Responses of ‘Golden Delicious’ Apples to 1-MCP Applied in Air or Water

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Abstract. The efficacy of the ethylene action inhibitor 1-methylcyclopropene (1-MCP) applied in water to slow ripening of ‘Golden Delicious’ [Malus sylvestris var. domestica (Borkh.) Mansf.] apples was evaluated in comparison with 1-MCP applied in air. The material was applied by dipping fruit in 1-MCP water solutions (0.03, 0.3, or 3 mmol·m⁻³) for 4 min or by exposing fruit to 1-MCP gas (0.42, 4.2, or 42 μmol·m⁻³) in air for 12 h. Fruit were held in air at 20 °C for 25 days after treatment or stored at 0.5 °C in air for up to 6 months followed by 7 days in air at 20 °C. Application of 1-MCP in water or air delayed the increase in respiration and ethylene production associated with fruit ripening and reduced the amount of fruit softening, loss of acidity, and change in peel color. Treatments applied in water required a 700-fold higher amount of active ingredient compared with treatments applied in air to induce similar physiological responses. Fruit responses to 1-MCP varied with treatment concentration, and the maximum effects were obtained at concentrations of 4.2 or 42 μmol·m⁻³ in air and 3 mmol·m⁻³ in water. Peel color change was impacted less than retention of firmness and titratable acidity compared with treatments applied in air to induce similar physiological responses. Fruit treated with 1-MCP and stored in air at 0.5 °C developed a peel disorder typified by a gray-brown discoloration that is unlike other disorders previously reported for this cultivar. Symptoms were present when fruit were removed from cold storage and no change in symptom appearance was observed during a 7-d holding period at 20 °C.

The ethylene action inhibitor 1-methylcyclopropene (1-MCP) prevents plant tissues from responding to ethylene (Sisler and Blankenship, 1996) by combining with ethylene receptors (Sisler and Serek, 1997). Impacts of 1-MCP demonstrated for many apple cultivars include reduction of ethylene production and respiration rates, slower progression of ripening processes, including softening, loss of acidity, yellowing, volatile production, and reduced development of some physiological disorders (Fan et al., 1999a, 1999b; Ferencz et al., 2006; Johnson, 2003; Lurie et al., 2002; Mattheis et al., 2005; Pre-Aymard et al., 2005; Rupasinghe et al., 2000; Watkins et al., 2000). Responses to 1-MCP for apples cold-stored in air can be comparable with that of midterm (4 to 6 months) CA storage of apples not treated with 1-MCP, and the combination of 1-MCP treatment followed by CA storage can extend the effectiveness of 1-MCP treatment (DeLong et al., 2004; Mattheis et al., 2005; Watkins et al., 2000; Zanella, 2003).

Efficacy of 1-MCP for delaying apple fruit ripening is concentration and treatment duration-dependent (Argenta et al., 2005; Dauny and Joyce, 2002; DeEII et al., 2002; Fan et al., 1999a; Jayanty et al., 2004; Pre-Aymard et al., 2005; Rupasinghe et al., 2000). Maximum benefits from 1-MCP treatment occur when apple fruit are harvested and treated at the preclimacteric stage of development (Argenta et al., 2005; Mir et al., 2001; Moran and McManus, 2005). Maximum control of fruit softening, superficial scald, and senescent breakdown for apples is achieved by treatment as soon after harvest as possible (Argenta et al., 2005; Watkins and Nock, 2005).

The low boiling point (less than 10 °C) of 1-MCP (Sisler and Serek, 1997) allows the material to be applied as a gas in a sealed space, including commercial CA storage rooms. However, application of 1-MCP in water would enable postharvest treatments as a dip or line spray during the packing process. Application of 1-MCP in water could allow application when sealed rooms are not available, when room filling will be delayed, or to treat only fruit that will be packed after nonpackable fruit has been segregated. Field application of an aqueous 1-MCP solution applied to citrus reduces ethylene-induced leaf abscission with little effect on fruit detachment force (Pozo et al., 2004). The efficacy of a postharvest application of 1-MCP in water to delay ripening of a climacteric fruit has not been reported.

The objective of this study was to compare the efficacy of postharvest application of 1-MCP in air or water to alter ripening of ‘Golden Delicious’ apples during storage at 20 °C or after CA.

Materials and Methods

‘Golden Delicious’ apples were harvested from the USDA, ARS Columbia View Experimental Plots near Wenatchee, WA. Application of 1-MCP gas (0.42, 4.2, or 42 μmol·m⁻³ = 0.01, 0.1, or 1 μL·L⁻¹) in air was performed at 20 °C for 12 h 1 d after harvest. Generation of 1-MCP was performed at room temperature by mixing cycloextrin-1-MCP powder (EthylBloc; BioTechnology for Horticulture, Burrr Ridge, IL) and 10 mL of water in a 150-mL flask sealed with a rubber stopper. After mixing, the flask and the 230-L treatment chamber were connected with Tygon tubing. The water solution in the flask was purged for 15 min by passing air from the treatment chamber while the headspace 1-MCP gas from the flask was pumped into the treatment chamber in a closed loop. The concentration of 1-MCP in the treatment chamber was analyzed by gas chromatography using a 1-butene standard to generate a response factor of 42 μmol·m⁻³ = 5.2 area counts (Fan et al., 1999b). Application of 1-MCP in water at 20 °C (water and fruit temperature) was performed by dipping fruit for 4 min during the first 30 min after the solution was prepared in a ventilated open room. For each 1-MCP concentration made up in a final volume of 4 L, five samples of 20 fruit were dipped sequentially. Concentrations of the 1-MCP solutions (0.03, 0.3, or 3 mmol·m⁻³) were based on the amounts of 1-MCP (1.6, 16, or 162 μg·L⁻¹) added to the water along with 0.05% Tween 20. Addition of 0.05% Tween 20 was based on preliminary experiments that compared efficacy of 1-MCP solutions prepared in water alone or with various surfactants. The 1-MCP solutions were disturbed only when fruit were added and removed to minimize off-gassing of 1-MCP. Untreated controls were held for 12 h in air without 1-MCP in a similar steel chamber or dipped for 4 min in water containing 0.05% Tween 20. After treatment, all fruit were held in air at 20 °C for 25 d or stored in air at 0.5 °C for 3 or 6 months plus 7 d at 20 °C. Fruit quality was analyzed on the day after harvest, after 25 d at 20 °C, or after 3 or 6 months of cold storage plus 1 or 7 d at 20 °C. Fruit firmness, starch index (1 = 100% starch coverage; 6 = 0% starch), titratable acidity (TA), soluble solids content, internal ethylene concentration (IEC), respiration, and ethylene production were analyzed as described earlier (Fan et al., 1999a; Mattheis et al., 1998). Peel color was measured with a colorimeter (Minolta CR-200, Osaka, Japan) using CIE illuminant C and an 8-mm-diameter...
aperture. Color values a* and b* were converted to hue angle (h°) (McGuire, 1992). Peel color was also rated visually (1 = green, 5 = yellow) using a ‘Golden Delicious’ color chart (USDA, 1929). Superficial scald was visually assessed (1 = no scald; 7 = dark scald and greater than 60% of the fruit surface affected) (Fan et al., 1999b).

A peel disorder of unknown etiology manifested as a diffuse, grayish brown discoloration and observed during fruit evaluation was visually assessed as clear or affected. There was no difference in incidence of peel discoloration resulting from days of ripening after storage; therefore, data from day 1 and day 7 after removal from storage were pooled.

The experiment followed a completely random design with 20 single fruit replicates for each combination of treatment, storage temperature, and storage duration, except for determination of the peel discoloration disorder that followed a randomized complete block design with four replicates of 10 fruit. Data were analyzed by analysis of variance using SAS (SAS Institute, Raleigh, NC). Data for peel color and disorder ratings and percent incidence were transformed to SQRT(x + 0.5) before analysis of variance. Effects of 1-MCP concentration were analyzed using orthogonal contrasts. Significance of orthogonal polynomials was calculated by the F-test procedure. Treatment mean differences were identified using Fisher’s protected least significant difference for time course rates of respiration and ethylene production.

**Results**

**Fruit maturity at harvest.** Fruit internal ethylene concentration at harvest was below 25 μmol·m⁻³, and firmness and starch index averaged 65 ± 5.4 N and 3.7 ± 0.9, respectively.

**Fruit ripening at 20 °C.** Control fruit respiration increased during 25 d ripening at 20 °C (Fig. 1A). Maximum production of CO₂ was detected between 9 and 15 d after harvest. Respiration by apples treated with 1-MCP in air was similar to or lower than the rate at harvest throughout the 25-d period. Ethylene production by control fruit increased asymptotically, whereas ethylene production by 1-MCP fruit was reduced compared with control fruit (Fig. 1B). Ethylene production by fruit exposed to 4.2 or 42 μmol·m⁻³ 1-MCP was not detectable for 21 or 18 d after treatment, respectively. Application of 1-MCP in water also resulted in altered respiration and ethylene production compared with control fruit (Fig. 1C–D). Ethylene production by fruit dipped in 0.3 or 3 mmol·m⁻³ 1-MCP in water was lower compared with control fruit through 25 d at 20 °C after treatment.

After 25 d at 20 °C, all fruit treated with 1-MCP in air or fruit treated in water at 0.3 or 3 mmol·m⁻³ were firmer, greener, and had higher TA compared with control fruit (Table 1). Concentration effects fit quadratic models indicating that fruit responses to 1-MCP were maximized at concentrations lower than 42 μmol·m⁻³ in air and lower than 3 mmol·m⁻³ in water.

**Discussion**

Responses of ‘Golden Delicious’ apples to 1-MCP are influenced by 1-MCP concentration, application in air or in water, storage temperature after treatment, and storage duration. Fruit exposed to 0.42 μmol·m⁻³ 1-MCP in air began to produce ethylene sooner after treatment than fruit treated at 4.2 or 42 μmol·m⁻³. Exposure of ‘Golden Delicious’ apples to 1-MCP in air at 4.2 or 42 μmol·m⁻³ was similarly effective for maintenance of firmness, TA, and peel color, regardless of storage period, indicating that maximum benefit for fruit quality retention...
Table 1. Firmness (Firm), titratable acidity (TA), peel hue angle, and peel color index (1 = green; 5 = yellow) of 'Golden Delicious' apples treated with 1-MCP (n = 20).^a

| 1-MCP in air | 1-MCP in water |
|--------------|----------------|
| Concentration | Concentration |
| (µmol·m⁻³)   | (µmol·m⁻³)     |
| 0            | 0              |
| 0.42         | 0.03           |
| 4.2          | 0.3            |
| 42           | 3              |
| L            | ***            |
| Q            | ***            |

Color ratings were transformed to SQRT(x + 0.5) before analysis of variance.
^aApples were treated the day after harvest with 1-MCP in air for 12 h or by dipping in 1-MCP solutions for 4 min. Fruit were held at 20 °C for 25 d after treatment. NS, *, **, ***Not significant or significant linear (L) and quadratic (Q) orthogonal polynomials at P < 0.05, 0.01, or 0.001, respectively. N = newtons; 1-MCP = 1-methylcyclopropene.

Table 2. Firmness (Firm), titratable acidity (TA), peel hue angle, peel color index (1 = green; 5 = yellow), and internal ethylene concentration (IEC) of 'Golden Delicious' apples treated with 1-MCP (n = 20).^a

| 1-MCP in air | 1-MCP in water |
|--------------|----------------|
| Concentration | Concentration |
| (µmol·m⁻³)   | (µmol·m⁻³)     |
| 0            | 0              |
| 0.42         | 0.03           |
| 4.2          | 0.3            |
| 42           | 3              |
| L            | ***            |
| Q            | ***            |

Color ratings were transformed to SQRT(x + 0.5) before analysis of variance.
^aApples were treated the day after harvest with 1-MCP in air for 12 h or by dipping in 1-MCP solutions for 4 min. Fruit were held at 20 °C for 3 and 6 mo plus 7 d at 20 °C. NS, *, **, ***Not significant or significant linear (L) and quadratic (Q) orthogonal polynomials at P < 0.05, 0.01, or 0.001, respectively. N = newtons; 1-MCP = 1-methylcyclopropene.

Table 3. Superficial scald severity of 'Golden Delicious' apples (n = 20).^a

| 1-MCP in air | 1-MCP in water |
|--------------|----------------|
| Concentration | Concentration |
| (µmol·m⁻³)   | (µmol·m⁻³)     |
| 0            | 0              |
| 0.42         | 0.03           |
| 4.2          | 0.3            |
| 42           | 3              |
| L            | ***            |
| Q            | ***            |

Scald ratings were transformed to SQRT(x + 0.5) before analysis of variance.
^aApples were treated the day after harvest with 1-MCP in air for 12 h or by dipping in 1-MCP solutions for 4 min. Fruit were held at 20 °C for 3 and 6 mo plus 7 d at 20 °C. NS, *, **, ***Significant linear (L) and quadratic (Q) orthogonal polynomials at P < 0.05, 0.01, or 0.001, respectively. N = newtons; 1-MCP = 1-methylcyclopropene.

is likely to be obtained in this concentration range. These results are consistent with previous studies in which the saturation concentration of 1-MCP in apple fruit has been estimated at ~42 µmol·m⁻³ (Argenta et al., 2005; Dauny and Joyce, 2002; Fan et al., 1999a; Jayanty et al., 2004; Pre-Aymard et al., 2003; Rupasinghe et al., 2000; Watkins et al., 2000).

Similar to 1-MCP applied in air, application of 1-MCP in water delayed onset of the climacteric and fruit ripening during storage at 20 °C or 0.5 °C. However, a 700-fold higher amount of cyclodextrin-1-MCP powder was necessary to achieve similar fruit responses from 1-MCP applied in water compared with treatments applied in air. Although the amounts of cyclodextrin-1-MCP added in the water treatments were high relative to treatments in air, some of the 1-MCP generated from cyclodextrin-1-MCP was likely released from water into the treatment room as the solution was disturbed. 1-MCP efficacy was the same for fruit dipped 5 min or 2 h after preparation in water (data not shown) indicating the concentration of 1-MCP available in water established rapidly after preparation of the solution and may remain stable in undisturbed water for extended time periods. Reduced efficacy on a concentration basis for 1-MCP applied in water may occur as a result of a lower rate of diffusion of 1-MCP in water. Exposure of apples to 1-MCP in air for 1 to 4 h effectively prevented ripening (Argenta et al., 2005; DeEll et al., 2002; Pre-Aymard et al., 2003), indicating that 1-MCP in air readily diffuses into fruit, possibly through lenticels as noted for CO₂ and ethylene (Burg and Burg, 1965). The rate of entry of 1-MCP in water into fruit may be reduced by slower diffusion of 1-MCP in water.
4.2 solutions for 4 mm. There were four replicates of 10 fruit for each treatment and storage duration. Disorder incidence percentages were transformed to $\sqrt{x + 0.5}$ before analysis of variance. Arithmetic means are presented with standard error. Differences were considered significant at $p \leq 0.05$. NS = nonsignificant difference.

Table 4. Peel discoloration disorder incidence (%) of 'Golden Delicious' apples after 3 and 6 mo of storage at 0.5 °C.

| 1-MCP in air | 1-MCP in water |
|--------------|----------------|
| (mmol-m⁻²)   | (mmol-m⁻²)     |
| 3 mo         | 6 mo           |
| 0            | 0              |
| 0.42         | 0              |
| 0.42         | 0.05           |
| 0.50         | 0              |
| 0.50         | 0.3            |
| 0.50         | 19             |
| 0.75         | 45             |
| Concentration | **            |
| Q            | NS             |

Disorder incidence percentages were transformed to $\sqrt{x + 0.5}$ before analysis of variance. * = significant difference. ** = very significant difference.

Application of 1-MCP in water delays the climacteric rise in respiration, ethylene production, softening, loss of acidity, peel color changes, and prevents development of superficial scald in 'Golden Delicious' apples as it does when applied as a gas in air. However, 1-MCP applied in air is more efficient than 1-MCP applied in water based on the amount of a.i. necessary to induce similar physiological responses. Effects of 1-MCP in water or air are concentration- and storage temperature-dependent.

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Fig. 2. ‘Golden Delicious’ apple exhibiting symptoms of peel disorder. Apple was treated the day after harvest with 42 μmol·m⁻² 1-methylcyclopropene in air for 12 h and then held in air at 0.5 °C for 3 months. Image was taken 1 d after removal from cold storage.
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