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IMPROVING COLOR PRESERVATION OF FROZEN SUGAR APPLE BY VACUUM PACKAGING

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Abstract. **Vacuum packaging as a means for improving color preservation of frozen sugar apple was investigated. Individually quick frozen (IQF) with vacuum packed and non-vacuum packed frozen sugar apple were stored at -18°C for 12 months. Little discoloration was observed for vacuum packaged whereas serious discoloration was observed for the non-vacuum packaged. Color values of vacuum packaged were significantly different from those of non-vacuum packaged samples. The activity of polyphenol oxidase (PPO) was measured. The main phenolic compounds were (+) catechin, chlorogenic acid, eugenol and gallic acid. The values of those four compounds were higher in frozen sugar apple with vacuum packaged than those of non-vacuum packaged samples. In addition, the values of chlorophyll in the pericarp of the vacuum packaged were higher than those of discolored non-vacuum packaged frozen samples. It was concluded that the discoloration of the pericarp of frozen sugar apple was oxygen-involved browning. IQF is a feasible method for extending shelf-life and vacuum packaging is an effective method to reduce browning and to preserve better color of frozen sugar apple.**

Sugar apple (or sweet sop) is a tropical fruit. It is reported that the planting acreage of sugar apple in Taiwan is over 11,100 acres (Taiwan Dept. of Agriculture and Forestry, 1996). Taiwan is an island country. Its north to south geography is equivalent to a latitude between Miami in Florida and Havana in Cuba. Sugar apple has distinct taste and flavors and is a preferred fruit by the oriental ethnics. Planting of sugar apple in Florida is just beginning. It is estimated that there are only 25 acres of commercial groves in south Florida (Crane, 1996). There is a potential large market for growing sugar apple in the United States.

Sugar apple can become soft within 3-4 days after harvest, and some will crack open. If the fruit is not consumed within a few days after harvest, it begins to decay, the pericarp turns to brown or black color, and loses its commercial value (Ke et al., 1983; Tsay and Wu 1989a, b). The cause of the fruit senescence is not well understood, and its control and prevention methods remain to be developed. Furthermore, the sugar apple is very sensitive to chilling injury (Tsay and Wu, 1989b; 1990). The critical temperature of chilling injury for the fruit is around 15°C and, when stored at this temperature, the shelf-life for the fresh fruit is around 10 days. Frozen preservation is an effective method for extending shelf -life of sugar apple from approximately 10 days to 12 months (Lin and Yang, 1988). One of the problems with freezing sugar apple arises since the fruit is frozen without blanching and then becomes brown easily as a result of enzymatic browning (Ma et al., 1992).

The objective of this study was to evaluate the method of vacuum packaging as a means to reduce browning and to improve color preservation of frozen sugar apple.

Materials and Methods

Materials. Samples of sugar apple (*Annona squamosa* L.) were harvested at a commercial orchard in Taitung County (South Taiwan). The fruits were classified into different stages according to softness. Soft fruit (about 70-80% ripe) was considered as optimal maturity.

Frozen samples. Optimal maturity fruits weighing an average of 290 ± 10 g were selected. Samples were frozen by three different methods: (1) individually quick frozen (IQF) by convection at -45°C in an IQF system, (2) statically frozen at -15°C, and (3) frozen by liquid nitrogen (-195.8°C). Some were vacuum packed with k-nylon bags after freezing and some were non-vacuum packed. The progress of browning was investigated to determine which method was most effective in inhibiting the browning reaction.

Color measurements. A PC-based color difference meter (JP 7100F JUKI Assoc. Japan) was used for measuring the Hunter L.a.b. values.

Enzyme activity assay. (a) Crude enzyme extract: A crude enzyme extract was prepared by taking 10 g of fruit pericarp from each sample, grounded in 40 ml of citrate-phosphate buffer (pH 4.5) with 2% polyvinylpyrrolidone by mortar pestle on ice bath, and the homogenate was centrifuged at 13,000 × g for 30 min. The supernatant was dialyzed overnight and used for enzyme assays (Ma et al., 1992). (b) Polyphenol oxidase (PPO) assay: The enzyme activity was assayed by using catechol (Sigma Chemical Co.) as substrate (Ma et al., 1992; Tsay and Wu, 1989a). The enzyme activity was calculated on the basis of the slope of the linear portion of the curve plotted with A 420 against time (up to 3 min). One unit of enzyme activity was defined as 0.001 A 420 min^{-1} (mg of protein).

Analysis of phenolic compounds. (a) Preparation of phenolic compounds extract: 1 g of L-ascorbic acid was added to the 50 g of pericarp sample. Added 70 ml of MeOH to prevent oxidation of phenolic compounds during extraction, the samples were homogenized to make a suspension and centrifuged to remove the residue. The supernatant was condensed to 20 ml, filtered with a 0.5 μm membrane and injected into a HPLC for analysis of phenolic compound (Wang and Huang, 1993). (b) Procedure for phenolic compounds analysis: High performance liquid chromatography was carried out with an Hitachi liquid chromatography pump equipped with a model L-6200A solvent delivery system, and UV detector with the wavelength set at 280 nm. A 1008 mm internal diameter Radial-Pak C_{18} column with water-acetic

acid (95:5) solution as a mobile phase was employed to separate phenolic compounds.

Analysis of chlorophyll. (a) Extraction of chlorophyll: Took 60 g of the fruit pericarp and added 100 ml of acetone, blended using a Waring blender for 2 min, filtered with Whatman No. 1 filter paper and washed the residues until the color of the liquid turned transparent, combined the supernatants and diluted to 200 ml for use. In the process of analysis, the influences of light and heat should be prevented (Gross, 1991). (b) Quantitative analysis and calculation: Put the sample liquid (30 ml) into a 50 ml-sized bottle, added 80% acetone to a volume of 50 ml, and detected optical density with a double beam spectrophotometer (Hitachi, UV-200, Japan) at 663 and 645 nm.

Total chlorophyll (mg/L) = 20.2 O.D. at 645 nm + 8.02 O.D. at 663 nm. The unit of chlorophyll can be changed into ppm (Gross, 1991).

Data treatment. Statistical analysis was performed using General Linear Model of Statistic Analysis System.

Results and Discussion

Comparison of color retention. The fruit cracked using liquid nitrogen immersed freezing and discolored in the process of statically contacted freezing; the IQF fruit remained unchanged. Therefore, the IQF samples were selected for further tests. Table 1 presents the measured color changes of the pericap during storage of frozen sugar apple at -18°C. The color changed in both non-vacuum packaged and vacuum packaged samples; however, the color was better retained by the latter treatment. This is reflected by higher 'L' and 'b' values, and lower 'a' values for vacuum packaged samples.

Changes in polyphenol oxidase activity. No blanching was used to prevent browning reactions since the quality of blanched sugar apple is unacceptable. The activity of PPO increases during ripening (Tsay and Wu, 1990). The PPO activity continued to decrease in frozen sugar apple with or without vacuum packaging during 12 months of storage at -18°C (Table 2). However, PPO activity remained higher in vacuum-packaged fruit as compared to those with non-vacuum packaging.

Phenolic compounds in the pericarp of frozen stored sugar apple. Since the phenolic compounds are substrates of PPO, it is of interest to determine phenolic compounds in the pericarp of the fruit. The major four categories of phenolic compounds in the sugar apple are (+) catechin, chlorogenic acid, gallic acid and eugenol. The contents of these phenolic compounds decrease during ripening (Tsay and Wu, 1990; Mowlah and Itoo, 1982). We found these phenolic compounds also continued to decrease during storage of frozen sugar apple at -18°C (Table 3). This indicated that the phenolic compounds in the pericarp of sugar apple fruit were oxidized by PPO activation and produced brown color products. Levels of these PPO substrates remained higher in vacuum-packaged fruit, however, suggesting that they were not oxidized as efficiently by PPO as were the non-vacuum packaged samples.

Changes in chlorophyll in the pericarp of frozen stored sugar apple. The content of chlorophyll of fresh sugar apple is over 100 ppm. Chlorophyll levels showed a similar pattern to that of phenolic compounds in that they continued to decrease during storage of frozen sugar apple at -18°C and that the rate of decrease in the non-vacuum packaged fruit was faster than in the vacuum-packaged fruit (Table 4).

Table 1. Changes in the color of the pericarp during storage of frozen sugar apple stored at -18°C.

	Storage time (months)	Hunter color values		
		L.	a.	b.
Non-vacuum packaging	0	37.5 g[z]	-5.7 g	14.4 g
	3	34.6 d	-2.5 c	11.4 d
	6	33.4 e	0.4 b	8.6 e
	12	31.5 f	4.5 a	2.7 f
Vacuum packaging	0	37.5 g	-5.7 g	14.4 g
	3	37.0 a	-4.2 e	14.1 a
	6	36.5 b	-3.5 d	13.5 b
	12	35.4 c	-2.5 c	12.4 c

[z]Mean values of those samples with same letter within a row are not significantly different (P > 0.05), df = 8, MSE = Mean Square of Error.

Table 2. Changes in the polyphenol oxidase activity (PPO) of the pericarp during storage of frozen sugar apple at -18°C.

	Storage time (months)	Relative PPO activity
Non-vacuum packaging	0	87.3 f[z]
	3	75.0 b
	6	67.7 c
	12	51.3 d
Vacuum packaging	0	87.3 f
	3	82.3 a
	6	79.3 b
	12	68.7 c

[z]Mean values of those samples with same letter within a row are not significantly different (P > 0.05), df = 8, MSE = Mean Square of Error.

Table 3. Changes in the phenolic compound of the pericarp during storage of frozen sugar apple at -18°C.

		Phenolic compound (ppm)			
	Storage time (months)	gallic acid	eugenol	catechin	chlorogenic acid
Non-vacuum packaging	0	11.7 g[z]	12.0 g	151.9 g	65.6 g
	3	9.5 b	10.4 c	114.0 c	54.8 c
	6	7.5 c	9.5 d	103.6 d	47.8 e
	12	4.3 d	8.1 e	81.2 e	34.1 f
Vacuum packaging	0	11.7 g	12.0 g	151.9 g	65.6 f
	3	10.5 a	11.6 a	146.7 a	60.0 a
	6	9.4 b	11.3 ab	140.3 ab	58.2 b
	12	7.5 c	11.1 b	134.3 b	53.5 c

[z]Mean values of those samples with same letter within a row are not significantly different (P > 0.05), df = 8, MSE = Mean Square of Error.

Table 4. Changes in the chlorophyll of the pericarp during storage of frozen sugar apple at -18°C.

	Storage time (months)	Total chlorophyll (ppm)
Non-vacuum packaging	0	110.6 f[z]
	3	95.1 c
	6	83.4 d
	12	60.8 e
Vacuum packaging	0	110.6 f
	3	104.7 a
	6	100.9 b
	12	94.0 c

[z]Mean values of those samples with same letter within a row are not significantly different (P > 0.05), df = 8, MSE = Mean Square of Error.

In conclusion, individually quick frozen (IQF) is a suitable method for freezing sugar apple. The green color of sugar apple continued to decrease during 12 months of frozen storage. The phenolic compounds, namely (+)-catechin, chlorogenic acid, eugenol and gallic acid also continued to decrease as a result of increased activity of the polyphenol oxidase (PPO). The discoloration of sugar apple fruit was a combined result of the oxidation of phenolic compounds and the degradation of the chlorophyll. Vacuum packaging was an effective method for inhibiting the PPO activity by removal of oxygen from the packaged fruit and, therefore, vacuum-packaged fruit were capable of retaining better color than the non-vacuum packaged fruit.

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ENZYME-PEELED CITRUS IN MODIFIED-ATMOSPHERE PACKAGING COMPARED WITH WHOLE FRUIT

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Abstract. **Peeled oranges and grapefruit had about the same respiration rate as whole fruit. Using packaging film having O_2 permeance of 6600 ml/m²day, bags of peeled fruit had headspace O_2 and CO_2 concentrations similar to the levels found in the interior gas of fruit coated with candelilla wax. With porous film having permeance of 37,000 ml/m²day the headspace gas concentrations were similar in composition to interior gases of uncoated fruit. Ethanol increased at a much faster rate for peeled fruit than for whole fruit. After 5 weeks storage the peeled fruit had yeast populations of about 1×10^6 cfu/g and appeared inedible.**

Citrus sections prepared by aseptic slicing have been shown to have good stability for about 12 days with respect to color, pH and sugar content, although vitamin C decreased by about 22% and the product lost its characteristic citrus flavor (Rocha et al., 1996).

A similar product developed in our laboratory consists of citrus sections whose peeling has been loosened by vacuum infusion of pectinases (Bruemmer, 1981; Baker and Bruemmer, 1988). Enzyme-peeled citrus is being produced commercially (Baker and Grohmann, 1995), although more information is needed on its storage properties. For example, some leakage of liquid occurs from stored sections, although this was reduced by application of a coating (Baker and Hagenmaier, 1997).

It would seem that gas concentrations might be as important for storage of enzyme-peeled citrus sections in modified atmosphere packaging as they are for storage of whole fruit. Whole citrus fruit has been shown to develop ethanolic off-flavor when stored under conditions of reduced O_2 and elevated CO_2 (Ke and Kader, 1990). Similarly, seal-wrapped whole citrus fruit sometimes develops off-flavor when stored under conditions that cause O_2 depletion or CO_2 build-up (Miller and Risse, 1988).

The present study aims to examine the storage stability of enzyme-peeled citrus in modified atmosphere packaging, from the point of view of how its storage relates to whole citrus fruit.

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