Postharvest Treatments for Degreening of 'Villa franca' Lemons

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SUMMARY. 'Villa franca' is the main lemon (Citrus limon) variety in Israel, also cultivated in several other citrus-growing countries. In winter, the fruit turns yellow naturally, but during the summer and autumn, it remains green on the tree and requires postharvest ethylene treatment to stimulate color change from green to yellow. However, 'Villa franca' lemons are very sensitive to ethylene, which enhances development of reddish/brown peel blemishes known as red blotch. In the present study, we provide three different methods for postharvest degreening of 'Villa franca' lemons without causing red blotch. First is a slow process, involving natural degreening during 4–5 weeks of storage at 13 °C without ethylene exposure. Second is a moderate "under-degreening" process, involving a short 48-hour exposure to ethylene followed by 2 weeks of storage at 13 °C. Third is a fast process involving degreening with ethylene for up to 4 days at a constant high conditioning temperature of 30 °C or a combination of 24 hours of ethylene treatment at 30 °C followed by additional 72 hours of exposure to ethylene at 25 °C. Overall, 'Villa franca' lemon growers, packers, and exporters may now choose to use any of these proposed degreening procedures, according to commercial needs and market demands.

'Illa franca' lemons are visually similar to 'Eureka', which is the most important cultivated lemon variety worldwide; they have a smooth but thick rind, medium-large fruit, and an ovate-oblong shape (Ladaniya, 2008; Sinclair, 1984).

Lemons set fruit all year round, but natural color break occurs only during the winter when temperatures drop below 12.8 °C, which elicits a small peak in ethylene evolution (Cooper et al., 1969; Young and Erickson, 1961). However, during the summer and autumn, the fruit remain green on the tree and need to be degreened after harvest by exposure to ethylene. The typical recommended degreening treatment for lemons includes exposure to $\approx 4 \,\mu\text{L}\cdot\text{L}^{-1}$ ethylene for 3 to 5 d at 25 °C (Cohen et al., 1988; Porat, 2008). However, 'Villa franca' lemons are very sensitive to ethylene, which enhances development of superficial reddish/brown

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peel blemishes known as red blotch (Calero et al., 1981; Klotz, 1973).

In a previous study, Cohen et al. (1988) suggested that appearance of red blotch on lemons may result from stimulation of oxidative processes; they indicated that development of red blotch could be prevented by degreening the fruit at a high temperature of 30 °C or by dipping the fruit in an antioxidant solution before the ethylene degreening treatment. Unfortunately, although degreening at 30 °C efficiently prevents red blotch, this approach is not widely used commercially, especially since heating enhances costs, and because of concern that the process would cause excessive weight loss. Furthermore, dipping the fruit in antioxidant solutions only partially prevents red blotch and, therefore, is not suitable in commercial practice. It should be noted that red blotch symptoms become apparent already during or immediately after exposure to ethylene, and symptom incidence does not become worse following subsequent storage and marketing in an ethylenefree environment (unpublished data).

Overall, the goals of the current study were to develop alternative post-harvest treatments for degreening of 'Villa franca' lemons without causing red blotch symptoms. In addition, identification of such treatments may be adopted in the future also for degreening of other citrus species that are also sensitive to ethylene, such as 'Fallglo' tangerines [Citrus reticulata (Petracek and Montalvo, 1997)] and early-season 'Satsuma' mandarins [C. reticulata (Porat, 2008)].

Materials and methods

PLANT MATERIAL AND STORAGE CONDITIONS. Green 'Villa franca' lemons were harvested from commercial plantations from July through October during the 2009 and 2010 growing seasons. For postharvest storage experiments, fruit were stored at 13 °C and ≈90% relative humidity until the external peel turned yellow. In one treatment, part of the fruit was dipped in a commercial polyethylene-based citrus wax coating (Zivdar; Safe-Pack Products, Kfar Saba, Israel) before storage at 13 °C. Each treatment comprised at least 30 fruit and all experiments were repeated three times.

ETHYLENE DEGREENING. Fruit were selected for uniformity of size and color, divided into two lots, and exposed to air or ethylene for 24, 48, 72, or 120 h at 20 °C. In other experiments, fruit were exposed to ethylene for 1 or 5 d at a high temperature of 30 °C by placing them in a large 400-L heating incubator (model PIF400; Carbolyte, Sheffield, England). Fruit were exposed to ethylene by placing them in 30-L airtight sealed plastic tanks into which appropriate amounts of pure ethylene were injected to achieve a final concentration of 4 $\mu L \cdot L^{-1}$. Ethylene concentrations were verified by gas chromatography as described by Porat et al. (1999). The tanks were flushed daily to ensure that accumulated carbon dioxide levels did not exceed 0.2%. Control fruit were held in the same storage room at 20 °C, but without ethylene. Each treatment included 30 fruit and all experiments

Units			
To convert U.S. to SI, multiply by	U.S. unit	SI unit	To convert SI to U.S., multiply by
3.7854	gal	L	0.2642
1	ppm	$\mu L \cdot L^{-1}$	1
$(^{\circ}F - 32) \div 1.8$	°F	°C	$(1.8 \times {}^{\circ}\text{C}) + 32$

were repeated three times with similar results. Red blotch incidence was evaluated after 5 d.

COLOR MEASUREMENT. For color measurements, 15 green fruit were circled with a black marker and the peel color within these circles was determined by measuring the hue angles with a model CR-200 Chromo Meter (Minolta, Osaka, Japan). A hue angle of 120° represents green and of 90° represents yellow. In addition, peel color was rated visually on a scale of 1 to 5, with 1 = dark green, 2 = light green, 3 = color break, 4 = yellow, and 5 = gold.

JUICE TSS, ACIDITY, AND VITAMIN C CONTENTS. Total soluble solids (TSS) content in the juice was determined with a digital refractometer (model PAL-1; Atago, Tokyo, Japan), and acidity percentages were measured by titration to pH 8.3 with 0.1 M sodium hydroxide by an automatic titrator (model CH-9101; Metrohm, Herisau, Switzerland). Each measurement comprised five replications, each using juice collected from three different fruit (i.e., a total of 15 fruit per measurement).

Ascorbic acid (vitamin C) content in citrus juice was determined by titration with 2,6-dichlorophenolindophenol, according to Hiromi et al. (1980). Ascorbic acid levels were determined by comparing the titration volumes of citrus juices with that of 0.1% ascorbic acid (Sigma-Aldrich, St. Louis, MO).

STATISTICAL ANALYSIS. One-way analysis of variance and Tukey's honestly significant difference pairwise comparison tests were applied by the SigmaStat statistical software (Jandel Scientific Software, San Rafael, CA) and Excel (Microsoft Office; Mircosoft, Redmond, WA) programs.

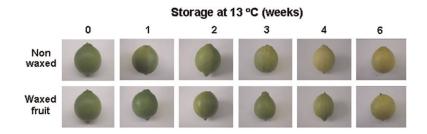
Results

EFFECTS OF POSTHARVEST STORAGE ON PEEL COLOR. Since exposure to ethylene enhances development of red blotch (Fig. 1), we first examined how long it would take for green lemons to turn yellow naturally without exposure to ethylene during postharvest storage at 13 °C. The results showed that, depending on the initial color of the fruit, green 'Villa franca' lemons turned yellow naturally (from hue angle of 118° to $\approx 95^{\circ}$) after about 4–5 weeks of storage at 13 °C (Fig. 2). However, if the fruit were waxed before storage, it took at least 6





Fig. 1. Appearance of reddish/brown peel blemishes (red blotch) on degreened 'Villa franca' lemons. The photographs were taken after 5 d of exposure [4 μ L·L⁻¹ (ppm) at 20 °C (68.0 °F)].



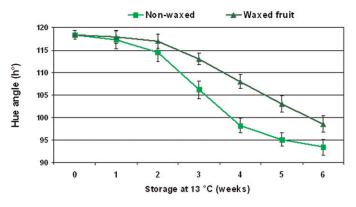


Fig. 2. Effects of storage duration and waxing on degreening of 'Villa franca' lemons. Green fruit remained untreated or were dipped in Zivdar wax solution (Safe-Pack Products, Kfar Saba, Israel) and afterward stored at 13 °C (55.4 °F) until achieving a minimum acceptable yellow color. Photographs show fruit color changes at weekly intervals. A hue angle of 120° represents green and of 90° represents yellow.

weeks for them to turn yellow and became suitable for marketing (Fig. 2). Increasing the storage temperature from 13 to 20 °C increased weight loss and shrinkage but barely shortened the time required for yellowing (data not shown).

EFFECTS OF DURATION OF ETHYLENE EXPOSURE ON PEEL COLOR AND DEVELOPMENT OF RED BLOTCH. Another strategy tested was the "underdegreening" approach; i.e., to test whether short exposures to ethylene might enhance peel color break without causing red blotch. It was found that as the duration of exposure to ethylene was increased from 24 to 48,

72, and 120 h the fruit became more vellow, but also developed more red blotch symptoms (Fig. 3). For example, fruit exposed to ethylene for just 48 h turned from green to color break and exhibited only 4% red blotch, whereas those exposed to ethylene for 120 h turned yellow but 65% of them exhibited red blotch (Fig. 3). Accordingly, we further examined how long it would take for the fruit to turn yellow following 24, 48, and 72 h exposures to ethylene followed by storage at 13 °C. It was found that short ethylene exposures of 24, 48, or 72 h reduced the time required for yellowing from 4 to 5 weeks in

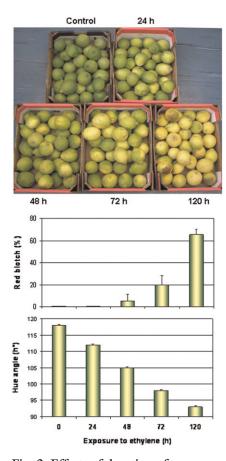


Fig. 3. Effects of duration of exposure to ethylene on peel color and development of red blotch on 'Villa franca' lemons. Green fruit were exposed to ethylene at 4 μL·L⁻¹ (ppm) and 20 °C (68.0 °F) for various periods of 0, 24, 48, 72, or 120 h, and afterward they were kept at the same temperature but without ethylene. Color and red blotch incidence were analyzed at the end of the experiment after 120 h. The photographs show the visual appearance of the fruit; the graphs illustrate changes in peel color and development of red blotch. Data are means ± ses of three replications, each including 30 fruit. A hue angle of 120° represents green and of 90° represents yellow.

untreated fruit to just 3, 2, or 1.5 weeks, respectively (Fig. 4). In light of these findings, we suggest a moderately time-consuming approach for degreening of 'Villa franca' lemons with minimum development of red blotch that includes a short exposure of just 48 h to ethylene, followed by 2 weeks more of postharvest storage at 13 °C to enable complete yellowing before marketing. Red blotch incidence did not increase or become worse following consequent postharvest storage without ethylene (data not shown).

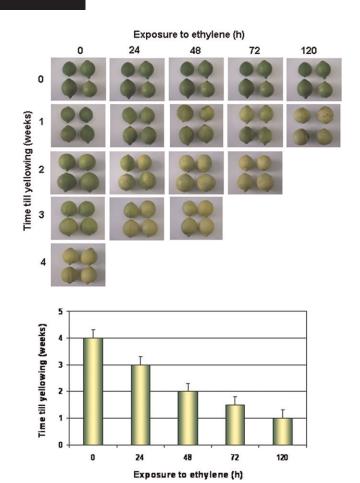


Fig. 4. Effects of exposure to ethylene and subsequent storage at 13 °C (55.4 °F) on degreening of 'Villa franca' lemons. Green fruit were exposed to ethylene at 4 μ L·L⁻¹ (ppm) and 20 °C (68.0 °F) for 0, 24, 48, 72, or 120 h, and the time to yellowing was recorded. The photographs show fruit color changes at weekly intervals; the graph bars shows storage time at 13 °C required until the fruit achieved the minimum acceptable yellow color, as related to ethylene exposure duration. Data are means ±SES of three replications, each involving 30 fruit.

Effects of degreening TEMPERATURES ON PEEL COLOR AND DEVELOPMENT OF RED BLOTCH. Cohen et al. (1988) found it possible to prevent red blotch development by degreening fruit at a high temperature of 30 °C. In the present study, we confirmed this finding and showed that exposure of green fruit to ethylene for 5 d at 30 °C promoted color change (yellowing) without causing red blotch (Fig. 5). Moreover, we showed that it was not necessary to actually degreen the fruit for 4-5 d at a constant high temperature of 30 °C, but rather that it was sufficient to use a temperaturecombination approach that comprised degreening at 30 °C for just 24 h and then completing the degreening process by continuing the exposure to ethylene at a moderate temperature of 25 °C (Fig. 5). This proposed new "temperature-combination" approach

enables reduction of the time required for degreening at a high temperature of 30 °C, thus reducing heating costs and preventing weight loss and shrinkage. It should be noted that none of these suggested degreening treatments had any significant effects on internal fruit quality parameters, such as juice TSS, acid, and vitamin C contents (data not shown).

Discussion

External color is a major quality parameter in lemons; in fact, consumers and supermarket chains are no longer willing to purchase green lemons but demand yellow fruit. Therefore, lemon growers and packers must treat lemons with ethylene following harvest to promote color change from green to yellow (Wardowsky et al., 2006).

In the present study, we examined three different procedures for

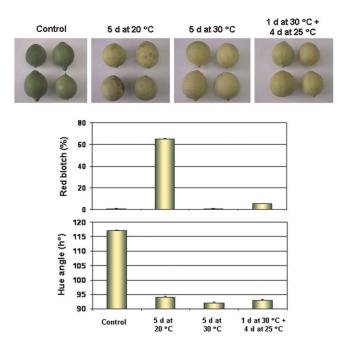


Fig. 5. Effects of different temperatures during ethylene degreening on peel color and development of red blotch on 'Villa franca' lemons. Green fruit were exposed to ethylene at $4~\mu L \cdot L^{-1}$ (ppm) for 5 d at either 20 or 30 °C (68.0 or 86.0 °F) or at a combination of 24 h exposure to ethylene at 30 °C followed by additional 96 h exposure to ethylene at 25 °C (77.0 °F). The photographs show the visual appearance of the fruit; the graphs illustrate changes in peel color and development of red blotch immediately after the 5-d degreening treatments. Data are means \pm SES of three replications, each including 30 fruit. A hue angle of 120° represents green and of 90° represents yellow.

postharvest degreening of 'Villa franca' lemons, a variety known to be sensitive to ethylene: 1) a slow degreening process-involving natural degreening during 4-5 weeks of storage at 13 °C without exposure to ethylene; 2) a moderate degreening process involving a short exposure of just 48 h to ethylene followed by 2 weeks of storage at 13 °C in air; and 3) a fast degreening process—involving exposure to ethylene for up to 4-5 d at a constant high conditioning temperature of 30 °C, or a combination of 24 h exposure to ethylene at 30 °C followed by an additional 72-96 h exposure to ethylene at 25 °C.

These proposed postharvest treatments provide 'Villa franca' lemon growers, packers, and exporters with three different strategies for postharvest degreening, without concern about red blotch. Choice among these treatments would depend on commercial needs and market demands. For example, if growers must harvest fruit

that reached their optimal size at the beginning of the summer, when they still have many winter-harvested yellow lemons in cold rooms, they might prefer the slow degreening process that takes a few weeks, which would enable them meantime to clear the stocks of fruit from their cold rooms. In contrast, if there is a good market price and high demand for lemons, growers and exporters might prefer the fast degreening procedure, which takes just a few days, so as to send the fruit to market as soon as possible.

Finally, it should be noted that although the present study dealt with 'Villa franca' lemons, the resulting degreening strategies might be usefully adopted also for degreening of other ethylene-sensitive citrus species. For example, a more or less similar "underdegreening" approach that included 24–48 h of exposure to ethylene followed by storage at 15 °C was previously recommended for commercial degreening of ethylene-sensitive 'Fallglo'

tangerines (Petracek and Montalvo, 1997).

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