Project Year: 2009-10 Anticipated Duration of Project: 1 year

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Project Title: **Use of SmartFresh after harvest to improve fruit quality after long-distance shipment and storage**

Keywords: ethylene action inhibitor, fruit ripening
Commodity: 'Bartlett' pear

Problem and Its Significance:

1-methyl-cyclopropane (1-MCP) is an ethylene action inhibitor that delays ripening of pear fruit. 1-MCP has been registered as SmartFresh™ for postharvest gas application in sealed rooms or tents and as Harvista™ for field application to fruit trees. The Harvista formulation is still under development and further work was included this year.

We have been investigating the use of SmartFresh for Bartlett pears for several years to improve post-storage quality and allow Bartlett pears to be shipped to distant markets such as Brazil. While results have been promising, a continued challenge is the balance between storage benefits and eventual ripening of the fruit for marketing. Our focus in recent years has been to reduce the fruit's response to SmartFresh through modifications in fruit handling at harvest prior to treatment. In 2008, some commercial trials of SmartFresh were conducted in California by AgroFresh Inc. The response of the fruit was quite variable in these trials and in many cases fruit did not respond or a much higher concentration was required to see an effect. It was not at all clear the reason for these treatment failures and the reasons for the widely variable results between the laboratory and the field trials. The rate of fruit cooling and the fruit pulp temperature at the time of SmartFresh application could be a factor.

Recent studies conducted in our laboratory indicate that exposure of fruit to ethylene prior to SmartFresh application results in a moderated effect on fruit ripening, resulting in delayed ripening at harvest, but full ripening after a period of cold storage.

Objectives:

1. Determine the influence of pre and post-treatment temperature exposure on Bartlett pear response to SmartFresh.

2. Determine the effect of the presence of ethylene gas during SmartFresh treatment on pear fruit response.

3. Explore the influence of exposure to ethylene gas after harvest on the response of Bartlett pears to SmartFresh^{TM}.

Materials and Methods

Temperature Conditioning Experiment prior to 1-MCP

Early-season 'Bartlett' pears were harvested on July 13, 2009 and packed on July 14, 2009 by Greene & Hemley Inc., Courtland, CA. Pear boxes, size 110, were transported in a cargo van to the postharvest laboratory at UC Davis, randomized and divided in groups held for 1, 3, 5, and 7 days at 32, 42, 50, 58 and 68F. After each time combination, half of fruits from each temperature conditioning were treated in cardboard boxes with 300ppb SmartFresh^{TM} in a 4,000L plastic tent at 32F for 12 hours while the other half was kept as an untreated control. Fruit condition was evaluated during ripening at 68F immediately after harvest and after 1.5 months storage at 32F.

Fruit and Room Temperature during 1-MCP Treatment

Mid-season 'Bartlett' pears, size 110 from Mendocino, CA, were harvested and packed on August 10, 2009 at Scully Packing Co. LLC in Finley, CA. Fruits were transported in a cargo van to the postharvest laboratory at UC Davis, randomized and divided into two groups to be held at either 68 or 32F. The following day after all the fruit reached either 68 or 32F, half of fruits for each temperature were treated in cardboard boxes with 300ppb SmartFresh^{TM} in a 4,000L plastic tent at either 68 or 32F for 12 or 24 hours while the other half was kept as an untreated control. Fruit condition was evaluated during ripening at 68F immediately after harvest and after 1.5 months of storage at 32F. Fruit from all treatments ripened immediately after harvest was warmed up to 68F and then treated with 100ppm ethylene for 24 hours to induce uniform ripening capacity.

Fruit Temperature after 1-MCP treatment

Mid-season 'Bartlett' pears, size 110, from Mendocino, CA were harvested and packed on August 4, 2009 by Scully Packing Co. LLC in Finley, CA. Fruit were transported in a cargo van to the postharvest laboratory at UC Davis, randomized and stored for 2 days at 32F. After this time, when all of the fruit had reached 32F, half of the fruit were treated in cardboard boxes with 300ppb SmartFresh^{TM} in a 4,000L plastic tent at 32F for

24 hours while the other half was kept as an untreated control. Fruit were then either ripened with 100ppm ethylene for 24 hours after harvest or stored for 1.5 months at 32, 42 or 50F. Fruit condition was evaluated during ripening at 68F immediately after harvest, or every other week during 1.5 months of storage.

Ethylene Competition during 1-MCP treatment

Mid-season 'Bartlett' pears, size 110 from Mendocino, CA, were harvested and packed on August 10, 2009 at Scully Packing Co. LLC in Finley, CA. Fruits were transported in a cargo van to the postharvest laboratory at UC Davis. After randomization, all of the fruit were placed at 32F for 3 days until they were thoroughly cooled. At this time, half of the fruit were treated in cardboard boxes with 300ppb SmartFresh^{TM} for 24 hours at 32F in two separate 4,000L plastic tents having either 61 (at the end of the treatment) or 275ppb (constantly) ethylene, while the other half was kept as an untreated control sample. Fruit condition was evaluated during ripening at 68F immediately after harvest and after 1.5 months of storage at 32F. Fruit from all treatments ripened immediately after harvest was warmed up to 68F and then treated with 100ppm ethylene for 24 hours to induce uniform ripening capacity.

Pre-Ethylene Exposure before 1-MCP treatment

Mid-season 'Bartlett' pears, size 110, from Mendocino, CA were harvested and packed on August 4, 2009 by Scully Packing Co. LLC in Finley, CA. Fruit were transported in a cargo van to the postharvest laboratory at UC Davis. After randomization, fruit were subdivided in five subgroups. Two of these subgroups were treated with 100ppm ethylene for either 4 or 8 hours at 68F and the other three subgroups were kept in regular air at 68F as an untreated control. After ethylene treatment, fruit from all subgroups were kept for an additional 12 hours in air to allow enough time for excess ethylene to diffuse out of the fruit in ethylene-treated fruit. Once 12 hours had passed, 4 hour and 8 hour ethylene-treated fruit were paired with the air-treated control fruit (identified as 4 hour or 8 hour air, respectively). Four hour (ethylene and air) and 8 hour-treated fruit were treated independently with 300ppb SmartFresh^{TM} in a 4,000L plastic tent at 68F for 12 hours while the fifth subgroup was kept as an untreated control. Fruit condition was evaluated during ripening at 68F immediately after harvest and after 1.5 months storage at 32F. After the 1-MCP treatment, fruit from all treatments ripened immediately after harvest were treated with 100ppm ethylene for 24 hours to induce uniform ripening.

Experimental Designs and Fruit Evaluations

The experimental design was completely randomized with 3 or 4 replications by treatment (3 replications for the temperature conditioning and 4 replications for the other experiments). Data were analyzed using SAS statistical software (version 9.1, SAS Institute Inc., USA). Means were compared using contrast or LSD to calculate the least significant differences for each data set ($\alpha = 0.05$). Power, log or arcsine transformations were used to fulfill Anova assumptions of ethylene production, firmness or color data.

Six to eight fruits per replication were assessed for color and firmness at each evaluation time. In addition, six fruit per replication were used to determine ethylene and CO_2 production (respiration rate) daily during each treatment or every other day during the ripening period at 68F. Firmness was measured objectively using a Gűss FTA Penetrometer (Gűss, South Africa) fitted with an 8 mm probe. CO_2 and ethylene production rates were measured by placing these six fruits from each treatment and replication into a 3.8 L jar and sealing it for 10 to 90 min. The headspace gas was analyzed for CO_2 and ethylene concentrations using rapid gas analysis (VIA510, Horiba, Japan) and flame ionization gas chromatography (Model 211 Series S, Hach-Carle Co., Fullerton, CA), respectively. Color was measured objectively using the Minolta Colorimeter CR 300 (Osaka, Japan). Internal browning and scald (including storage scald and senescent scald) severity were evaluated subjectively using the following scale: 0 = none; 1 = slight; 2 = moderate; 3 = severe.

Results

Temperature Conditioning prior to 1-MCP

At harvest and after storage, 1 day conditioned, untreated-fruit ripened normally after 6d at 68F while the 1-MCP treated fruit took approximately 2-4 additional days to have a ready to eat texture (Figure 1C, D). 1-MCP treated fruit had lower ethylene production (1A, B) and higher hue angle (less yellow) (Figure 1E, F), reflecting slower ripening rates. The relationship of flesh temperature and 1-MCP treatment efficacy was only analyzed after storage because of the differences between treatments in the rate of warming during ripening at harvest. After storage, there was significantly lower firmness after 6d at 68F (Fig. 1D) in the 1-MCP-treated fruit that were held at colder temperatures during conditioning. Only during this evaluation, however, was this difference observed, and such a difference was not clearly detected in other ripening parameters such as ethylene production (Fig. 1B) or external color (Fig. 1C).

1-MCP-treated fruit that were conditioned for 3 days showed minor or no difference in ripening relative to their untreated counterpart. Distinct ripening behavior among treatments observed at harvest might have been produced by a combination of distinct warming rates after the exposure to conditioning temperatures and some ripening delay provided by 1-MCP (Fig. 2A, C, E). However, 1-MCP- treated fruit showed no differences in firmness or external color compared with the untreated counterpart after 3 days of ripening in fruit stored at 32F (Fig. 2D). This minor to absent effect provided by 1-MCP was reproduced in fruit conditioned 5 and 7 days (data not shown) and was likely due to a combination of high ethylene in the tent during 1-MCP treatments (1.6, 5.8, and 12ppm at the end of the 3, 5, and 7 day conditioned followed by 1-MCP treatments, respectively, compared with 0.10ppm for the 1 day treatment) and the application of 1-MCP to fruits in advanced ripening stages, especially those conditioned at 68 and 58F. As is shown later in this report, ethylene competition with 1-MCP greatly decreases the efficacy of the latter.

Fruit and Room Temperature during 1-MCP treatment

At each evaluation, 1-MCP-treated fruit exhibited decreased ethylene production, softening rates, and yellow color development relative to the untreated fruit (Figure 3). Fruit treated with 1-MCP at 32F had a significantly faster induction of ripening than those treated at 68F, indicating fruit pulp and/or treatment temperature plays an important role in efficacy. The magnitude of ripening delay provided by 1-MCP was dependant on the duration of the treatment when the fruit were treated at 32F, but not at 68F. This suggests that ethylene receptors might be saturated at faster rates at warmer temperatures. Therefore, pulp/treatment temperature might be used to modulate the 1-MCP response. In this experiment, however, there were slightly different ethylene concentrations in the plastic tent at the end of the 1-MCP treatment (51, 26, 23, and 16 ppb for 12 hours at 32F, 24 hours at 32F, and 12 hours at 68F and 24 hours at 68F, respectively). It is unknown what effect this might have had on the results. Nevertheless, fruit treated with 1-MCP for 24 hours at 32F and 12 hours at 68F had almost the same ethylene concentration at the end of the treatment and still there were statistically significant differences in ripening attributes at all the evaluations favoring stronger ripening inhibition when fruit were treated with 1-MCP at warmer temperatures (Figure 3).

Fruit Temperature after 1-MCP treatment

At each evaluation, 1-MCP-treated fruit exhibited reduced ethylene production, softening rates, and yellow color development relative to the untreated fruit (Figure 4). Throughout storage, 1-MCP-treated fruit held at 32F after treatment had very low ethylene production, slight changes in firmness and moderate changes in color (Fig. 4A, B, C) while 1-MCP-treated fruit held at 42F after treatment produced similar amounts of ethylene to the untreated fruit held at 32F, but slightly higher than the 1-MCP-treated fruit held at 32F (Fig. 4A). Firmness in this fruit (1-MCP-treated held at 42F) remained very similar to both untreated and 1-MCP-treated fruit stored at 32F, but development of yellow color was more pronounced in fruit held at 42F. Untreated fruit held at 50F ripened in 2 weeks while 1-MCP-treated fruit held at the same temperature took approximately 4 weeks to become fully ripe, but was unmarketable by the end of the 1.5 month period.

After 1.5 months of storage at these temperatures, fruit were transferred to 68F to induce ripening (Fig. 5). While 1-MCP-treated fruit held at 32F took approximately 20 days to ripen (Fig. 5B, D, F), the same treatment stored at 42F softened after 6 days at 68F, similar to the untreated fruit stored at 32F, but with lower ethylene production. These results suggest that 1-MCP-treated fruit is able to tolerate some temperature abuse during transport to market. This temperature stress in fact might help to induce

nearly normal ripening during display in the supermarket. Flavor attributes were not characterized in this study, but it will be important to further characterize flavor in the future.

Ethylene Competition during 1-MCP treatment

At each evaluation, 1-MCP-treated fruit showed reduced ethylene production, softening rates, and yellow color development relative to the untreated fruit (Figure 6). Ethylene (275ppb) in the tent during 1-MCP treatment considerably reduced ripening inhibition by 1-MCP as determined by ethylene production, firmness and color change at most of the evaluations both at harvest and after 1.5 months of storage at 32F.

Pre-Ethylene Exposure before 1-MCP treatment

At each evaluation, 1-MCP-treated fruit showed reduced ethylene production, softening rates, and yellow color development relative to the untreated fruit (Figure 7). Fruit exposure to ethylene for 8 hours prior to 1-MCP treatment showed a significant effect in inducing slightly higher ethylene production and softening rates relative to the 8 hour air and 4 hour (air and ethylene) – treated fruit, but this effect was only observed during ripening at harvest, but not during ripening after cold storage.

Conclusions

Our study suggests that 1-MCP-treated fruit can be stimulated to ripen effectively under a variety of conditions which had been our main challenge in previous seasons. In fact, our research shows that fine tuning the ethylene concentration and temperature during treatment together with fruit temperature during storage or transport to final markets after treatment is important to assure beneficial effects. Treatments performed with low ethylene concentrations in the atmosphere, at warmer temperatures (68F), and followed by fruit storage at low temperatures (32F) had strong ripening inhibition. Further research is required to determine the specific combination of these factors under a variety of maturities and growing regions needed to consistently deliver pears with the capacity to ripen and with good flavor attributes. Preliminary research in our laboratory suggests the amount of fruit in the tent/room during treatment also plays an important role in fruit response to 1-MCP treatment. This will likely be another factor to include in the variables for further study this next season.

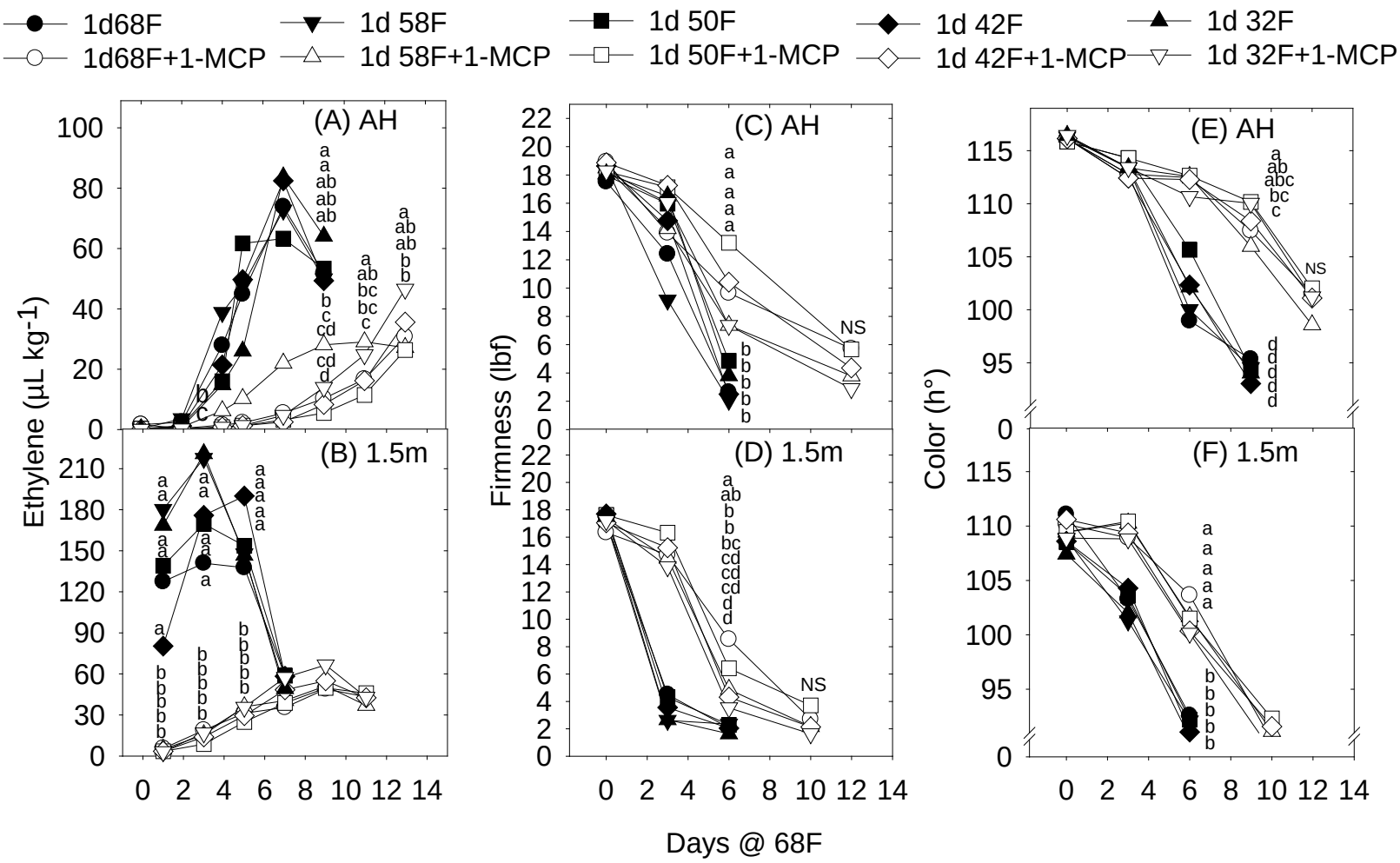


Figure 1. Ethylene production, firmness and color (hue angle decreases as the fruit change from green to yellow) during ripening immediately after harvest (AH) and after 1.5 months of storage at 32F (1.5m) of fruit temperature-conditioned for 1 day then treated with 1-MCP or maintained as untreated control. Distinct letters within each evaluation represent means with statistically significant differences using LSD (α≤0.05). Ethylene concentration in tent was 0.10ppm after 1-MCP treatment.

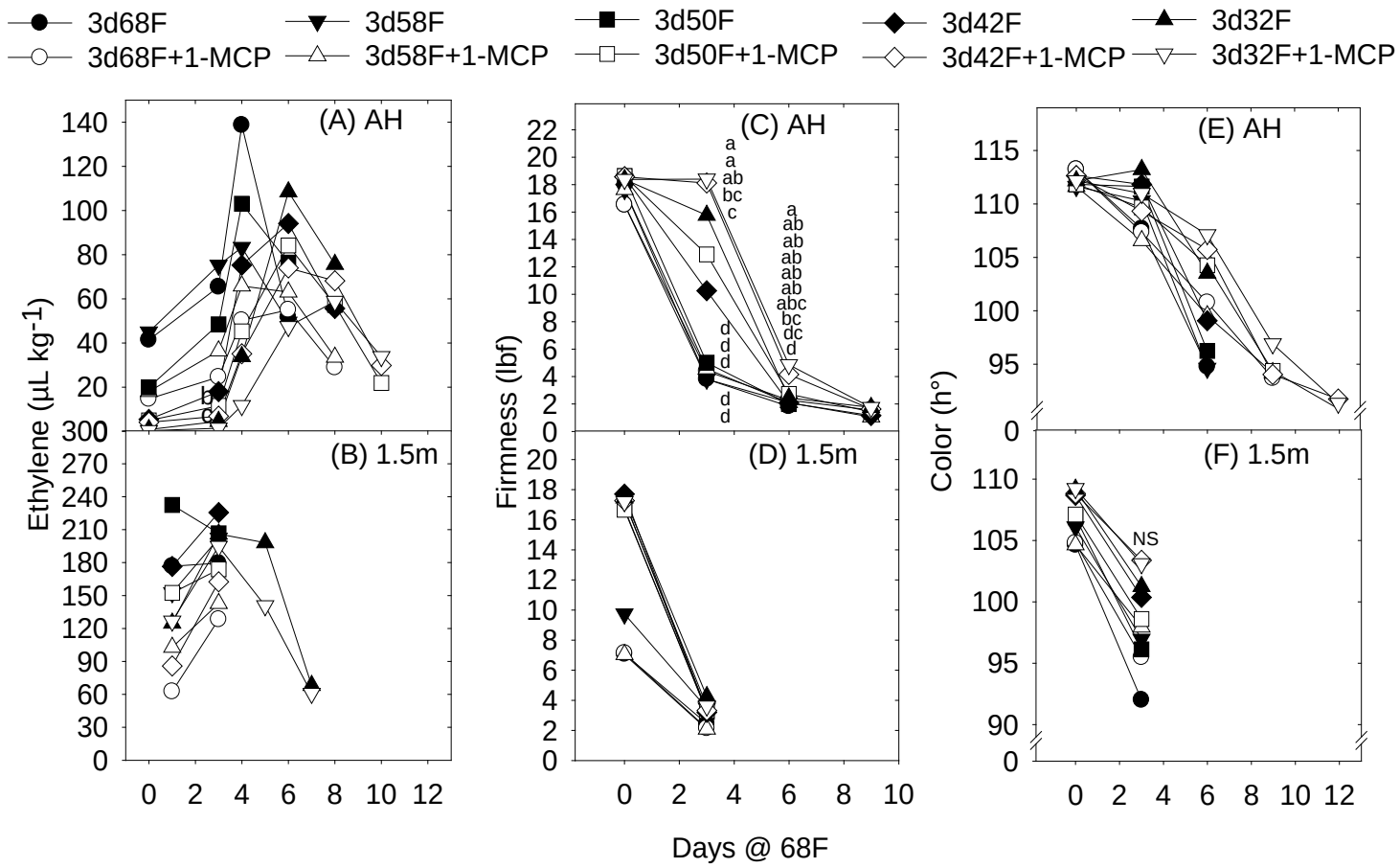


Figure 2. Ethylene production, firmness and color (hue angle decreases as the fruit change from green to yellow) during ripening immediately after harvest (AH) and after 1.5 months of storage at 32F (1.5m) of fruit temperature conditioned for 3 days then treated with 1-MCP or maintained as untreated control. Distinct letters within each evaluation represent means with statistically significant differences using LSD ($\alpha \leq 0.05$). Ethylene concentration was 1.6ppm in the tent after 1-MCP treatment.

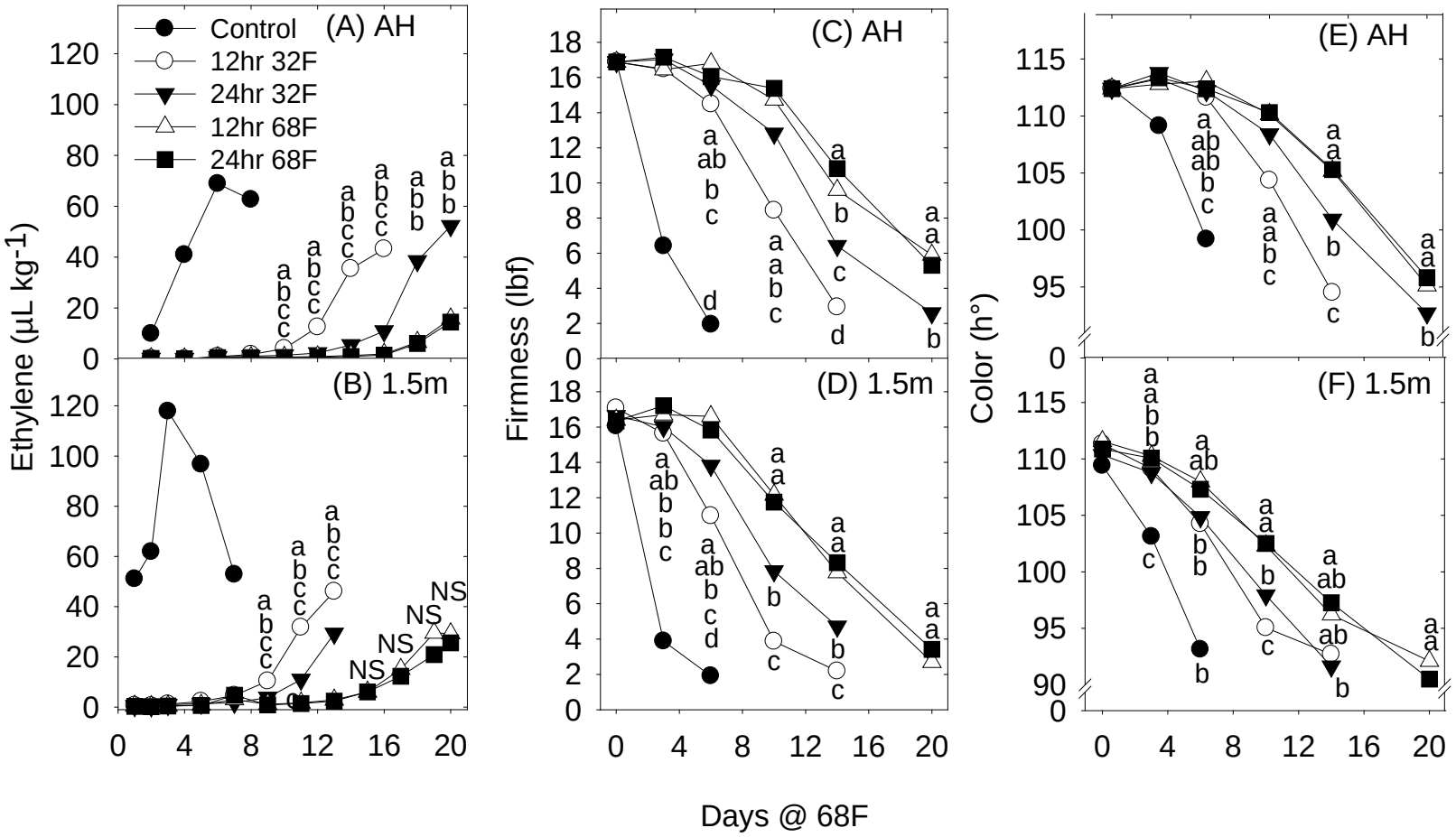


Figure 3. Ethylene production, firmness and color (hue angle decreases as the fruit change from green to yellow) during ripening immediately after harvest (AH) and after 1.5 months of storage at 32F (1.5m) of fruit treated with 1-MCP 12 and 24 hours at 32 or 68F. Distinct letters within each evaluation represent means with statistically significant differences using LSD ($\alpha \leq 0.05$). Ethylene concentration in the tent after 1-MCP treatment was 51, 26, 23, and 16 ppb for 12 hours at 32F, 24 hours at 32F, 12 hours at 68F and 24 hours at 68F, respectively.

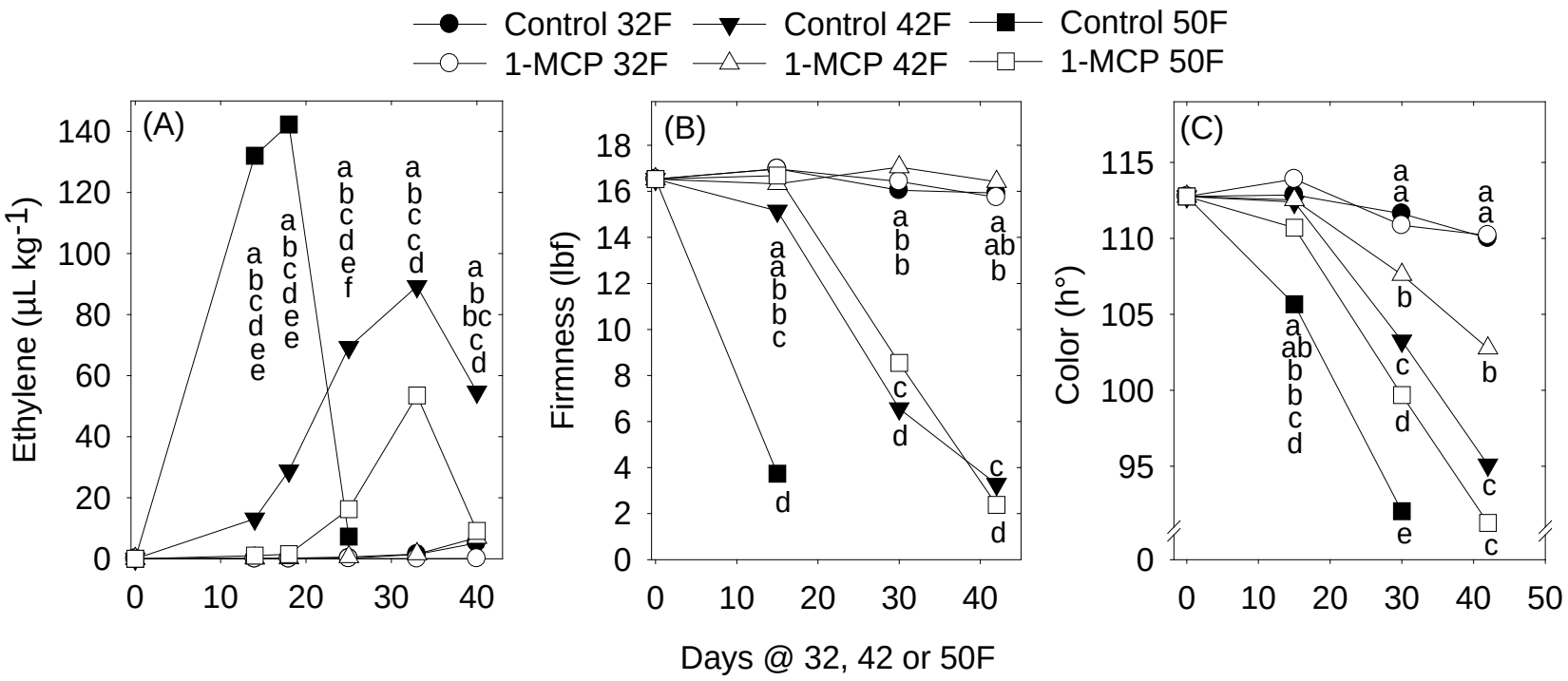


Figure 4. Ethylene production, firmness and color (hue angle decreases as the fruit change from green to yellow) during 1.5 months of cold storage of untreated and 1-MCP-treated fruit stored at 32, 42, or 50F after treatment. Distinct letters within each evaluation represent means with statistically significant differences using LSD (α≤0.05). Ethylene concentration was 44ppb in the tent after 1-MCP treatment.

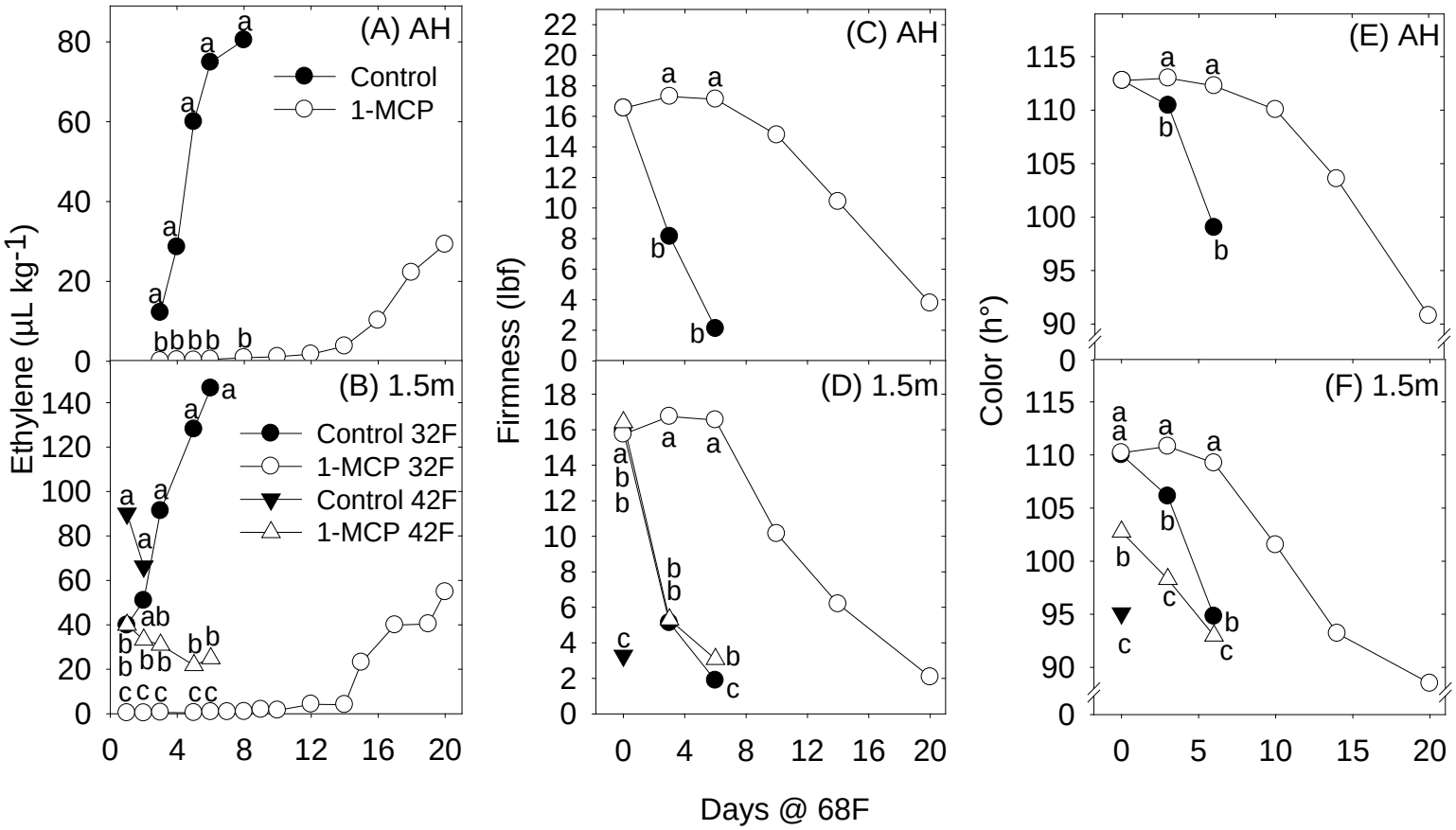


Figure 5. Ethylene production, firmness and color (hue angle decreases as the fruit change from green to yellow) during ripening immediately after harvest (AH) and after 1.5 months of storage (1.5m) of fruit treated with 1-MCP and then stored at 32, 42 or 50F for 1.5 months. Distinct letters within each evaluation represent means with statistically significant differences using LSD (α≤0.05). Fruit stored at 50F ripened before 1.5 month storage and thus are not shown in this graph. Ethylene concentration was 44ppb in the tent after 1-MCP treatment.

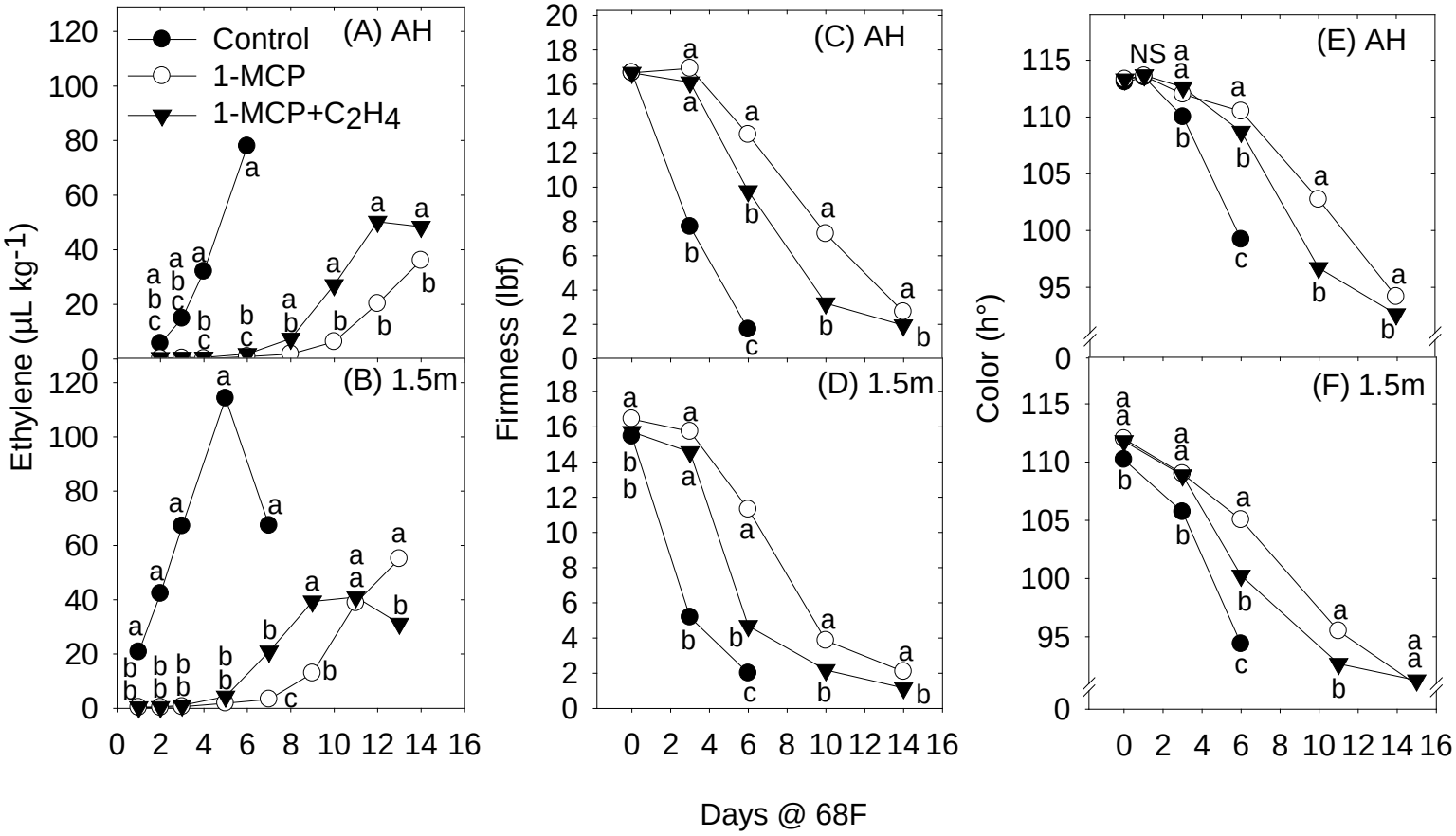


Figure 6. Ethylene production, firmness and color (hue angle decreases as the fruit change from green to yellow) during ripening immediately after harvest (AH) and after 1.5 months of cold storage (1.5m) of untreated and 1-MCP- treated fruit. 1-MCP was applied with low ethylene competition (1-MCP; 61ppb ethylene at the end of the treatment) and high ethylene competition (1-MCP+C_2H_4; 250ppb ethylene throughout treatment). Distinct letters within each evaluation represent means with statistically significant differences using LSD ($\alpha \leq 0.05$).

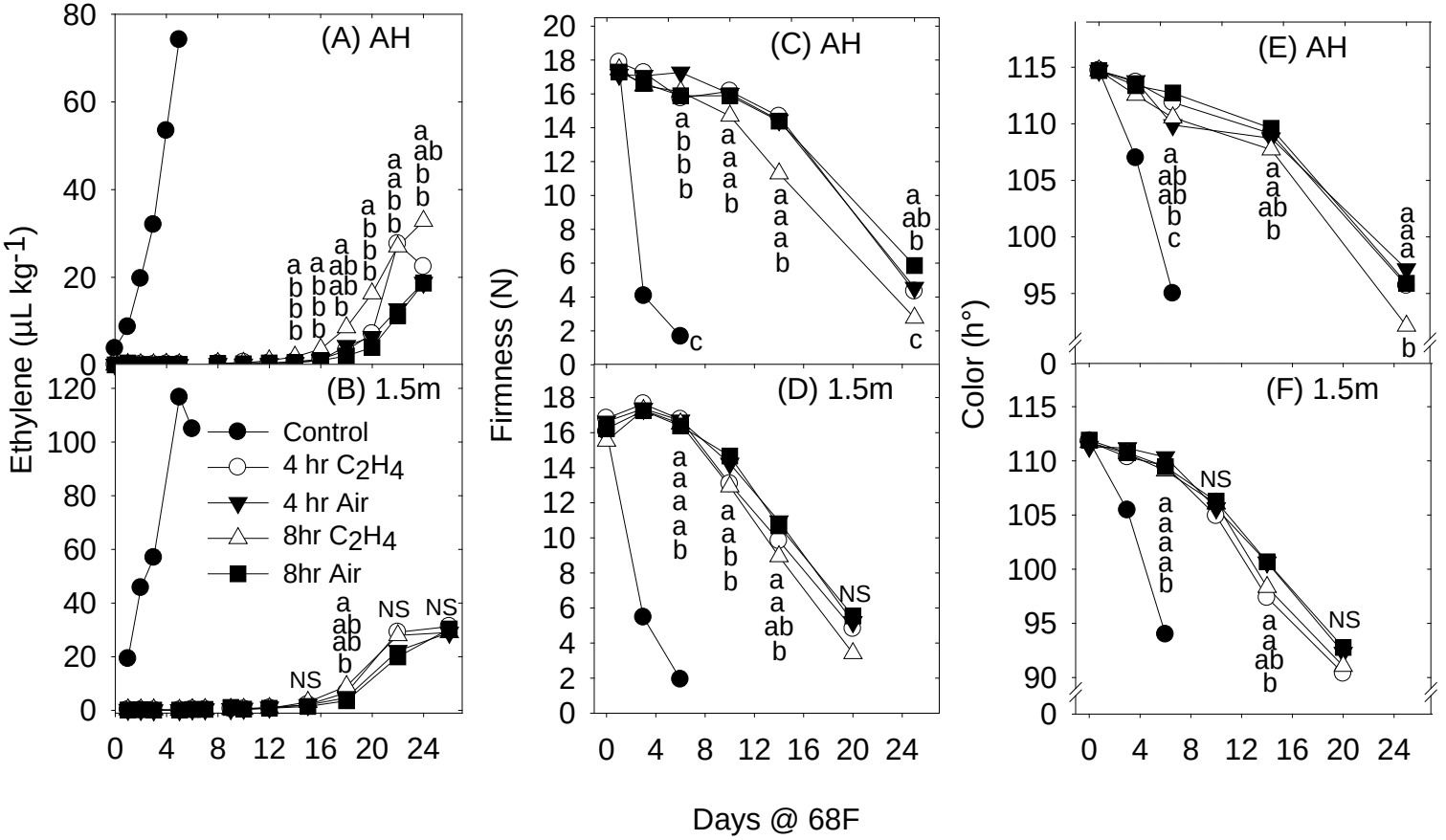


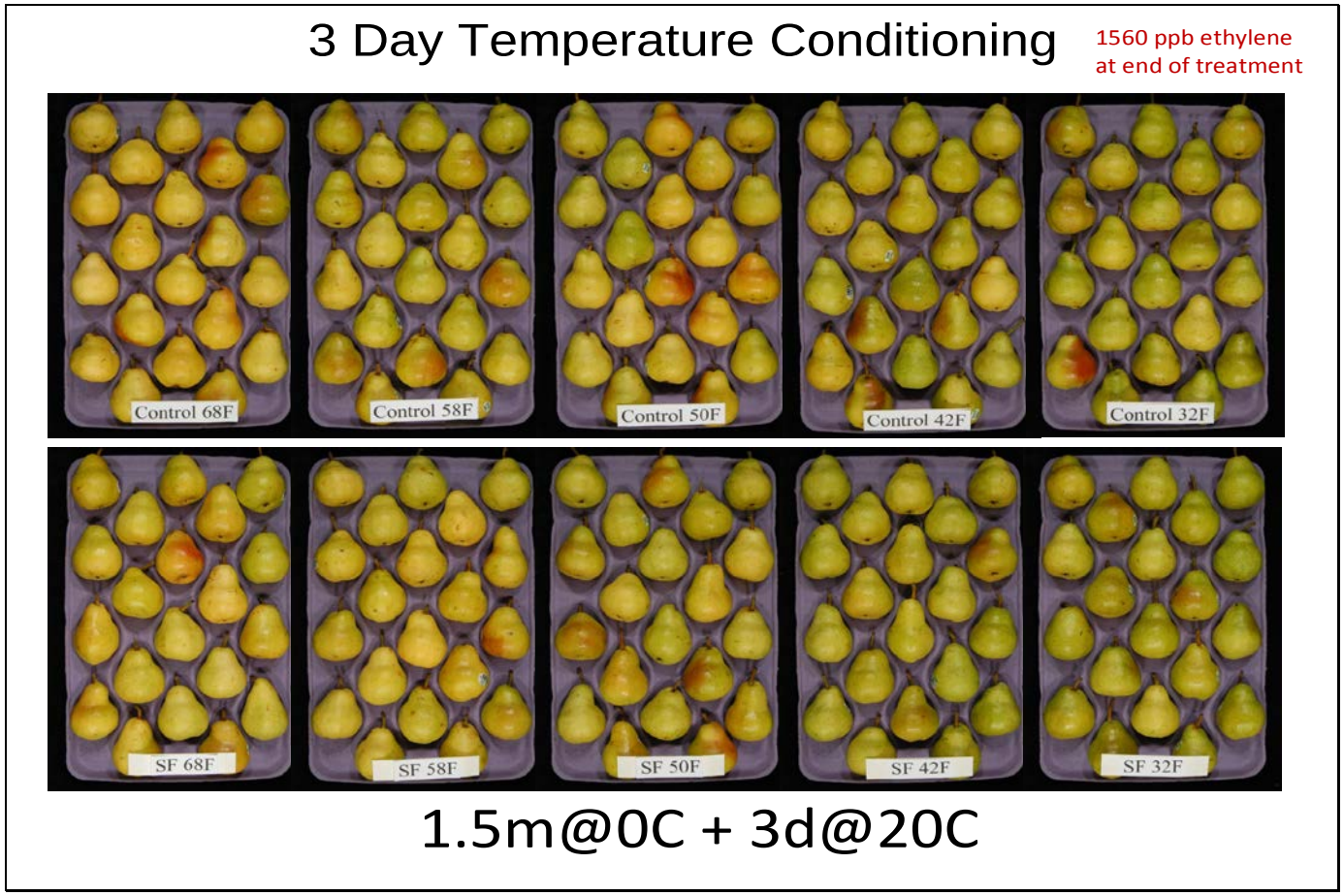
Figure 7. Ethylene production, firmness and color (hue angle decreases as the fruit change from green to yellow) during ripening immediately after harvest (AH) and after 1.5 months of storage at 32F (1.5m) of untreated and 1-MCP- treated fruit exposed to ethylene or air prior to 1-MCP treatment. Distinct letters within each evaluation represent means with statistically significant differences using LSD ($\alpha \leq 0.05$). Ethylene concentrations were 43 and 13ppb in the tent after 1-MCP treatment for fruit previously exposed for 4 hours (air and ethylene-treated) or 8 hours, respectively.

1 Day Temperature Conditioning

102 ppb ethylene measured in tent at the end of 12hr 1-MCP treatment (produced by fruit)

1.5m@0C + 6d@20C

There is a slight effect of temperature during 1-MCP treatment. Notice SF 32F is riper than SF 68F. We saw this effect in another experiment later.



(Very mild to no effect provided by 1-MCP. We think the main reason for this mild effect was the high concentration of ethylene in the tent 1560ppb) at the end of the 1-MCP treatment.

1-MCP Treatment Temperature

After Harvest - 14d@20C

1-MCP 12hr 0C = 51 ppb C2H4 at end of treatment.
1-MCP 24hr 0C = 26 ppb C2H4 at end of treatment.
1-MCP 12hr 20C = 23 ppb C2H4 at end of treatment.
1-MCP 24hr 20C = 16 ppb C2H4 at end of treatment.

Box in red shows ethylene concentration at the end of the 1-MCP treatment. 1-MCP treatment at lower temperatures seem to be less effective.

Storage Temperature after 1-MCP

Condition after 1.5m at 0, 5 or 10C

Control fruit at 10C was ripe after 2 weeks

Temperature plays an important role on how fruit recover after 1-MCP treatment.

Ethylene Competition

Ripened 6d@20C

Ripened 10d@20C

1-MCP+C2H4 means aprox. 300 ppb ethylene + 300 ppb 1-MCP in tent. Ethylene competition decreases 1-MCP efficacy.