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MATURITY AND HEAT PRETREATMENT INFLUENCE ON STORAGE QUALITY AT LOW TEMPERATURE OF SOME LOCAL VARIETIES OF AVOCADO *Persea americana* Mill IN TAIWAN

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The purpose of this research is to determine the quality changes of avocado fruits of local varieties during low temperature storage. Two local varieties with three maturity levels were selected to analyze the relation between maturity and storage quality under low temperature condition. Quality changes were studied by analyzing the skin colour, flesh colour, firmness, chilling injury, and rotting characteristics. Samples with high maturity showed better storage quality for both varieties. At 1℃, 'zhangan' avocado can be stored for 21 days and 'qing jing er hao' for 14 days with fruit retaining full ripening ability at room temperature. Pretreatments at 38℃ in water for 5, 15, 30, and 60 minutes were conducted to decrease the low temperature injury of avocado during 1℃ storage. Thirty-minute immersion resulted in better quality of avocado after 28 days of low temperature storage. Only pretreatment for 6 hours below 38℃ warm air is recommended for reducing chilling injury of avocado during 1℃ storage.

INFLUENCIA DE MADUREZ Y PRETRATAMIENTO DE CALOR EN LA CALIDAD DE ALMACENAMIENTO A BAJA TEMPERATURA DE ALGUNAS VARIEDADES LOCALES DE PALTA (Persea americana Mill) EN TAIWÁN

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El propósito de esta investigación fue determinar los cambios en la calidad de variedades locales de palta durante almacenamiento a baja temperatura. Se seleccionaron dos variedades locales con tres estados de madurez para analizar la relación entre madurez y la calidad del almacenamiento a baja temperatura. Los cambios en la calidad fueron investigados mediante el análisis de color de la piel, el color de la pulpa, la resistencia al daño por frío, y pudrición. Las muestras con una mayor madurez demostraron una mejor calidad en el almacenamiento para ambas variedades. La palta 'zhangan' puede almacenarse a 1° C por 21 días, y en el caso de la palta 'qing ping er hao' por 14 días donde la palta retiene totalmente a temperatura ambiente su capacidad de maduración posterior. Se realizaron pretratamientos en agua a 38° C por 5, 15, 30, y 60 minutos para disminuir los daños causados por el frío durante el almacenamiento a 1° C. La inmersión por treinta minutos entregó los mejores resultados en calidad después del almacenamiento a baja temperatura por 28 días. Sólo se recomienda el pretratamiento de 6 horas con aire caliente a 38° C para reducir daños en la palta causados por el frío durante el almacenamiento a 1° C.

1. Introduction

Maturity at harvest is a very important factor that determines storage-life and fruit quality to the consumer. Immature fruits are more likely to shrivel and are of

inferior quality when ripe. Overripe fruits are likely to become soft and mealy soon after harvest. Fruits picked either too early or too late in the season are more likely to have a shorter storage-life than those picked at the proper maturity. Maturity has been broadly studied in relation to the low temperature tolerance of horticultural crops. 'Hass' avocado fruits with high maturity or after ripening showed better storage ability under low temperature conditions (Hoffman et al., 2002). Chilling injury can be reduced, thus permitting extended storage time. A more advanced maturity is desirable for kiwifruit destined for long term storage at 0℃ (Gordon Mitchell et al., 1992). However, it is not always true for all. For example, 'Jonathan' apple, the most advanced in maturity at harvest, the highest incidence of low-temperature breakdown and the lowest incidence of storage scald was found. Apples were the least advanced in maturity at harvest, had the lowest incidence of low-temperature breakdown and the highest incidence of scald (Blanpied and Little, 1991).

As a postharvest treatment, heat treatments have been shown to be generally effective in reducing chilling injury in tomato fruit (McDonald et al., 2000), maintaining storage quality of strawberries (Vicente et al., 2002), mangos (Pepsis, et al., 1997), grapefruits (Porat, et al., 2000), avocados (Woolf, et al., 1995), pears (Abreu et al., 2003) and peaches (Zhau et al., 2002) during cold storage. Tolerance to low temperature in avocado can be increased by pretreatment at temperature such as 38℃ by pretreatment with hot air (Woolf et al., 1995; Florissen et al., 1996) and hot water treatments (Woolf, 1997) in 'Hass' avocados. Heat treatment could protect the ultrastructure of the pericarp cells in the heat -pretreated grape berries under chilling stress (Zhang et al., 2005). Heat pretreatment not only showed a significant reduction of chilling injury, but also effective decreased the decay of fruit during low temperature storage. Sabehat et al. (1998) showed that a 2-day 38℃ pretreatment of tomato fruit reduced chilling injury, including rots, in fruit stored at 2℃. It is also successful in controlling microbial levels or decay in citrus (Porat, et al., 2000). Those results are very useful guides for us to prolong the low temperature storage limits of the local variety of avocados in Taiwan. Effects of maturity and heat pretreatments on the low temperature storage quality of avocado will be focused on this work. Two local varieties of avocado with tree maturities are investigated for their quality changes at 1℃ within a 30 days' storage.

2. Material and methods

2.1. Materials

Two avocado varieties, 'Chanan' and 'Ching-Jin 2', were selected for this experiment. Avocado fruits were harvested from the orchard of local farmer in Cha-I county, Taiwan. Sufficient fruits were hand-picked from the trees treated by the same agricultural practices. Fruits were packed in carton boxes and transferred to laboratory within 12 hours at 4-7. Fruits were graded for size by weight. Free from damage and sunburn avocados were randomized for further analysis.

2.2. Methods

2.2.1. Low temperature storage for different maturity fruits

Avocado fruits were harvested at 10 day intervals over a 30 day period from the end of July to the end of August (high–season) in 2005 and 2006. Fruits were packed in carton boxes and promptly cooled and air stored at 1℃ without ethylene and monitored at 0, 7, 14, 21 and 28 days. After treatment the fruits were allowed to ripen at 21℃.

2.2.2. Hot water pretreatment

Fruits were immersed in a water bath tank set at 38℃ for the following periods: 0, 5, 15, 30 and 60 min. Thermometer was placed into tank and was used to monitor temperature changes during the heat treatment. Following heat pretreatment, fruits were allowed to recover at 25℃ for 2 h. Before low temperature storage (1℃), all fruits were wiped out water adhered on the skin by clean tissues individually.

2.2.3. Hot air pretreatment

Fruits were loaded in an air convection oven set at 38℃ for heat pretreatment by air for the following periods: 0, 6, 12, 24 and 48 h. To prevent water loss during the heat treatment fruits were individually sealed in perforated polyethylene bags. Following heat pretreatment, fruits were allowed to recover at 25℃ for 2 h. Polyethylene bags were removed before stored at 1℃.

2.2.4. Quality analysis

For all experiments, quality analysis was based on 3 replicates of 3 fruits. Quality parameters including color of skin, color of flesh, and hardness of flesh,
symptoms of chilling injury and decay of the fruits were assessed objectively.

2.2.4.1. Measurement of Color

Skin color of the fruits were measured using a color differential meter (ZE-2000 Nippon Denshoku Japan) to determined Hunter Lab's L value (lightness or brightness), a value (redness or greenness), and b value (yellowness or blueness) by averaging four measurements taken around the fruit equator. Color of flesh was determined on the surface of flesh, 0.5 cm beneath the skin. Measurements were taken for three samples and the average of L, a and b values were obtained. The colorimeter was warmed up for 30 min and calibrated with a white standard tile: L=95.87, a=-0.86 and b=2.47.

2.2.4.2. Measurement of Hardness

Texture measurements were made using a texture analyzer (TA-XT2 Texture analyzerStable Micro Systems(SMS)England). Samples were subjected to a puncture test at a constant speed of 2mm/sec, using a 5mm diameter round tipped puncture probe. Four measurements were taken on each fruit at different location, 0.5 cm beneath the skin, around the fruit equator. Measurements were taken for three samples and the average values were obtained.

2.2.4.3. Assessment of chilling injury

The degree of ripening of each fruit was determined using a subjective

assessment of softness determined by hand (Florissen et al., 1996). Once ripe, the fruit was checked the appearance first, then cut in half longitudinally and examined for symptoms of chilling injury. Chilling injury was rated on a relative scale of 0-3 as described by Woolf and Lay-Yee (1997), where 0 is for no occurrence; 0.5, <10%; 1.0, 10-20%; 1.5, 21-50%; 2, 51-75%; 2.5, 76-90%; 3.0, >90% of the fruit surface.

2.2.5. Statistical analysis

One way analysis of variance (ANOVA) was used in order to detect significant differences among avocado samples with different treatments. The significant level used was p~0.05. Duncan's multiple range tests were conducted to compare the mean values in different storage days. Statistical analysis was carried out using SAS software (SAS Institute, Cary, NC, USA).

3. Results and discussion

3.1. Effect of harvest maturity on the low temperature storage quality

Avocado fruits with different maturities were stored at 1℃. After taking out every the seventh, fruits were left at 21℃ to ripen for at least 3 days in order to observe the symptoms of chilling injury and analyze the quality of ripening. Figure 1 showed the color changes of flesh of 'Chanan' avocados with low (L), mid (M) and high (H) maturity at 21℃ for 3 days after 1℃ storage. For fruits with low and mid maturity of 'Chanan' variety, after preserving for 7 days and 14 days under the low temperature, there is no serious chilling injury happening in the avocado fruits (Table 2). Fruits were able to keep the good conditions both on appearance and flesh during ripening, and normal ripening was found. After three-week storage, the appearance of fruit was still good, however, serious chilling symptoms were found in the internal part of fruit, including internal browning and unable to soften normally. Not only the color of flesh became dark, but also the peel had obvious brown spot occurred. Then the situation was more serious for the samples taking out from storage on the day 28th. Fruit of the high maturity, after the low temperature storage for 7, 14 and 21 days, there is no serious chilling symptom happened. Moreover, three days after stored at 21℃, the overall quality of avocados is still good. No matter the appearance or the flesh could keep the good condition and the ripening of fruit was normally done. On the day 28th, fruits were found under good condition of appearance when they were taken out of storage room. But serious chilling injury symptoms occurred during ripening at 21℃. Internal browning and unable softening of flesh were major symptoms of chilling injury, small spots with dark brown color could also be found on the skin. Due to the unable fully softening, the variation of hardness of flesh became obviously resulted in the high standard deviations obtained in table 1.

For 'Ching-Jin 2' fruits with low maturity, changes of color were not obvious in the internal of fruits stored at 1℃ for 7 days and 3 days at 21℃ (Fig.1). Normal softening resulted in the decline of hardness was observed (Table2). Both observations showed that serious chilling injury was not found among samples. Storage for two weeks resulted in serious chilling injury in flesh three days after removed to 21℃ condition even the skin quality was excellent at the moment taking out from cold room. To extend the period of storage caused more serious chilling symptoms including internal browning and unable softening occurred during ripening. Brown spots were found on the skin, too. High standard deviations of the hardness of flesh were also found for samples with serious chilling injury

(Table 2). With the increase of maturity, fruits showed good ripening quality after 21 days for mid maturity and 14 days for high maturity samples stored at 1℃. Chilling injury became significant if the storage period reached 28 days for avocado fruits with mid maturity. Fruits with mid maturity showed the best low temperature storage ability as compared to fruits with other maturities.

Maturity had a dramatic effect on the incidence of vascular browning of cold stored fruits. Cutting et al., (1988) pointed out that the potential and often the incidence of the physiological disorders, vascular browning and mesocarp discoloration, were both increasing with increasing maturity of 'Fuerte' avocado. Browning disorders was very serious in fruits harvested early in the season, thereafter the incidence of the disorder declined but rose rapidly as the fruits reached high maturity (Cutting and Wolstenholme, 1992). There is a coincidence of the effect of maturity on the chilling injury of cold stored between 'Ching-Jin 2'avocado and their results. However, for 'Chanan' avocado, those symptoms above are reduced with the decreasing of fruit maturity which is not correspondent with their results.

Preharvest factors influencing postharvest chilling injury are mostly those to do with temperatures experienced during fruit development. Woolf et al., (1999) found flesh temperatures of avocado fruit growing in New Zealand, exposed to direct sunlight on the tree, frequently exceed 35℃. These exposed fruit had lower levels of chilling injury than fruit from shaded parts of the tree, when stored at 0℃. They believed the avocado fruits exposure to high temperatures on the tree, particularly close to or at harvest, may induce tolerance to low temperatures in postharvest storage. Fruits with high maturity it means that those have more chances to expose to sunshine on the tree. The field temperature in the center of Taiwan area during summer time is always above 35℃. It will be very easy to rise the flesh temperature of avocado exceeding 35℃. That may be one of the reasons for the relatively high matured fruits can be stored long under low temperature conditions in my experiments. Bramlage and Weis (1997) have concluded that reduced chilling injury (scald) susceptibility in 'Delicious' apples can be related to late season high temperatures.

High temperatures, both in terms of diurnal fluctuations and long-term sunlight exposure, can result in differences in internal quality properties such as sugar contents, tissue firmness, and oil levels, as well as in mineral content differences. Oil content is known to increase and water content is to decrease with increasing maturity (Pearson, 1975). Those differences will also respond differently to postharvest low temperatures (Woolf and Ferguson, 2000). The activity of polyphenol oxidase (Kahn, 1975) and the concentration of abscisic acid (Cutting et al., 1988) both increase with the increasing maturity of avocado fruits. Fruits under different maturity will have different quality properties contents. To understand more about the correlation between low temperature storage ability of avocado and each property factors should be very helpful on finding the optimum maturity for low temperature storage of avocados.

3.2. Effect of hot water pretreatment on storage quality at 1℃

'Ching-Jin 2' fruits were selected for the hot water pretreatment due to their bad low temperature storage tolerance. Fruits were immersed in 38℃ hot water before stored at °C condition. 5 minutes hot water treated fruits were found with good

quality after 7, 14 and 21 days storage. The color of skin was very similar to the fruits before storage. Fruits were ripe normally 3 days after removed to 2℃ condition. No chilling injury symptom was found both outside and inside of the fruits. Although the appearance of fruits taking out of cold room at the 28th day was still good, however, serious chilling injury symptoms mentioned above were found 3 days after ripening at 2℃. Fruits pretreated at 3℃ water bath for 15 min were found with good quality on the day 7th and day 14th but day 21st and day 28th. Some of the fruits taking out on the day 21st were found with chilling injury symptoms during ripening at 2℃. All fruits showed serious chilling injury symptoms after 3 days at 2℃ if the storage period extended to four weeks. No chilling injury symptom occurred on the samples pretreated at 3℃ water bath for 30min. Avocados kept good outlook and internal qualities after a 28 days' storage at °C. Fruits went through normal ripening resulted in a similar texture as well as color of flesh and skin to the samples without storage. Hot water pretreatment at 3℃ for 60 min also prolonged storage life of avocado fruits at °C. It could be extended up to 21 days without serious chilling injury symptom occurred after fully ripe. Samples taking out of storage on the 28th day began to show serious symptoms, internal browning and unable to ripening, during 3 days' storage at 2℃. To sum up the above, 30min hot water pretreatment at 3℃ is the optimum condition for 'Ching-Jin 2' avocado fruits to prolong the storage period to 4 weeks.

Hot water treatment significantly reduced skin damage of 'Hass' avocado caused by cold disinfestation, with 40 degrees C for 30 min, 4℃ for 20-30 min and 42℃ for 25-30 min giving the greatest reduction. Hot water treatments also reduced body rots in ripe fruit, with 40 and 4℃ for 30 min being consistently the most effective (Hofman et al., 2002). Woolf (1996) found that 'Hass' avocado fruit heated in water at 3℃ for 60 minutes was the optimal duration that eliminated external chilling injury, while stored at 0.5℃ for up to 28 days. As compared to above, we found that the optimal conditions for 'Hass' avocado to reduced chilling injury were longer heating time or higher heating temperature than those of 'Ching-Jin 2'. By the consideration of size, the average weight of 'Hass' is about 250g which is less than the average weight of 'Ching-Jin 2', about 650g. It suggested that the heat sensitivity of 'Ching-Jin 2' should be higher than that of 'Hass' avocado.

3.3. Effect of hot air pretreatment on storage quality at 1℃

Fruits were loaded in an air convection oven at 3℃ for 6, 12, 24 and 48h. Results showed that all pretreated fruits were not able to undergo normal ripening 3 weeks after °C storage. The shorter time of pretreatment, the longer time of storage was observed. Pretreated the avocados with hot air for 6hr can still keep quite good ripening quality after 21 days' storage at °C. For 12h-pretreated samples, internal tissue disruption was found on the 7th day of storage and serious internal browning was observed 14 days after storage at °C. Avocados pretreated for 24 and 48hr showed serious browning of the skin and tough texture of flesh which were totally lost of commercial value. Therefore, they are not necessary for further storage. Florissen et al., (1996) demonstrated that short heat treatments of 6-12h applied to pre-climacteric 'Hass' avocado fruit provides partial protection from chilling. For 'Sharwil' avocado heat pretreatment period of 8 to 12 hours is effective in reducing chilling injury symptoms when the pulp is at less than or equal to 2. during 16 days of storage (Nishijima et al., 1995). Chilling injury symptoms were reduced when 'Sharwil' avocados were held at 37 to 3℃ for 17 to 18 hours

and then air-cooled at 2 °C for 4 hours before storage at 1.℃ for greater than or equal to 14 days (Sanxter et al., 1994). However, from our studies we found protection effect from chilling injury observed in samples treated for 6h in 3℃ air only. The protection effect was lost gradually if fruits were heated for longer time. A similar result was found by Woolf et al., (1995). For 'Ching-Jin 2' avocado, 6h heated at 3℃ is the optimal duration which is much shorter than that of 'Hass' or 'Shirwil' avocado. No matter hot water or hot air treatment, 'Ching-Jin 2' avocado showed a shorter preheating time or lower preheating temperature than other varieties of avocados for the best maintaining fruit quality at low temperature. We may believe that 'Ching-Jin 2' is a very heat sensitive avocado.

Florissen et al., (1996) used short heat treatments to overcome the

effects of chilling injury was investigated by subjecting the avocado fruits to 3 °C for 6, 12, 24, 36, or 48h prior to transferring them to 0℃ for 7, 14, or 21 days. Results showed that heating for 6-12 h provided a significant degree of protection from chilling injury. For 'Ching-Jin 2' avocado, the symptoms observed in fruits heat-treated in water for up to 60min were ascribed to chilling injury. The optimum treatment for protection was 30min immersed in 3 °C water, since time of greater duration exacerbated the level of chilling injury. The serious browning on the skin and tough texture of flesh observed in fruits right after treated by 3℃ air for 24 or 48h should not be correlated to chilling injury. It could be ascribed to heat injury. Florissen et al., (1996) believed that there is a 'crossover' point after which the protection from chilling injury is lost and the heat treatment has a detrimental effect. In their trial the 'crossover' point was between 12 and 24 h at 3℃ which coincided with that of samples treated by 3℃ air in our experiment. For samples heat-treated by water the 'crossover' point was between 30 and 60 min.

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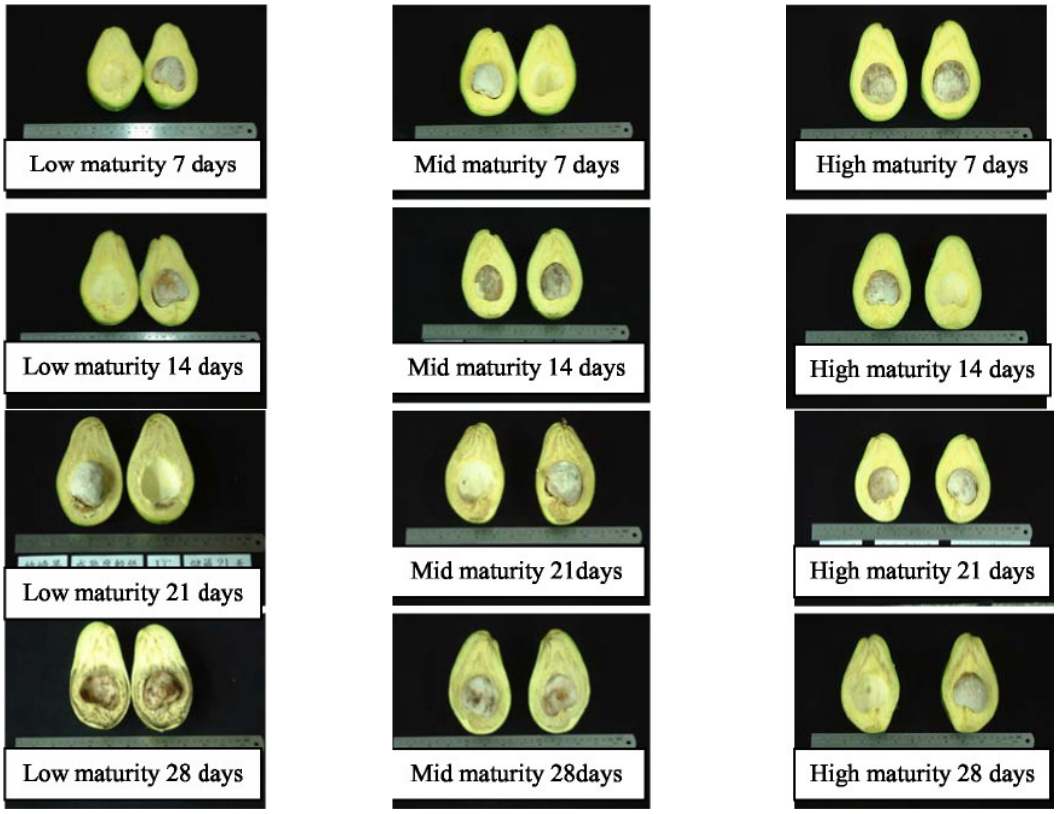


Fig.1. Changes of internal conditions of 'Chanan' avocados with low, mid and high maturity at 2℃ for 3 days after℃ storage.

Fig.1. Cambios de condiciones internas de aguacates 'Chanan' con madurez baja, mediados de y alta en 21℃ por 3 días después del almacenaje 1℃

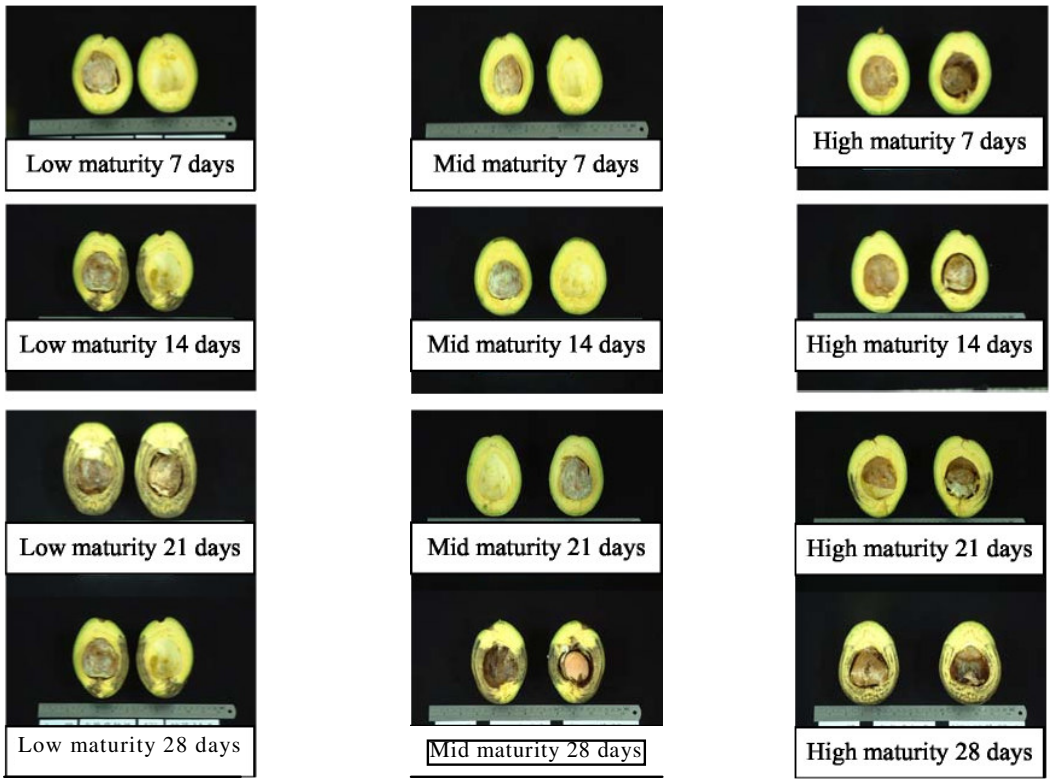
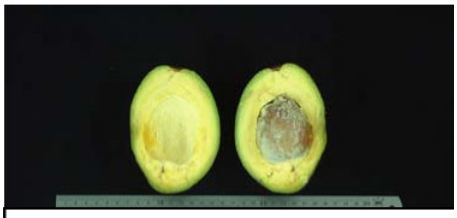


Fig.2. Changes of internal conditions of 'Ching-Jin 2' avocados with low, mid and high maturity at 2℃ for 3 days after℃ storage.

Fig.2. Cambios de condiciones internas de aguacates 'Ching-Jun 2' con madurez baja, mediados de y alta en 21℃ por 3 días después del almacenaje 1℃

5 min 38℃ Water Preheating
21 days 1℃ Storage

30 min 38℃ Water Preheating 28 days 1℃ Storage



15 min 38℃ Water Preheating
14 days 1℃ Storage

60 min 38℃ Water Preheating 21 days 1℃ Storage

Fig.3. Storage limits of 'Ching-Jin 2' avocado fruits preheated in 3 °C water for 5, 15, 30 and 60 minutes and stored at °C for 4 weeks as well as 3 days at 2℃. Fig.3. Los límites del almacenaje de los aguacates precalentados en 38℃ riegan para 5, 15, 30 y 60 minutos y at1℃ almacenado por 4 semanas así como 3 días en 21℃.

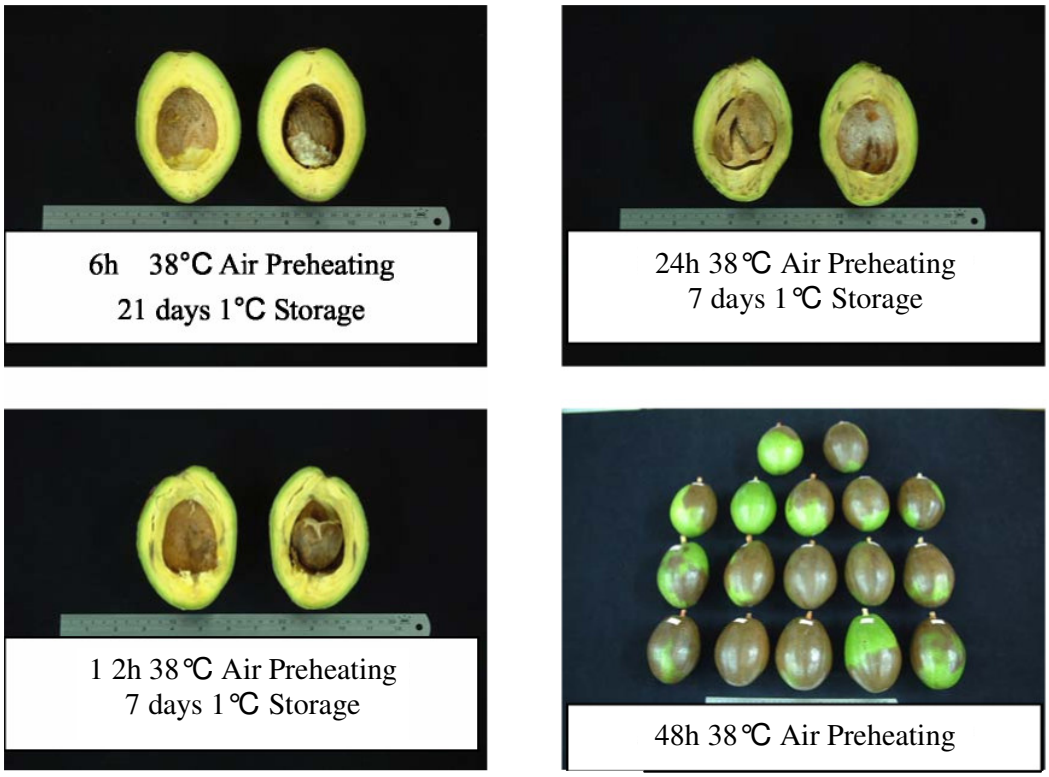
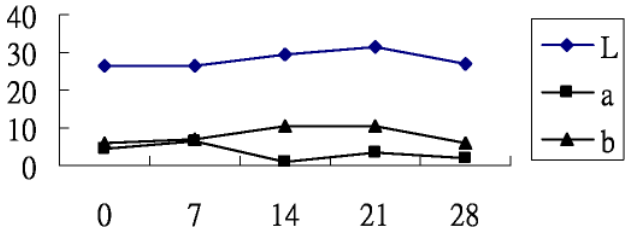
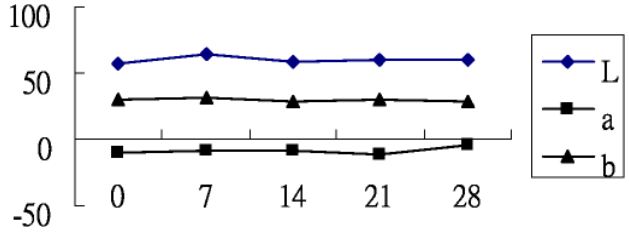


Fig.4. Storage limits of 'Ching-Jin 2' avocado fruits preheated by 3℃ air for 6, 12, 24 and 48 hours and stored at °C for 4 weeks as well as 3 days at 2℃

Fig.4. Los límites del almacenaje de los aguacates precalentados por 38℃ ventilan por 6, 12, 24 y 48 horas y almacenado en 1℃ por 4 semanas así como 3 días en 21℃



(A)



(B)

Fig.5. Skin (A) and flesh (B) color changes of 'Ching-Jin 2' avocado fruits preheated in 3 °C water for 30 minutes followed by 7, 14, 21 and 28days at℃ as well as 3 days at 2℃.

Fig.5. Color de la piel (a) y color de la carne (B)changes de los aguacates precalentados en el agua 38℃ por 30 minutos siguió por 7, 14, 21 y 28days at1℃ así como 3 días en 21℃.

Table1. Hardness of ‘Chanan’ and ‘Ching-Jin 2’ avocados with low (L), mid (M) and high (H) maturity at 2℃ for 3 days after℃ storage.
Table1. Dureza de aguacates con (l) bajo, (m) mediados de y alta (h) madurez en 21℃ por 3 días después del almacenaje 1℃.

	Hardness (g/cm^2)					
	Chanan			Ching-Jin 2		
Time(day)	L	M	H	L	M	H
7	108.8±28.4	82.5±6.8	74.6±3.8	106.4±11.6	79.0±11.4	74.4±121.9
14	172.5±24.3	99.7±9.0	302.3±42.9	142.6±58.9	62.0±15.0	72.0±15.9
21	131.1±30.0	178.1±53.3	187.8±53.7	591.0±522.5	95.1±3.0	81.2±210.9
28	660.6±403.5	867.4±770.5	163.3±76.8	658.4±356.4	336.3±397.8	740.6±584.0

Values are expressed as average ± standard deviation (n=3).

Table2. Chilling injury of ‘Chanan’ and ‘Ching-Jin 2’ avocados with low (L), mid (M) and high (H) maturity at 2℃ for 3 days after℃ storage.
Table2. Lesión que se enfría de aguacates con (l) bajo, (m) mediados de y alta (h) mad urez en 21℃ por 3 días después del almacenaje 1℃.

	‘Chanan			‘Ching-Jin 2’		
Time(day)	L	M	H	L	M	H
7	0.67^{ay}	0.67_{ax}	0.25_{ax}	0.58^{by}	0.25_{ax}	0.08_{ax}
14	1.83_{by}	0.58_{ax}	0.33_{ax}	1.33^{by}	0.41^{ax}	0.42_{ax}
21	1.83^{by}	1.33_{by}	0.33^{ax}	1.25^{bx}	0.25_{ax}	0.58^{ax}
28	3.00_{cz}	1.75^{by}	1.08^{ax}	1.67^{bx}	1.08_{ax}	1.83^{bx}

$_{a, b, c}$ Different superscripts within same column indicate significant different at 5% level.
$_{x, y, z}$ Different superscripts within same row indicate significant different at 5% level.

Table3. Hardness of 'Ching-Jin 2' avocado fruits preheated at 3℃ water or air followed by 7, 14, 21 and 28days at℃ as well as 3 days at 2℃.
Table3. La dureza de los aguacates precalentados en el agua 38℃ o el aire siguió por 7, 14, 21 y 28days at1℃ así como 3 días en 21℃.

	Hardness g/cm^2					
	Water				Air	
Time(day)	5min	15min	30min	60min	6h	12h
7	109.97	121.41	119.76	105.24	120.99	200.42
14	70.41	73.93	80.88	71.74	69.13	1263.10
21	80.40	202.60	81.69	74.08	81.63	1248.06
28	163.50	293.30	74.97	298.13	294.30	1257.78