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Effect of Phytosanitary Irradiation on the Quality of Two Varieties of Pummelos (*Citrus maxima* (Burm.) Merr.)

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ABSTRACT

30 Phytosanitary treatments prevent the introduction of pests such as fruit flies into pest
31 free zones, and are often required for international trade. Irradiation is increasingly
32 being considered as an alternative to cold and chemical phytosanitary treatments, such
33 as methyl bromide. . In this study, the effect of low dose gamma irradiation on the post-
34 harvest quality of two varieties of pummelos (*Citrus maxima* (Burm.) Merr.), an
35 emerging crop of interest in the US was evaluated. Two varieties of pummelos grown
36 in California were irradiated at the phytosanitary target dose of 150 Gy and a higher
37 dose of 1000 Gy to exaggerate and hence confirm the effects of treatment. The fruit was
38 stored at 12 °C for 3 weeks and at 20 °C for the 4th week to reflect three weeks of sea
39 shipment at the ideal temperature for storage of pummelos and an additional week of
40 retail under ambient conditions. Neither irradiation nor storage affected juice content,
41 organic acids, sugars, peel or pulp color and consumer sensory preference, although
42 numerous volatiles increased in concentration as a result of irradiation treatment.
43 Irradiation caused an immediate reduction in whole fruit and pulp firmness in
44 'Chandler' but not 'Sarawak' pummelos at both 150 Gy and 1000Gy. The quality of
45 irradiated pummelos stored at refrigerated temperature for 3 weeks was similar to
46 untreated pummelos, however, physical handling and exposure to higher temperature

47 resulted in increased peel pitting of irradiated fruit compared to non-treated fruit. The
48 results suggest that irradiation could serve as a potential phytosanitary treatment for
49 Chandler and Sarawak pummelos, provided that the fruit is subjected to minimal
50 handling and not temperature abused.

51 ***Keywords: Citrus, Postharvest quality, Ionizing irradiation, Chemical composition, Peel***
52 ***damage***

53 **1 INTRODUCTION**

54 Pummelo (*Citrus maxima* (Burm.) Merr.) is one of the largest citrus fruits by size.
55 This fruit is popular throughout Asia and Europe for their desirable taste, flavor and juicy
56 texture and is gaining popularity in the U.S. In August 2014, the United States Animal and
57 Plant Health Inspection Service (APHIS) proposed the importation of five species of citrus
58 fruits, including pummelos, from China into the U.S. (USDA-APHIS, 2014) provided that
59 adequate phytosanitary measures are taken to ensure quarantine pest free fruit shipment.
60 APHIS has identified 22 pests including eight species of Bactrocera fruit flies as quarantine
61 pests that might enter U.S. through importation of citrus fruits from China (USDA-APHIS,
62 2015). Phytosanitary treatments allowed by APHIS on citrus include chemical fumigation,
63 irradiation, and cold and heat treatments (USDA-APHIS, 2015), although the commercial
64 phytosanitary treatments prevalent in the citrus industry are cold treatment and methyl
65 bromide fumigation (MeBr). Cold treatments (0.56-1.67 °C for 22-18 days) are very
66 effective in eliminating certain insect infestations (USDA-APHIS, 2015), however, the
67 optimum temperature for storage of pummelos is 12 °C, and storage at significantly lower
68 temperature can adversely affect the external appearance and color of pummelos resulting in
69 damage and loss of market quality (International Tree Fruit Network, 2011). Methyl bromide

70 (MeBr) has a depleting effect on the ozone layer and pre-plant use has been mostly phased
71 out as required by the 1987 Montreal Protocol (Kuijpers et al., 2014). Due to lack of viable
72 alternatives, MeBr is currently exempted from the ban under the Montreal Protocol for post-
73 harvest phytosanitary purposes (Hallman and Thomas, 2011), although continuous efforts
74 are being made to phase out MeBr completely (USDA-EPA, 2013). The Methyl Bromide
75 Technical Options Committee under the United Nations Environmental Programme has
76 recognized irradiation as a potential phytosanitary alternative to MeBr fumigation (Kuijpers
77 et al., 2014).

78 Irradiation is a highly efficacious phytosanitary treatment which utilizes ionizing
79 radiation from radioisotopes (gamma rays), electron beam or x-rays to disrupt the genetic
80 material of the pest or microorganism infesting food, causing either sterilization or death of
81 the target organisms. Irradiation is used commercially to treat several fruit including mangos,
82 guavas, litchi, dragon fruit and rambutan in various countries, although it is not currently
83 utilized for citrus products. USDA-APHIS (2015) is considering allowing citrus from China
84 to be treated at 150 Gy, a dose effective against all fruit flies from the family Tephritidae,
85 and in particular the Oriental fruit fly (*Bactrocera dorsalis*).

86 The effect of irradiation depends upon the variety or cultivar of fruit, irradiation dose,
87 ripening/maturity stage of the fruit, harvest season and post-treatment storage conditions of
88 fruit such as temperature (McDonald et al., 2013; Bustos and Mindeta, 1988; Patil et al.,
89 2004; Miller et al., 2000; Nagai and Moy, 1985). Some fruits such as Valencia and
90 ‘Ambersweet’ oranges can tolerate irradiation but peel pitting is observed on irradiated navel
91 and ‘Hamlin’ oranges, and ‘Fallglo’, ‘Sunburst’ and ‘Temple’ hybrid mandarins (McDonald
92 et al., 2013; Miller et al., 2000). Irradiation also causes dose dependent softening of Valencia

93 oranges treated at 300 Gy and higher (Nagai and Moy, 1985), navel oranges treated at 600
94 Gy and higher (McDonald et al., 2013) , and ‘Murcott’, Minneola and ‘Temple’ fruit (Miller
95 et al., 2000) whereas other varieties such as ‘Hamlin’, ‘Sunburst’ and ‘Ambersweet’ are more
96 radiotolerant (Miller et al., 2000). .

97 The effect of low-dose gamma irradiation on the post-harvest quality of pummelos
98 has not been documented. Thus, the objective of the research was to observe the effects of
99 low-dose gamma irradiation (150 Gy and 1000 Gy) on the post-harvest quality of two
100 varieties of Pummelos available in California, Chandler (red flesh) and Sarawak (white
101 flesh), after storage at 12°C for 3 weeks, the time required for sea shipment between the US
102 and Asia, followed by one week at 20°C to reflect retail conditions. The dose of 150 Gy was
103 selected since it is the specified minimum dose in the PPQ Treatment Manual (APHIS, 2014).
104 1000 Gy is the maximum dose allowed by the FDA to treat fresh fruits and vegetables and
105 was selected to exacerbate the negative effects and allow recognition of symptoms of
106 phytotoxicity.

107 **2 MATERIALS AND METHODS**

108 **2.1 Fruit Procurement**

109 Pummelos were harvested on January 15th, 2015 in Tulare County, CA. The fruits
110 were packed on January 17th, 2015 in Orosi, CA following standard commercial practices.
111 The fruits were first washed using 50-150 mg/L chlorine at the point of dumping, then using
112 a mixture of 100-200 mg/L chlorine and 1-2% sodium bicarbonate in a high pressure washer
113 at 862 kPa (125 psi) followed by a fresh water rinse. After washing, the fruits were treated
114 with 200-300 mg/L heated imazalil. In addition to treatment with imazalil, the fruits were

115 waxed with carnauba-based wax that had 2000 mg/L imazalil and 3500 mg/L thiabendazole
116 mixed in it. Pummelos were bulk packed in 24 kg cartons, with approximately 12 pummelos
117 in each carton. After packaging, the cartons were stored at 5° C prior to shipping to Santa Fe
118 Springs, CA (350 km) in a refrigerated truck. The cartons of pummelos were picked up from
119 the distributor and transported in a van to Sterigenics, Inc., Tustin CA (20 km) for gamma
120 irradiation.

121 2.2 Gamma Irradiation:

122 Upon arrival at Sterigenics, the cartons were labeled either control, 150 Gy, or 1000
123 Gy. Dose mapping was conducted using eight cartons of pummelos in the exact configuration
124 as the sample treatment with 150 Gy and 1000 Gy. Eight boxes were stacked in two rows of
125 four a fixed distance from the Co₆₀ source (~278Bq). Dose mapping was conducted by
126 placing 16 alanine pellet dosimeters (FarWest Technology, In, Goleta, CA) at various
127 locations in the cases. The dose rate was determined to be 0.637 Gy s⁻¹. Eight cases of
128 pummelos were placed exactly in the same configuration as the dummy cases to receive
129 treatment at a target dose of 150 and 1000 Gy (4.6-5.5% uncertainty) and Dmax/Dmin ratio
130 of 1.33. The eight cartons used as control fruit did not receive any treatment. After treatment
131 was complete, all the cartons were transported to Chapman University, Orange, CA in a van,
132 covering a distance of approximately 24 miles. Upon arrival, all the cartons were stored at
133 12° C for 3 weeks to simulate shipment to and from Asian countries and at 20° C for the 4th
134 week to simulate one week at retail temperatures. Temperature was verified using LogTags®
135 (Auckland, New Zealand) placed in cartons of pummelos during storage. Analytical and
136 chemical tests were performed on samples taken at day 1 after irradiation, then after 3 and 4
137 weeks of storage.

138 2.2.1 Peel pitting and external damage

139 Ten pummelos per treatment were labeled (control, 150 Gy and 1000 Gy) and used
140 for determining external damage during storage. The fruit was evaluated during storage for
141 surface damage such as peel pitting, scarring bruising and discoloration by three panelists
142 using a five-point scale reflecting the percentage of surface area impacted by damage. The
143 scale was as follows: 0=0% damage, 1=1-4%, 2=5-9%, 3=10-12%, 4=13-15% and 5=16%
144 or more external damage.

145 2.2.2 Weight Loss

146 The same ten pummelos used for determining peel pitting and external damage were
147 also used for measuring change in weight. To measure weight loss, ten whole fruits per
148 treatment were weighed together on each evaluation day. The fruits were weighed using a
149 Mettler Toledo SD32000 scale (Columbus, OH) and percentage weight loss was calculated.

150 2.2.3 Texture

151 Pulp firmness was measured using a Kramer Shear probe attachment on a TA-XT2
152 Stable Micro Texture Analyzer equipped with Exponent software (Stable Microsystems,
153 Godalming, Surrey, U.K and Texture Technology Corp. Scarsdale, N.Y.). Six pummelos per
154 treatment were first scored with a knife and then hand peeled. Pummelo segments were
155 separated carefully by hand to ensure fruit firmness was not compromised then cut into
156 halves. Aliquots weighing approximately 150 g were placed in the holding cell of the Kramer
157 shear press. Blades were positioned 80 mm above the bottom of the holding unit platform;
158 the test speed was 1 mm/s with a pre and post-test speed of 5 mm/s. The maximum force (N)

159 required to pierce through the pulp and the area (N.mm) under the curve was recorded. Eight
160 measurements were made for each treatment and averaged.

161 Whole fruit firmness was measured using a puncture probe. Six pummelos per
162 treatment were each punctured six times along the equatorial region to a depth of 35 mm
163 using a 3 mm puncture probe at a speed of 3 mm/sec with a pre and post speed of 5 mm/sec.
164 The maximum force (N) required to penetrate and the area (N.mm) under the curve were
165 recorded.

166 2.2.4 Peel and pulp color measurements

167 Hunter Lab values of the external peel of six pummelos from each dose was measured
168 at four equidistant points along the equatorial axis using a Konica Minolta
169 Spectrophotometer CM-2500d (Ramsey, NJ). The same fruits were cut into half at the
170 equatorial axis, and two measurements for internal color were made on each half; a total of
171 four measurements per fruit. Hue values were calculated using the following formula:

$$172 \quad h = \tan^{-1}(b^*/a^*)$$

173 where h is hue value, a* represents the red/green opponent colors and b* represents
174 the yellow/blue opponent colors on the L*a*b* color space.

175 2.2.5 Juice content

176 Six whole pummelos per treatment were weighed and peeled. The juice was
177 extracted from each pummelo using a TS-738 Elite Gourmet Maxi-Matic Juice Extractor
178 (City of industry, CA) and % juice was calculated using the following formula:

$$179 \quad \% \text{ Juice Content (whole fruit basis)} = [\text{Juice weight} \times 100] / [\text{Whole fruit weight}]$$

180 2.3 Chemical Analysis

181 Fifteen pummelos per treatment were juiced as described above. The juice was
182 centrifuged (20000 g/ 20 °C/10 min) and the supernatant divided into five samples which
183 were individually evaluated for total soluble solids content (TSS), titratable acidity (TA),
184 individual sugars, individual organic acids, individual and total phenolic compounds, and
185 volatile compounds.

186 2.3.1 Total soluble solids

187 A few drops of the centrifuged juice were placed on the prism of a Digital “Pocket”
188 Refractometer PAL (Atago Co., Ltd, Tokyo, Japan). Five readings per treatment were
189 recorded and average total soluble solids were determined.

190 2.3.2 Titratable acidity

191 Five mL of the centrifuged juice was combined with 50 mL of di-ionized water and
192 titrated to an end point of 8.2 (pH) using 0.1 N NaOH. Five readings per treatment were
193 recorded. Percentage citric acid was calculated using:

$$194 \quad \% \text{ Acid} = [(\text{ml NaOH}) \times (0.1 \text{ N NaOH}) \times (0.064) \times (100)] / \text{ml of sample}$$

195 2.3.3 Organic acids

196 Organic acids were analyzed according to Ornelas-Paz et al. (2013) with some
197 modifications. One mL of centrifuged juice was diluted with three mL of HPLC grade water.
198 The diluted juice was filtered using a 45 µm pore size polyethylene membrane (Sigma-
199 Aldrich, St. Louis, MO, USA) and injected (20 µL) into an Agilent 1100 series HPLC system
200 equipped with a diode array detector (Agilent Technologies, Santa Clara, CA). The organic
201 acids were separated in an Aminex HPX-87H ion exchange column (7.8 x 300 mm) (Bio-

202 Rad, Hercules, CA) at 60 °C. The mobile phase was composed of 5mM H₂SO₄ and
203 acetonitrile (90:10, v/v) at a flow rate of 0.4 mL/min. Citric, malic, oxalic, tartaric, quinic,
204 fumaric and succinic acids were monitored at $\lambda=210$ nm. The ascorbic acid was monitored
205 at $\lambda=260$ nm. Identification and quantification of organic acids was carried out using
206 reference compounds.

207 2.3.4 Sugars

208 Glucose, fructose and sucrose were analyzed according to Ornelas-Paz et al. (2013)
209 with some modifications, using the HPLC system described above but using refractive index
210 detection. One hundred μ L of filtered juice was mixed with 2 mL of HPLC grade water and
211 filtered using a 0.45 μ m pore size polyethylene membrane. Five μ L of the mixture were
212 injected into the HPLC system. The separation was carried out at 80 °C in a Sugar SC 1821
213 (8.0 x 300 mm, 6 μ m) ion exclusion column fitted to a Sugar SC-LG precolumn (6.0 x 50
214 mm, 10 μ m) (Shodex, Tokyo, Japan). Water was used as mobile phase at a flow rate of 0.8
215 mL/min. The identification and quantification of sugars was performed using standard
216 compounds.

217 2.3.5 Individual and total phenols

218 The analysis of individual and total phenols was simultaneously performed. The juice
219 was filtered with a 0.45 μ m membrane and directly injected (100 μ L) into the HPLC
220 described previously. The separation of phenolic compounds was performed in a Kinetex
221 C18 (4.6 x 100 mm) (Phenomenex, Torrance, CA, USA) at 30 °C. The phenolic compounds
222 were monitored at $\lambda= 280, 320, 350$ and 520 nm. The mobile phase consisted of 2% acetic
223 acid (A), and acetonitrile (B), according to the following gradient: 100% A at 0 min, 93% A

224 at 12 min, 89% A at 20 min, 86% A at 35 min, 84% A at 36 min, 82% A at 41 min, 79% A
225 at 44, 0% A from min 55 to 60. The flow rate was 1 mL/min. The phenolic compounds were
226 identified and quantified by using reference compounds. The UV-Vis spectrum of individual
227 phenols was also used for identification purposes.

228 For total phenols content, 100 μ L of filtered juice were mixed with 100 μ L of Folin-
229 Ciocalteu, 3 mL of deionized water and 100 μ L of 20% Na_2CO_3 . The mixture was vigorously
230 shaken for 1 min and incubated for 1h in the darkness. The absorbance was evaluated five
231 times at 765 nm using a FLUOstar Omega microplate reader (BMG LABTECH Inc.; Cary,
232 NC, USA). The absorbance values were corrected with those generated with blank
233 reactions. Quantification was performed using a calibration curve constructed with several
234 sets of dilutions of gallic acid. The results were expressed as mg GAE per liter of juice.

235 2.3.6 Aroma Volatiles

236 Five samples of juice (5 mL each) were placed into headspace vials (12 x 32 mm)
237 and then 5 mL of saturated sodium chloride and 1-pentanol at a final concentration of 490
238 $\mu\text{g L}^{-1}$ as an internal standard were added. The vials were sealed with a cap with a Teflon-
239 coated septum. Immediately prior to being run the samples were held at 4 $^{\circ}\text{C}$ in a water-
240 cooled rack. Analysis of volatile headspace components was conducted using solid phase
241 microextraction (SPME) with a 75- μm carboxen/polydimethylsiloxane fiber (Supelco, St.
242 Louis, MO, USA). The equipment utilized and analytical run conditions were exactly as
243 detailed in Ummarat et al. (2015) again using a FID detector for quantification.
244 Identification of compounds utilized mass spectrometry with comparison to Wiley/NBS
245 library spectra, retention times of standards when available and retention indices using *n*-
246 alkanes. Volatile concentrations were semi-quantified from the FID detector data by

247 utilizing calibration curves produced from standards purchased from Sigma-Aldrich (St.
248 Louis, MO, USA) placed into deodorized citrus juice. The standard used to quantify each
249 compound class is as follows: ethanol (ethanol), ethyl acetate (esters), 3-methyl butanol
250 (alcohols), E-2-hexenal (aldehydes), α -pinene (terpenes), 4-terpineol (terpene alcohols),
251 and carvone (ketones). Differences in SPME or GC detector response were adjusted for by
252 using the internal standard (1-pentanol).

253 2.4 Consumer testing

254 Consumer testing of pummelos was conducted at Chapman University on day 26
255 (during the fourth week of storage at 20 °C). Forty panelists including faculty, staff and
256 students from Chapman University participated in the testing. Six randomly selected
257 pummelos per treatment were peeled and segmented. The membrane surrounding the
258 segments was removed and the segments were further cut into halves. Each panelist was
259 provided with 2-3 half segments of pummelos per treatment in soufflé cups, labeled with a
260 3-digit code. Panelists were also provided with water cup and unsalted cracker to cleanse
261 their palates between samples. Consumers rated the degree of liking for overall liking,
262 overall flavor, sweetness, bitterness and juiciness on a 9-point hedonic scale (Peryam and
263 Pilgrim, 1957). Tests were administered in individual booths with samples and their
264 corresponding question sets presented in random order using SIMS 2000 Sensory
265 Evaluation software (Berkley Heights, N.J., U.S.A.) to prevent positional bias.

266 2.5 Statistical analysis using JMP statistical software

267 Two-way analysis of variance with replication was used to determine effects of irradiation
268 dose and storage time in a randomized complete block design. When F-values were

269 significant, least significant differences (LSD) at a significance level of 0.05 were
270 calculated using a Tukey-Kramer test. Statistical analysis was performed using JMP
271 statistical software ver. 8.0 (SAS Institute, Inc., NC, USA).

272 **3 RESULTS AND DISCUSSION**

273 **3.1 Firmness of pummelo fruit and segments**

274 The pulp of the Chandler pummelos was twice as firm as compared to the Sarawak
275 pummelos, which could be a difference in variety or stage of maturity. Irradiation caused an
276 immediate softening of pulp in Chandler but not in Sarawak pummelos (Fig. 1). During
277 storage, the pulp of Chandler pummelos from the control fruit continued to soften, but
278 Sarawak pummelos did not show consistent pulp softening due to irradiation or storage.

279 The puncture test profile indicated that the peak force was recorded as the probe
280 punctured through the peel of the pummelos, which is the outer 5-10 mm of the fruit. The
281 peak force measured using the puncture probe were similar for the Chandler and Sarawak
282 pummelos indicating that the peels of the two varieties had similar firmness. Similar to pulp
283 texture measured using a Kramer Shear, irradiated Chandler pummelos, but not Sarawak,
284 had significantly lower peel firmness on day one after irradiation (Fig. 2). During the fourth
285 week of storage, however, the peel of the 1000 Gy samples of both varieties were softer than
286 the control and 150 Gy treated fruit, although it is important to note that peel firmness
287 increased for all Chandler and control Sarawak pummelos during that time. This increase in
288 fruit firmness might be a result of moisture loss from the surface of the fruit (peel) due to
289 transpiration as well as decrease in relative humidity during ambient temperature storage.

290 The peel of pummelos is considered a rich source of pectin and other structural
291 polysaccharides such as cellulose (Methacanon et al., 2014). Pectin content in flavedo and

292 albedo layers of pummelos is about 15 and 19% (Chaidedgumjorn et al., 2009). Radiation-
293 induced free radical activity (Kovács and Keresztes, 2002) as well as increased
294 polygalacturonase and pectinmethylesterase activity (Ladaniya et al., 2003) can accelerate
295 degradation of cell wall pectin and other structural polysaccharides causing breakdown of
296 the middle lamella in the cell wall, ultimately resulting in reduced firmness. The high pectin
297 content in the flavedo layer of pummelos suggests that this layer will be likely impacted by
298 irradiation, and while loss of firmness of the peel was readily observed in the irradiated
299 Chandler pummelo, in the Sarawak, a significant decrease ($P<0.05$) was seen only after three
300 weeks of storage.

301 3.2 Juice Content

302 No effect of irradiation or storage was observed on the juice content of Sarawak pummelos
303 (~33%) based on total fruit weight (data not shown). An initial small but significant
304 ($P<0.05$) increase in juice content based on fruit weight was observed for the 1000 Gy
305 Chandler pummelos (from 24 to 27%) but the difference was not maintained during storage.
306 Sensory testing also corroborated that consumer perception of juiciness was not affected.
307 Similarly, Ladaniya et al. (2003) and Mitchell et al. (1991) also reported that treatment with
308 gamma irradiation up to 1500 Gy and 600 Gy did not affect juice yield of Mosambi oranges,
309 Nagpur mandarins and Valencia oranges, respectively.

310 3.3 Weight Loss

311 In the first three weeks of storage, the weight loss in Chandler pummelos was ~3%
312 increasing to ~11% by the end of the fourth week (data not shown). The Sarawak pummelos
313 lost ~3% of their weight in the first three weeks, and ~8% by the end of the fourth week

314 (data not shown). In both varieties, there was no impact of treatment. As previously noted,
315 juice content was unaffected during storage, which suggests that the weight loss was
316 primarily due to loss of moisture from the peel of the fruit. Exposure to warm temperature
317 along with storage at low relative humidity (~60%) was most likely the cause of weight loss.
318 Pummelo peels are thick (0.5-1 cm) and can amount to up to 50% of the weight of the whole
319 fruit, thus moisture loss from the surface of the peel can result in significant weight loss.

320 3.4 Peel and Pulp Color

321 Neither irradiation nor storage affected the color of peel or pulp in the pummelos
322 (data not shown). Previous studies have also observed minimal effects on citrus fruit and
323 pulp color at low dose levels. McDonald et al. (2013) and Ladaniya et al. (2003) reported
324 that storage or irradiation up to 600 Gy and 1500 Gy did not affect the color of the pulp in
325 navel oranges, Nagpur mandarins or Mosambi oranges.

326 3.5 Titratable Acidity and Organic Acids

327 Irradiated Chandler pummelos had a lower titratable acidity as compared to the
328 control ($P < 0.05$) after three weeks and after subsequent storage at ambient temperature
329 (Table 1). There was no consistent effect of treatment or storage on Sarawak pummelos
330 (Table 1). McDonald et al. (2013), Ladaniya et al. (2003), and O'Mahony and Goldstein
331 (1987) also reported lower titratable acidity in irradiated navel oranges, Mosambi
332 oranges, Nagpur mandarins and Kagzi limes in comparison to their control
333 counterparts.

334 Citric acid followed by quinic then succinic and tartaric acids were the predominant
335 acids in Chandler and in the Sarawak pummelos, citric was followed by succinic then

336 quinic acids (Table 2). Citric acid in the 1000 Gy Chandler pummelos decreased after four
337 weeks of storage, consistent with the change in TA, but not in the Sarawak, also consistent
338 with TA results. However, in general, acid concentrations remained fairly stable during the
339 four weeks of storage and there was no consistent effect of irradiation or storage. Small
340 differences in concentrations can be attributed to fruit variability rather than an effect of
341 irradiation or storage. In a study of three cultivars of pummelos, Sun et al. (2012) found
342 citric acid to be the predominant acid followed by malic, fumaric and aconitic acids.
343 During storage for 100 days, the changes in acid content were small and inconsistent,
344 similar to the pattern observed in our study. Previous studies suggest that effect of
345 irradiation on the organic acid content of citrus fruit varies depends upon the dose, storage
346 time and temperature and mainly on the species and cultivar of fruit. Macfarlane and
347 Roberts (1968) and Rouse et al. (1966) reported a dose dependent decrease in citric acid
348 content of Valencia oranges treated with 250 Gy-2000 Gy; a possible explanation could be
349 the excessive utilization of citric acid as substrate during accelerated respiration in
350 irradiated fruits (Ladaniya, 2008) and resultant lowered TA. Irradiation-induced loss of
351 ascorbic has been observed in grapefruit, Mosambi oranges, Nagpur mandarins, Kagzi acid
352 limes and lemons (Vanamala et al., 2005; Ladaniya et al., 2003; Maxie et al., 1964). In our
353 study, loss of ascorbic acid was observed in the 1000 Gy Sarawak pummelos but not in the
354 Chandler pummelos.

355 3.6 Total Soluble Solids and Sugars

356 Chandler pummelos had higher TSS values than Sarawak pummelos (Table 1), and
357 during storage, the TSS values of the control fruit of both varieties increased ($P < 0.05$) but
358 there was not a consistent effect of irradiation. Similarly, sucrose concentrations, but not

359 glucose and fructose, in the Chandler variety were higher than the Sarawak pummelos and
360 concentrations fluctuated among treatments and during storage (Table 1). Sun et al., (2012)
361 also observed higher initial sucrose concentrations in three varieties of pomelos as compared
362 to glucose and fructose, but there were no consistent patterns in changes in the sugar
363 concentrations during storage. The only consistent pattern in our data was an increase in
364 glucose, fructose and sucrose concentrations in the control and 150 Gy Chandler pummelos
365 during storage, but no other irradiation related effect is discernible. Patil et al. (2004)
366 reported that irradiation up to 700 Gy did not affect soluble solids in early 'Rio Red'
367 grapefruit. However, irradiation has also been shown to decrease soluble solid content in
368 'Ambersweet' oranges, navel oranges and 'Sunburst' mandarins treated with 300, 400 and
369 450 Gy of gamma radiation respectively (McDonald et al., 2013; Miller et al., 2000). Citrus
370 fruits utilize sugars as substrates for respiration, therefore it is expected that the total soluble
371 solids and sugar content will decrease with time. Also, irradiation can cause oxidative stress,
372 which can result in a higher respiration rate and consumption of simple sugars during the
373 process of respiration, leading to a decrease in sugar content (Ladaniya et al., 2003), however
374 no such decrease was observed in our study.

375 3.7 Total and Individual Phenols

376 Total phenolics ranged between 425 mg GAE L⁻¹ and 650 mg GAE L⁻¹ in the two
377 pummelo varieties (Fig. 3). In both sets of pummelos, naringin was the most abundant
378 phenolic compound (Fig. 4), about 10 fold higher than the next most abundant compounds
379 which were hesperidin and chlorogenic acid in the Chandler pummelos. Rutin, nairutin,
380 and ferulic acid were also detected in small quantities. In Sarawak pummelos, rutin and

381 chlorogenic acid were more abundant than hesperidin. In addition to small amounts of
382 narirutin and ferulic acid, very small amounts of p-coumaric acid were also detected.

383 Literature on phenolic compounds in pummelos is limited. Li et al. (2012) reported
384 that the phenolic content of honey pomelos was 55 mg/100 g fruit which decreased during
385 storage. Mäkynen et al. (2013) evaluated the phenolic compounds in six varieties of
386 pummelos found in Thailand and found that total phenols ranged between 101.32 to 113.73
387 mg gallic acid equivalent/g extract and the predominant phenolic compounds were
388 naringin, hesperidin, neohesperidin, neohesperidin dihydrochalcone, naringenin, and
389 hesperitin. The relative amounts of these phenolic compounds varied between the six
390 varieties.

391 Irradiation increased total phenolics in Chandler pummelos soon after treatment. After
392 three weeks, there were no differences between the treatments, but after the fourth week at
393 ambient temperature, the irradiated fruit had higher total phenolics. In the Sarawak
394 pummelos, the 1000 Gy fruit had the highest total phenolic concentrations soon after
395 treatment. During storage, the 400 Gy samples had the highest concentrations ($P < 0.05$),
396 but the differences between the treatments was small. Irradiation increased ($P < 0.05$)
397 naringin concentrations in both sets of pummelos. There is no information on effect of
398 irradiation on phenolics in pummelos, however, irradiation-induced stimulation of PAL
399 activity leading to increased phenolic concentrations has been documented in citrus fruit
400 such as Clementines and mandarins (Rojas-Argudo et al., 2012; Shen et al., 2013).

401 3.8 Aroma Volatiles

402 Sixty-four volatiles were identified in samples from both of the pumelo varieties
403 consisting of aldehydes, alcohols, esters, ketones, and various classes of terpenes

404 (data not shown). Gonzalez-Mas et al. (2011) used HS-SPME-GC-MS analysis to
405 characterize the volatile compounds in different citrus fruit (Powell Navel orange,
406 Clemenules mandarin, Fortune mandarin and Chandler pummelos). The volatile
407 profile of the pummelos was unique in that mostly terpenic compounds (β -
408 caryophyllene, (*Z*)-ocimene, (*E,E*)-2,4-nonadienal, (*Z*)- and (*E*)-linalool oxides, *p*-cymene)
409 were identified, almost exclusively in Chandler pummelos. Cheong, Liu, Zhou, Curran,
410 and Yu (2012) evaluated the flavor profile of two varieties of Malasian pummelos, *Citrus*
411 *grandis* (L.) Osbeck PO 51 and PO 52 (red and white fleshed, respectively). They found
412 that the pink pomelo juice shared a closer resemblance to grapefruit chemically in contrast
413 to the white pummelo and that the higher terpenic content of the white pomelo juice with
414 milder acidity and lower amounts of volatiles made it unique among citrus fruit.

415 Of the 64 compounds identified in this study, 39 changed significantly ($p \leq 0.05$)
416 due to irradiation treatment in 'Chandler' (Table 3) and 48 in 'Sarawak' (Table 4). In
417 both varieties the most common trend was for irradiation to increase volatile
418 concentrations relative to the controls, with 1000 Gy often causing the greatest
419 increase. This effect of irradiation to enhance citrus volatile concentration had also
420 been seen in oranges treated at doses as high as 600 Gy, with ethanol, esters and
421 aldehydes being the primary volatiles altered in amount by irradiation in that study
422 (McDonald et al., 2013). Although the effect of irradiation on volatile components was
423 similar for both varieties there were some differences in how specific classes of
424 compounds were altered. For instance, terpenes in 'Chandler' prior to storage
425 increased as a result of irradiation while in 'Sarawak' these compounds, with the
426 exception of α -thujene, either did not significantly change or decreased in

427 concentration. Storage impacted the treatment effect in 'Chandler' as significant
428 concentration differences between the control and 150 Gy treatment became much
429 less common following storage at both 3 weeks at 5 °C and after an additional 1 week
430 at 20 °C. While the overall impact of the 1000 Gy treatment was also lessened by
431 storage, there were still elevated concentrations relative to the untreated control for
432 many of the volatile compounds, particularly in the case of ethanol and in the esters
433 and ketones. Ethyl acetate in particular was strongly increased in the irradiated fruit
434 following storage. For 'Sarawak' there was still considerable impact of the 150 Gy
435 treatment after storage, particularly in the elevated levels of aldehydes and esters
436 following 3 weeks at 5 °C. Treatment with 1000 Gy and storage led to even higher
437 concentrations in volatiles from these two classes of compounds as well as increases
438 in four of the thirteen terpenes that were quantified. As is commonly practiced
439 commercially, the pummelos used in this study had a wax coating applied to the peel
440 surface after harvest and prior to irradiation treatment and storage. Waxing citrus can
441 result in the enhancement of volatile accumulation in the fruit that can sometimes
442 alter flavor (Obenland et al., 2011). In both 'Chandler' and 'Sarawak' concentrations
443 of many of the volatiles were higher as a result of storage in the untreated fruit, this
444 being a primary reason for why there was sometimes significant differences observed
445 as a result of irradiation immediately after treatment but not following storage.

446 3.9 Consumer Testing

447 Consumer testing of Chandler and Sarawak pummelos indicates that irradiation did
448 not affect consumer liking of the two pummelo varieties (data not shown). Overall, the scores
449 for all attributes ranged between 5 (Neither Like or Dislike) and 6 (Like Slightly). The reason

450 for overall low scores might be the lack of familiarity with pummelos. Although changes in
451 aroma-active juice volatiles as a result of irradiation were noted in this study, panelists were
452 evidently unable to detect the altered aroma volatile profile of the irradiated fruit.
453 McDonalds et al. (2013) also reported that the panelists did not observe any significant
454 differences in overall liking, flavor and juiciness among control navel oranges and oranges
455 irradiated at 200, 400 and 600 Gy. In contrast, some studies have reported development of
456 off-flavor in irradiated citrus treated with doses ranging from 60 Gy to 1000 Gy (Nagai and
457 Moy, 1985; O'Mahony et al., 1985; Mitchell et al., 1991; Miller et al., 2000).

458 3.10 Peel Damage

459 Both varieties had small but noticeable amount of dents and scars upon receipt,
460 presumably due to postharvest handling of the fruit. During storage, fruit developed
461 significant brown colored stippling (small brown spots), loss of gloss, bruising and areas of
462 softening. In irradiated fruit, we also observed larger, sunken brown areas (Fig. 5) that have
463 been reported in the literature as peel pitting, generally associated with areas of peel that
464 collapse and discolor and not limited to oil glands (Ritenour et al., 2003). In general, peel
465 damage was greater and developed more quickly in irradiated pummelos and became even
466 more severe when the fruit was stored at ambient temperature for a week. In Chandler
467 pummelos, eight out of ten of the 150 Gy and nine out of ten of the 1000 Gy Chandler
468 pummelos showed evidence of pitting, bruising and spot development covering more than
469 5% of the total surface area of each pummelo as compared to only two out of ten control
470 pummelos after the first week of storage (Fig. 6). No pitting, bruising or spotting was evident
471 in Sarawak pummelos during the first week. The extent of damage increased gradually in
472 both varieties of pummelos during the three week storage at 12 °C, however, storage at

473 ambient temperature (20 °C) for one week severely exacerbated peel damage in irradiated
474 pummelos. By the end of the fourth week, 70 and 80% of the irradiated Chandler and
475 Sarawak fruit, respectively, showed 16% or more damaged surface area, as compared to only
476 10% of the control Chandler and none of the control Sarawak. Pummelos with pitting
477 covering greater than 16% of the peel surface were considered unmarketable. Thus, the
478 combination of irradiation, physical manipulation and high temperature storage was highly
479 detrimental to the quality of the pummelos. Mold growth was observed in both varieties of
480 irradiated pummelos, mainly because cell injury caused by irradiation provided a suitable
481 environment for mold growth.

482 Peel pitting is a disorder that generally occurs during postharvest storage of citrus
483 fruits under environmental conditions, such as low relative humidity, that can cause damage
484 to cells in the flavedo and albedo, ultimately affecting nearby oil glands present in the fruit
485 peel (Alferez et al., 2008). Low internal oxygen concentrations within the fruit might be
486 another possible explanation for non-chilling peel pitting observed in citrus fruits (Ritenour,
487 2013). Peel pitting as a result of irradiation has been observed in several citrus fruits
488 (McDonald et al., 2013; Ladaniya et al., 2003; Miller et al., 2000; Dennison et al., 1966).
489 One of the causes of peel pitting in irradiated citrus fruit may be irradiation-induced
490 accumulation of phenolic compounds in the flavedo cells of the peel which oxidize resulting
491 in cell death (Riov, 1975). Irradiation can also elevate respiration rate of fresh fruit resulting
492 in increased ethylene production (Riov et al., 1972), and subsequent increase in PAL activity
493 (Benoit et al., 2000). Enhanced PAL activity leads to accumulation of phenolic compounds
494 in irradiated fruits (Mahrouz et al., 2002). In this study, an immediate irradiation-induced
495 increase in naringin was seen in both varieties of pummelos. While respiration rate was not

496 measured in this study, the TA was lower in the 1000 Gy Chandler pummelos suggesting
497 that respiration rate of these fruit may be affected. Additionally, the high ethanol levels
498 measured in this study, particularly in the Sarawak pummelos after 20 C storage, are
499 indicative of low internal oxygen concentrations which can also lead to peel pitting
500 (Ritenour, 2013). Using a wax that allows adequate oxygen diffusion rate may help in
501 reducing peel pitting (Ritenour, 2013). Heat conditioning treatment at 38 and 42° C for 2
502 hours reduced PAL activity, which further resulted in reduced peel pitting by 8 and 10%
503 respectively, in grapefruit irradiated at 1000 Gy and stored at 10 °C for 4 weeks (McDonald
504 et al., 2000). Thus, heat conditioning might prove helpful in case of pummelos as well.

505 **4 CONCLUSION**

506 Radiation impacted the quality of the two varieties differently with a greater impact
507 observed on Chandler pummelos, which could reflect varietal differences. The differences
508 could also be attributed to maturity level at harvest, however that can only be confirmed if
509 the same variety was studied at different maturity levels.

510 In general, 1000 Gy had a greater impact on pummelo quality than 150 Gy. One
511 impact of irradiation was manifested as softening of the fruit, an effect observed in most
512 irradiated pummelos, although irradiation-induced peel damage was the most significant
513 effect of irradiation. Peel damage in the irradiated fruit increased gradually during the three
514 weeks of storage under ideal conditions. However, when the conditions were less than ideal
515 in terms of ambient temperatures and excessive handling, the fruit showed signs of
516 phytotoxicity. Our results show that 150 Gy could be a feasible dose for Chandler and

517 Sarawak pummelos but they would need to be handled minimally and stored under ideal
518 temperatures.

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526

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647

648

649 Table 1. Titratable acidity, total soluble solids and sugars in irradiated Chandler and Sarawak

Treatment	TA (% citric acid)	TSS (%)	Sugars (g/L)			
			Glucose	Fructose	Sucrose	
Chandler						
Day 1	Control	0.76 Bxy	11.58 Cz	10.73 By	14.17 y	69.85 Cy
	150 Gy	0.84 Ax	13.10 Ax	12.20 Ay	14.74 y	78.18 Ax
	1000 Gy	0.79 Bx	12.44 Bx	12.27 A	14.92 y	72.57 B
After 3 weeks	Control	0.78 Ax	12.40 Ay	12.79 x	15.51 Ax	76.28 x
	150 Gy	0.57 Bz	12.06 Cz	12.34 y	14.92 By	74.78 y
	1000 Gy	0.58 Bz	12.30 By	12.58	15.14 ABx	73.69
After 4 weeks	Control	0.73 Ay	12.72 Ax	12.78 x	15.66 x	77.36 Ax
	150 Gy	0.66 By	12.40 By	13.20 x	16.30 x	74.90 Ay
	1000 Gy	0.66 By	11.72 Cz	12.99	15.95 x	72.03 B
Sarawak						
Day 1	Control	0.740 A	10.50 Bz	16.71 Ax	16.92 Ax	48.62 By
	150 Gy	0.75 Ax	10.62 A	15.86 B	16.04 B	51.08 Ax
	1000 Gy	0.71 B	10.52 By	16.34 ABy	16.69 Ax	50.89 A
After 3 weeks	Control	0.72 AB	10.58 Ay	16.05 Ay	16.45 Ay	51.33 Cx
	150 Gy	0.75 Ax	10.60 A	15.90 A	16.27 A	50.81 Bxy
	1000 Gy	0.70 B	10.44 By	15.18 Bz	15.75 By	50.41 B
After 4 weeks	Control	0.69	10.70 Bx	15.97 By	16.33 By	50.68 x
	150 Gy	0.68 y	10.62 C	15.61 B	15.99 B	49.81 y
	1000 Gy	0.68	10.86 Ax	16.96 Ax	17.16 Ax	50.81

650 pummelos stored for three weeks at 12°C and for an additional week at 20°C. Statistically
 651 significant differences ($P < 0.05$) among treatments within the same time point are denoted by
 652 letters A-C; and across time points for the same treatment by letters x-z.

653 Table 2. Organic acids in irradiated Chandler and Sarawak pummelos stored for 3 weeks at 12 °C and after an additional week at 20 °C. Statistically
654 significant differences (P<0.05) among treatments within the same time point are denoted by letters A-C; and across time points for the same
655 treatment by letters x-z. OA-Oxalic acid, CA-Citric acid, TA-Tartaric acid, MA-Malic acid, AA-Ascorbic acid, QA-Quinic acid, SA-Succinic acid,
656 FA-Fumaric acid

Time	Treatment	Organic acids							
		OA	CA	TA	MA	AA	QA	SA	FA
Chandler									
Day 1	Control	109.84 Bx	12143.97 Cy	1683.17 Ax	815.18 Axy	494.74 Ax	2514.41 By	1734.33 Ay	12.19 Ay
	150 Gy	119.78 Ax	13558.84 Ax	1710.36 Ax	682.76 Cy	477.98 By	2909.36 Az	1487.87 Bz	10.64 Bz
	1000 Gy	101.95 Cy	12394.95 By	1620.00 Bx	753.31 Bz	506.50 Ax	2807.91 By	1469.76 By	11.80 By
After 3 weeks	Control	100.66 By	12516.30 Ax	1582.55 Bz	842.48 Bx	475.99 Ax	3314.61 x	1994.52 x	13.33 x
	150 Gy	103.95 Aby	12259.55 By	1631.56 Az	931.11 Ax	471.60 Ay	3271.30 y	2104.62 y	14.27 y
	1000 Gy	104.96 Ax	12594.32 Ax	1577.73 By	787.71 Bx	473.33 Ay	3356.93 x	2160.43 x	13.36 x
After 4 weeks	Control	110.89 Ax	12625.31 Ax	1629.80 By	775.55 Cy	433.10 By	3436.22 Bx	2119.94 Bx	13.70 Bx
	150 Gy	105.39 By	12051.15 Bz	1677.13 Ay	885.38 Ax	504.39 Ax	3719.23 Ax	2323.57 Ax	15.373 Ax
	1000 Gy	104.42 Bx	12074.80 Bz	1607.40 Bx	806.70 Bx	498.81 Ax	3285.17 Cx	2271.21 Ax	13.53 Bx
Sarawak									
Day 1	Control	151.20 Ay	11757.35 y	1403.31 Bx	842.07 Cz	316.64 Ax	3801.45 Ay	6012.78 Ay	13.83 Abx
	150 Gy	138.26 By	11723.57 y	1365.78 Cx	914.17 Bz	290.68 Bx	3562.29 Bz	5159.71 Cz	12.87 Bx
	1000 Gy	136.49 Bz	11710.07 z	1444.21 Ax	952.98 Ay	271.46 Cx	3828.69 Ax	5365.34 Bz	14.179 Ax
After 3 weeks	Control	142.36 Bz	12073.39 Bx	1320.87 y	997.40 Cx	245.06 Ay	4723.92 Cx	6127.47 By	9.75 By
	150 Gy	149.11 Ax	12415.31 Ax	1335.24 y	1079.11 Ax	237.65 Bz	4898.32 Bx	6377.26 Ax	10.674 Ay
	1000 Gy	142.43 By	12256.39 Ax	1321.35 y	1017.71 Bx	232.63 By	5053.08 Ax	5923.80 Cy	10.20 By
After 4 weeks	Control	159.99 Ax	11749.47 By	1291.39 By	945.85 By	245.27 By	4732.49 Bx	6610.734 Ax	10.27 y
	150 Gy	151.94 Bx	11525.89 Cz	1380.50 Ax	960.94 By	259.12 Ay	4765.30 By	5961.60 By	10.72 y
	1000 Gy	154.50 Bx	11956.54 Ay	1314.27 By	1009.67 Ax	226.76 Cx	5203.20 Ax	6574.14 Ax	10.40 y

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661 Table 3 Aroma volatile concentrations ($\mu\text{g L}^{-1}$) in irradiated Chandler pummelos during storage for 3 weeks at 12 °C and after an additional
 662 week at 20 °C. Values followed by different letters within a storage regime represent significant differences ($P \leq 0.05$). Shading indicates that
 663 there is at least a two-fold difference from the control (0 Gy) dose. nd = not detectable.

Treatment	Day 1			After 3 weeks			After 4 weeks		
	Control	150 Gy	1000 Gy	Control	150 Gy	1000 Gy	Control	150 Gy	1000 Gy
Alcohols									
ethanol ^a	145.90c	279.66b	317.04a	321.76b	434.87b	863.19a	522.58b	462.04b	894.44a
2-butanol	1.75b	1.81b	2.48a	19.09a	9.82b	10.53b	8.67a	3.58b	3.75b
1-penten-3-ol	2.44c	4.13a	3.56b	8.89a	7.24ab	6.48b	9.45a	9.33a	7.17b
3-methylbutanol	0.95a	0.51b	0.81ab	3.02c	4.64b	8.00a	4.11b	3.78b	7.17a
cis-3-hexenol	12.44c	24.78a	16.84b	49.05a	38.86a	24.15b	52.81a	40.75b	16.37c
hexanol	17.32c	25.66a	20.79b	35.89a	28.87b	29.17b	46.20a	34.50a	15.40b
Aldehydes									
acetaldehyde	47.23b	50.81ab	51.78a	50.96a	57.73a	55.64a	56.17a	52.41a	58.47a
2-methylpropanal	1.52b	1.542b	2.15a	6.07a	3.57b	4.17b	3.58a	2.93b	3.53a
3-methylbutanal	1.84b	1.84b	2.17a	3.25b	3.66b	7.21a	4.03b	3.67b	5.99a
2-methylbutanal	1.78b	1.97b	2.33a	2.59b	3.69ab	5.16a	2.75a	2.22b	2.73a
pentenal	4.34b	5.10a	4.64b	15.29b	20.25a	22.33a	15.46a	15.38a	15.20a
E-2-pentenal	0.95a	0.71b	0.92b	2.00a	2.12a	2.05a	1.78a	1.77a	1.89a
hexanal	22.84c	28.00a	25.65b	95.78a	103.85a	117.72a	84.96a	71.06a	65.42b
heptanal	1.83a	1.94a	1.86a	4.92b	6.74a	7.87a	4.73a	4.52a	4.80a
2,4-hexadienal	1.10a	1.11a	1.20a	4.31a	2.54b	2.05b	2.88a	2.85a	2.18b
E-2-heptanal	1.20a	1.33a	1.39a	3.14b	3.38b	3.82a	2.91a	2.88a	3.25a
octanal	0.81a	0.83a	0.91a	1.86a	1.51a	1.51a	1.41a	1.41a	1.40a

nonanal	1.64a	1.58a	1.72a	2.79b	3.26ab	3.82a	3.08a	3.14a	3.57a	664
Esters										
ethyl acetate	1.35c	2.89b	3.53a	22.89b	36.09b	428.25a	42.97c	93.69b	853.27a	
ethyl propanoate	0.55a	0.64a	0.76a	2.68a	4.51a	8.31a	4.22b	5.50b	12.78a	
ethyl-2-methylpropanoate	nd	nd	nd	0.35b	0.50b	2.02a	0.60b	0.61b	2.32a	
ethyl-2-methylbutanoate	0.24a	0.20a	0.24a	0.46b	0.70b	3.36a	0.79b	1.03b	4.93a	
ethyl hexanoate	0.69c	1.19b	1.62a	1.40b	1.32b	1.93a	1.54b	1.64b	2.42a	667
Ketones										
1-penten-3-one	40.33a	32.83b	33.73b	76.05b	80.34b	96.89a	74.23b	80.15b	104.06b	668
4-methyl-2-heptanone	11.42a	11.63a	9.97a	15.24b	14.24b	18.27a	21.86b	19.95b	28.65a	
methyl-heptanone	19.07b	17.37c	21.32a	33.56b	36.61ab	42.82a	34.30b	40.07ab	45.49a	669
dihydrocarvone	0.99a	1.32a	1.40a	1.71a	1.88a	2.22a	2.54b	2.49b	3.38a	
carvone	1.30b	1.88a	2.24a	3.90ab	3.67b	5.28a	4.15a	4.81a	4.72a	670
piperitone	0.91a	0.98a	1.07a	2.45b	2.97ab	4.13a	2.65a	3.14a	4.23a	
Terpenes										
α -pinene	0.15c	0.27b	0.35a	0.40a	0.41a	0.43a	0.48a	0.33b	0.37b	671
camphene	0.04b	0.06ab	0.08a	0.10a	0.08a	0.09a	0.09a	0.08a	0.08a	672
β -pinene	0.28b	0.31b	0.44a	0.67a	0.70a	0.69a	0.66a	0.54b	0.72a	
β -myrcene	0.46c	0.76b	1.02a	1.17a	1.14a	1.43a	1.21a	1.04a	1.16a	673
α -terpinene	0.13b	0.17a	0.19a	0.22a	0.21a	0.24a	0.18a	0.17a	0.18a	
p-cymene	1.05a	1.40b	1.81a	3.64a	2.34a	3.90a	4.16a	3.90a	2.38a	674
limonene	12.93c	32.02b	48.50a	40.03a	39.86a	46.56a	47.68a	32.04b	36.45ab	
β -elemene	0.06c	0.10b	0.14a	0.13b	0.12b	0.22a	0.17b	0.19b	0.23a	675
trans-caryophyllene	0.19c	0.25b	0.32a	0.46a	0.47a	0.54a	0.58b	0.64ab	0.84a	
Terpene alcohols										
linalool	1.41c	2.16b	4.10a	3.02a	2.97a	2.45b	5.53a	4.20ab	4.06b	676
										677

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681 Table 4. Aroma volatile concentrations ($\mu\text{g L}^{-1}$) in irradiated Sarawak pummelos during storage for 3 weeks at 12 °C and after an additional week
682 at 20 °C. Values followed by different letters within a storage regime represent significant differences ($P \leq 0.05$). Shading indicates that there is at
683 least a two-fold difference from the control (0 Gy) dose. nd = not detectable.

Treatment	Day 1			After 3 weeks			After 4 weeks		
	Control	150 Gy	1000 Gy	Control	150 Gy	1000 Gy	Control	150 Gy	1000 Gy
Alcohols									
ethanol ^a	380.98a	322.50a	428.91a	410.95b	500.67b	813.69a	1053.10a	1019.55a	1020.96a
2-butanol	9.72b	15.05a	13.77a	18.62a	14.51b	17.27a	12.54a	10.60a	10.90a
2-methyl-3-buten-2-ol	107.68a	84.01b	82.31b	112.57a	95.42a	70.77b	120.66a	100.83a	94.41a
1-penten-3-ol	7.55b	8.27b	9.59a	6.22a	6.41a	6.14a	4.53b	5.01ab	5.60a
3-methylbutanol	4.76b	4.64b	7.10a	6.79b	8.70b	12.68a	45.75a	48.66a	44.54a
cis-3-hexenol	63.41b	69.07a	51.95c	54.62a	59.38a	46.73b	29.47ab	27.32b	35.95a
hexanol	39.36a	39.94a	31.84b	32.51b	39.56a	38.27a	21.77b	20.31b	30.44a
Aldehydes									
butanal	1.56b	3.51a	3.66a	3.89ab	3.64b	4.35a	3.17a	3.36a	3.72a
2-methylpropanal	6.00b	21.19a	20.60a	25.02ab	19.98b	29.65a	16.26c	21.37b	24.97a
3-methylbutanal	2.49b	3.18a	3.79a	4.61a	4.08a	5.64a	7.15a	7.07a	6.92a
2-methylbutanal	1.85a	1.99a	2.10a	2.52b	3.12b	4.42a	3.67b	4.34a	4.77a
pentenal	8.63b	56.80a	50.39a	63.69b	67.24b	85.09a	41.03b	56.27a	62.25a
hexanal	139.44b	370.36a	312.37a	463.49b	487.20b	663.74a	315.88b	476.74a	509.81a
heptanal	4.17b	21.60a	16.81a	25.02b	26.67b	37.84a	17.79b	26.32a	27.13a
2,4-hexadienal	4.68a	6.45a	4.84a	5.56a	7.06a	4.73a	2.76a	5.40a	4.47a
E-2-heptenal	2.88b	4.51a	4.42a	5.50a	6.02a	5.90a	3.96b	5.33a	5.45a
benzaldehyde	1.78b	1.89ab	2.22a	2.10a	2.23a	1.95a	2.06a	2.11a	2.13a
octanal	3.14b	6.34a	5.67a	7.20b	7.63b	10.40a	6.09b	8.84a	7.69ab
nonanal	3.12b	5.68a	4.39ab	6.41b	6.98ab	9.38a	5.91a	8.29a	7.03a
decanal	0.63a	1.43a	1.26a	1.81a	1.68a	2.28a	1.45b	2.10a	1.72ab
Esters									
ethyl acetate	18.36b	13.98c	20.40a	18.69b	17.55b	39.79a	27.16c	42.95b	104.92a

ethyl propanoate	1.07b	1.85b	5.01a	1.21b	3.95a	4.45a	2.52b	4.28ab	5.95a	a=mg L-1	
ethyl-2-methylpropanoate	0.36a	0.32ab	0.27b	0.37b	0.46b	0.78a	1.16b	1.45b	2.24a		
ethyl-2-methylbutanoate	0.36a	0.43a	0.45a	0.62b	0.59b	1.64a	1.27c	2.39b	4.35a		
E-2-hexenal	24.99a	26.58a	23.06a	35.82a	39.06a	29.26a	20.08b	31.51a	23.96b		
ethyl hexanoate	1.30b	2.13a	2.70a	2.05b	1.88b	2.87a	2.48b	3.94a	3.33a		
Ketones											
1-penten-3-one	45.97c	75.02b	94.98a	100.47a	83.92a	96.07a	184.47b	201.02a	114.86c		
gamma-butyrolactone	11.86b	17.72ab	21.782a	15.52a	15.21a	14.58a	12.43a	15.49a	16.65a		
methyl-heptanone	24.06a	29.12a	30.79a	32.10b	36.35ab	40.28a	31.51b	43.88a	36.81ab		
dihydrocarvone	2.77a	3.30a	2.63a	3.37b	5.01a	5.33a	3.31a	3.59a	4.58a		
carvone	5.01b	9.17a	7.09ab	11.24b	20.13a	26.97a	13.81b	10.73b	21.17a		
piperitone	3.24b	6.35a	4.58ab	7.54a	9.26a	9.79a	8.34a	9.89a	9.59a		
Terpenes											
α-thujene	0.85b	0.88b	1.42a	1.09c	1.60b	1.88a	1.35b	0.99c	1.97a		
α-pinene	4.98ab	4.09b	5.76a	5.27b	8.33a	9.31a	7.29b	4.25ab	9.31a		
camphene	0.19a	0.17a	0.20a	0.20b	0.27a	0.29a	0.23ab	0.19b	0.30a		
β-pinene	2.94a	2.32a	3.22a	2.80b	4.62a	5.18a	4.39b	2.55c	5.73a		
β-myrcene	2.69a	1.57b	2.11ab	2.27a	2.78a	2.94a	2.34a	2.10a	2.82a		
α-terpinene	1.42a	0.63b	0.91b	0.79b	1.64a	1.65a	1.36ab	0.79b	1.63a		
1-menthene	0.86a	0.34b	0.57b	0.43a	0.88b	0.80b	0.79a	0.41b	0.85a		
p-cymene	38.78a	35.76a	51.77a	42.01b	79.21a	94.39a	59.87b	40.34c	84.05a		
limonene	106.23a	80.03a	100.60a	104.85a	190.84a	235.15a	173.44a	100.76b	229.32a		
γ-terpinene	14.45a	4.45b	7.99b	5.77b	13.60a	12.85a	11.11ab	5.67b	12.75a		
β-elemene	1.23a	0.70b	1.06a	0.87a	0.95a	0.95a	0.97a	0.87a	0.90a		
trans-caryophyllene	4.05a	2.16b	2.86b	2.93a	3.27a	3.11a	3.02a	2.63a	2.97a		
α-humulene	0.72a	0.36b	0.49b	0.50a	0.58a	0.55a	0.55a	0.46a	0.50a		
Terpene alcohols											
linalool	9.39a	8.22b	10.11a	10.99c	14.88a	12.40b	17.82a	10.53c	13.42b		
4-terpineol	12.65a	11.78ab	10.21b	13.27b	22.20a	19.98a	19.24a	11.74b	18.49a		
β-fenchyl alcohol	12.94a	13.28a	11.92a	16.36b	23.15a	21.88a	22.66a	16.88b	20.98ab		

686 List of Figures:

687 Figure 1. Pulp firmness of (a) Chandler and (b) Sarawak pummelos stored for three
688 weeks at 12°C and for an additional week at 20°C measured using a Kramer shear
689 press. Statistically significant differences ($P<0.05$) among treatments within the same
690 time point are denoted by letters A-C; and across time points for the same treatment by
691 letters x-z.

692

693 Figure 2. Fruit firmness of (a) Chandler and (b) Sarawak pummelos stored for three
694 weeks at 12°C and for an additional week at 20°C measured using a puncture
695 probe. Statistically significant differences ($P<0.05$) among treatments within the same
696 time point are denoted by letters A-C; and across time points for the same treatment by
697 letters x-z.

698

699 Figure 3. Content total phenols in (a) Chandler and (b) Sarawak pummelos. A-
700 immediately after irradiation; B-after 3 weeks of storage at 12 °C and C-after
701 additional week at 20 °C.

702

703 Figure 4. Content of several phenolic compounds (NG, naringin; H, hesperidin; CA,
704 chlorogenic acid; R, rutin; NR, narirutin; FA, ferulic acid) in (a) Chandler and (b)
705 Sarawak pummelos. A-immediately after irradiation; B-after 3 weeks of storage at 12
706 °C and C-after additional week at 20 °C.

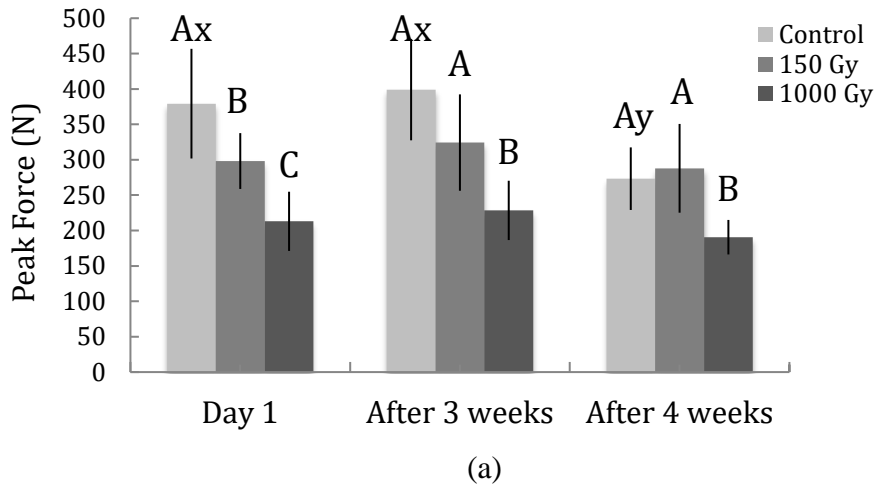
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708 Figure 5. Peel damage in 150 Gy Chandler pummelo after 4 weeks of storage, 3 weeks
709 at 12 °C and 4th week at 20 °C.

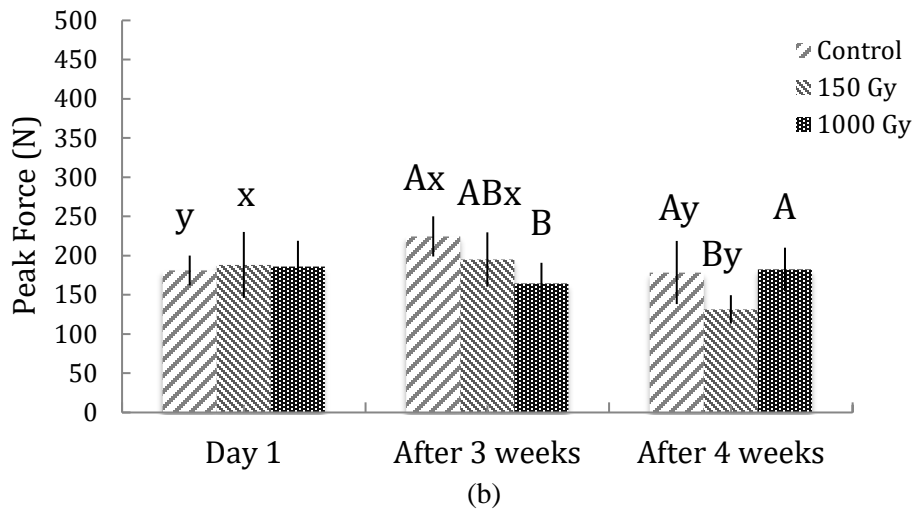
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711 Figure 6. Peel damage in (a) Chandler and (b) Sarawak, control and irradiated (150 Gy
712 and 1000 Gy) pummelos stored for 3 weeks at 12 °C and for an additional week at 20
713 °C. 0=0%, 1=1-4%, 2=5-9%, 3=10-12%, 4=13-15% and 5=16% or more external
714 damage, M=Moldy pummelos.

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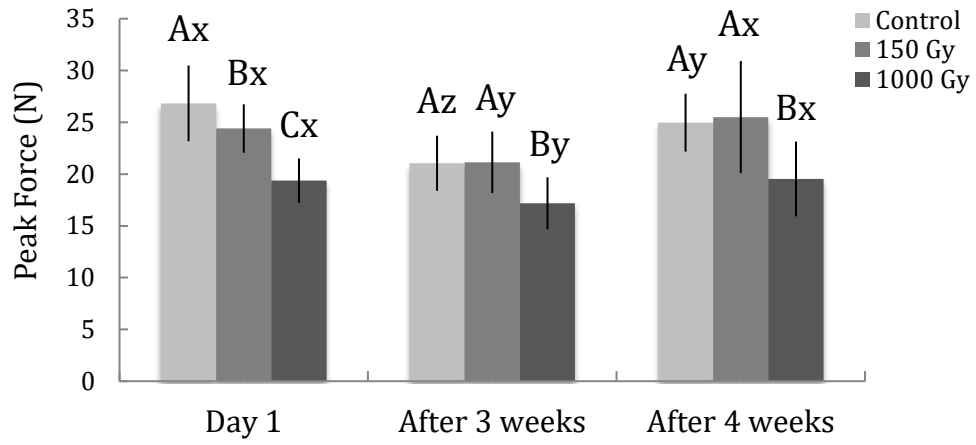
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725 Fig. 1

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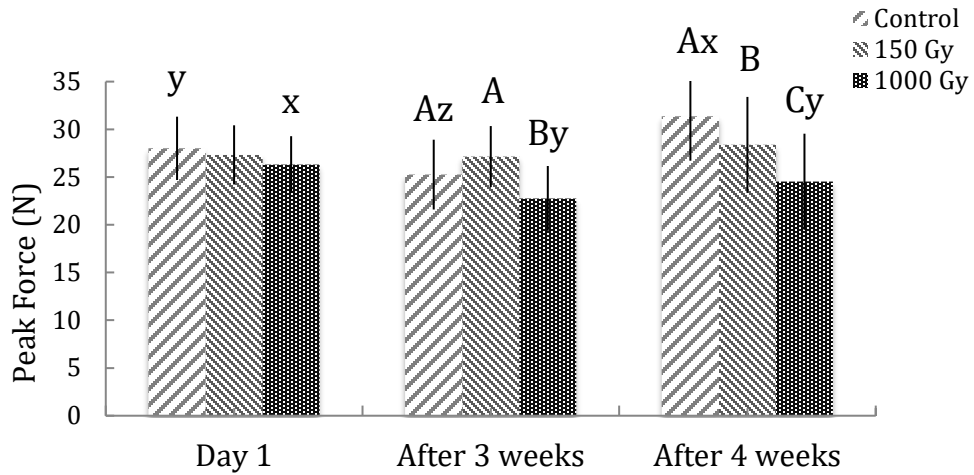
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(a)



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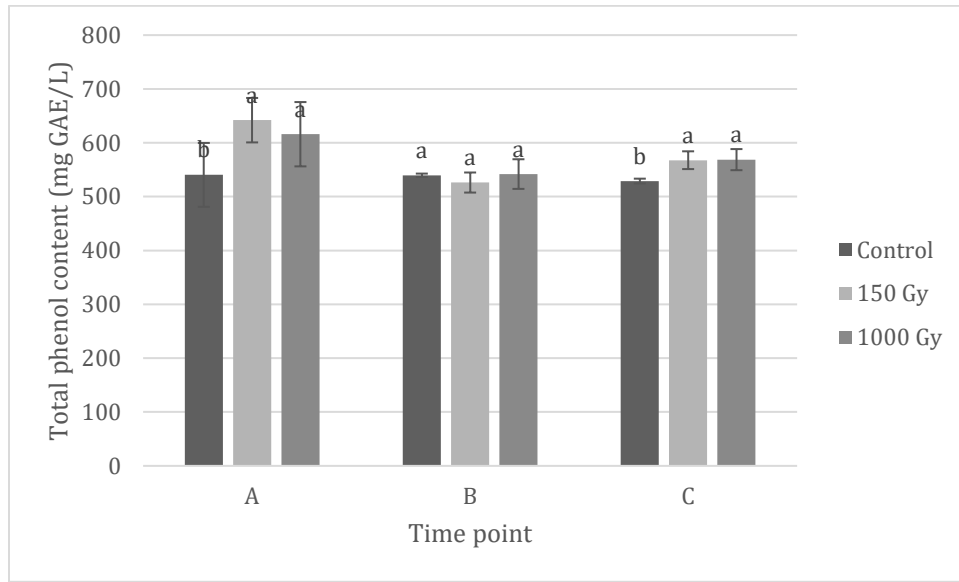
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(b)

753 Fig. 2

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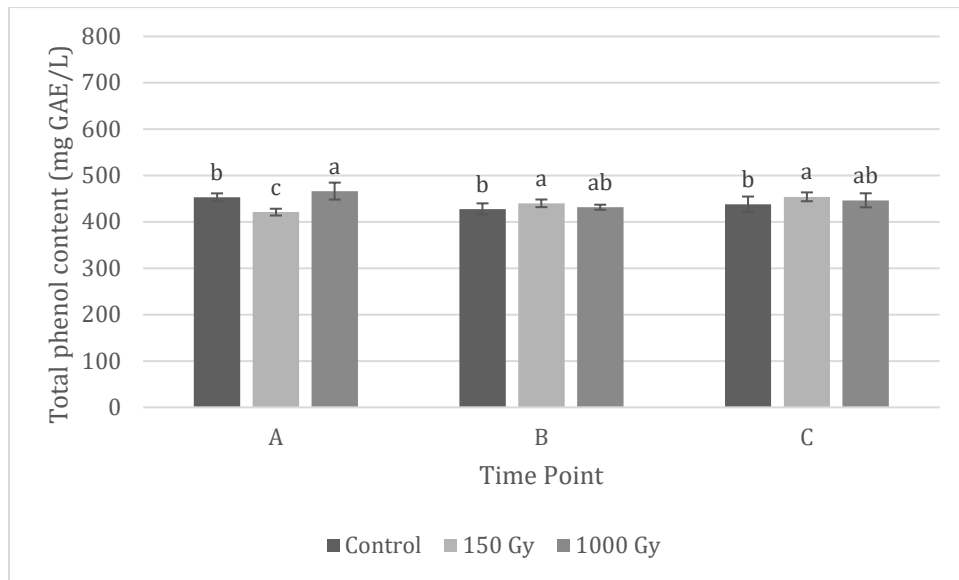


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(a)

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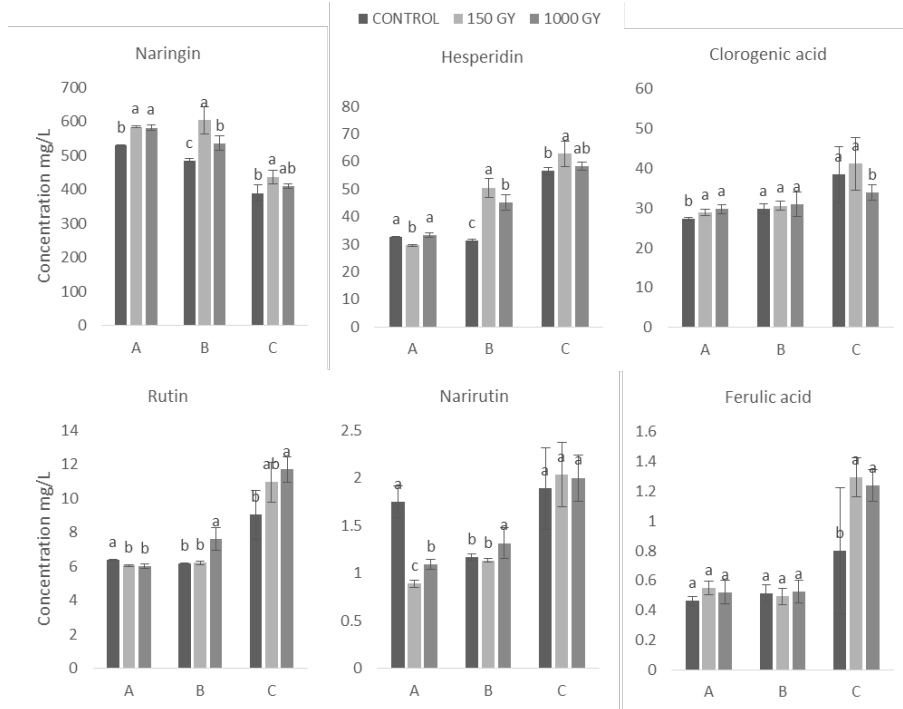
(b)

761 Fig. 3

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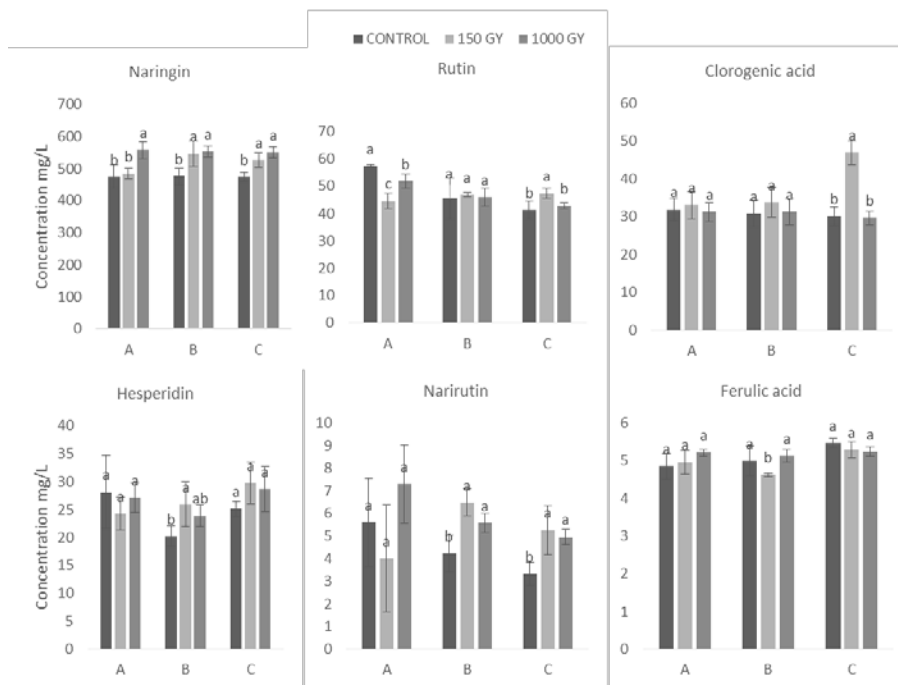
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765

766 (a)



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768 (b)

769 Fig. 4

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773 Fig. 5

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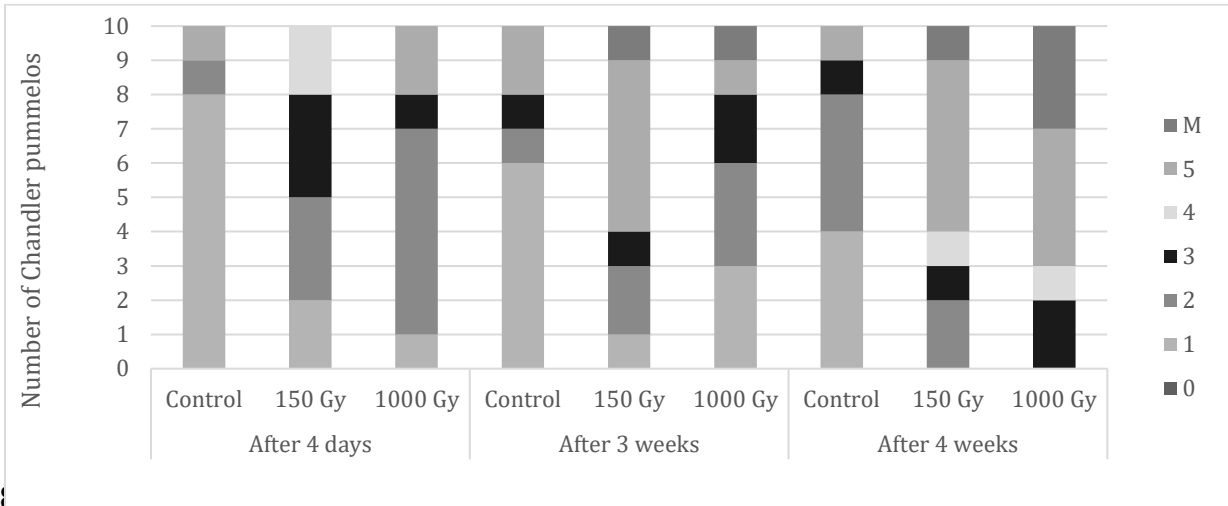
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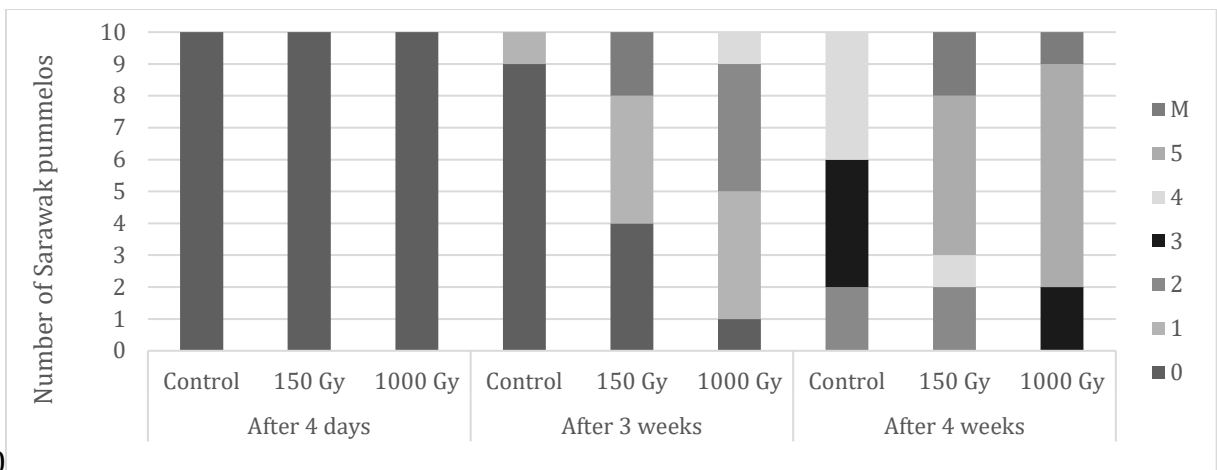
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(a)



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(b)

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Fig. 6