Effect of Treatment Time of 1-MCP on Ripening of ‘Fuji’ Apples Stored at Low and Room Temperature for a Long Period

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Abstract The effect of 1-methylcyclopropene (1-MCP) treatment at 0, 2, and 4 d after harvest (0 DAH, 2 DAH, and 4 DAH) of apples [*Malus sylvestris* (L.) Mill var. *domestica* (Borkh.) Mansf.] was evaluated on several fruit quality parameters for 180-d at 0.5°C in the air, followed by 28-d at room temperature. The 1,000 nl l⁻¹ 1-MCP treatments were adequate to maintain fruit titratable acidity (TA) and firmness, inhibiting the rates of ethylene production and respiration during the storage of both cold and the room temperatures, regardless of the 1-MCP treatment time.

Keywords: apple, CO₂, ethylene, fruit quality, 1-MCP

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1. Introduction

The 1-methylcyclopropene (1-MCP) has been used to retard ripening or senescence of climacteric apple fruit, binding to the sites of ethylene reception of the fruit for a while [1,2]. Loss of 1-MCP inhibition of ripening apple fruit was associated with softening and development of the physiological storage disorder [1,2,3,4]. Instant or delayed application of 1-MCP affected several attributers of postharvest apple fruit quality [2,3,4,5,6,7,8] on various apple cultivars having different initial ethylene production from harvest, and reviewed by Watkins [2]. Apple growers or the distributors typically preserved fruits at low temperature without the application of 1-MCP or by delaying application of 1-MCP for several days from harvest until the consumers need the fruit on the market. This phenomenon has also not being observed for prolonged-cold storage, followed by room temperature storage, the latter which is the normal storage condition for a large number of apples in Korea. Little information is available about the responses of delayed 1-MCP treatment on fruit ripening of ‘Fuji’ apples during cold storage [6,8].

‘Fuji’ apples comprise approximately 64% of the total apple production area in 2007 [9], which ranks the first of the apple cultivars in Korea. The fruit is large-size and late-season cultivar (mid-October), with a storage life of between 30 - 50 d at room temperature or approx. 150-d in the air at 0.5°C. ‘Fuji’ apples kept in the cold storage over six months declined their storability during the shelf life

periods of 7 d at 20°C, resulting in decreased marketability to the other Asian countries. Fruit quality of early ‘Tsugaru’ apple cultivar was compared with 1-MCP versus ethylene right after 1-MCP treatment in cold storage for long periods, followed by a shelf life period of a month [10], which has not been hitherto evaluated for a late season of apple cultivar, such as ‘Fuji’.

The objective of this study was to investigate the responses of 1-MCP treatment time on the quality of ‘Fuji’ apples in the air at 0.5°C for six months and on subsequent fruit ripening for 28 d at 20°C in the room temperature.

2. Materials and Methods

2.1. Plant Source and Sampling

Fruits from mature ‘Fuji’ apple (*Malus × domestica* Borkh.) trees were harvested on the orchard farm at Kongju National University at Yesan, South Korea (36°N, 126°E). Any malformed fruit or injured fruit were excluded from the study, and the cleaned fruit were then cooled to 0°C for 12 h (85 - 90% RH). Experiment included the effects of treatment time of 1-MCP on the fruit quality and rates of ethylene production and respiration for the 180-d cold storage at 0.5°C. We then evaluated the fruit quality of the apples applied with those above treatments during the 28-d at 7 d intervals at room temperature (20±2°C; 80 - 85% RH).

Ten individual ‘Fuji’ apple fruit per storage time were randomly removed from the storage to measure their

soluble solid contents (SSC), titratable acidity (TA), and fruit firmness. The other ten individual fruit were taken to measure the rates of fruit ethylene and respiration.

2.2. Treatment Condition

1-MCP as a powder (SmartFreshSM, AgroFresh, Spring House, PA, USA; 0.14% active ingredient by weight) was allowed to release a final gas concentration of 1,000 nl l⁻¹ into the Erlenmeyer flasks. Fruit were pre-cooled overnight and were then either untreated (control) or treated with 1,000 nl l⁻¹ 1-MCP 0 d, 2 d, and 4 d after harvest (0 DAH, 2 DAH, and 4 DAH). The 1-MCP was supplied from the sealed 135-L plastic containers at 10°C for 24 h and then ventilated at a rate of 30 ml min⁻¹. 1-MCP from the powder was liberated by adding water of it. Untreated-fruit were also applied from the same plastic containers used for 1-MCP-treated fruit under the same stored condition. All harvested fruit were then stored at 0.5°C in air for up to 6 months followed by room temperature for 28 d, and some of fruit were taken out of the storage whenever the fruit needed to be investigated for ripening.

2.3. Ethylene Production, CO₂, and Fruit Quality

The 1.0 ml of internal headspace gas was extracted from the interior of each apple fruit in order to determine the ethylene and CO₂ concentrations. Ethylene production in the fruit samples were analyzed using a gas chromatography (GC, Hewlett-Packard 5890 series II; Agilent Co., Wilmington, DE, USA), which was fitted with a stainless steel needle column packed with activated 60/80 mesh alumina F-1 (2 m × 2 mm, i.d.). The detection was via a flame ionization detector [11]. Each fruit was placed into a 1 L plastic container with an air flow rate of 30 ml min⁻¹, and the containers were fitted with a septum, and the 1 ml gas samples were withdrawn slowly from the apples at each measurable time point. The gas samples were then injected into the GC meter, and the quantified values obtained from the GC were calculated for the ethylene concentrations with the external standards. The 1.0 ml of each headspace gas was also removed to measure its CO₂ content using a GC (Varian 3400; Varian Inc., Santa Clara, CA, USA) equipped with a 2.4 mm (i.d.) × 2-m long column filled with 60/80-mesh active charcoal. CO₂ concentrations were measured using a thermal conductivity detector at 110°C, with a carrier gas of helium at a flow rate of 30 ml min⁻¹.

Soluble solid contents (SSC; °Brix) in flesh were measured with a hand-held digital refractometer (RA-520N; Kyoto Electronic Co., Kyoto, Japan). For measurement of titratable acidity (TA), fruit juice of each sample was titrated with 0.1 N-sodium hydroxide to an end point of pH 8.2 and converted to malic acid (%). Fruit were assessed manually on the opposite paired sides of each apple using a TA-XT2 pressure tester (Stable Micro system Ltd., Godalming, England) fitted with a 5.0 mm cylindrical end probe for measuring flesh firmness (N).

2.4. Statistical Analysis

The study was arranged in a completely randomized design with ten fruit replications of each treatment per

storage time. All data were subjected to ANOVA using Minitab software v. 14.1 (Minitab, Inc., State College, PA, USA). Means were separated using Duncan's New Multiple Range Test at *P* = 0.05. Data over time were also shown as means ± standard errors.

3. Results and Discussion

Figure 1. Soluble solid contents (SSC; A), titratable acidity (TA; B), and firmness (C) of 'Fuji' apple fruits untreated (control) or treated with 1,000 nl l⁻¹ 1-MCP of delays of 0, 2, and 4 days after harvest (DAH) in 0.5°C cold storage for 180 d after treatment. Bars represent error of the means (S.E.M.; n = 10), when larger than the dimension of the symbol. Different lower-case letters adjacent to each datum point at each sampling date indicate differences as determined by Duncan's MRT (*P* ≤ 0.05).

Fruit SSC increased during the 180-d cold storage at control or 1-MCP treatments, presumably due to the conversion of starch to sugars [12]. The SSC were not affected by time of 1-MCP treatment (Figure 1A) as summarized from several reports for most apple cultivars [13]. Fruit SSC and TA in the long storage type apples, such as 'Fuji', were not affected by 1-MCP until 3 months cold storage [1]. However, fruit TA decreased during the storage time, and the control fruit had lower TA content

from 150-d storage compared with those of 1-MCP-treated fruits (Figure 1B) as observed in other 'Fuji' apples 6. Flesh firmness in control fruit decreased rapidly during the storage (Figure 1C), and the difference between the control and 1-MCP was significant with advanced storage time, which was agreement with other observations on different apple cultivars [14]. Time of 1-MCP treatment did not affect fruit firmness during the storage.

Figure 2. Rates of ethylene production (A) and respiration (B) of 'Fuji' apple fruits untreated (control) or treated with 1,000 nl l⁻¹ 1-MCP of delays of 0, 2, and 4 days after harvest (DAH) in 0.5°C cold storage for 180 d after treatment. Bars represent error of the means (S.E.M; n = 10), when larger than the dimension of the symbol. Different lower-case letters adjacent to each datum point at each sampling date indicate differences as determined by Duncan's MRT ($P \leq 0.05$)

Control fruit had higher rates of ethylene production and fruit respiration rate than those of 1-MCP-treated fruits during the storage (Figure 2), with not being observed for the significant difference of the above variables from time of 1-MCP-treatment. Apple fruit is considered as a climacteric fruit, rapidly increasing rates of ethylene production and respiration after the onset of climacteric phase with increasing storage periods [15]. Less delay of tomato ripening by 1-MCP was associated with higher ethylene concentration in the fruit at the time of treatment [14,16]. Responses of apple cultivar to 1-MCP treatment widely varied due to the different binding ability to ethylene receptor sites [12,14,17]. On the other hand, the 1,000 nl l⁻¹ 1-MCP application, the maximum allowed concentration in USA, between 3 - 7 DAH was effective in saturating the 1-MCP response for most apple cultivars [1]. Four days of delayed 1-MCP treatment were sufficient to maintain low level of ethylene concentration in 'Fuji' apples. This was well documented for 1-MCP-treated 'Cortland' and 'Low Rome' apples within 7 DAH

in air at 0.5°C for six months but was not observed after that for fruit associating with the development of fruit softening and senescence breakdown [4].

Figure 3. Soluble solid contents (SSC; A), titratable acidity (TA; B), and firmness (C) of 'Fuji' apple fruits untreated (control) or treated with 1,000 nl l⁻¹ 1-MCP of delays of 0, 2, and 4 days after harvest (DAH) at 20°C for 28 d after removal from 0.5°C cold storage for 180 d. Bars represent error of the means (S.E.M; n = 10), when larger than the dimension of the symbol. Different lower-case letters adjacent to each datum point at each sampling date indicate differences as determined by Duncan's MRT ($P \leq 0.05$)

The differences of SSC, TA, and firmness of the fruit in cold storage between the treatments were similarly observed at 20°C for 28 d. Fruit SSC was not markedly affected by 1-MCP treatment time and control (Figure 3A). Loss of fruit TA was inhibited by 1-MCP treatments (Figure 3B) as reported in other 'Fuji' apples [6,8]. Fruit firmness was the lowest for control, having a low TA, with overall no difference observed for both TA and firmness between time of 1-MCP treatment (Figure 3C). High fruit TA resulted in firm fruit of various apple cultivars [13]. Lower ethylene concentration at harvest date of 'Fuji' apples would have less affected fruit TA or firmness, regardless of 1-MCP treatment time [14]. High ethylene concentration initiated at late harvested 'Empire'

apples increased fruit softening in cold air plus shelf life with increasing storage periods, showing significant effects of 1-MCP treatment time up to 8 d after harvest [14]. This would be because late season of 'Fuji' cultivar has low initial ethylene concentration occurring at the harvest [18], catalyzing by 1-aminocyclopropane-1-carboxylic acid (ACC) synthase and the ACC oxidase [19].

Prolonged-cold-stored 'Fuji' apples could be commercially transported for 28 d at room temperature when applied with 1,000 nl l⁻¹ 1-MCP within 4 d of harvest. The 1-MCP-treated fruits retained more than 14 N fruit firmness value (10% reduction of initial firmness) during long-term storage, keeping shelf-life of fruit quality in Korean market. Flexibility of 1-MCP treatment time would provide a remarkable benefit for growers during the harvest season by maintaining good fruit quality. Further studies are required to evaluate the long-term responses to extended intervals of 1-MCP treatment from harvest on postharvest fruit quality.

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