Differential Responses in Color Changes and Softening of ‘Florida 47’ Tomato Fruit Treated at Green and Advanced Ripening Stages with the Ethylene Antagonist 1-Methylcyclopropene

Brandon M. Hurr, Donald J. Huber1, and James H. Lee

ADDITIONAL INDEX WORDS. Lycopersicon esculentum, 1-MCP, ripening, immature-green, mature-green, fruit quality, maturity

SUMMARY. In this study, ripening characteristics, including color change and softening, were determined for tomato (Lycopersicon esculentum ‘Florida 47’) fruit at immature-green through light red stages of development and subsequently treated with 1 µL·L⁻¹ 1-methylcyclopropene (1-MCP). Special attention was directed at comparing the responses of immature and mature-green fruit. Surface color and whole fruit firmness were measured every other day. 1-MCP delayed or slowed color changes and softening in fruit of every maturity class, with differences between control and treated fruit evident immediately following 1-MCP application for 24 h at 20 °C. Fruit treated with 1-MCP at early maturity stages (immature-green, mature-green, and breaker) exhibited an extended delay in external red pigment accumulation compared with control fruit. Fruit of all maturity classes developed acceptable final hue values (hue angle ≤55°), and the time required to reach these values declined with advancing fruit maturity. Immature-green fruit treated with 1-MCP did not attain an acceptable degree of softening during the specified storage periods examined before deteriorating due to shriveling and pathogen proliferation. 1-MCP-treated mature-green and breaker stage fruit did recover to acceptable firmness (5–10 N) and hue values but exhibited a severely reduced storage life thereafter compared with untreated fruit of equal maturity. Fruit at turning and more advanced stages exhibited reduced rates of softening and color development when treated with 1-MCP, yet they attained firmness and color values within the range of acceptability for commercial use. Fruit treated with 1-MCP at pink and light-red stages of ripening developed normal external color and exhibited significantly extended postharvest life due largely to a significant retention in firmness when compared to control fruit. Based on the studies described for ‘Florida 47’ tomato fruit, 1-MCP would appear to be of little benefit and possibly detrimental if applied to early maturity fruit, most notably greens and breakers, due to irreversible limitations in the capacity of these fruit to soften to acceptable values. In sharp contrast, more advanced stage fruit, particularly pink and light red, responded to 1-MCP with significantly extended shelf-life due to retention of firmness.

The tomato industry in Florida is devoted nearly exclusively to fresh market production, with emphasis on fruit harvested green and subsequently induced to ripen by exposure to ethylene gas (Chomchalow et al., 2002). Fruit harvested green are more tolerant to handling and considerably more resistant to bruising (Kader et al., 1977). Furthermore, fruit harvested green require longer periods to attain acceptable ripening status allowing for longer transportation and storage periods at either the re-packing or retail level. The largest problem encountered with tomato fruit harvested green is the lack of easy, effective techniques for distinguishing mature-green from immature-green fruit (Brecht et al., 1991). Unlike mature-green fruit, fruit harvested prior to completion of full maturation and seed development are severely suppressed in their capacity to ripen normally or completely, resulting in compromised final fruit quality. Depending on season and market conditions, the frequency of immature specimens in green harvests in Florida can range from 20% to as high as 80% of the total fruit count (S.A. Sargent, personal communication).

Fruit of more advanced maturity (turning through red) comprise a relatively minor proportion of the total specimens in a typical commercial harvest regime; however, these fruit are potentially of higher quality due to their extended development prior to harvest (Kader et al., 1977). Fruit showing degrees of red pigmentation at harvest are segregated at the packinghouse, either manually or through use of optical instrumentation, and marketed separately. Broader adoption of harvesting tomato fruit at advanced stages of ripening, while perhaps economically or logistically problematic, offers the advantage of higher fruit quality, and could ultimately increase consumer satisfaction and demand. Disadvantages include a shortened postharvest life due to earlier completion of ripening, reduced tolerance to handling due to softening (Kader et al., 1977), and necessity for multiple harvests.

Ripening in tomato fruit is characterized by lycopene accumulation, chlorophyll loss, softening, and changes in aroma and other compositional properties. Ethylene, applied either exogenously (i.e., to green fruit), or produced endogenously upon initiation and progression of ripening, plays a pivotal role in coordinating these events (Saltveit, 1999). Continuous requirement for ethylene perception throughout ripening was first demonstrated through the use of

Horticultural Sciences Department, University of Florida, IFAS, Gainesville, FL 32611.
Journal Series No. R-15066 of the Florida Agricultural Experiment Station. This research supported in part by contributions from Agro-Fresh a division of Rohm and Haas and by a grant from the USDA program in Tropical & Subtropical Agricultural Research (T-STAR). Special thanks to West Coast Tomato Inc. and Pacific Tomato Inc. for supplying tomato fruit.

1To whom reprint requests should be addressed. E-mail address: djh@ifas.ufl.edu
ethylene response inhibitors, including silver ions (Ag+), applied either as silver nitrate (AgNO3) or silver thiosulphate (STS), a series of cyclic olefins, such as norbornadiene, diazacyclopentadiene (DACP) and, more recently, 1-MCP (Blankenship and Dole, 2003). 1-MCP has been approved for postharvest use with a number of fruit and vegetables (Blankenship and Dole, 2003). Applied as a gas at low concentrations (≤1 µL·L−1) and short durations (≤24 h), 1-MCP has been shown to significantly delay the onset of ripening in fruit harvested pre-ripe and, in some fruits, to decelerate the progression of ripening once initiated. Reports for 1-MCP treatment of partially ripe tomato (Hoeberichts et al., 2002; Mir et al., 2004), apple (Malus sylvestris) (Mir et al., 2001), ‘Galia’ melon (Cucumis melo) (Ergun et al., 2005) and papaya (Carica papaya) (Ergun and Huber, 2004) fruits are consistent with the idea that ethylene perception is obligatory for the coordinated completion of ripening.

The objectives of this study were to examine the effectiveness of 1-MCP on ‘Florida 47’ tomato, a cultivar widely produced in Florida, when applied to fruit at varying developmental stages including immature-green, mature-green, breaker, turning, pink, and light red. Similar studies with tomato have been reported (Hoeberichts et al., 2002; Mir et al., 2004), yet the distinction between immature- and mature-green specimens was not addressed, and information on criteria to classify fruit as mature-green fruit were not presented. In this study, emphasis was directed toward the capacity of fruit of a range of developmental categories to recover from the ripening decelerating effects of 1-MCP.

Materials and methods

PLANT MATERIAL. ‘Florida 47’ tomatoes at the immature-green, mature-green, breaker and turning stage, commercially harvested and packed, were obtained three times throughout the spring/summer season of 2003 in Palmetto, Fla., or Quincy, Fla. Fruit were not exposed to ethylene. The fruit were immediately transported to the Postharvest Horticulture Laboratory at the Horticultural Sciences Department, University of Florida. Fruit were washed for 60 s in NaOCl (200 µL·L−1, prepared from a 6% commercial bleach solution), rinsed with tap water, and air-dried. Fruit were sorted for freedom from defects and uniformity of size and shape. Pink and light-red stage fruit were selected from fruit harvested at mature-green and breaker stage and ripened at 20 °C until these stages were reached (~7 d).

The experiments described were performed in triplicate using fruit from separate harvests. Similar results were obtained in all replications and data from one experiment are presented.

Maturity classification and distinction between immature- and mature-green fruits. Green fruit (both immature and mature) exhibited no external signs of red pigmentation, breaker-stage fruit exhibited a small amount of red coloration (<10% of the total surface area), turning fruit from 10% to 30% of the surface area, pink fruit from 30% to 60%, and light red from 60% to 90% (USDA, 1991). Since we were unable to visually classify immature- and mature-green fruit, the distinction between these developmental stages was established retrospectively based on postharvest ripening behavior. Green fruit that failed to exhibit external red pigment within 7 (control fruit) or 14 d (1-MCP-treated fruit), respectively, of harvest were designated immature-green. Green fruit that did exhibit external red color during these time intervals were defined as mature-green. Slicing of selected specimens to expose the seed and locule tissues of the putative maturity classifications confirmed the accuracy of our approach based on timing of appearance of external color. Based on the criteria employed, the percentage of immature- and mature-green specimens among green fruit obtained directly from the packinghouses was 40% and 60%, respectively.

1-MCP treatment. Two groups of 76 fruit, each consisting of 40 green, 20 breaker, and 16 turning, were treated with 1-MCP or air within 12 h of harvest. Other fruit at the breaker and turning stage were maintained for approximately 7 d at 20 °C until they reached pink and light-red stages of ripening. At this time, groups of pink (40) and light red (16) fruit were treated with 1-MCP or air as described below for fruit of all maturation/ripening categories. 1-MCP was applied at 1 µL·L−1 by addition of a commercial powder (0.14% 1-MCP formulation; AgroFresh Inc., Philadelphia) to 40 mL tap water in a 125-mL Erlenmeyer flask that was capped and swirled gently to dissolve the powder. The flask was unsealed and placed into the treatment chamber that was immediately sealed. 1-MCP exposure was performed twice for 12 h each (total 24 h exposure) at 20 °C, with the chambers opened and vented with forced air for 5 min prior to the second addition of 1-MCP. Control fruit (not exposed to 1-MCP) were maintained under identical storage conditions. The control chamber was vented before the 1-MCP treatment chamber to prevent cross contamination. After the 24 h exposure to 1-MCP (or air for controls), the fruit were covered with 3-mil black plastic film to reduce moisture loss, and stored at 20 °C in the dark. Fruit were stored for up to 26 d depending on fruit maturity at the time of treatment. Individual fruit of each maturity class were monitored for decay, oversofterning (firmness <5 N), or excessive water loss (shriveling). When approximately 30% of the fruit of a given treatment or maturity class exhibited one or more of these symptoms, the treatment was terminated.

COLOR. Immediately after 1-MCP treatment (day 0) individual fruit of each maturity stage were marked at two equidistant points on the equatorial axis. Color was measured with a Minolta Chroma Meter (model CR-200; Minolta Camera Co. Ltd., Tokyo) every other day at these markings starting after 1-MCP treatment and recorded as lightness (L*), Chroma (C*), and Hue angle (°). Hue angle is the most indicative of the color status of tomato fruit during ripening from green (120°) to overripe (40°) and, for simplicity, is the only color component reported. For the purposes of this study, a hue of 55° or lower was considered as acceptably red.

FIRMNESS. Immediately after 1-MCP treatment (day 0) individual fruit of each maturity stage were marked at a single point on the equatorial axis over a locule chamber, and firmness was measured every other day at this point using an Instron Universal Testing Instrument (model 4411; Canton, Mass.). After establishing zero force contact with the surface of the fruit, a 15.5 mm convex probe was driven a distance of 2.5 mm (crosshead speed 50 mm·min−1, 5-kg load cell). The maximum force achieved over the 2.5-mm travel was recorded in Newtons. Informal sensory analysis performed
by postharvest personnel found fruit with firmness above 10 N were “unacceptably hard” and below 5 N the fruit were “oversoft.” The range of 5 to 10 N is referred to as the “range of acceptability.”

Results and discussion

**TOMATO FRUIT COLOR DEVELOPMENT.** The effect of 1-MCP on postharvest color development in ‘Florida 47’ tomato fruit varied significantly with fruit maturity at time of application (Fig. 1). Both 1-MCP-treated and control green fruit showed little change in color for up to 12 d (immature-green, Fig. 1A) or 6 d (mature-green, Fig. 1B), respectively, after which time the control fruit (no 1-MCP) exhibited a sharp decline in hue angle associated with rapid accumulation and increased surface coverage of external red color. Red color development was significantly impaired in 1-MCP-treated green fruit, with little change in hue angle noted until after about 18 (immature-green) or 14 d (mature-green) of storage. Following the initial appearance of external red pigmentation in these maturity classes, the ensuing color development in 1-MCP-treated fruit occurred at approximately the same rate as noted for control fruit (Fig. 1A–B).

Fig. 1. Epidermal hue angle (°) of ‘Florida 47’ tomato fruit at different stages of development at 20 °C (68.0 °F) treated with (●) and without (○) 1 µL·L⁻¹ 1-methylcyclopropene (1-MCP) for 24 h. Vertical bars represent standard error of the mean. The horizontal line represents the acceptable color threshold.
Even so, immature- and mature-green fruit treated with 1-MCP required up to 24 and 20 d, respectively, to reach uniform, red external color. Although color values (~45°) eventually attained by green fruit were acceptable (≤55°) for fully ripe fruit, 1-MCP-treated immature- and mature-green fruit showed a high incidence of decay and water loss (data not shown) in these extended storage periods. The color development of control breaker fruit followed a pattern quite similar to that for control mature-green fruit, differing by only 2 d in time to achieve acceptable color. 1-MCP-treated breaker and mature-green fruit both achieved acceptable color after 20 d of storage (Fig. 1B–C).

Fruit at more advanced stages of development (turning, pink and light red) showed no delay in continued color development (Fig. 1D–F) in response to 1-MCP treatment, but the rate of hue angle loss (increase in red color) decelerated within 2 d compared with the respective controls. Control fruit at turning and pink maturities required 6 and 2 d, respectively, to achieve acceptable hue values, whereas 1-MCP treated fruit of these maturity classes required 12 and 6 d, respectively. The extension in time required for 1-MCP-treated turning and pink fruit to reach acceptable color was considerably less than noted for fruit of early maturities but was not accompanied by the enhanced decay and shriveling noted for the less mature fruit. Light red fruit, the most advanced maturity stage examined in this study, showed no significant delay in achieving acceptable hue values in response to 1-MCP.

The data collectively show that the efficacy of 1-MCP at inhibiting the postharvest color development of ‘Florida 47’ tomato fruit declined with increasing fruit maturity at the time of treatment. Hoeberichts et al. (2002) and Mir et al. (2004) have reported similar observations for the tomato cultivars ‘Prisca’ and ‘Neptune.’ In the present study, fruit treated with 1-MCP at all stages of development, with the exception of immature-green fruit, exhibited final hue angles equal to their respective controls. The ability of 1-MCP-treated fruit to reach final hue values equivalent to untreated fruit appears to be cultivar dependent. Mir et al. (2004) found that with the exception of mature-green and red fruit, 1-MCP-treated (250 nL·L⁻¹, 16 h at 22 °C) fruit did not, within the storage periods examined, achieve final color scores equal to those of control fruit. Mostofi et al. (2003), however, reported that mature-green ‘Rhapsodie’ tomato fruit subjected to 250 nL·L⁻¹ 1-MCP (16 h at 20 °C) and stored at 20 °C eventually attained color scores comparable to those of control fruit. Moretti et al. (2002) noted that breaker stage ‘Santa Clara’ fruit treated with 250 or 500 nL·L⁻¹ 1-MCP (12 h at 22 °C) achieved a final a*/b* ratio equal to that of untreated fruit; however, fruit treated with higher 1-MCP levels (1000 nL·L⁻¹) did not. In the same study color development of some fruits treated with 1-MCP was atypical, beginning at the stem rather than blossom end. Unusual spatial patterns including blotches of color development and stem end ripening before the blossom end were also observed in the present study with ‘Florida 47’ fruit, but were noted only for fruit designated ‘immature.’

**Tomato fruit firmness.** Fruit firmness changes during storage were significantly influenced by 1-MCP treatment, the effects of which were strongly dependent on fruit maturity at the time of treatment (Fig. 2). In contrast to fruit color changes (Fig. 1), however, the rates of softening and the final firmness values attained within the storage periods examined were significantly reduced in 1-MCP treated fruit compared with control fruit. The irreversible effect of 1-MCP on fruit softening within the storage period examined was particularly acute for immature fruit (Fig. 2A), which after 26 d had softened to only about 14 N compared with about 6 N for control fruit. The high firmness retention in immature-green fruit rendered the fruit unacceptable, in spite of the acceptable color score attained by these fruit (Fig. 1A). The acceptable but slightly firm (9 N) condition and color (Fig. 1B) of 1-MCP-treated mature-green fruit attained by 22 d of storage belies the fact that upon reaching an acceptable stage of ripeness based on color and firmness, these fruit could only be stored for 4 d due to decay proliferation and excessive shriveling. The trends of softening of breaker stage control and 1-MCP-treated fruit were similar to those observed for mature-green fruit, with 1-MCP-treated fruit exhibiting a greatly attenuated (in excess of 21 d) capacity to reach acceptable firmness values. In spite of the inability of 1-MCP-treated, early maturity fruit to soften adequately, it is noted that control fruit of all early maturity fruit attained nearly identical firmness values of approximately 6 N.

Fruit at advanced maturities (turning, pink, and light red) at the time of 1-MCP treatment exhibited an initial arrest in softening followed within several days by resumption at a reduced rate, most notably in pink and light-red fruit (Fig. 2E–F). Unlike fruit of earlier maturity stages, however, all advanced stage fruit treated with 1-MCP softened to values within the range of acceptability (5–10 N) and, in the case of pink and light red fruit, eventually softened to values below the range of acceptability (<5 N). The time in which turning and pink fruit remained in the range of acceptability for firmness, 7 and 9 d, respectively, did not differ between control and 1-MCP-treated fruit. The time required for 1-MCP-treated fruit to reach the range of acceptability was delayed approximately 6 and 4 d, respectively, for turning and pink fruit (Fig. 2D–E). Fruit at the light red maturity stage were within the range of acceptable firmness values at the start of the experiment, averaging about 9 N. Thereafter, control fruit softened to values below the range of acceptability by 6 d, whereas, 1-MCP-treated fruit remained within the acceptable range of firmness through 16 d.

It was observed in the present experiments with ‘Florida 47’ and in studies of other cultivars (Huber and Hurr, unpublished) that early maturity fruit treated with 1-MCP exhibited consistent and significant transient increases in firmness, typically within the period of 5 to 10 d of storage (Fig. 2A–D). The basis of this firmness increase is unknown, but differential responses of different tissue types (i.e., pericarp versus placenta tissues) to 1-MCP treatment may be involved. 1-MCP has been shown in other fruits, including ‘Galia’ melon (Ergun et al., 2005) and papaya (Ergun and Huber, 2004), to exert differential effects on different tissue types, an observation that suggests that different tissue types (e.g., epidermal vs. mesocarp) exhibit differential ethylene sensitivities.

The data demonstrate that interference with ethylene perception in early maturity ‘Florida 47’ tomato fruit,
in particular those at immature- and mature-green stages of development, adversely and irreversibly affects ripening, suppressing the ability to fully recover the capacity to soften. Since both immature- and mature-green fruit were adversely affected, especially in their ability to soften, a non-invasive approach to distinguishing these development stages at harvest would appear to be of little benefit in implementing 1-MCP treatment protocols. ‘Florida 47’ fruit with initial external color (breaker stage) at the time of 1-MCP treatment were incapable of reaching firmness values within the range of acceptability until after prolonged periods of storage (22 d). With advancing maturity, however, the capacity to recover from the effects of 1-MCP became more pronounced, with the ethylene antagonist extending potential holding periods largely due to maintaining fruit firmness.

To our knowledge, no studies addressing the influence of 1-MCP on green tomato fruit have attempted to distinguish the response of immature- and mature-green specimens. Hoeberichts et al. (2002), Wills and Ku

Fig. 2. Whole fruit firmness of ‘Florida 47’ tomato fruit at different stages of development at 20 °C (68.0 °F) treated with (●) and without (○) 1 µL·L⁻¹ 1-methylcyclopropene (1-MCP) for 24 h. Vertical bars represent standard error of the mean. The two horizontal lines represent the upper (10 N) and lower (5 N) limits of acceptable whole fruit firmness as determined by informal sensory analysis (1 N = 0.2248 lbf).
RESEARCH REPORTS

(2002), Mostofi et al. (2003) and Mir et al. (2004) utilized “mature-green” fruit; however, no information was provided as to how these fruit were defined and differentiated from immature specimens. Mir et al. (2004) also reported that aroma profiles of tomato tissue from the different maturity stages of tomato breeding lines 308 and 311 were minimally impacted; however, in our study, based on informal sensory analyses, aroma profiles were negatively and irreversibly affected in fruit treated with 1-MCP, most notably in green and breaker fruit.

The results of this study provide strong evidence that early maturity tomato fruit including green (immature and mature) and breaker are unsuitable candidates for 1-MCP application in commercial settings. Immature-green fruit fail to recover from 1-MCP treatment, resulting in fruit that develop color abnormally and that do not soften to an acceptable level. Fruit at both mature-green and breaker maturities treated with 1-MCP do eventually achieve acceptable color and firmness, but have prolonged periods of delay in color development and firmness often resulting in seriously compromised fruit quality due largely to pathogen incidence and water loss.

The application of 1-MCP appears most suitable for fruit beyond the breaker stage. Turning, pink, and light-red fruit exposed to 1-MCP recover completely in terms of color development and also attain acceptable firmness values. The capacity of fruit at advanced stages of ripening to recover from 1-MCP treatment has potential to extend the useful postharvest life without compromising handling, shipping, and repacking operations, and also to extend the period of retail display. An additional advantage would be the possibility to delay harvest beyond the green stage, permitting more complete “on-vine” ripening and attainment of higher quality attributes.

Literature cited


Wills, R.B.H. and V.V.V. Ku. 2002. Use of 1-MCP to extend the time to ripen of green tomatoes and postharvest life of ripe tomatoes. Postharvest Biol. Technol. 26:85–90.