タンゴール‘清見’果実のこはん症発生と品質に及ぼす収穫後処理の効果

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A Comparative Study of the Effects of Postharvest Treatments on Occurrence of Kohansho and Quality of 'Kiyomi' Tangor

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The effects of postharvest treatments, namely, perforated film packaging (PFP), hot water dip (HWD), and hot calcium dip (HCD), on the occurrence of "Kohansho (rind disorder)" and the quality attributes of 'Kiyomi' Tangor (*Citrus unshiu* Marc. × *C. sinensis* Osb.) fruit were compared. Samples of the fruit were treated, packed in corrugated fiberboard boxes, and stored at 10℃ for up to 4 weeks. Weight loss was significantly lower and firmness was significantly higher in the fruit treated by PFP than in those treated by other postharvest treatments or in the control fruit. All postharvest treatments reduced the occurrence of Kohansho compared with control, and PFP and HWD treatments were more effective than HCD in reducing the occurrence of Kohansho. None of the postharvest treatments impaired quality attributes tested (total soluble solids [TSS], titratable acidity [TA], TSS/TA ratio, ascorbic acid content, and ethanol content), when compared with the attributes of the control. We conclude that PFP treatment, which can maintain a high relative humidity and a modified atmosphere except in the presence of high CO_2 levels (> 8%) and/or low O_2 levels (< 5%) was the most effective and commercially practical method to control Kohansho and preserve quality. (Received Nov. 15, 2010 ; Accepted May 30, 2011)

Key words : *postharvest treatments, perforated film packaging, hot water dip, kohansho, 'Kiyomi' Tangor*
収穫後処理，有孔フィルム包装，温湯浸漬，こはん症，タンゴール清見

'Kiyomi' Tangor (*Citrus unshiu* Marc. × *C. sinensis* Osb.) is recognized as a high-quality late-season citrus fruit. Unfortunately, this variety is vulnerable to Kohansho, a serious, non-chilling physiological rind disorder that lowers quality during storage, transportation, and marketing periods. Kohansho initially appears as small pitting, later developing into a brown sunken area. The disordered fruit is usually rejected by the market.

Individual seal -packaging reduces the occurrence of Kohansho[1], and storage under low temperature and high relative humidity conditions without prestorage conditioning reduces the occurrence of rind injury in 'Kiyomi' fruit[2]. In our previous studies, we observed that Kohansho can be suppressed by reducing fruit weight loss during storage[3], and it was recommended that in order to prevent Kohansho and preserve high quality, 'Kiyomi' fruit should be stored under modified atmosphere conditions, with high relative humidity, and should not be exposed to high CO_2 levels (> 8%) and/or low O_2 levels (< 5%)[4].

Hot water dip (HWD) treatment (50℃ for 3 ~ 5 min) reduced the occurrence of Kohansho in Navel orange[5], Natsudaidai[5] and Hassaku[5,6]. Similar HWD treatment (50 ~ 53℃ for 2 ~ 3 min) reduced the incidence of chilling injury in 'Star Ruby' grapefruit[7] and various cultivars of blood oranges[8] without impairing quality. Calcium dip treatment was reported to lessen the damage due to many physiological disorders and preserve fruit quality by maintaining firmness and reducing weight loss and decay in a variety of fresh fruits and vegetables[9]. A combined calcium and hot water (hot calcium dip, HCD) treatment[10] is therefore expected to be more effective than either HWD or calcium dip treatment

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alone.

However, there is no information about the effect of HWD and HCD treatment on 'Kiyomi' Tangor. The aim of the present study was to compare the effects of perforated film packaging (PFP), HWD, and HCD treatments on the occurrence of Kohansho and the quality of 'Kiyomi' Tangor fruit.

Materials and Methods

1. Plant material

'Kiyomi' Tangor fruit (diameter, 7.3～8.0 cm) harvested on March 19, 2008, from Misaki, Ehime, were transported to the Laboratory of Agricultural Process Engineering, Ehime University. The fruit without Kohansho were used for the experiment on the following day (March 26, 2008).

2. Postharvest treatments

The fruit were treated with different postharvest treatments as follows.

(1) **PFP treatment** The fruit were sealed in a perforated low-density polyethylene film package (a 0.019-mm film of dimensions 60 × 50 cm, with 15 perforations, each 5mm in diameter) and packed in a corrugated fiberboard box (dimensions 36 × 25 × 15 cm). This perforated package was designed in accordance with the modified atmospheric conditions recommended by TECHAVISES and HIKIDA[4].

(2) **HWD treatment** The fruit were dipped in a hot water bath at 53℃ for 2 min, dried at room temperature for about 2 h, and packed in a corrugated box. The temperature in the hot water bath was maintained at 53 ± 0.5℃ using an electronic thermostat.

(3) **HCD treatment** The fruit were dipped in hot 1% (w/v) calcium chloride solution at 53℃ for 2 min, dried at room temperature for about 2 h, and packed in a corrugated box.

(4) **Control** The fruit were packed in a corrugated box without treatment.

Three replicates of 5kg of 'Kiyomi' fruit (about 28 fruits) were used in each treatment. All corrugated boxes were stored at 10℃ for up to 4 weeks. The temperature and relative humidity (RH) inside the boxes, inside the packages in the PFP treatment, and inside the storage chamber were monitored throughout the storage period by using temperature and humidity sensors. The vapor pressure deficit (VPD) between the fruit surface and the surrounding air, a major driving force of transpiration, was calculated by subtracting the vapor pressure of the surrounding air from 98% of saturated vapor pressure at the surrounding air temperature.

3. Evaluation of Kohansho

Three replicates of 16 fruits in each treatment were checked for Kohansho by visual inspection at 1-week intervals. The number of fruit and the diameter of affected areas were measured and rated using the following index: 0 = no occurrence of Kohansho; 1 = one spot of less than 5 mm; 2 = one spot of 6～20 mm; 3 = 2～3 spots of 20 mm; 4 = more than 4 spots of 20 mm.

The results were expressed as the percentage and degree of Kohansho:

$$\text{Percentage of Kohansho}(\%) = \frac{\text{total number of fruit incurring Kohansho}}{\text{total number of fruit}} \times 100$$

$$\text{Degree of Kohansho} = \frac{\Sigma(\text{Kohansho index} \times \text{number of fruit incurring Kohansho in each index})}{4 \times \text{total number of fruit}} \times 100$$

4. Determination of quality attributes

Three replicates of 16 fruits in each treatment (the same samples that were used to evaluate Kohansho) were weighed at 1-week intervals to assess percentage weight loss. The firmness, and the content of total soluble solids (TSS), titratable acidity (TA), ascorbic acid (AA), and ethanol were determined in 3 replicates of 4 fruits each, on day 0 (fresh) and day 28 (end of storage). Fruit firmness was determined with a fruit hardness tester using a 1-kg weight. Juice TSS content was measured with a digital refractometer, juice TA was measured by titration with 0.1 M NaOH, and the results were expressed as citric acid percentages. Juice AA content was determined by the indophenol titration method[11].

Ethanol, the dominant fermentative metabolite, can be used to indicate fermentative metabolism for mandarin fruit[12]. The acetaldehyde and ethanol that accumulate during fermentation can lead to the development of off-flavors, thus impairing fruit quality[13]. Juice ethanol content was determined by incubation of 10-mℓ aliquots of juice in 50-mℓ glass vials sealed with an aluminum cap at 30℃ for 30 min following SCHIRRA *et al.*[8]. Aliquots of 10-mℓ each of ethanol standards of 1,000, 2,000, 3,000, 4,000, and 5,000 (μℓ / ℓ) were used to generate a calibration curve. After incubation, a 1-mℓ gas sample was withdrawn from the headspace and analyzed with a gas chromatograph (GC; model GC-8 A,

Shimadzu, Japan) coupled with an FID and a stainless steel column (length and internal diameter, 200 × 0.2 cm) containing 15% Carbowax and Chromosorb W 80/100 mesh. The temperatures of the injector, column, and detector were 100, 90, and 100℃, respectively, and nitrogen was used as the carrier gas.

5. Atmosphere analysis

A gas sample of 0.5 mℓ was taken from inside each PFP package and from inside the storage chamber at 3 - day intervals. O_2 and CO_2 concentrations were determined using a GC (model GC-8 A; Shimadzu, Japan) coupled with a TCD and a stainless steel column (length and internal diameter, 180 cm × 0.6 cm) containing WG-100. The temperatures of the injector, column, and detector were 80, 50, and 80℃, respectively. Helium was used as the carrier gas.

6. Statistical analysis

All data were analyzed by one-way analysis of variance (ANOVA) by the SPSS software (Windows version 11.5; SPSS Inc., Chicago, IL, USA). The significance of differences between means was determined by the Tukey HSD test ($P<0.05$).

Results and Discussion

The 'Kiyomi' Tangor fruit in all treatments lost weight during storage; the weight loss was significantly lower in the PFP treatment than in other treatments and control (Table 1). This was because the notably higher RH led to lower vapor pressure deficit (VPD) in the PFP treatment than in the other postharvest treatments or control (Table 2). VPD is a major driving force for weight loss, since higher VPD results in higher transpiration.

The fruit that underwent all the postharvest treatments showed lower percentage and degree of Kohansho than the control fruit, but the PFP and HWD treatments were more effective than the HCD treatment on reducing the occurrence of Kohansho (Table 3). In the PFP treatment, the average O_2 and CO_2 concentrations inside the package during the storage period were 20.40% and 0.97%, a slight change from normal atmospheric level ($P<0.05$). According to our previous study[4], such a slight change does not affect the occurrence of Kohansho. Therefore, the effectiveness of PFP treatment results from the generation of high RH

Table 1 Weight loss of 'Kiyomi' Tangor fruit during storage at 10℃ under different postharvest treatments

Treatment	% Weight loss			
	Week 1	Week 2	Week 3	Week 4
PFP	0.17±0.01 c	0.30±0.01 b	0.48±0.01 b	0.66±0.02 b
HWD	1.74±0.04 ab	2.95±0.06 a	4.40±0.08 a	5.89±0.11 a
HCD	1.82±0.04 a	3.09±0.07 a	4.61±0.10 a	6.13±0.13 a
Control	1.65±0.04 b	2.93±0.08 a	4.54±0.12 a	6.14±0.15 a

Data are represented as mean±S.E. of 3 replicates of 16 fruits each.
Column means followed by different letters are significantly different at $P<0.05$.

Table 2 Average temperature and RH inside the corrugated fiberboard box (HWD, HCD, control), inside the PFP package, and inside the storage chamber throughout the storage period, and the vapor pressure deficit (VPD), in each treatment

Treatment	Temperature (℃)	RH (%)	VPD* (kPa)
PFP	10.67	99.21	0.01
HWD	10.06	79.16	0.27
HCD	10.25	79.85	0.26
Control	9.82	82.35	0.22
Chamber	9.73	49.17	-

*VPD was calculated by subtracting the vapor pressure of the surrounding air from 98% of saturated vapor pressure at the surrounding air temperature.

Table 3 Effect of postharvest treatments on occurrence of Kohansho in 'Kiyomi' Tangor fruit during storage at10℃

Treatment	Percentage of Kohansho (%)				Degree of Kohansho			
	Week 1	Week 2	Week 3	Week 4	Week 1	Week 2	Week 3	Week 4
PFP	0.00	0.00	2.08	2.08	0.00	0.00	0.52	0.52
HWD	0.00	0.00	0.00	2.08	0.00	0.00	0.00	1.04
HCD	0.00	0.00	2.08	6.25	0.00	0.00	1.04	2.60
Control	2.08	2.08	4.17	14.58	0.52	0.52	1.56	5.73

Data are means of 3 replicates of 16 fruits.

surrounding the fruit; this result is consistent with our previous findings[3),4)]. The HWD treatment was as effective as the PFP treatment (Table 3). Similar effects of the HWD treatment have also been reported in Navel orange[5)], Natsudaidai[5)], and Hassaku[5),6),14)]. There was no significant difference in weight loss between the HWD and control groups; therefore, other mechanisms may act to limit Kohansho. For example, MAKITA[5)] reported that the HWD treatment of Navel oranges enhanced the reducing sugar content in the peel, which is associated with Kohansho tolerance. The fruit that underwent the HCD treatment also showed reduced occurrence of Kohansho compared with the control fruit but was not as effective as the PFP and HWD treatments (Table 3). This implies that calcium was not as effective as expected; in contrast, the calcium treatment reduced the effectiveness of HWD. MANAGO[6)] also reported increased occurrence of Kohansho in Hassaku after dipping in a 2% $CaCO_3$solution. These results suggest that the best results can be achieved by a combination of the PFP and HWD treatments.

Firmness was the highest in fresh fruit (determined on day 0) and decreased during storage in all treatments, but it was significantly higher in the PFP treatment group at the end of storage. This result is consistent with the observation that the weight loss was lower in the PFP treatment group than in the other treatment or control groups (Table 4). Our previous study[4)] found that use of modified atmospheric conditions maintains the firmness of 'Kiyomi' Tangor via the maintenance of a high RH inside the package. BEN-YEHOSHUA *et al*[15)]. also reported that the high humidity of the micro-atmosphere created inside modified atmosphere packaging maintains the turgidity of the tissue and markedly inhibits water loss and softening in citrus fruits.

The TSS content did not change, but the TA content tended to decrease, resulting in an increase in the TSS/TA ratio after 4 weeks of storage in all treatment and control groups, compared with the ratio in the fresh fruit (Table 4). The AA content decreased slightly and the ethanol content tended to increase in all the treatment and control groups compared with the contents in the fresh fruit (Table 4). Ethanol was seen to accumulate during the storage period even in the control group, consistent with our previous observation[4)]. However, there were no significant differences in TSS, TA, TSS/TA ratio, AA content, and ethanol content between any

Table 4 Effect of postharvest treatments on the quality attributes of 'Kiyomi' Tangor fruit

Treatment	Firmness (kg)	TSS (%)	TA (%)	TSS/TA ratio	AA (mg/100mℓ)	Ethanol* (μℓ / ℓ)
Fresh	0.922±0.005 a	12.4±0.4 a	1.09±0.05 a	11.6±0.6 a	58.3±1.8 a	1568±215 a
PFP	0.887±0.009 b	12.5±0.2 a	0.93±0.04 ab	13.6±0.6 a	48.7±1.6 b	1817±340 a
HWD	0.847±0.005 c	12.2±0.3 a	0.98±0.11 ab	15.6±3.2 a	51.1±1.4 b	2533±227 a
HCD	0.857±0.005 c	12.3±0.3 a	0.81±0.06 b	16.2±1.4 a	52.6±1.6 ab	2596±304 a
Control	0.833±0.006 c	11.9±0.5 a	0.79±0.05 b	15.4±0.9 a	50.9±1.7 b	2000±250 a

Data are represented as mean±S.E. of 3 replicates of 4 fruits each.
In the case of the 'fresh' condition, all parameters were determined on day 0 (initial day of experiment), while in the case of all other treatments, the parameters were determined on day 28 (end of storage).
Column means followed by different letters are significantly different at $P<0.05$.
*Ethanol is an indicator of fermentative metabolism.

of the postharvest treatment groups and the control group after 4 weeks of storage.

On the basis of the results obtained and considering the recommended modified atmosphere conditions[4], the PFP treatment, which maintains high RH and a modified atmosphere except in the presence of high CO_2 levels (> 8 %) and/or low O_2 levels (< 5 %), appears to be the most effective means to control Kohansho and preserve quality.

Conclusions

The results clearly showed that PFP is better than other postharvest treatments in reducing weight loss and incidence of Kohansho, as well as in retaining firmness and other quality attributes. Since Kohansho is caused mainly by fruit weight loss[3], we conclude that PFP treatment reduces Kohansho by restricting water loss. The HWD and HCD treatments reduce the occurrence of Kohansho even when the fruit lose weight, and these treatments are effective during storage, transportation, and marketing periods where some water loss is expected. Since the PFP treatment is more convenient and economical than the HWD or HCD treatment, the PFP which can maintain a high RH and a modified atmosphere except in the presence of high CO_2 levels (> 8 %) and/or low O_2 levels (< 5 %), may be preferable as a practical commercial method of Kohansho prevention and quality maintenance in 'Kiyomi' Tangor.

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有孔フィルム包装 (PFP), 温湯浸漬 (HWD), 温湯カルシウム浸漬 (HCD) の収穫後処理が‘清見’果実のこはん症発生と品質に及ぼす効果を比較した。3種類の処理を施した果実を段ボール箱に収納し, 10℃で4週間貯蔵した。PFP処理区では, ほかの処理および対照区(無処理) と比較して, 重量減少が明らかに小さく, 果実硬度が明らかに高い結果が得られた。すべての処理はこはん症の発生を減少させたが, PFPとHWD処理はHCD処理より高い減少効果を示した。また, これらの処理は対照区と比較して, 糖度 (TSS), 滴定酸度 (TA), TSS/TA比, アスコルビン酸含量, エタノール含量を損なうことはなかった。これらの結果より, 高湿度条件を保ち, かつ8％以上のCO_2濃度環境と5％以下のO_2濃度環境を避けたPFP処理はこはん症の抑制と品質保持において最も効果的で, 商業的に実用的な方法であると考えた。