muskmelon 'Solar King' was affected by 1-MCP treatment, but did not affect pH, total titratable acidity, total soluble solid content, total soluble sugar content, mass loss and external appearance (DE LIMA et al., 2004). An earlier research also indicated that 1-MCP retards the decline in fluorescence parameters of apple during storage (MIR et al., 2001), in agreement with the present study. Our data show that 1-MCP delayed the ripening of four melon cultivars, but 1-MCP treatment did not inhibit the microbial development on the melon surface. However, another report found that the combination of 1-MCP and cold storage decreased rot of 'Orange Flesh' melons (ALVES et al., 2005). Another study also indicated that 1-MCP could decrease the decay by inhibiting the fruit softening of 'New Queen' melon. Delaying the fruit softening could slow down the development of microorganisms in the fruit flesh, thus decrease the decay incidence of melon (ZHANG et al., 2019). It was concluded in that study that fruit softening is favorable for the reproduction and spreading of microorganisms.

Results of this study on the four melon cultivars clearly indicate that treatment temperature did not affect the efficacy of 1-MCP. Presented results differ from a previous report (PERZENLAN et al., 2014), which concluded that application of 1-MCP at 20 °C was more effective in reducing softening of 'Galia' melon than at 5 °C or 10 °C. This might be due to varying responsivity of the different melon species, varieties and cultivars to 1-MCP. A prior experiment with two cultivars in one season showed no effect of 1-MCP treatment temperature in the range of 5-20 °C (NGUYEN et al., 2018).

The statistical analysis of the obtained results determined significant contribution of the melon type to the responses to 1-MCP exposure. This suggests that it is not possible to draw a simple general conclusion, and findings may be limited to certain groups of melon types.

Conclusions

Application of 625-650 nL L^{-1} gaseous 1-MCP at 5, 10 and 20 °C prolonged the shelf-life of fruit of four melon cultivars. The treatment with 1-MCP delayed the ripening of melons during 10 d of shelf-life. In addition, fruit of the cultivars 'Celestial', 'Centro', 'Donatello' and 'Lillo' showed similar patterns of quality changes as analyzed by variations in ethylene release, respiration, stiffness, rind color and chlorophyll fluorescence. Treatment temperatures did not affect significantly any of these parameters and their changes during storage.

This finding is in contrast with earlier reports obtained with 'Galia' melons. This discrepancy and our results suggest that different melon varieties, types and cultivars may differ in their response to 1-MCP.

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Conflict of interest

No potential conflict of interest was reported by the authors.

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