Combined postharvest UV-C and 1methylcyclopropene (1-MCP) treatment, followed by storage continuously in low level of ethylene atmosphere improves the quality of Tahitian limes

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23 Abstract

- The green Tahitian limes (*Citrus latifolia*) were exposed to 7.2 kJ/m² UV-C and 0.5 μ L 24 L^{-1} 1-methylcyclopropene (1-MCP) treatments both separately and in combination. 25 After treatment, fruit were stored in ethylene free (ie air containing $< 0.005 \ \mu L \ L^{-1}$) or 26 27 $0.1 \ \mu L \ L^{-1}$ ethylene at 20°C and 100% RH. The results showed that UV-C treatment 28 delayed skin degreening and reduced endogenous ethylene production compared to 29 untreated control fruit, however these effects reduced over the storage time. As 30 expected, 1-MCP inhibited ethylene production, reduced calyx abscission and retained 31 peel greenness during the storage. Both of the combination treatments, 1-MCP + UV-C 32 and UV-C + 1-MCP reduced endogenous ethylene production and delayed skin 33 vellowing. In all treatments, UV-C and 1-MCP resulted in lower fruit respiration rates 34 than untreated control fruit, however this effect diminished during 7 and 14 days storage for fruits stored in air and 0.1 μ L L⁻¹ ethylene atmosphere, respectively. There was no 35 36 difference in weight loss, SSC, TA and SSC/TA ratio between the treatments and 37 storage conditions. The results suggest that a pre-storage UV-C treatment, followed by 38 storage at low level of ethylene improves the quality of limes, with the additional 39 improvement when combined with 1-MCP treatment prior or after UV-C irradiation. 40
- 41 Keywords : Citrus latifolia; quality; ethylene; respiration; colour; calyx abscission

42 Introduction

43 Green peel colour is an important quality attribute of the storage of Tahitian lime 44 (Citrus latifolia) where postharvest degreening of the peel can significantly downgrade 45 consumer acceptance. UV treatment has been reported to have beneficial effect on 46 maintaining postharvest quality of many horticultural produce. For example treatment with UV-C (100 -280 nm) has been reported to delay ripening and senescence in non-47 48 climacteric table grapes (Cantos et al. 2002), oranges (D'hallewin et al. 1999) grapefruit 49 (D'Hallewin et al. 2000) and climacteric mangoes (Gonzalez-Aguilar et al. 2007) and 50 tomatoes (Liu et al. 2012). UV-C irradiation has also been reported to prevent yellowing 51 of broccoli (Buchert et al. 2011). Specifically UV-B irradiation (280 -315 nm) treatment 52 has been shown to maintain lime peel colour (Kaewsuksaeng et al. 2011; Srilaong et al. 53 2011).

54 The recommended storage temperature for limes is 10°C (Burns 2016) and 55 storing fruit at higher temperatures can accelerate fruit senescence, where the main 56 deterioration is turning the peel colour from green to yellow. Although citrus fruit only 57 normally produce only low levels of ethylene, Goldschmidt (1998) suggested that even 58 these small amounts may play a role in the endogenous regulation of maturation and 59 senescence in citrus. Ethylene is a ubiquitous in the horticulture supply chain where the ethylene levels in the supermarkets have been shown to be 0.017-0.035 μ L L⁻¹ in the 60 wholesale markets and greater than 0.06 μ L L⁻¹ and distribution centres (Wills et al. 61 62 2000). 1- Methylcyclopropene (1-MCP) treatment has been shown to be very effective 63 in delaying yellowing and in extending the shelf life of West Indian limes (Citrus 64 aurantifolia, Swingle) (Win et al. 2006). They reported that limes treated with 250 or 500 nL L⁻¹ 1-MCP effectively delayed yellowing for 21 days at ambient storage (24-65

66 31°C and 73-81% RH). Also, 1-MCP treatment has also been reported to delay

67 yellowing in other horticultural produce such as on broccoli, where showed delayed 68 yellowing during storage after broccoli were exposed to 2.5 μ L L⁻¹1-MCP (Xu et al. 69 2016).

The effect of UV-C irradiation combined with 1-MCP treatment followed by
storage in air containing low level ethylene to stimulate the normal supply chain
conditions at 20°C on postharvest senescence of limes was studied in this experiment.
The aim of the experiment was to examine the single and combined effects of UV-C
and 1-MCP on lime quality at 20°C in air containing low levels of ethylene (0.1 µL L⁻¹).

75

76 Materials and methods

77 **Produce**

Commercial green Tahitian limes (*Citrus latifolia*) of uniform colour, shape, and size
and were free from damage were used in this experiment. The experiment was repeated
two times with different batches of fruit with three replicates within each batch.

81

82 1-Methylcyclopropene (1-MCP) and UV-C treatment and storage conditions

83 The UV-C treatments were conducted using a custom made light proof box fitted with

84 two germicidal lamps (Sahkyo Denki Co. Ltd G20T10 20 Watt, Low Pressure

85 Mercury). A SED008/W detector with PIR Irradiance Calibration at 254 nm was used

- 86 to monitor UV-C intensity. UV-C intensity was determined prior to treatment by
- 87 measuring the light intensity (kJm⁻²) using an International Light Technologies 1700
- 88 series research radiometer. The applied dose (kJm⁻²) was calculated by multiplying the
- 89 emitting UV light intensity with treatment time in seconds. Light intensity was

90	evaluated several times during the experiments to ensure consistent output. The limes
91	were placed approximately 20 cm from the UV-C light sources on one side then rotated
92	180°C and exposed again to ensure complete coverage. During the six minute treatment
93	the samples received 7.2 kJm ⁻² of radiation and no increase in peel temperature was
94	recorded using TinyTag data loggers. UV-C irradiation treatment was carried out at
95	room temperature ($20 \pm 1^{\circ}$ C) and relative humidity of about 80%, unless otherwise
96	stated. 1-MCP (0.5 $\mu L \ L^{\text{-1}})$ was applied in a 60 L sealed drum for 24 hours at 20°C and
97	85% RH, using SmartFresh TM powder (AgroFresh Solutions Inc., Philadelphia, PA,
98	USA) containing 0.34% 1-MCP as active ingredient.
99	Control fruits were not treated with UV-C or 1-MCP application, UV-C
100	application of 7.2 kJm ⁻² as a single treatment or was combined with 0.5 μ L L ⁻¹ 1-MCP
101	fumigation. For the combined treatments, 1-MCP fumigation was applied first followed
102	by UV-C treatment 24 hours later (1-MCP + UV-C). Another treatment, UV-C was
103	applied first and then 1-MCP was applied 24 hours later (UV-C + 1-MCP). After
104	treatment, all fruit were stored inside the containers with continuously exposed to air
105	(less than 0.005 μ L L ⁻¹ ethylene) in a flow through system (100 mL min ⁻¹) at 20°C and
106	100% RH or stored inside the containers with continuously exposed to 0.1 μ L L ⁻¹
107	ethylene in a flow through system (100 mL/min) at 20°C 100% RH.
108	

109 Determination of fruits quality attributes

110 Fruit were removed from storage at 7, 14, 21 and 28 days and assessed for weight loss,

111 calyx detachment, skin colour, respiration rate, ethylene production, soluble solids

112 content (SSC), titratable acidity (TA) and overall acceptability.

The weight loss percentages were calculated based on the initial weight of the fruit and weight after storage. Calyx detachment was assessed based on the scoring of its attachment to the fruit (1) or detachment (0). Peel colour was measured using a Minolta colorimeter (Minolta CR-400, Osaka) by hue angle value. Before measuring, the colorimeter was calibrated with a white standard calibrate plate. Each fruit, the hue value were measured the average of two points from calyx to blossom end.

119 The ethylene production and respiration rate was measured according to 120 Pristijono (2007), where limes were transferred to a sealed 1500 mL glass jar at 20°C 121 and after 2 hours incubation, a gas sample (1 mL) was collected in a syringe and the 122 ethylene and carbon dioxide content were analysed. Ethylene was measured by injecting 123 a gas sample into a gas chromatograph (Gow-Mac 580, Bridgewater NJ) and expressed 124 as $\mu L C_2 H_4 kg^{-1} h^{-1}$. Carbon dioxide concentration was measured to within 0.1% using 125 an ICA40 series low volume gas analysis system (International Controlled Atmosphere 126 Ltd., Kent, UK) and expressed as mL CO₂.kg⁻¹.h⁻¹.

Soluble solid content (SSC), expressed as °Brix, was measured from the pressed
juice of fruit with a digital refractometer (ATAGO Inc., Bellevue, WA, USA).

129 Titratable acidity (TA), expressed as % citric acid, was determined by titrating 1 mL

juice to pH 8.2 with a 0.1 N NaOH solution using an automatic titrator (Mettler ToledoT50, Switzerland).

The lime overall acceptability index were assessed visually based on the skin
colour, skin glossiness or/and calyx attachment, using the following scores of 1 = severe
degreening or calyx detached; 2 = severe degreening, dull skin or calyx detached; 3 =
slight degreening, shiny skin and calyx detached; 4 = green, shiny skin and calyx intact;
and 5 = fresh as just harvested. The overall acceptability index was calculated according

to Wang et al. (2015) with slight modifications. The calculation as overall acceptability
index (%) = ∑[(acceptability score) × (number of fruit at this level)] / (highest level ×
total number of fruit in the treatment) × 100.

140

141 Statistical analysis

142 The experiment was performed in a completely randomized design with three

143 replications in each of the two batches. The initial colour of the limes of the two batches

- 144 were similar, as measured by the hue angle which show no significant differences
- 145 (p < 0.05) denoting homogeneity in colour between the batches. Therefore the data from
- both batches were combined and analysed together for a total of six replicates for the
- 147 experiment. Each replication consisted of five treatment units of untreated control

148 (without UV-C or 1-MCP), UV-C alone, 1-MCP alone, 1-MCP + UV-C and UV-C + 1-

- 149 MCP. Each treatment unit consisted of 20 fruits. The two-way ANOVA and the Least
- 150 Significance Difference (LSD) tests were conducted using the SAS software (SAS Ver.
- 151 9.4, USA). Differences among means were analysed at a significance level of p < 0.05.

152

153 Results and discussion

154 The initial quality of the limes at the beginning of the experiment was excellent with

- uniform green peel colour ; hue value of skin 118.3 ± 0.3 , ethylene production rate
- $156 \qquad 0.014 \pm 0.001 \ \mu L \ C_2 H_4. kg^{-1}. h^{-1}, \ respiration \ rate \ 12.18 \pm 0.47 \ m L \ CO_2. kg^{-1}. h^{-1}, \ SSC \ 8.4$

157 ± 0.2 °Brix and TA 5.86 ± 0.27 % citric acid.

158

159 Calyx abscission

160	The presence of the calyx (button) on the fruit is a good indicator of quality for many
161	consumers. The effect of postharvest 1-MCP, UV-C and ethylene treatment on calyx
162	retention is presented in Table 1, and the results show that in general, calyx detachment
163	was significantly affected by UV-C, 1-MCP and ethylene treatments .After 21 days
164	storage at 20°C, the percentage of intact calyx for fruits treated with UV-C combined
165	with 1-MCP was higher than untreated fruits in both storage atmospheres. Comparing
166	the different storage atmospheres, fruit treated with the combination UV-C and 1-MCP
167	and stored in 0.1 μ L L ⁻¹ ethylene had higher calyx retention than fruits stored in air (less
168	than 0.005 μ L L ⁻¹ ethylene) during storage for 21 days.
169	
170	Weight loss
171	In general, there was no difference between the different pre-storage treatments on
171 172	In general, there was no difference between the different pre-storage treatments on weight loss from the limes during storage. Limes treated with UV-C and 1-MCP both
172	weight loss from the limes during storage. Limes treated with UV-C and 1-MCP both
172 173	weight loss from the limes during storage. Limes treated with UV-C and 1-MCP both separately and in combination did not significantly affect the weight loss during storage
172 173 174	weight loss from the limes during storage. Limes treated with UV-C and 1-MCP both separately and in combination did not significantly affect the weight loss during storage (Table 1). As expected, the different storage atmospheres did not contribute to water
172 173 174 175	weight loss from the limes during storage. Limes treated with UV-C and 1-MCP both separately and in combination did not significantly affect the weight loss during storage (Table 1). As expected, the different storage atmospheres did not contribute to water loss for all treatments, as all atmospheres were at 100% RH which maintained fruit
172 173 174 175 176	weight loss from the limes during storage. Limes treated with UV-C and 1-MCP both separately and in combination did not significantly affect the weight loss during storage (Table 1). As expected, the different storage atmospheres did not contribute to water loss for all treatments, as all atmospheres were at 100% RH which maintained fruit weight during storage. The time in storage was a significant factor affecting weight loss,
172 173 174 175 176 177	weight loss from the limes during storage. Limes treated with UV-C and 1-MCP both separately and in combination did not significantly affect the weight loss during storage (Table 1). As expected, the different storage atmospheres did not contribute to water loss for all treatments, as all atmospheres were at 100% RH which maintained fruit weight during storage. The time in storage was a significant factor affecting weight loss, where the longer time in storage resulted in the greatest weight loss through respiration
172 173 174 175 176 177 178	weight loss from the limes during storage. Limes treated with UV-C and 1-MCP both separately and in combination did not significantly affect the weight loss during storage (Table 1). As expected, the different storage atmospheres did not contribute to water loss for all treatments, as all atmospheres were at 100% RH which maintained fruit weight during storage. The time in storage was a significant factor affecting weight loss, where the longer time in storage resulted in the greatest weight loss through respiration

181 Limes are classified as a non-climacteric fruit which characteristically do not exhibit

- 182 significant a burst of ethylene production after harvest (Burns 2016). Although non-
- 183 climacteric fruits do not exhibit any clear increases in ethylene production rates during

184 ripening, in certain cases, exposure to exogenously applied ethylene may stimulate 185 certain ripening-related processes, such as degreening of citrus fruit (Reid 2002). 186 In this study, untreated fruit produced significant higher in ethylene production during storage than all other treated fruits (Fig.1). Treating limes with 7.2 kJm⁻² UV-C 187 188 alone had the higher ethylene production than other treatments, whilst ethylene 189 production rates in fruit treated with 1-MCP alone and in combination with UV-C 190 treatment resulted in low of ethylene production rates (Fig.1). These results show that 191 UV-C treatment suppressed ethylene production and the additional of 1-MCP futher 192 suppresed ethylene production, regardless the application of 1-MCP prior or after UV-C 193 treatment. These results also show that UV-C effect associated with the ethylene 194 synthesis due to UV-C treatment alone without ethyelene interference by combined with 195 1-MCP provided greater effect, especially when treated fruits were stored in ethylene-196 free atmosphere.

197 Combining the storage time data, the result showed that storage time 198 significantly (p<0.05) affected the endogeneous ethylene production, where the ethylene 199 production increased significantly after 7 days storage, and remained at the level of 0.08 μ L C₂H₄.kg⁻¹.h⁻¹ for 28 days storage. Moreover, there was significant difference in the 200 201 ethylene production rates between the two storage atmospheres, where fruits were stored in air produced higher ethylene than fruits were stored at 0.1 μ L L⁻¹ ethylene 202 atmosphere, with the overall ethylene production of 0.074 and 0.054 μ L C₂H₄.kg⁻¹.h⁻¹ 203 for fruits that were stored in air and 0.1 μ L L⁻¹ ethylene, respectively. These results 204 suggest that exogenous ethylene application (0.1 μ L L⁻¹ ethylene) supressed 205 206 enodgenous ethylene production rates during storage for 28 days.

208 Skin colour

209

210 colour of peel as this is a key determinant of consumer preference. (Kaewsuksaeng et

al., 2015) Peel colour as measured by hue angle was significantly influenced by storage

The most important factor for marketing of Tahitian limes is the retention of the green

time and pre-storage treatment, where both UV-C and 1-MCP treatment applied

213 separately and in combination maintained green colour of the skin during storage (Fig.

214 2). UV-C treatment has been reported to delay de-greening of horticultural produce.

215 For example Costa et al. (2006) showed that broccoli treated with 10 kJm⁻² UV-C

216 delayed yellowing after storage at 20°C for 6 days. In this experiment, UV-C treated

217 fruits had significantly higher in hue value (greener peel colour) than untreated fruits.

218 The retention of peel green colour was significantly greater (p<0.05) when UV-C

treatment was combined with 1-MCP.

220 For the first 14 days storage, there were no significant different between the 221 treatments, where all fruits had similar green colour. In the later stage of storage, the 1-222 MCP treated fruits (alone or in combination with UV-C) maintained peel green colour. 223 Fruits treated with UV-C alone (without 1-MCP) resulted in quicker yellowing peel 224 colour than 1-MCP treated fruits included UV-C+1-MCP and 1-MCP+UV-C. This 225 indicated that although UV-C delayed degreening, this effect was enhanced with 1-226 MCP fumigation (either prior or after UV-C treatment). However, 1-MCP treatmet 227 alone was effcetive in maitaining peel colour. The results in agreement with previous reports by Win et al. (2006) who found that Western Indian limes treated with 500 nL L⁻ 228 229 ¹1-MCP retained their green peel (hue angle value 110.7) at 12 days. Other studies have 230 also been reported that 1-MCP treatment delayed degreening in other horticultural 231 produce such as on broccoli florets (Gómez-Lobato et al. 2012; Xu et al. 2016).

In this study, the highest hue value was obtained by application of 1-MCP prior UV-C treatment (1-MCP+UVC). The results suggest that the skin degreening may be partially ethylene dependent since 1-MCP+UV-C treated fruit had low ethylene production but produced high hue value. These results an agreement with the report by Barsan et al. (2010) and Kahlau and Bock (2008) who found that tomato skin colour changes are regulated by ethylene.

238 Comparing the storage conditions, the rate of green colour loss from untreated peel was relatively high and occurred more greatly in fruits stored in 0.1 µL L⁻¹ethylene 239 240 atmosphere (Fig.2). The minimum acceptable hue value for Tahitian limes is 108 (refer 241 to score 3 for acceptability index). In this study, the lime to reach unacceptable peel colour was 3 days guicker in fruits stored in 0.1 µL L⁻¹ ethylene atmosphere than stored 242 243 in air. These results showed that exogenous ethylene affected the peel colour changes 244 during storage. This result differ with previous reported by Porat et al. (1999) who 245 reported that exogenous ethylene applied to promote degreening peel colour in citrus. 246 The result suggests that fruits stored in atmosphere containing 0.1 μ L L⁻¹ ethylene 247 continuously affect the treatment of UV-C and 1-MCP both separately and in 248 combination on degreening of lime peel.

249

250 **Respiration rate**

251 The ripening of non-climacteric fruit such as citrus are characterised without any

252 increase in fruit respiration rate (Eaks 1970). This was also observed in this experiment

253 (Table 3), where respiration rates across all treatments and storage times ranged from

- 254 12.6 to 19.5 mL CO_2 .kg⁻¹.h⁻¹. After 7 days storage, the untreated fruit had significantly
- 255 higher respiration rates than fruit treated with 1-MCP or 1-MCP+UVC, in both storage

256	atmospheres. These effects remained after 14 days for fruits stored in 0.1 μ L L ⁻¹
257	ethylene, however, there was no pre-storage treatment effects when the fruit were
258	stored in air (less than 0.005 μ L L ⁻¹ ethylene) atmosphere. This result was expected,
259	since if the 1-MCP blocks the ethylene receptor, the respiration remained low for fruits
260	were stored in 0.1 μ L.L ⁻¹ ethylene atmosphere. For fruits stored in less than 0.005
261	μ L.L ⁻¹ , there was no difference in respiration rate between untreated and all treated
262	fruits. These results suggest that the respiration increased with the presence of ethylene.
263	Respiration rate was not greatly affected by UV-C treatment apart from a
264	significant decrease in rate after treated fruits were stored for 14 days in air at 20°C with
265	13.74 ml CO ₂ .kg ⁻¹ .h ⁻¹ . While UV-C treated fruits were store in 0.1 μ L.L ⁻¹ ethylene, the
266	respiration rate was significantly lower than untreated limes, however these effects
267	reduced over the storage time. Even though the effects of UV-C treatment alone on
268	respiration rate were not as marked as the effect of ethylene production, these results
269	suggest that UV-C treatment combined with 1-MCP followed by storage in air
270	containing 0.1 μ L.L ⁻¹ ethylene at 20°C maintained limes quality by maintaining
271	respiration rate during storage as a natural ripening of citrus fruit.

273 SSC, TA and SSC/TA ratio

274 UV-C treatment has been reported to influence the SSC or TA in a range of horticultural

produce. For example Charles et al. (2016) reported that tomatoes treated with 3.7 kJm⁻²

276 UV-C followed by storage at 15°C for 15 days resulted in lower sugar content and

277 higher in acid titre than untreated fruits. The results from this study showed that in

- 278 general SSC and TA were not affected by UV-C treatment alone or in combination with
- 279 1-MCP (Table2). These results are consistent with previous reports that showed

exposure to 1-MCP did not affect internal properties (SSC and TA) in citrus fruit (Dou
et al. 2005; Kluge et al. 2003; Porat et al. 1999; Salvador et al. 2006).

282 The SSC/TA ratio is an important parameter related with quality characteristics 283 of citrus fruits (Barros et al. 2012). In this study, comparing the storage conditions, 284 there was no difference in SSC, TA and SSC/TA ratio between limes that were stored in air (less than 0.005 μ L L⁻¹) and 0.1 μ L L⁻¹ ethylene atmospheres. These results suggest 285 that UV-C treatment alone or in combination with 1-MCP, followed by storage under 286 287 low level ethylene can be applied without affecting the SSC or TA. Thus, UV-C alone 288 or in combination with 1-MCP is a potential postharvest treatment for the maintaining 289 of limes' quality during storage in actual supply chain conditions.

290

291 Acceptability index

The overall cosmetic acceptability of the limes index were assessed visually based on the skin colour, skin glossiness or/and calyx intact. The effect of UV-C and 1-MCP both separately or in combination is presented in Table 3 and the results show that fruit treated with UV-C and 1-MCP alone or in combination had higher overall acceptability than untreated fruits in both storage atmospheres.

Within the treated fruit, UV-C treatment resulted in fruit with significantly
lower acceptability index than fruits treated by 1-MCP alone or in combination with
7.2 kJm⁻² UV-C after 21 days storage in both storage atmospheres. The higher
acceptability index during the earlier stages of storage (up to 21 days), may be
associated with the peel colour, since after 21 days storage, UV-C treated limes were

302 more yellow (lower hue angle). These results show that limes treated with UV-C

303	maitained a better acceptability after 21 days storage, the greater acceptability index
304	when combined with 0.5 μ L L ⁻¹ 1-MCP prior or after UV-C treament.

306 Conclusions

Our study showed the application of 7.2 kJm⁻² UV-C and 0.5 µL L⁻¹ 1-MCP separately 307 308 or in combination, followed by storage at 20°C in low level of ethylene atmosphere 309 improved lime fruit quality compared to untreated fruit. The UV-C treatment alone 310 improved lime fruit quality by delaying peel yellowing and this effect was greater when 311 combined with 1-MCP. There was no significant difference effect of 1-MCP applied 312 prior or after UV-C treatment on lime quality. The application UV-C and 1-MCP did 313 not affect weight loss, SSC nor TA. Overall, the UV-C treatment combined with 1-314 MCP resulted in improved fruit quality by delaying the peel degreening, maintaining the 315 attachment of the calyx, maintained low ethylene production and improved the 316 acceptability index. More study is required to assess the effect of application of UV-C 317 combined with 1-MCP, followed by storage in different temperatures (such as 10°C) to 318 determine if the mode of action of UV-C is similar with this study. 319 320 Acknowledgements

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326 References

- 327 Barros HR, Ferreira TA, Genovese MI (2012). Antioxidant capacity and mineral
- 328 content of pulp and peel from commercial cultivars of citrus from Brazil. Food Chem
- 329 134:1892-1898. doi: 10.1016/j.foodchem.2012.03.090
- 330 Barsan C, Sanchez-Bel P, Rombaldi C, Egea I, Rossignol M, Kuntz M, Zouine M,
- 331 Latché A, Bouzayen M, Pech JC (2010). Characteristics of the tomato chromoplast
- revealed by proteomic analysis. J Exp Bot 61: 2413-2431. doi: 10.1093/jxb/erq070
- 333 Buchert AM, Civello PM, Martinez GA (2011). Effect of hot air, UV-C, white light and
- 334 modified atmosphere treatments on expression of chlorophyll degrading genes in
- 335 postharvest broccoli (Brassica oleracea L.) florets. Sci Hortic 127: 214-219. doi:
- **336** 10.1016/j.scienta.2010.11.001
- 337 Burns JK (2016). Lime. In The Commercial Storage of Fruits, Vegetables, and Florist
- and Nursery Stocks, N.C. Gross, C.Y. Wang, and M. Salveit, eds. (USDA, ARS), pp.
 339 390-391.
- 340 Cantos E, Espin JC, Tomas-Barberan FA (2002). Postharvest stilbene-enrichment of red
- and white table grape varieties using UV-C irradiation pulses. J Agric Food Chem 50:
- 342 6322-6329. doi: 10.1021/jf020562x
- 343 Charles MT, Arul J, Charlebois D, Yaganza ES, Rolland D, Roussel D, Merisier MJ
- 344 (2016). Postharvest UV-C treatment of tomato fruits: Changes in simple sugars and
- 345 organic acids contents during storage. Lwt-Food Sci Technol 65: 557-564. doi:
- 346 10.1016/j.lwt.2015.08.055
- 347 Costa L, Vicente AR, Civello PM, Chaves AR, Martinez GA (2006). UV-C treatment
- delays postharvest senescence in broccoli florets. Postharvest Biol Technol 39: 204-210.
- 349 doi:10.1016/j.postharvbio.2005.10.012

- 350 D'hallewin G, Schirra M, Manueddu E, Piga A, Ben-Yehoshua S (1999). Scoparone and
- 351 Scopoletin Accumulation and Ultraviolet-C Induced Resistance to Postharvest Decay in
- 352 Oranges as Influenced by Harvest Date. J Am Soc Hortic Sci 124: 702-707
- 353 D'Hallewin G, Schirra M, Pala M, Ben-Yehoshua S (2000). Ultraviolet C Irradiation at
- 354 0.5 kJ/m² Reduces Decay without Causing Damage or Affecting Postharvest Quality of
- 355 Star Ruby Grapefruit (C. paradisi Macf.). J Agric Food Chem 48: 4571-4575. doi:
- 356 10.1021/jf000559i
- 357 Dou H, Jones S, Ritenour M (2005). Influence of 1-MCP application and concentration
- 358 on post-harvest peel disorders and incidence of decay in citrus fruit. J Hortic Sci
- 359 Biotechnol 80: 786-792. doi: 10.1080/14620316.2005.11512015
- 360 Eaks IL (1970). Respiratory Response, Ethylene Production, and Response to Ethylene
- 361 of Citrus Fruit during Ontogeny. Plant Physiol 45: 334-338.
- 362 Goldschmidt EE (1998). Ripening of citrus and other non-climacteric fruits : a role for
- 363 ethylene. Acta Hortic 463: 335-340. doi: 10.17660/ActaHortic.1998.463.42
- 364 Gómez-Lobato ME, Hasperué JH, Civello PM, Chaves AR, Martínez GA (2012). Effect
- 365 of 1-MCP on the expression of chlorophyll degrading genes during senescence of
- 366 broccoli (Brassica oleracea L.). Sci Hortic 144: 208-211. doi:
- 367 http://dx.doi.org/10.1016/j.scienta.2012.07.017
- 368 Gonzalez-Aguilar GA, Zavaleta-Gatica R, Tiznado-Hernandez ME (2007). Improving
- 369 postharvest quality of mango 'Haden' by UV-C treatment. Postharvest Biol Technol 45:
- 370 108-116. doi: 10.1016/j.postharvbio.2007.01.012
- 371 Kaewsuksaeng S, Tatmala N, Srilaong V, Pongprasert N (2015). Postharvest heat
- 372 treatment delays chlorophyll degradation and maintains quality in Thai lime (Citrus

- aurantifolia Swingle cv. Paan) fruit. Postharvest Biol Technol 100: 1-7. doi:
- 374 http://dx.doi.org/10.1016/j.postharvbio.2014.09.020
- 375 Kaewsuksaeng S, Urano Y, Aiamla-or S, Shigyo M, Yamauchi N (2011). Effect of UV-
- 376 B irradiation on chlorophyll-degrading enzyme activities and postharvest quality in
- 377 stored lime (Citrus latifolia Tan.) fruit. Postharvest Biol Technol 61: 124-130. doi:
- 378 http://dx.doi.org/10.1016/j.postharvbio.2011.02.014
- 379 Kahlau S, Bock R (2008). Plastid Transcriptomics and Translatomics of Tomato Fruit
- 380 Development and Chloroplast-to-Chromoplast Differentiation: Chromoplast Gene
- 381 Expression Largely Serves the Production of a Single Protein. Plant Cell 20: 856-874.
- 382 doi: 10.1105/tpc.107.055202
- 383 Kluge RA, Jomori MLL, Jacomino AP, Vitti MCD, Padula M (2003). Intermittent
- 384 warming in 'Tahiti' lime treated with an ethylene inhibitor. Postharvest Biol Technol
- 385 29: 195-203. doi: http://dx.doi.org/10.1016/S0925-5214(03)00022-X
- 386 Liu C.H, Cai LY, Lu XY, Han XX, Ying TJ (2012). Effect of Postharvest UV-C
- 387 Irradiation on Phenolic Compound Content and Antioxidant Activity of Tomato Fruit
- 388 During Storage. J Integr Agric 11: 159-165. doi: http://dx.doi.org/10.1016/S1671-
- 389 2927(12)60794-9
- 390 Porat R, Weiss B, Cohen L, Daus A, Goren R, Droby S (1999). Effects of ethylene and
- 391 1-methylcyclopropene on the postharvest qualities of 'Shamouti' oranges. Postharvest
- 392 Biol Technol 15: 155-163. doi: http://dx.doi.org/10.1016/S0925-5214(98)00079-9
- 393 Pristijono P (2007). Use of nitric oxide to extend the postharvest life of fresh-cut and
- intact fruits and vegetables. (PhD). The University of Newcastle, Australia, pp. 223.

- 395 Reid SM (2002). Ethylene in postharvest technology (Oakland, CA: Postharvest
- 396 Technology of Horticultural Crops, Regents of the University of California, Division of
- 397 Agricultural and Natural Resources).
- 398 Salvador A, Carvalho CP, Monterde A, Martinez-Javega JM (2006). Note. 1-MCP
- 399 effect on chilling injury development in 'Nova' and 'Ortanique' mandarins. Food Sci
- 400 Technol Int 12: 165-170. doi: 10.1177/1082013206063736
- 401 Srilaong V, Aiamla-or S, Soontornwat A, Shigyo M, Yamauchi N (2011). UV-B
- 402 irradiation retards chlorophyll degradation in lime (Citrus latifolia Tan.) fruit.
- 403 Postharvest Biol Technol 59: 110-112. : doi:
- 404 http://dx.doi.org/10.1016/j.postharvbio.2010.07.006
- 405 Wang J, You Y, Chen W, Xu Q, Wang J, Liu Y, Song L, Wu J (2015). Optimal
- 406 hypobaric treatment delays ripening of honey peach fruit via increasing endogenous
- 407 energy status and enhancing antioxidant defence systems during storage. Postharvest
- 408 Biol Technol 101: 1-9. doi: http://dx.doi.org/10.1016/j.postharvbio.2014.11.004
- 409 Wills RBH, Warton MA, Ku VVV (2000). Ethylene levels associated with fruit and
- 410 vegetables during marketing. Aust J Exp Agric 40: 465-470.
- 411 Win TO, Srilaong V, Heyes J, Kyu KL, Kanlayanarat S (2006). Effects of different
- 412 concentrations of 1-MCP on the yellowing of West Indian lime (Citrus aurantifolia,
- 413 Swingle) fruit. Postharvest Biol Technol 42: 23-30. doi:
- 414 10.1016/j.postharvbio.2006.05.005
- 415 Xu F, Wang H, Tang Y., Dong S, Qiao X, Chen X, Zheng Y (2016). Effect of 1-
- 416 methylcyclopropene on senescence and sugar metabolism in harvested broccoli florets.
- 417 Postharvest Biol Technol 116: 45-49. doi:
- 418 http://dx.doi.org/10.1016/j.postharvbio.2016.01.004
- 419

420 Table 1. Weight loss and calyx intact percentage of limes after treated with UV-C

421 and/or 1-MCP, followed by storage up to 28 days at	20°C.
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Storage /		Weight	loss (%)			Calyx in	ntact (%)	
Treatments	day 7	day 14	day 21	day 28	day 7	day 14	day 21	day 28
$< 0.005 \ \mu L.L^{-1} \ e$	ethylene							
Control	0.3 ^a	0.3 ^{ab}	0.5 ^a	0.7 ^a	79 ^a	67 ^a	75 ^a	58 ^a
1-MCP	0.2 ^a	0.4 ^a	0.5 ^a	0.7 ^a	96 ^b	92 ^b	88 ^a	83 ^{ab}
UVC	0.2 ^a	0.4 ^a	0.5 ^a	0.6 ^a	96 ^b	96 ^b	88 ^a	75 ^{ab}
1-MCP+UVC	0.2 ^a	0.3 ^{ab}	0.5 ^a	0.6 ^a	100 ^b	92 ^b	92 ^b	92 ^{ab}
UVC+1-MCP	0.2	0.2 ^b	0.5 ^a	0.7 ^a	100 ^b	92 ^b	92 ^b	83 ^a
<u>0.1 $\mu L.L^{-1}$ ethylene</u>								
Control	0.2 ^a	0.3 ^a	0.5 ^a	0.8 ^a	88 ^b	71 ^a	88 ^b	79 ^{ab}
1-MCP	0.2 ^a	0.3 ^a	0.4 ^b	0.6^{b}	100 ^b	96 ^b	92 ^a	79 ^{ab}
UVC	0.2^{a}	0.4^{a}	0.5 ^a	0.6^{b}	100 ^b	96 ^b	71 ^b	75 ^b
1-MCP+UVC	0.2 ^a	0.2 ^a	0.5 ^a	0.6^{b}	100 ^b	100 ^b	100 ^a	79 ^{ab}
UVC+1-MCP	0.2 ^a	0.3 ^a	0.5 ^a	0.6 ^b	100 ^b	100 ^b	100 ^a	92 ^a

Values are the mean of 6 replicates. Letters indicate mean values at the same columns, treatments and storage time that are statistically different (P < 0.05)

Storage /	SSC (°Brix)				TA (% citric acid)			
Treatments	day 7	day 14	day 21	day 28	day 7	day 14	day 21	day 28
$< 0.005 \ \mu L.L^{-1} \ e$	<u>thylene</u>							
Control	8.5 ^a	8.2 ^a	8.8 ^{ab}	8.9 ^a	6.28 ^a	6.40 ^a	6.54 ^a	6.60 ^a
1-MCP	8.8 ^a	8.9 ^b	8.9 ^a	8.8 ^a	6.37 ^a	6.35 ^a	6.69 ^a	6.60 ^a
UVC	8.7 ^a	8.9 ^b	8.4 ^b	8.9 ^a	6.65 ^a	6.36 ^a	6.53 ^a	6.56 ^a
1-MCP+UVC	8.5 ^a	8.7 ^{ab}	8.7 ^{ab}	8.7 ^a	6.45 ^a	6.00 ^a	6.37 ^a	6.50 ^a
UVC+1-MCP	9.0 ^a	9.2 ^b	8.9 ^a	9.1 ^a	6.44 ^a	6.18 ^a	6.35 ^a	6.75 ^a
<u>0.1 µL.L⁻¹ ethyle</u>	e <u>ne</u>							
Control	8.6 ^a	8.7 ^a	8.7 ^a	8.8 ^a	6.28 ^a	6.43 ^a	6.48 ^a	6.53 ^a
1-MCP	8.5 ^a	8.8 ^a	8.7 ^a	9.0 ^a	6.37 ^a	6.27 ^a	6.54 ^a	6.40 ^a
UVC	8.7 ^a	8.9 ^a	8.9 ^a	8.9 ^a	6.34 ^a	6.52 ^a	6.15 ^a	6.66 ^a
1-MCP+UVC	8.8 ^a	9.0 ^a	8.7 ^a	9.0 ^a	6.33 ^a	6.61 ^a	6.82 ^a	6.44 ^a
UVC+1-MCP	8.8 ^a	9.0 ^a	8.7 ^a	9.0 ^a	6.36 ^a	6.47 ^a	6.45 ^a	6.64 ^a

426 Table 2. Soluble solids content (SSC) and titratable acidity (TA) of limes after treated

427 with UV-C and/or 1-MCP, followed by storage up to 28 days at 20°C.

Values are the mean of 6 replicates. Letters indicate mean values at the same columns, treatments and storage time that are statistically different (P < 0.05)

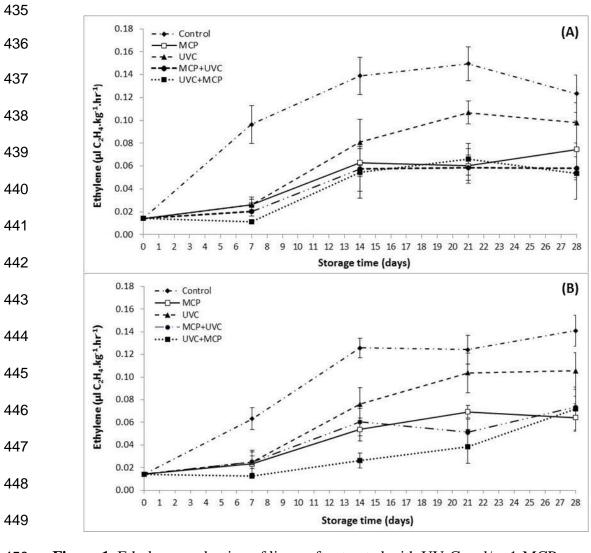
Table 3. Respiration rate and acceptability index of limes after treated with UV-C

	431	and/or 1-MCP, followed	d by	storage up to	28 days at 20°C.
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Storage /	Respiration rate (mlCO ₂ .kg ⁻¹ .hr ⁻¹)				Acceptability index (%)			
Treatments	day 7	day 14	day 21	day 28	day 7	day 14	day 21	day 28
$< 0.005 \ \mu L.L^{-1} \ ethylene$								
Control	18.61 ^a	14.78^{a}	16.82 ^a	18.65 ^a	60 ^a	43 ^a	33 ^c	22 ^c
1-MCP	13.68 ^b	13.83 ^a	15.45 ^a	16.00 ^{ab}	74 ^b	68 ^b	67 ^a	43 ^a
UVC	14.66 ^{ab}	13.74 ^a	15.73 ^a	16.88 ^{ab}	79 ^b	66 ^b	50 ^b	38 ^b
1-MCP+UVC	13.98 ^b	13.51 ^a	14.12 ^a	13.95 ^b	77 ^b	75 ^b	64 ^a	43 ^a
UVC+1-MCP	15.19 ^{ab}	13.89 ^a	15.23 ^a	16.05 ^{ab}	85 ^b	77 ^b	68 ^a	43 ^a
<u>0.1 μL.L⁻¹ ethylene</u>								
Control	19.06 ^a	16.84 ^a	16.69 ^a	19.46 ^a	54 ^a	35 ^c	30 ^c	23 ^c
1-MCP	13.80 ^b	13.57 ^b	15.52 ^a	17.37 ^{ab}	78 ^b	66 ^b	58 ^a	39 ^{ab}
UVC	14.98 ^b	13.66 ^b	15.53 ^a	17.42 ^{ab}	80 ^b	68^{ab}	48 ^b	34 ^{bc}
1-MCP+UVC	14.10 ^b	12.63 ^b	15.37 ^a	15.32 ^b	77 ^b	73 ^{ab}	63 ^a	51 ^a
UVC+1-MCP	17.88 ^{ab}	14.73 ^{ab}	15.60 ^a	18.35 ^a	81 ^b	77 ^a	62 ^a	45 ^{ab}

Values are the mean of 6 replicates. Letters indicate mean values at the same columns, treatments and storage time that are statistically different (P<0.05)

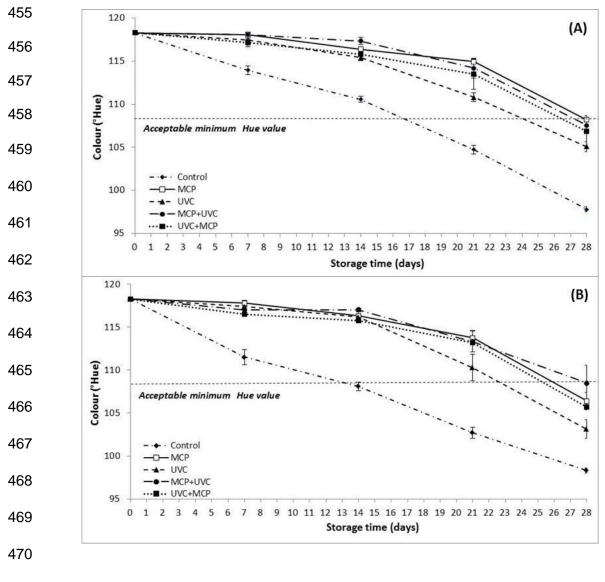


450 Figure 1 Ethylene production of limes after treated with UV-C and/or 1-MCP,

451 followed by storage in air containing (A) < 0.005 μ L.L⁻¹ ethylene and (B) 0.1 μ L.L⁻¹

452 ethylene at 20° C.

453



471 Figure 2 Peel colour (°Hue) of limes after treated with UV-C and/or 1-MCP, followed 472 by storage in air containing (A) < $0.005 \ \mu L.L^{-1}$ ethylene and (B) $0.1 \ \mu L.L^{-1}$ ethylene at 473 20°C.