Mango *(Mangifera indica L.)* Lyophilization Using Vacuum-Induced Freezing

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Abstract—Lyophilization, also called freeze-drying, is an important dehydration technique mainly used for pharmaceuticals. Food industry also uses lyophilization when it is important to retain most of the nutritional quality, taste, shape and size of dried products and to extend their shelf life. Vacuum-Induced during freezing cycle (VI) has been used in order to control ice nucleation and, consequently, to reduce the time of primary drying cycle of pharmaceuticals preserving quality properties of the final product. This procedure has not been applied in freeze drying of foods. The present work aims to investigate the effect of VI on the lyophilization drying time, final moisture content, density and reconstitutional properties of mango (*Mangifera indica* L.) slices (MS) and mango pulp-maltodextrin dispersions (MPM) (30% concentration of total solids). Control samples were run at each freezing rate without using induced vacuum. The lyophilization endpoint was the same for all treatments (constant difference between capacitance and Pirani vacuum gauges). From the experimental results it can be concluded that at the high freezing rate (0.4°C/min) reduced the overall process time up to 30% comparing process time required for the control and VI of the lower freeze rate (0.1°C/min) without affecting the quality characteristics of the dried product, which yields a reduction in costs and energy consumption for MS and MPM freeze drying. Controls and samples treated with VI at freezing rate of 0.4°C/min in MS showed similar results in moisture and density parameters. Furthermore, results from MPM dispersion showed favorable values when VI was applied because dried product with low moisture content and low density was obtained at shorter process time compared with the control. There were not found significant differences between reconstitutional properties (rehydration for MS and solubility for MPM) of freeze dried mango resulting from controls, and VI treatments.

Keywords—Drying time, lyophilization, mango, vacuum induced freezing.

I. Introduction

FRUITS are highly perishable products mainly because their water and soluble solids content promotes the microbiological growth in post-harvest handing, storage and distribution [1].

Dehydrated products have become an alternative to increase the shelf life, allowing better commercialization of fruits in international markets. However, the quality losses related to the structural changes generated during fruits conventional drying process has driven the search for milder dehydration techniques. Freeze-drying or lyophilization minimizes structural changes, preserves bioactives and flavors of dried product and becomes an attractive alternative for the design of new fruit value-added derivatives [2].

Lyophilization, a drying process based in water sublimation, usually has three steps: product freezing, primary drying or removing of the free-frozen water by direct sublimation under reduced pressure, and the secondary drying in which the unfrozen water is released by desorption and diffusion [3].

During ice formation can be differentiated three stages: 1) Nucleation or initial crystal seed formation. 2) Propagation or growing of ice crystals and 3) Recrystallization or maturity [4]. Water crystallization is affected by the conditions of the freezing which in turn defines the structure of the frozen sample and its behavior throughout freeze-drying. Despite of this, freezing step is frequently not properly controlled in the research or industrial food lyophilization practice, and there are a lack of studies regarding the effect of the freezing variables on the lyophilization and the final quality of the product.

Kramer et al. [5] studied the reduction of the absolute chamber pressure (Vacuum-Induced during freezing cycle VI) during the freezing stage of solutions (manitol, sucrose and glycerin) as a suitable technique for the reduction of the primary dying time. Liu et al. [6] and Oddone et al. [7], who used VI and annealing during freezing, also found significant effects on the primary step rate and the freeze-dried product quality.

The aim of this work was to investigate the effect of VI and freezing rate in the freezing of fresh fruit and encapsulated fruit pulp on the lyophilization drying time, final moisture content, apparent density and structural properties of mango (*Mangifera indica* L.) slices and mango pulp-maltodextrin dispersions (30% concentration of total solids).

II. Materials and Methods

A. Sample Preparation

Fresh mango, Tommy Atkins variety, was purchased from a local market in Manizales, Colombia. The fresh fruits were selected according the color, and apparent hardness. After cleaning and sanitization they were cut in slices (MS) (ca. 6.5 g, 0.006 m height and 0.040 m diameter).

For the preparation of mango pulp-maltodextrin dispersions (MPM), the selection of the fruit was carried out at the same conditions for fresh mango. The dispersion was prepared with fresh mango pulp and maltodextrin (DE 20, MOR-REX® 1920, Corn Products (Brazil)) in order to reach 30% concentration of total solids in the encapsulated pulp samples

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to be used in freeze drying assays.

B. Vacuum Induced Freezing Procedure

Freeze drying process was carried out in a Virtis Pilot Lyophilizer (Genesis SQ XL-70). Sample freezing was carried out at three freezing rates (0.1, 0.25 and 0.4°C/min). VI was applied when shelf temperature reached -2°C using three different final pressures (500, 700 and 900 mTorr). The procedure was carried out according to [5] with some modification and the test conditions are summarized in Table I.

MS and MPM samples were placed on shelves at 20°C and their temperature was lowered to -2°C. At this instant the chamber pressure controller was set to the vacuum fixed value (500, 700 or 900 mTorr), and the condenser was then cooled to -60°C while the valve between chamber and condenser remained open. After the final chamber pressure set was reached, the vacuum was released as rapidly as possible (<3 min). Subsequently, the shelf temperature, was programmed to decreased until -40°C (at the freezing rate of the assay) to complete the freezing step at atmospheric pressure. Control tests at each freezing rate were run for comparisons.

C. Freeze Drying Procedure

After the freezing step was completed, primary and secondary drying for all the tests was performed by following the same temperature profile. One thermocouple probe was used to control and monitor product temperature near to the tray bottom during drying. The end point was determined by Capacitance Manometer/Pirani differential and was the same for all assays.

Lyophilized samples with and without VI were packed in individual high barrier plastic bags under vacuum, and then stored in the dark at 4^0C until their analysis. All sample analysis was performed as triplicate.

D. Statistical Analysis

To evaluate the freezing rate and VI effect on the final properties of the dried product, a factorial design 3 K with two factors and three replicates was used. The results were evaluated with an ANOVA of 95% of confidence

TABLE I
OPERATING CONDITIONS USED DURING VI FREEZING

Conditions		
Freezing rate (°C/min)	Temperature[*] (°C)	Pressure (mTorr)
	-2	900
0.1	-2	700
	-2	500
	-2	900
0.25	-2	700
	-2	500
	-2	900
0.4	-2	700
	-2	500

*At which vacuum was applied

E. Moisture Content

The initial and final moisture content of MS and the final moisture content of MPM were determined using a moisture balance (MOC-120H, Shimadzu Corporation Japan) at 100°C.

F. Apparent Density

Apparent density of dried MS and MPM was measured using the pycnometer method [8]. A pycnometer of 5 mL was weighed using an electronic balance (w_1), then it was filled with toluene and weighed again (w_2). A piece of freeze dried mango was weighed (w_3) and coated with wax and re-weighed (w_4), then it was transferred to the pycnometer and re-filled with toluene. Finally, the set was weighed (w_5) and the apparent density was calculated as (1):

$$\rho_{app} = \frac{w_3}{\frac{v(w_2 - w_5 + w_4)}{w_2 - w_1} - \frac{w_4 - w_3}{\rho_w}} \qquad (1)$$

where, v is the pycnometer volume and ρ_w the wax density

G. Rehydration on Mango Slices (MS)

Rehydration experiments were performed according [8], with some modification. 1g of dried MS was immersed in 100mL of distilled water at 25ºC. The samples were taken out at 1, 3, 5, 7, 9, 12 and 15 min, respectively, and drained over a mesh for 2 min to eliminate the superficial water, subsequently the sample weight was recorded. Final rehydration ratio (RR), defined as the ratio of rehydrated sample weight (15 min) to initial weight of dry sample, was calculated according to (2):

$$RR = \frac{mf}{mo} \qquad (2)$$

where RR is the final rehydration ratio, *mf* and *mo* are the ending and initial weights of MS sample.

H. Solubility

Solubility was determined as described by [9]. Water solubility was calculated using

$$solubility(\%) = \frac{100 \ (weight \ of \ dissolved \ solids \ in \ supernatant}{weight \ of \ sample} \qquad (3)$$

I. Energy Costs Estimation

The electricity costs of the lyophilization tests were calculated from wattmeter measurements of the power consumption of the freeze-drier equipment during the drying tests (local electricity retail price: ten US$ cents per kWh).

III. RESULTS

Freezing rate is the slope of the linear zone of the mango sample temperature profile, after initial freezing point. Process time include freezing time plus freeze drying time.

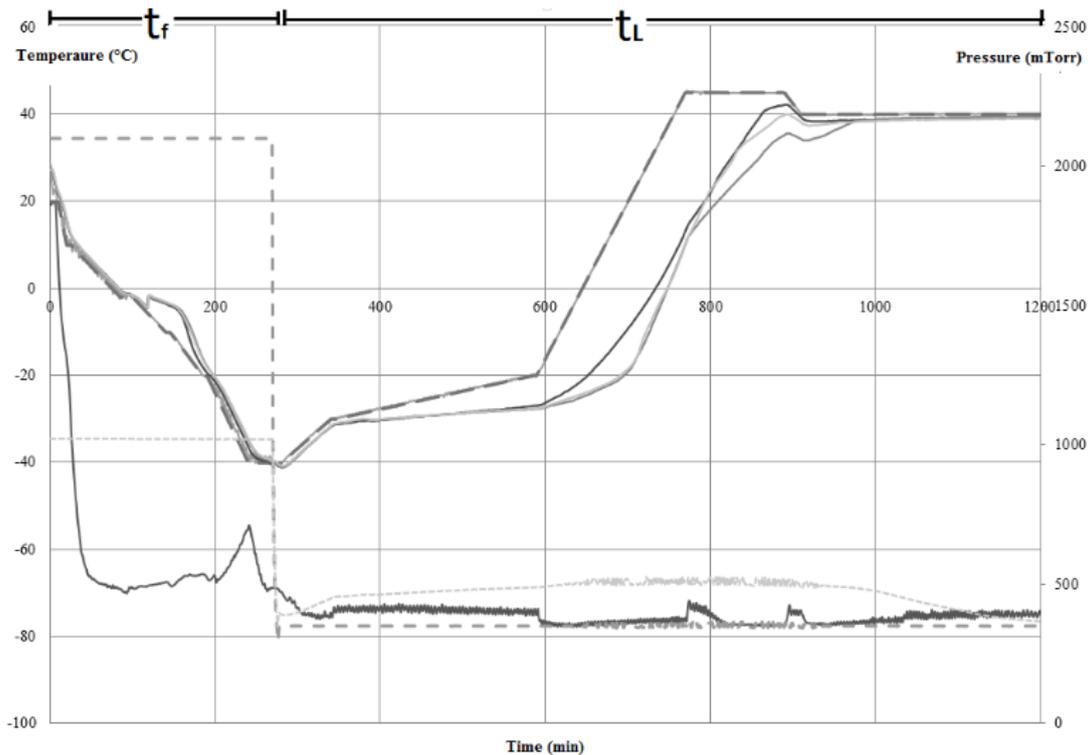Typical freezing and freeze drying run is shown in Fig. 1.

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Fig. 1 Typical freeze drying run for Mango samples

——Condenser Temperature; ——— Shelf Temperature; ━━ ¯Reference Temperature; ——Mango Temperatures; – – Capacitance gauge;
·········· Pirani gauge; t_F = Freezing time; t_L= lyophilization time; t_F + t_L = Overall rocess time

A. Influence of Freezing Rates on the Overall Process Time

Different freezing rates were applied to (MS) and (MPM) in order to find differences on the overall process time and the corresponding energy costs between them. Results are showed in Table II.

Among the tests that used different freezing times and VI treatments, as the freezing rate was increased, the overall process time decreased significantly. This very same result stands for the freezing rate of 0.4°C/min, when the final pressure of VI treatment was lowered.

In the set of constant 0.4°C/min freezing rate – VI treatments it was evident the reduction of the overall process time for all VI final pressure used compared with the same parameter observer for the controls. Furthermore, the same behavior was observed for MPM treatments.

Regarding the calculated energy costs from Table I, it can be concluded that there was a significant cost reduction- up to 30% -when freezing rate was done at 0.4°C/min respecting the controls and the treatments at 0.1°C/min.

B. Influence of Freezing Treatments on the Quality Parameters of Dried Mango Samples

Moisture content: Initial moisture content in fresh MS ranged between 80 to 88% while the final moisture of dried samples was 5.0% ± 2.0% (wet basis) for all freezing rates, VI and control tests. However, it can be observed from Fig. 2, that 700 and 900 mtorr VI treatments increased the water content in dried mango slices while the two fast freezing rate VI at 500 mtorr yielded the same output level of moisture than

the observed for the control tests. So, the use of VI treatments with the lowest final pressure-500 mtorr- and 0.4 °C/min gave the best combination for reductions in overall process time and costs, while keeping appropriate low output moisture content of freeze dried mango slices.

TABLE II
OVERALL PROCESS TIME AND ENERGY COST OF SLICES AT THE DIFFERENT OPERATING CONDITIONS

Conditions			Overall process time (min)	Energy cost USD kWh
Freezing rate (°C/min)	Temperature* (°C)	Pressure** (mTorr)		
Control***			1513	$ 16.31
	-2	900	1581	$ 17.05
0.1	-2	700	1513	$ 16.31
	-2	500	1578	$ 17.02
Control***			1181	$ 12.73
	-2	900	1221	$ 13.17
0.25	-2	700	1195	$ 12.89
	-2	500	1244	$ 13.41
Control***			1271	$ 13.71
	-2	900	1142	$ 12.31
0.4	-2	700	1127	$ 12.15
	-2	500	1114	$ 12.01

* Temperature where vacuum induced (VI) is applied. ** VI Pressure. *** Without VI.

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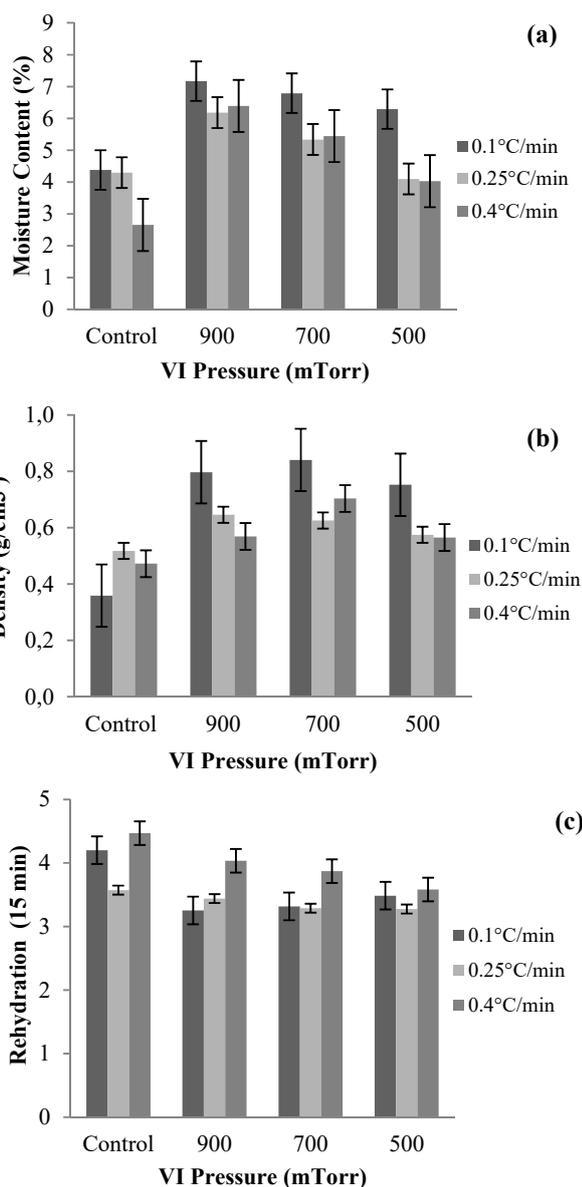International Journal of Biotechnology and Bioengineering
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Fig. 2 Quality attributes of freeze dried Mango Slices (MS) (a)
Moisture content (b) Apparent density (c) rehydration ratio

the solutions were foamed while the slices were shrunk in some degree as result of the pressure reduction during freezing. Further microscopic examination of these dried materials is necessary to verify these effects.
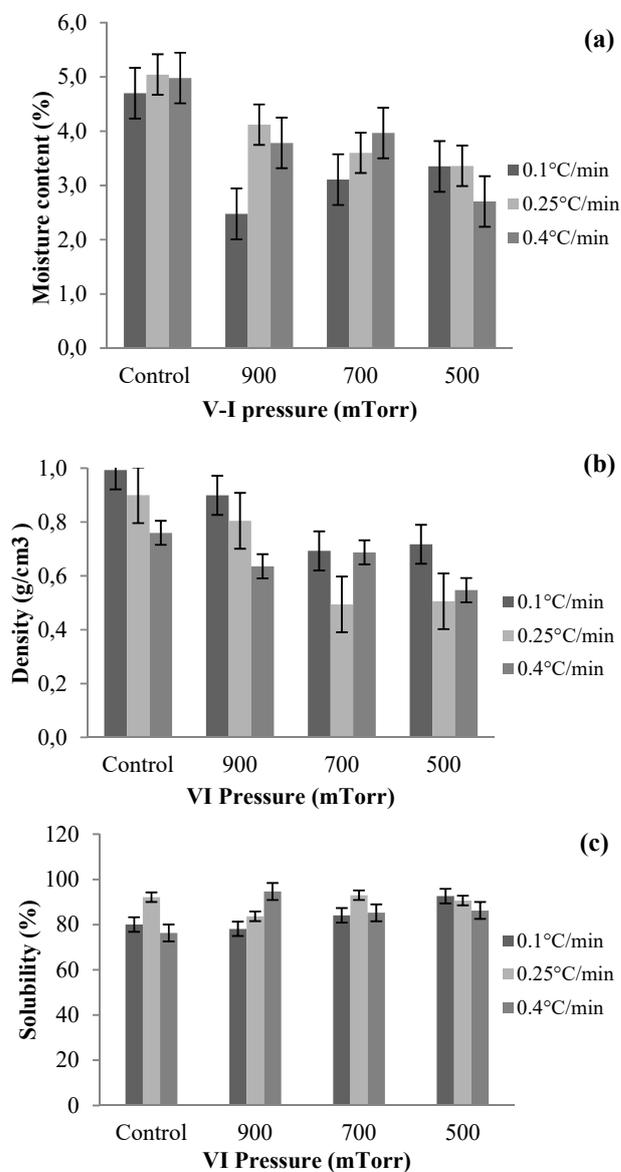

Fig. 3 Quality attributes of freeze dried mango pulp-maltodextrin dispersions (MPM) (a) Moisture content (b) Apparent density (c) Solubility (%)

IV. CONCLUSIONS

Freezing rate and sudden reduction in pressure during freezing stage affect overall drying time and properties of dried products in mango lyophilization.

Between the freezing conditions studied in this work, the higher freezing rate (0.4°C/min) reduced the overall process time (and, consequently, the operational cost) up to 30% compared with the required for the lower freezing rate (0.1°C/min) for both MS and MPM. Furthermore, results from MPM dispersion showed favorable values when VI was

The mango pulp-maltodextrin dispersions (MPM) showed an opposite behavior (Fig. 3 (a)). The higher values of final water content in the dried samples were observed for the control tests. For this material, the effect of VI treatments is the simultaneous reduction of overall freezing time (Table I), costs and final moisture content. Similar behavior were reported for pharmaceutical solutions by [5], [7].

Density: The differences between the responses of MS and MPM to VI treatments could be explained by opposite microstructural alterations as can be inferred from the analysis of Figs. 2 (b) and 3 (b). The densities of freeze-dried MS was higher than those of the correspondingly controls indicating possible superior porosities. On the other hand the dried MPM control samples showed higher densities respecting to those measured for 700 and 500 mtorr VI treatments. Apparently,

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applied because product with low moisture content and low density was obtained at shorter process time compared with the control.

Reconstitutional properties as rehydration for MS and solubility for MPM remained similar in controls, VI treatments, and different freezing rates.

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