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Physical and chemical stability of Refractance Window®–dried mango (Philippine 'Carabao' var.) powder during storage

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ABSTRACT

The effect of packaging atmosphere, storage temperature, and time on the physical and chemical stability of Refractance Window® (RW)-dried mango powder was evaluated over a period of 12 months. RW-dried mango powder with a moisture content of 0.037 ± 0.001 kg water/kg dry solids was stored at 5, 22 and 45°C for 12 months using air or nitrogen flush packaging. Headspace gas composition in the package, moisture content, color, ascorbic acid (AA), β-carotene, and microstructures of powder was measured at months 0, 6, and 12. The mango powder stored at 45°C suffered discoloration and ascorbic acid and β-carotene degradation after 6 and 12 months of storage in both air packaging and nitrogen flush packaging, while powder stored at 5 and 22°C had reduced nutrient degradation and preserved color of mango powder for both packaging conditions. Replacing the air inside the package with nitrogen was effective in preserving AA in mango powder stored at 22 and 5°C. Nitrogen flushing also reduced the percentage loss of β-carotene after 6 months, but no significant difference was found after 12 months. The microstructure of mango powder stored at 5 and 22°C was preserved after 6 and 12 months. Regardless of the packaging technique applied, the powder particles inside the package formed a single lump when stored at 45°C. The microstructures of mango powder at different storage conditions confirmed these results. Nitrogen flush packaging of mango powder is recommended to minimize degradation during long-term storage.

Introduction

The Carabao mango variety known as "Manila super mango" ranks third among the fruit crops produced in the Philippines after banana and pineapple, based on export volume and value with a total of 800,551 MT harvested in 2011.[1] In terms of total mango production worldwide, Philippines ranked sixth in 2010 as exporter.[2] Mango is rich in β-carotene, ranging from 800 to 13,000 µg/100 g depending on the cultivar,[3] and about 60% of all carotenoids found in mango is β-carotene.[4] It also contains ascorbic acid (AA), ranging from 7.8 to 172 mg/100 g of ripe mango pulp.[5,6] However, the problem confronting the Philippine mango industry is seasonality of production and unstable market situations. Mango is highly perishable, prone to mechanical damage, and microbial contamination during postharvest handling and processing.[7] Physical damage and chemical quality losses in mango may range between 5 and 87% depending on how the

product is handled.[8] In fact, untreated ripe mango fruit "Haden" suffered 100% fungal infection and severe decay during 18 days of storage at 25°C.[9]

Alternative products, such as mango powder, are gaining popularity as ingredients of other food products including health drinks, baby foods, sauces, marinades, confections, yogurt, ice cream, nutrition bars, baked goods, cereals[7] and in the pharmaceutical and cosmetic industry.[2] Currently, several drying technologies for commercial production of fruit flakes or powder are available, including Refractance Window® (RW) drying, a novel drying technique designed mainly to convert liquid and semiliquid foods or puree into powder or flakes. The principles and operations of this drying technology have been described.[10,11]

Refractance Window® drying offers many advantages and benefits when applied to fruits and vegetables. In experiments using carrots, strawberries, and squash, it was shown that the RW drying system maintains

the nutritional (vitamins) and sensory (color, aroma) attributes of the dried products.[10,12] It was also reported that the bright green color of RW-dried asparagus remained unchanged.[13] An earlier investigation by Abonyi et al.[14] showed that RW-dried carrots and strawberries were not significantly different when compared to their freeze-dried counterparts. Moreover, the study reported 94 and 90% retentions of ascorbic acid and β-carotene in strawberries through RW drying, comparable to freeze-dried products. Cunha et al.[15] found high β-carotene retention and decrease in browning during spout–fluidized bed drying of mango at high process temperatures (70°C) for a very short time. Most recently, Caparino et al.[11] concluded that the physicochemical properties of Refractance Window®–dried mango (RW-M) powder were comparable to the freeze-dried counterpart and better than drum- and spray-dried mango powder. Experiments conducted on a pilot-scale equipment revealed that RW dryers have higher thermal efficiency and low energy consumption compared with conventional dyers. It was reported previously[10,16] that the coliforms, *Escherichia coli* and *Listeria innocua* inoculated in pumpkin puree, were significantly reduced to minimum detection levels after RW drying, indicating that foods dried using this method are safe. Despite the reported positive results of fruit products derived from RW drying, there is no published literature showing thermal degradation, physical and nutritional changes of RW-M powder in storage. The objective of this study is to evaluate the physical and chemical stability of RW-M powder during storage as affected by packaging atmosphere and storage temperature over a period of 12 months to aid in the manufacture and handling of high-quality mango powder.

Materials and methods

Preparation of mango flakes

Frozen mango puree (Philippine 'Carabao' var.) was acquired from Ramar Foods International (Pittsburg, CA, USA). The mango puree was prepared following the same procedure as described by Caparino et al.[11] The average moisture content of the puree, determined using a standard oven method,[17] was 6.5 ± 0.1 kg water/kg dry solids, while the sugar content was $14.5° \pm 0.5°$Brix. Before drying, the samples were thawed overnight at ~23°C and blended (Oster blender, Mexico) to a uniform consistency using the lowest speed setting for 5 min. The puree was dried to 0.037 ± 0.001 kg water/kg dry solids using a pilot-scale RW dryer.[11]

During the drying operation, the mango puree was uniformly spread on the top surface of a plastic conveyor belt, with the bottom surface in contact with circulating hot water (95–97°C). Water temperature was maintained just below boiling point to minimize bubble formation, so that the hot water was constantly in contact with the belt to allow transmission of thermal energy directly into mango puree through the plastic conveyor belt.[10,14] The initial thickness of puree samples of 0.5–0.7 mm was controlled using a spreader bar located at the inlet section of the dryer. Moisture coming out of the product during drying was evacuated by suction air (22°C and 50–52% RH) flowing at an average velocity of 0.7 m/s. The dried mango was cooled just before exiting the dryer in form of flakes or thin sheets that were then collected and sealed in Ziploc® bags. The sealed bags were put inside multilayer aluminum laminated pouches. And to restrict oxidation, the packaging was flushed with nitrogen gas, heat sealed, and stored at −35°C until further analysis.

Sieving, packaging, and storage

Dried mango flakes or sheets obtained from RW drying were ground using a mortar and pestle and sieved using mesh sizes 10 and 12 (American Society for Testing and Materials, ASTM) to obtain mango powder with particle sizes ranging from 1.7 to 2.0 mm (International Organization for Standardization, ISO), respectively.[18] The mango flakes were packaged in multilayer aluminum-laminated pouches with ASTM test specification of zero oxygen (at 23°C and 0%RH) and zero water vapor (at 37.8°C and 100%RH) transmission rates (Osiopack® Retort Gold Pouch, Osiopack, Anaheim, CA, USA).

Thirty six pouches were prepared, each containing approximately 20 g of the dried mango powder. One half of the pouches were air packaged and the other half were flushed with nitrogen. Air-packaged pouches of mango powder were heat sealed using 16" impulse bag sealer (Powerseal, Anaheim, CA, USA), while an automatic vacuum sealer (UltraVac®250, KS) was used for the nitrogen-flushed counterparts by first sucking out the air from the packages at 26 kPa followed by purging with nitrogen gas for 3 s, and finally heat sealing for a standard time of 7 s. The air-packed and nitrogen-flushed samples were stored at 5°C (refrigerator), 22°C (ambient temperature), and 45°C (oven) for 12 months. Physical properties, nutritional qualities, and microstructures were evaluated in duplicate during 0, 6, and 12 months of storage.

Moisture content and water activity

The moisture content of mango puree and RW-M flakes/powder was determined using the standard oven

method at 70°C and at absolute pressure of 13.3 kPa for 24 h.[17] The water activity of the RW-M powder was measured in duplicate using Aqualab 4TE water activity meter (Decagon Devices, Pullman, WA, USA) set at $25 \pm 1°C$.

Physicochemical characteristics and nutritional quality

Headspace gas
The headspace carbon dioxide, oxygen, and nitrogen gas concentrations of the packaged mango flakes were measured with a gas analyzer (Fisher Gas Partitioner 1200, Fisher Scientific Co., Pittsburgh, PA, USA) equipped with a thermal conductivity detector. During the measurement, a 3-cm² adhesive septum with a 0.5-cm layer of dried silicone gel was attached to the surface of the package to avoid leakage when the gas sample was collected. A gas sample of 0.25 mL was withdrawn from the headspace of the package by a syringe through the septum. Gas concentration from each package headspace was determined by injecting the gas sample into a 6.096-m stainless steel column (3.18 mm i.d.) packed with 80/100 mesh Chromosorb P-AW and a 0.9144-m (4.76 mm i.d.) aluminum column packed with 60/80 mesh molecular sieve 13X connected in series and a thermal conductivity detector inside the gas analyzer. The flow of ultrapure helium carrier gas and oven temperature was adjusted to 10 mL/min and 40°C, respectively.[19] The instrument was calibrated by triplicate injections of both 10% CO_2 and 10% O_2 (Matheson standard gas, Fisher Scientific) and developing a simple % to area equation to calculate % gas in each sample. All readings were performed in duplicate at 23°C.

Color measurement
The International Commission on Illumination color parameter's $L*$, $a*$, and $b*$ values of stored mango powder were measured at different storage time intervals with a color meter (Minolta Chroma CR-200, Minolta Co., Osaka, Japan). The color meter was calibrated with a standard white ceramic plate ($L* = 95.98$, $a* = -0.13$, $b* = -0.30$). Prior to taking the readings, mango powder was poured into Petri dish to form a 10-mm thick layer and covered with transparent film (Saran™ Wrap, SC Johnson, Racine, WI). The average $L*$, $a*$, and $b*$ values were obtained from six readings taken from each of five locations of each sample. The hue angle, $H*$ and chroma, $C*$ expressed as $H* = \tan^{-1}\frac{b*}{a*}$ and $C* = \sqrt{a*^2 + b*^2}$, respectively, were calculated.

For color comparison with the original mango puree, 2.00 g each of the stored mango flakes with water content of 0.017 ± 0.001 kg water/kg dry solids were reconstituted by adding 12.10 g of distilled water. The reconstituted mango flakes had produced slurries with water content of 6.14 kg water/kg dry solids similar to the original mango puree. The reconstituted mango powder was stirred at 23°C using a vortex mixer (Mini Vortexer, Fisher Scientific) until the powder was completely dispersed. The $L*$, $a*$, and $b*$ values were immediately measured, and $H*$ and $C*$ were calculated following the same procedure used for mango flakes. The total change in color of the reconstituted mango powder with reference to the original puree was computed as: $\Delta E = \sqrt{(L_o^* - L^*)^2 + (a_o^* - a^*)^2 + (b_o^* - b^*)^2}$ where subscript "o" denotes the color of original puree.[13,20]

L-Ascorbic acid analysis
The L-ascorbic acid content of mango powder was determined by the 2,6-dichloroindophenol titrimetric method (AOAC Official Method 967.21) with modification. Filtered indophenol dye solution containing 50 mg of 2,6-dichlorophenolindophenol (Sigma-Aldrich, USA), standard ascorbic acid solution containing dissolved pure ascorbic acid in distilled water (1 mg/mL), and extracting solution (3% metaphosphoric acid (HPO_3) were prepared for the analysis. A calibration standard curve for L-ascorbic acid was established with six data points: 0.2, 0.4, 0.6, 0.8 mg/mL (ascorbic acid: HPO_3) and blank (HPO_3), and undiluted ascorbic acid were used as end points. A 2 mL solution placed in a 25-mL conical flask was titrated with indophenol solution until a faint pink color persisted for 30 s. The ascorbic acid concentration was expressed as milligram ascorbic acid equivalent to 1 mL of dye solution or milligram per milliliter. The L-ascorbic acid from mango powder was determined by blending 2.5 g of mango flakes with 25 mL extracting solution for 3 min at 23°C using a vortex mixer (Mini Vortexer, Fisher Scientific). The mixture was centrifuged at 3,000 g for 10 min at 4°C. Aliquots of the supernatant (2 mL) were transferred to 25-mL conical flasks and titrated rapidly with 2,6-dichlorophenolindophenol solution until a faint pink color persisted for 30 s. All measurements were performed thrice and the readings agreed within 0.1 mL deviation.

β-Carotene analysis
An extraction method used by Kaspar et al.[21] and Abonyi[22] was adopted in this study with modification. One gram of newly dried or stored mango powder (particle dimensions <2 mm) was first reconstituted with 2 mL of distilled water in a 50-mL centrifuge tube and mixed thoroughly. Then, 7.5 mL of acetone +0.1%

butylated hydroxytoluene (BHT) was added and vortexed (Mini Vortexer, Fisher Scientific). The resulting slurry was cold-saponified by adding 1.2 mL of 40% methanolic KOH solution (KOH:MeOH, w:v) and incubated in the dark for 16 h at 4°C. Following this overnight incubation, 7.5 mL of solvent mixture (hexane/ethyl acetate, (1:1, v/v) was added and vortexed for 1 min, followed by adding 10 mL of 10% Na_2SO_4 and vortexed for 1 min. The solution was allowed to stand in the dark for 1 h until the organic layer was separated. The upper organic layer was collected and transferred to a small tube and centrifuged at 600g for 8 min (DYNAC Centrifuge, Becton Dickinson, Sparks, MD). Two milliliters of supernatant were dried under nitrogen (Analytical Nitrogen Evaporator, Organomation Assoc. Inc., Berlin, MA) for analysis of carotenoids by high performance liquid chromatography (HPLC). All extraction procedures were performed under yellow light to minimize loss of carotenoids.

The dried extract was reconstituted in mobile phase (acetonitrile: methanol: chloroform, 47:47:6; v/v/v) and analyzed using HPLC. The HPLC system is consisted of a Waters 2690 separation module and a Waters 996 photodiode array detector (Waters Corporation, Milford, MA, USA). A C18, 5 μm × 150 mm column (Resolve, Waters Corporation), which was used to separate the β-carotene. The flow rate was 1 mL/min with an injection volume of 10 μL. Duplicate samples were analyzed for each treatment and the mean values were reported.

Glass transition temperature

The glass transition temperatures of RW-M powder with moisture content ranging from 0.074 to 0.097 kg water/kg mango powder were determined using differential scanning calorimetry (DSC) (DSC Q2000, TA Instruments, New Castle, DE, USA), as described by Syamaladevi et al.[23] The calorimeter was calibrated for heat flow and temperature using standard indium and sapphire. An empty aluminum pan was used as reference for each sample test. About 10 to 12 mg of the equilibrated mango powder was sealed in an aluminum pan (volume of 30 mL) and cooled from 25°C down to −90°C using liquid nitrogen and equilibrated for 10 min. The equilibrated samples were scanned to 70°C and then cooled down to 25°C. Scanning of all samples was performed using the same heating or cooling rate of 5°C/min.[23] To avoid condensation on the surface of the powder particles, a nitrogen carrier gas was purged at a flow rate of 50 mL/min. The onset- (T_{gi}), mid- (T_{gm}), and end-point (T_{ge}) values of the mango powder were determined by finding the vertical shift in the heat flow temperature diagram. All measurements were performed in duplicate.

Microstructures of mango powder

About 0.5 g of ground RW-M powder (180–250 μm) was obtained by passing the material through sieve sizes 60 and 80 (ASTM),[18] mounting on aluminum stubs, followed by coating with a fine layer of gold (15 nm) using a sputter gold coater (Technics Hummer V, Anatech, San Jose, CA, USA). The gold-coated samples were examined by a Quanta 200 F Environmental Scanning Electron Microscope (FEI, Field Emission Instruments, Hillsboro, OR, USA) at low vacuum mode (200 Pa). Observations were made at accelerated voltage of 30 kV and magnification of 100 × and 500 × at a scale of 1 mm and 200 μm, respectively.

Statistical analysis

All experiments were performed in duplicate or triplicate, and the results were analyzed in a full factorial design using SAS (SAS Institute Inc., Cary, NC) general linear model procedure, and the means were separated and compared by Tukey-honest significant difference test with a confidence interval of 95%.

Results and discussion

Moisture content and water activity

Table 1 shows the changes in the measured moisture content and water activity (a_w) of RW-M powder stored in air- and nitrogen-flushed packages after 6 and 12 months at different temperatures. Prior to storage, the water content and a_w values of the powder were 0.037 ± 0.001 kg water/kg dry solids and 0.126 ± 0.003, respectively. Regardless of packaging atmosphere, we observed no significant ($p < 0.05$) increase in water content of mango powder stored at 5 and 22°C between 6 and 12 months, but a significant increase ($p < 0.05$) was observed for the product stored at 45°C. The nitrogen-flushed mango powder stored at ambient temperature had the lowest percentage increase after 6 months (1.4%) and 12 months (2.1%), while air-packaged mango powder stored at 45°C showed the highest water gain at 17.5 and 22.6% after 6 and 12 months of storage, respectively. It is possible that the exposure of the amorphous powder to high temperature (45°C) for the longer storage period promoted crystallization and the pouch moisture barrier property was also affected somewhat—and those conditions subsequently caused the observed increases in moisture content. Al Mahdi et al.[24] reported that the release of water in stored dried skim milk was accelerated during crystallization process resulting in caking caused by

Table 1. Moisture content and water activity of mango powder after 6 and 12 months of storage at different packaging conditions and temperatures.

		6 months			12 months		
		5°C	22°C	45°C	5°C	22°C	45°C
Parameter	Initial value	Air packaging					
Moisture content (kg water/kg dry solids)	0.037 ± 0.006^a	$0.041 \pm 0.000^{a,c,f}$	$0.040 \pm 0.002^{a,c}$	$0.046 \pm 0.000^{b,c,g}$	$0.041 \pm 0.001^{a,c,f}$	$0.041 \pm 0.000^{a,c,f}$	$0.049 \pm 0.001^{b,g}$
Water activity	0.126 ± 0.003^a	0.154 ± 0.000^b	0.159 ± 0.000^b	0.190 ± 0.000^d	0.243 ± 0.001^c	0.241 ± 0.004^c	0.290 ± 0.003^e
		Nitrogen flushing					
Moisture content (kg water/kg dry solids)	0.037 ± 0.006^a	$0.039 \pm 0.002^{a,d}$	$0.038 \pm 0.002^{a,d}$	$0.043 \pm 0.001^{c-e,h}$	$0.039 \pm 0.000^{a,d}$	$0.039 \pm 0.001^{a,e}$	$0.045 \pm 0.001^{f-h}$
Water activity	0.126 ± 0.003^a	0.157 ± 0.000^d	0.155 ± 0.000^b	0.171 ± 0.000^h	$0.226 \pm 0.005^{f,g}$	$0.234 \pm 0.003^{c,g}$	0.279 ± 0.003^i

$^{a-i}$Means with the same superscript letters or numbers within a row across 6 and 12 months of storage indicate no significant differences ($p < 0.05$). Means were obtained from three replicates.

interparticle liquid bridges. The water activity values for all the samples tested did not exceed 0.3.

Headspace gas analysis

The initial concentration of oxygen and nitrogen inside the air-packaged mango powder was 19.8 and 80.2% by volume, respectively (Table 2). The oxygen level in the headspace of the air-packaged mango powder significantly decreased ($p < 0.05$) over the first 6 months of storage at 5, 22, and 45°C. However, the head space oxygen concentration of the samples at 5 and 22°C was the same between 6 and 12 months of storage, but further decreased beyond 6 months when stored at 45°C. On the other hand, the oxygen concentration significantly increased ($p < 0.05$) when the packaged mango powder was stored for 6 and 12 months at 5, 22, and 45°C using nitrogen-flushed packaging. But the level of oxygen for all samples (<4% by volume) was still in the range that would justify measuring its effect on the physical and chemical stability of mango powder during storage. In a study on the effect of nitrogen flushing and storage temperature of whole milk, Lloyd et al.[25] discarded nitrogen-flushed samples when the oxygen concentration inside the package exceeded 6.5% by volume.

Ambient air has a carbon dioxide (CO_2) concentration of 0.04% by volume. But 6.1–2.8% CO_2 by volume was detected inside the air and nitrogen-flush

packaging after 6 months of storage at 45°C, respectively. One possible reason for this was the presence of citric acid, which is the major organic acid found in mango. It is likely that during storage at higher temperature, the acetyl CoA molecule (the fuel for the citric acid cycle) was formed from the breakdown of starch and then completely oxidized to CO_2.[26]

Extending the storage time of air-packaged samples to 12 months at 45°C further reduced the level of oxygen to 12.8%, while the amount of CO_2 present in air and nitrogen-flush packaging further increased to 10.9 and 4.1% by volume, respectively. The replacement of air by nitrogen raised the initial headspace nitrogen level from 80.2 to 97.7% by volume. No carbon dioxide was traced in nitrogen-flushed mango powder at 5 and 22°C up to 12 months of storage, but it was detected at 45°C with a lower concentration compared to air packaging. It is apparent that regardless of packaging atmosphere, carbon dioxide evolved to a certain level at 45°C but was completely absent at 5 and 22°C.

Color stability

The RW-dried mango powder in air packaging and nitrogen-flushed bags showed no color change over 12 months storage at 5 and 22°C. But the powder stored at 45°C turned dark brown (Fig. 1a). The lightness (L^*) values of air-packaged and nitrogen-flushed mango

Table 2. Headspace gas concentration of RW-dried mango flakes after 6 and 12 months of storage, percentage by volume.

		Air packaging					
		6 months			12 months		
Components	Initial value	5°C	22°C	45°C	5°C	22°C	45°C
CO_2	0^a	0^a	0^a	6.1 ± 0.1^b	0^a	0^a	10.9 ± 0.08^c
O_2	19.8 ± 0.1^a	$19.0 \pm 0.2^{b-e}$	$18.9 \pm 0.1^{d,e}$	14.4 ± 0.1^f	$19.4 \pm 0.1^{c,e}$	19.0 ± 0.2^e	12.8 ± 0.0^g
N_2	80.2 ± 0.1^a	$81.0 \pm 0.2^{b,c}$	81.1 ± 0.1^c	79.6 ± 0.1^d	$80.6 \pm 0.2^{a,b}$	$81.0 \pm 0.1^{c,b}$	76.3 ± 0.0^e
		Nitrogen flushing					
CO_2	0^1	0^1	0^1	2.8 ± 0.0^2	0^1	0^1	4.1 ± 0.0^3
O_2	2.4 ± 0.0^1	2.7 ± 0.0^2	2.8 ± 0.0^2	2.7 ± 0.0^2	3.7 ± 0.2^3	3.6 ± 0.0^3	3.7 ± 0.0^3
N_2	97.7 ± 0.0^1	97.3 ± 0.0^2	97.2 ± 0.0^2	94.6 ± 0.0^4	96.3 ± 0.2^3	96.4 ± 0.0^3	92.2 ± 0.0^5

$^{a-f}$ and $^{1-5}$Means with the same superscript letters or numbers within a row across 6 and 12 months of storage indicate no significant differences ($p < 0.05$). Means were obtained from three replicates.
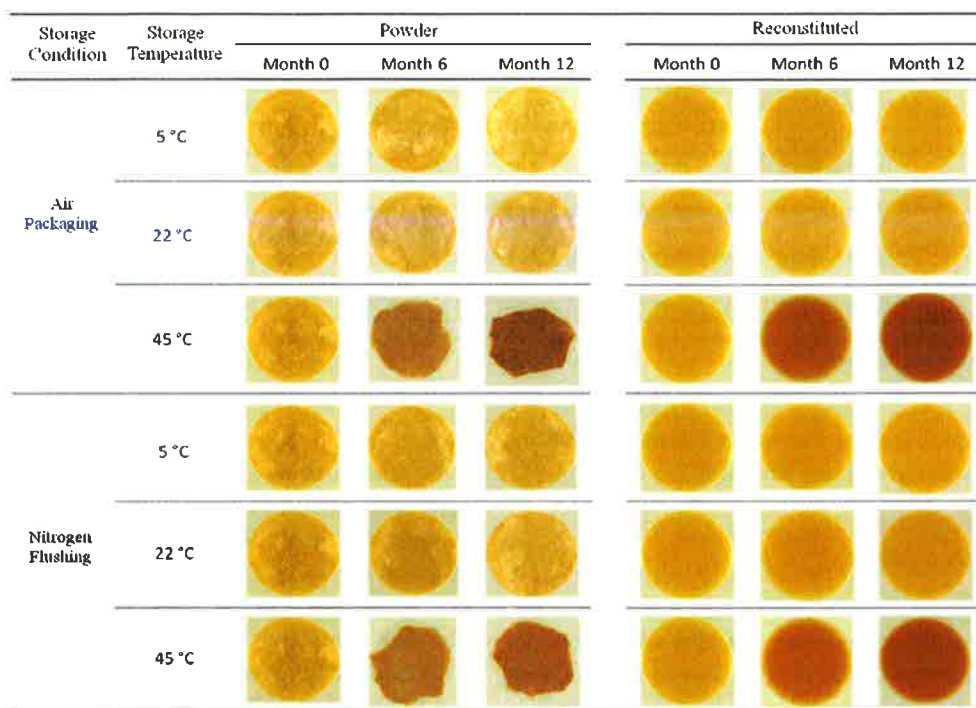
Figure 1. Photographs of RW-dried mango powder (a) and reconstituted mango puree (b) after 6 and 12 months of storage at different packaging atmospheres.

powder remained stable over 12 months of storage at 5 and 22°C (Table 3), confirming the visual appearance shown in Fig. 1a. The L^*-value was significantly reduced ($p < 0.05$) when mango powder was stored at 45°C, beginning at 6 months and further darkened after 12 months of storage. The degree of redness and greenness (a^*) values obtained in both air- and nitrogen-flushed packaging during the 12 months of storage increased while the corresponding L^*-values decreased. This can be explained by the higher oxidation of β-carotene, which affected the natural yellow color of mango powder turning it to brown. Similar relationship was

found between the a^*- and L^*-values for freeze-dried encapsulated β-carotene during storage.[27]

The darkening or formation of brown color as indicated by reduced L^*-value of mango powder stored at 45°C can be attributed to nonenzymatic browning (NEB). Sagar et al.[28] reported significantly higher NEB of air-dried mango powder stored at 35°C compared to mango powder stored at 7°C. Browning of vacuum-dried mango powder packaged in polyester poly (PP) and metalized polyester poly during 6 months of storage was associated with ascorbic acid degradation,[3] which was higher in PP; and this AA

Table 3. Hunter color parameters of mango powder stored in air or nitrogen atmosphere at 5, 22, and 45°C and evaluated after 6 and 12 months.

| | | Air packaging | | | | | |
| | | 6 months | | | 12 months | | |
Hunter color parameters	Initial value	5°C	22°C	45°C	5°C	22°C	45°C
L	63.7 ± 0.9^a	64.1 ± 0.1^a	64.3 ± 0.2^a	48.9 ± 0.1^c	67.4 ± 0.6^b	65.1 ± 0.11^a	40.2 ± 0.1^d
a^*	6.5 ± 0.2^a	4.8 ± 0.1^b	5.1 ± 0.1^b	6.7 ± 0.1^a	5.8 ± 0.3^c	6.0 ± 0.2^c	9.1 ± 0.3^d
b^*	32.9 ± 1.0^a	28.8 ± 0.3^b	28.3 ± 0.1^b	19.5 ± 0.6^c	32.2 ± 0.4^a	28.8 ± 0.5^b	22.0 ± 0.2^c
Chroma	$33.6 \pm 1.0^{a,e,f,h}$	$29.2 \pm 0.4^{b,j}$	$28.7 \pm 0.5^{b,j}$	23.0 ± 0.2^c	$32.7 \pm 0.4^{a,f}$	$29.4 \pm 0.1^{b,f,j}$	21.5 ± 0.7^c
Hue	78.7 ± 0.2^a	80.6 ± 0.1^b	79.8 ± 0.1^b	73.0 ± 0.1^c	79.7 ± 0.6^b	78.3 ± 0.3^a	65.0 ± 0.4^d
Components		Nitrogen flushing					
L	63.7 ± 0.9^a	63.6 ± 0.1^a	62.5 ± 0.1^a	49.2 ± 0.6^e	64.3 ± 0.7^a	64.5 ± 0.6^a	42.7 ± 0.8^f
a^*	6.5 ± 0.2^a	$6.3 \pm 0.1^{a,c}$	$6.3 \pm 0.1^{a,c}$	8.3 ± 0.1^f	7.5 ± 0.3^e	7.4 ± 0.2^e	10.6 ± 0.2^g
b^*	32.9 ± 1.0^a	35.6 ± 0.2^e	31.3 ± 1.3^a	24.7 ± 0.3^f	35.9 ± 0.8^e	$34.3 \pm 1.4^{a,e}$	25.2 ± 1.0^f
Chroma	33.6 ± 1.0^a	$36.2 \pm 0.2^{d,g}$	$31.9 \pm 1.4^{f,h}$	$26.0 \pm 1.0^{i,k}$	$35.9 \pm 0.8^{a,d}$	$35.1 \pm 1.3^{a,g}$	$27.4 \pm 0.2^{j,k}$
Hue	78.7 ± 0.2^a	80.0 ± 0.1^b	78.7 ± 0.4^a	71.4 ± 0.4^e	78.3 ± 0.2^a	77.9 ± 0.7^a	67.2 ± 0.3^f

[a–k]Means with the same superscript letters within a row from initial color values and after 6 and 12 months of storage indicate no significant differences ($p < 0.05$). Means were obtained from six readings taken from each of five locations per sample.

degradation was also the major mechanism causing browning in lemon juice.[29] Mango contains free amino acids that may react with sugars and cause the observed NEB. The RW-M powder stored at ambient or low temperatures for 12 months showed negligible color change and is therefore a good quality indicator.

The b^* value is a good indicator to distinguish differences in yellowness of mango powder as affected by storage condition. The b^* value of air-packaged mango powder was significantly different after storage for 6 months—being lower compared to the initial b^* values, and the b^* value of samples stored at 45°C dropped significantly ($p < 0.05$) as indicated by the darker color (Table 3). The b^* value of nitrogen-flushed mango powder stored at different temperatures was significantly greater than air packaging ($p \leq 0.05$). It is interesting to note that nitrogen flushing preserved the yellowness of mango powder stored at ambient temperature after 6 and 12 months, while powder color was significantly altered when stored at 5 and 45°C ($p \leq 0.05$). Also, the vividness in yellow color represented by chroma values of all stored samples significantly changed ($p \leq 0.05$) after 6 months of storage at different temperatures in both packaging atmospheres, except for samples stored under nitrogen at 22°C.

When the stored mango powder was reconstituted to the same water content as the original puree, their visual color appearance followed a similar trend found in the powdered form. Slight changes were observed after 6 and 12 months except in samples stored at 45°C, which turned brown both for air-packaged and nitrogen-flushed one (Fig. 1b). Apparently, regardless of storage temperature and packaging atmosphere, the L^*, a^*, b^* values of mango powder were significantly different among each other with the exceptions of samples stored with nitrogen flushing at 22°C, where yellowness was unchanged after 12 months (Table 4). Likewise, the yellow vividness (chroma) of reconstituted mango powder stored at 22°C revealed no significant difference ($p < 0.05$) from the original puree after 12 months of storage. Moreover, the hue angle of the same samples did not change after 6 months of storage at 22°C in both packaging. Interestingly, color change was more pronounced under cold storage than at ambient temperatures, an observation which needs further investigation. Discoloration of egg yolk, which contains similar yellow pigments, was more pronounced under refrigerated storage than at warmer temperature because the pH was higher at 4°C but did not change between 21 and 24°C.[30] The lower hue angle obtained in mango powder stored at 45°C explains the dull color.

The total deviation (ΔE) in color of reconstituted mango powder at different temperatures with respect to the original puree ranged from 1.87 ± 0.09 to 16.02 ± 0.10 in nitrogen-flushed and 5.93 ± 0.12 to 20.82 ± 0.09 in air-packaged samples, respectively (Table 4). The lowest deviation in color of reconstituted mango powder was detected in nitrogen-flushed samples stored at 22°C ($\Delta E = 1.87 \pm 0.09$), while the highest values were observed in air-packaged samples stored at 45°C having a ΔE value of 20.82 ± 0.09. In this study, it can be concluded in general that the color of mango powder and its reconstituted form can be well preserved with the aid of nitrogen flushing during storage for up to 12 months.

L-Ascorbic acid

The ascorbic acid content of mango powder obtained from RW drying decreased significantly ($p < 0.05$) over 12 months storage regardless of the packaging atmosphere (Fig. 2), and the percent loss of AA

Table 4. Hunter color measurements of reconstituted mango powder stored in air or nitrogen atmosphere at 5, 22, and 45°C and evaluated after 6 and 12 months.

Components	Initial value	Air packaging					
		6 months			12 months		
		5°C	22°C	45°C	5°C	22°C	45°C
L	44.0 ± 0.0ᵃ	46.9 ± 0.1ᵇ	45.9 ± 0.2ᶜ	38.7 ± 0.1ᵉ	45.9 ± 0.1ᶜ	45.2 ± 0.1ᵈ	28.1 ± 0.1ʲ
a*	4.2 ± 0.0ᵃ	2.5 ± 0.1ᵇ	3.5 ± 0.1ᵈ	5.8 ± 0.0ᶠ	2.7 ± 0.1ᶜ	3.72 ± 0.0ᵉ	10.2 ± 0.0ᵍ
b*	39.5 ± 0.2ᵃ	34.7 ± 0.1ᵇ·ᶜ	33.7 ± 0.2ᵈ	27.5 ± 0.1ᶠ	34.0 ± 0.2ᶜ·ᵈ	31.04 ± 0.1ᵉ	26.9 ± 0.1ᶠ
Chroma	39.8 ± 0.2ᵃ	34.8 ± 0.1ᵇ	33.9 ± 0.2ᵇ	29.3 ± 0.1ᵈ	34.1 ± 0.2ᵇ	32.7 ± 0.8ᶜ	27.5 ± 0.2ᵉ
Hue angle	83.9 ± 0.1ᵃ	85.9 ± 0.1ᵇ	77.9 ± 0.1ᵃ	77.9 ± 0.1ᵉ	85.4 ± 0.2ᶜ	83.5 ± 0.0ᵈ	69.7 ± 0.1ᶠ
ΔE	0.0ᵃ	5.9 ± 0.1ᵇ	6.2 ± 0.1ᵇ	13.8 ± 0.1ᵈ	6.0 ± 0.2ᵇ	7.2 ± 0.1ᶜ	20.8 ± 0.1ᵉ
		Nitrogen flushing					
L	44.0 ± 0.0ᵃ	45.7 ± 0.2ᶜ	46.2 ± 0.0ʰ	39.4 ± 0.0ʲ	44.5 ± 0.19ᵍ·ⁱ	44.39 ± 0.11ⁱ	30.6 ± 0.1ᵏ
a*	4.2 ± 0.0ᵃ	4.7 ± 0.1ʰ	4.4 ± 0.0ᵃ	6.8 ± 0.2ʲ	6.0 ± 0.2ʲ	5.60 ± 0.04ᶠ	11.5 ± 0.0ᵏ
b*	39.5 ± 0.2ᵃ	39.2 ± 0.2ᵍ	41.7 ± 0.1ⁱ	32.5 ± 0.5ʲ	38.3 ± 0.8ʰ	40.52 ± 0.42ᵃ	31.0 ± 0.3ᵇ⁻ᵈ
Chroma	39.8 ± 0.2ᵃ	38.6 ± 0.2ᶠ	40.8 ± 0.1ʰ	33.9 ± 0.5ⁱ	39.7 ± 0.8ᵍ	42.11 ± 0.41ᵃ	32.7 ± 0.3ʲ
Hue angle	83.9 ± 0.1ᵃ	83.0 ± 0.1ᵍ	83.9 ± 0.0ᵃ	77.6 ± 0.3ᵉ	82.4 ± 0.1ʰ	81.31 ± 0.12ⁱ	71.6 ± 0.1ʲ
ΔE	0.00ᵃ	2.2 ± 0.1ᶠ⁻ʰ	1.9 ± 0.1ᵍ	10.0 ± 0.4ⁱ	2.7 ± 0.6ᶠ·ʰ	2.45 ± 0.06ʰ	16.0 ± 0.1ʲ

ᵃ⁻ᵏMeans with the same superscript letters within a row from initial color values and after 6 and 12 months of storage indicate no significant differences ($p < 0.05$). Means were obtained from six readings taken from each of five locations per sample.
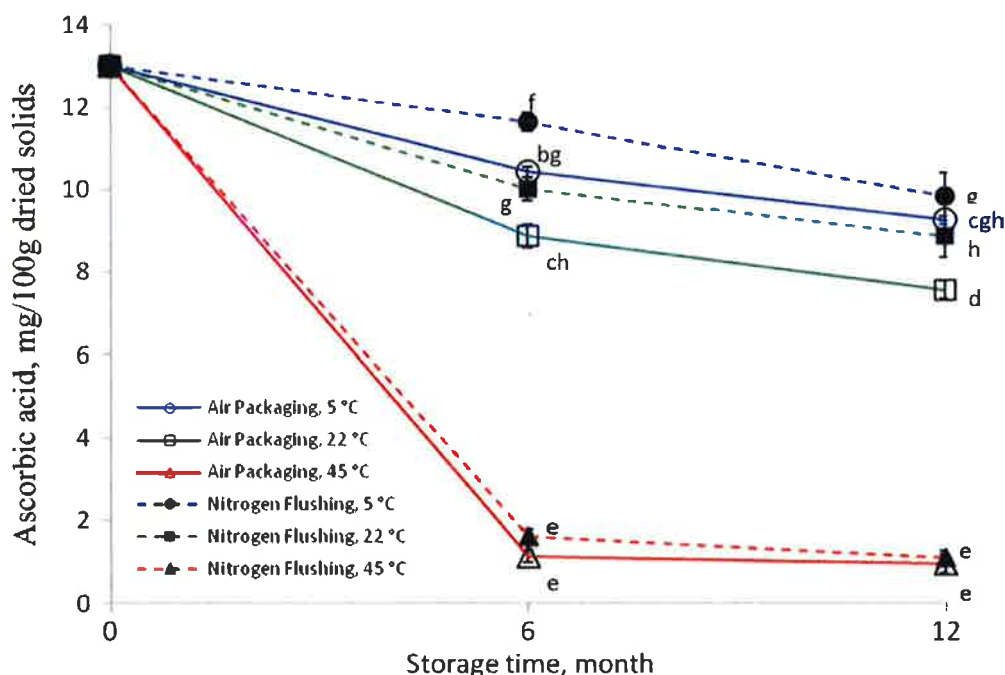
Figure 2. Influence of storage time, temperature, and packaging atmosphere on the ascorbic acid content of RW-dried mango powder. Different letters a–h indicate significant differences ($p < 0.05$).

also increased as storage temperature increased. At storage temperature of 45°C, ascorbic acid content of RW-M powder was reduced from 13 ± 0.2 mg/100 g dried solids at time 0 to 1.1 ± 0.1 mg/100 g dried solids or 91.3% loss and 1.6 ± 0.2 mg/100 g dried solids or 87.5% loss after 6 months of storage in air-packaging and nitrogen-flushed packages, respectively ($p < 0.05$). This result indicates that higher storage temperature affects AA retention in mango powder more than the packaging atmosphere. Several kinetic studies confirmed that ascorbic acid is a very heat-sensitive compound and its destruction increases as the temperature of products in dried or aqueous forms increases.[31–34] The slight increase in ascorbic acid degradation (91.4–92.6%) observed after 12 months of storage in both packaging atmospheres could also be explained by the small amount of AA left after 6 months of storage. The effect of packaging atmosphere on the loss of ascorbic acid after 12 months of storage at 45°C yielded no significant difference ($p < 0.05$).

The combined use of 5°C storage with nitrogen flushing led to the lowest proportional AA loss (10.5%) among all treatments over the period of 12 months. The loss of ascorbic acid in nitrogen-flushed mango powder at 22°C increased slightly (10.5–23.0%) but was significantly lower compared to air-packaged powder (31.7%) during 6 months of storage ($p < 0.05$). AA loss of mango powder further increased to 31.8% in

nitrogen-flushed packages and 41.7% in air package at the end of 12 months of storage.

The 31.8% loss of ascorbic acid in RW-dried mango powder stored in nitrogen-flushed packaging for 12 months at 22°C was about one-third lower when compared with 42.3% observed in a previous study on freeze-dried mango "Badami" powder packed in cans under nitrogen and stored at 24–28°C.[35] Also, the AA loss of RW-M powder in both air packaging and nitrogen flushing was less than one half observed with vacuum-dried[3] mango powder packed in a high oxygen permeable packaging material (82–83%) stored at 27–32°C. By considering freeze-dried mango powder as the standard for a quality product, we can conclude that AA loss noticed with RW-M powder was not due to the drying process but is dependent on the effect of storage conditions and the packaging material used. However, it was observed that RW-M powder is well preserved compared to freeze-dried and vacuum-dried mango powder. The variations may be attributed to variety, drying preparation, and packaging methods. This is a good indication that the physicochemical stability and quality attributes of RW-M powder during storage are superior compared to that of mango powder produced by the widely accepted standard process of freeze drying.

The loss of ascorbic acid in nitrogen-flushed packaging was found to be significantly ($p < 0.05$) lower than air-packaged mango powder stored at 5 and 22°C over

a 12-month period, indicating that the presence of oxygen in the headspace of package at 22°C and under 5°C greatly affects the degradation of ascorbic acid. Kirk et al.[31] reported that the destruction of ascorbic acid in a model dehydrated food system was dependent on oxygen and moisture contents. Dennison and Kirk[36] also made the conclusion that dissolved and gaseous oxygen are the primary factors that affect the storage of dehydrated food. Although investigating the effect of nitrogen flushing on ascorbic acid retention in orange juice during storage at 4, 22, and 37°C, Zhang et al.[37] found no significant difference ($p > 0.05$) between air and nitrogen in the headspace, suggesting that nitrogen flushing is not effective for orange juice. The type of packaging and different interaction mechanisms of headspace gas with juice and powder materials may explain these observed differences.

In general, the replacement of air inside the multi-layer package with nitrogen extended the shelf-life of mango powder kept at 22°C and below resulted in ~90% retention of ascorbic acid content after 12 months of storage. It is also apparent from the present study that temperature and time are the most critical factors that affect ascorbic acid degradation. This observation is in agreement with Trammell et al.[38] who reported that package headspace gas composition is not sufficient to prevent ascorbic acid degradation unless accompanied by lowering the storage temperature.

β-Carotene

The β-carotene content of the mango powder obtained from the RW drying process significantly decreased during 12 months storage regardless of the packaging atmosphere and temperature (Fig. 3). The high temperature (45°C) treatment severely degraded the β-carotene in mango powder in air packaging resulting in 82.4 and 98.5% loss after 6 and 12 months of storage, respectively. It was reported that the possible cause of carotenoid degradation in dehydrated products is due to an increase in porosity and surface area.[3] Also, loss of β-carotene can generally be associated with auto-oxidation, wherein the high susceptibility of unsaturated chemical structure in the product contributes to thermal degradation and oxidation.[39] The decrease in β-carotene can also be attributed to the formation of cis-isomers due to degradation of all-trans β-carotene.[27,40,41] Another factor in our study that might have increased the degradation of β-carotene is the low water activity ($a_w < 0.3$) of the mango powder regardless of storage temperature and duration (Table 1). Lavelli et al.[42] found a U-shaped response curve when β-carotene loss was plotted against water activity of freeze-dried carrots stored at 40°C. The loss of β-carotene was minimum at water activity ranging between 0.34 and 0.54 while the degradation increased outside those bounds. Likewise, Arya et al.[43] also
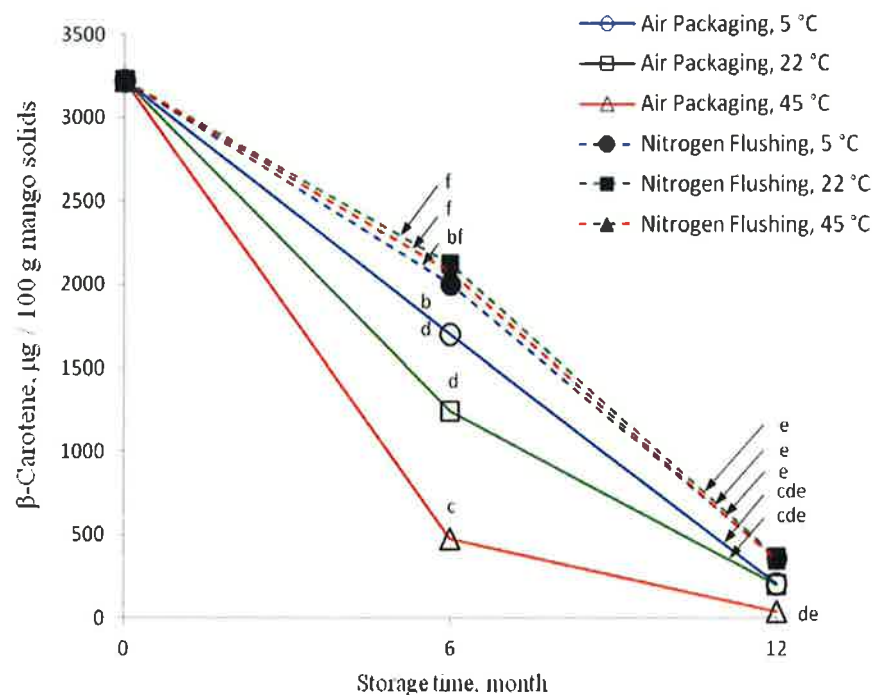

Figure 3. Influence of storage time, temperature, and packaging atmosphere on the β-carotene content of RW-dried mango powder. Different letters a–f indicate significant differences ($p < 0.05$).

found that β-carotene in dehydrated carrots was more stable at a_w in the range of 0.32–0.57.

The replacement of air in nitrogen-flushed mango powder packages yielded significantly lower loss of β-carotene with 26.07, 21.8, and 23.38% at 5, 22, and 45°C, ($p < 0.05$) after 6 months of storage, respectively. It can be noted that β-carotene losses at different temperatures are not significantly different, indicating the positive effect of nitrogen flushing in maintaining the stability of β-carotene even at a higher temperature. These findings are supported by Talcott and Howard[44] who observed less degradation of β-carotene in processed carrots when treated with nitrogen compared to control samples and those treated with oxygen.

However, there was considerable decline of β-carotene content in RW-M powder after 12 months of storage with nitrogen flush with losses ranging from 86.5 to 87.1% for all temperatures applied, and no significant difference was found compared to samples stored in air packaging. The increase in water content measured inside the package after 12 months of storage ranging from 7 to 22.6% in air packaging and 2.1 to 15.6% in nitrogen flushing (Table 1) may also have contributed to the degradation of β-carotene at longer storage periods. This

finding indicates that the application of nitrogen flushing of RW-M powder is not useful to prevent degradation of β-carotene after 12 months of storage.

Caking and glass transition temperature

One major observation in this study was the caking that affected the quality of mango powder after storage at high temperature. This caking is highly associated with glass transition temperature which affects product stability. In our previous study,[45] the onset glass transition temperature (T_{gi}) for RW-M powder containing 0.03 ± 0.001 kg water/kg dry solids was 30.6 ± 1.5°C. In the present study, it was determined that mango powder stored at 5 and 22°C (below T_g) were well preserved and free from structural transformations, while those stored at 45°C ($>T_g$) suffered caking, resulting in a hard and brittle assemblage of powder particles (Fig. 1a).

Caking normally occurs when amorphous sugar initially in the form of a rigid glass-like substance is transformed into a rubbery state.[46] It has been reported that physical and chemical stabilities of amorphous food powder are well protected when the product is stored at temperature below T_g, while deteriorative reactions such as stickiness and caking
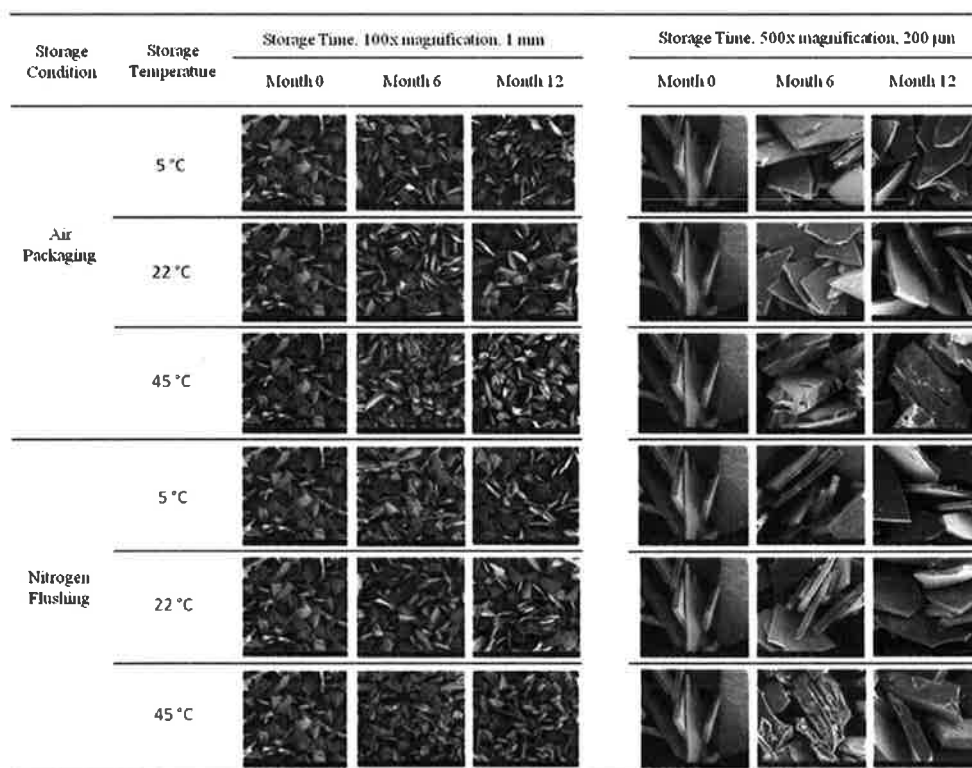

Figure 4. Field emission scanning electron micrographs of Refractance Window® (180–250 µm) stored for 1 year at different storage conditions (magnification of 100× and 500×, 30 kV).

occur at temperature above T_g.[47–49] It has also been shown that sugar-rich food powder become sticky at temperature range ($T–T_g$) between 10 and 20°C during storage.[50] The sticky point temperature of mango powder containing 0 and 5% water content (db) was 11.5 and 15.5°C ($T–T_g$), respectively. This was consistent with our study of RW-M powder stored at 45°C, which has a ($T–T_g$) value of 14.4°C.

Microstructures

The particle size of RW-M powder prior to and during storage was in the range of 1.7–2.0 mm. For the scanning electron microscopic study, all the samples were ground into smaller particles (180–250 µm) for better interpretation of the images. Initially, the microstructures of the powder under different storage conditions (Fig. 4) were smooth and flaky with uniform thickness, nearly the same as reported by Caparino et al.[45] The powder samples clearly show that their particles were unagglomerated. The two sides of every single particle were smooth indicating more flowability and less susceptibility to oxidation because of lesser surface area. These particle characteristics of RW-M powder or flakes were achieved with the aid of a spreader bar provided in the RW dryer, coupled with a controlled mechanism that allows uniform feeding of mango puree from the inlet section and as it moves toward the other end of the dryer.

The initial structure of mango powder was well preserved after 6 and 12 months of storage at 5 and 22°C in both air-packaging and nitrogen-flushing powder (Fig. 4). This suggests that RW-M powder remains flowable and is very stable during storage at ambient and lower temperatures with or without modification of the atmosphere inside the package. From economic point of view, the powder can be well protected just by air sealing the package and storing at ambient temperature, eliminating the need for refrigeration. The microscopic examination showed that mango powder stored at 45°C for 6 and 12 months was severely degraded regardless of packaging protocol used (Fig. 4). The degradation was excessive for air-packaging than in nitrogen-flushed samples. Figure 1a shows clearly that the original unagglomerated particles were strongly bonded together, forming large assemblages or lumps of particles after 6 and 12 months of storage. The flaky structure of the powder particles coupled with its fine and smooth surface characteristics may have contributed to greater interparticle forces that allowed formation of strong adhering particles. Likewise, these product lump formations can be associated with partial melting during storage, which enhances surface contact between particles through diffusion of molecules from one particle to another as affected by the elevated temperature.

Conclusion

The physical and nutritional quality changes in RW-M powder in air and nitrogen-flushed packages after 6 and 12 months of storage at 5, 22, and 45°C were determined. Regardless of the packaging atmosphere, the water content of mango powder was least ($p < 0.05$) affected except for those stored at 45°C where the increase was significant ($p < 0.05$). Reduction in headspace oxygen concentration for powder stored for 6 and 12 months at 45°C was also significant ($p < 0.05$). Storage at lower temperatures (5 and 22°C) did not affect the color of RW-M powder in either type of packaging, but powder darkening occurred at 45°C. L-Ascorbic acid also diminished greatly when the RW-M powder was stored at 45°C over 12 months. Replacing the headspace air with nitrogen preserved the color of reconstituted mango powder and improved the retention of AA by 68.2–89.5% for mango powder kept at 22 and 5°C for 6 and 12 months, respectively. The β-carotene in air-packaged mango powder was also severely degraded at 45°C (after 6 months), nitrogen flushing significantly ($p < 0.05$) reduced the loss by 21.8–26.1%. Extending the storage to 12 months increased β-carotene degradation (86.5–87.1%), regardless of headspace gas composition and storage temperature. Physical characteristics of mango powder stored below the glass transition temperature were well preserved, and free from structural transformations, while the powder kept at 45°C ($>T_g$) suffered intense caking, and became hard and brittle regardless of the packaging technique applied. These results will serve as a good reference in the production and storage of mango powder, particularly those obtained by the Refractance Window® drying process.

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