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Effect of osmotic dehydration and ultrasound pre-treatment on cell structure: Melon dehydration

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Abstract

The effect of osmotic dehydration and of ultrasound pre-treatment applied at atmospheric pressure for different lengths of time on melon tissue structure was evaluated. Osmotic dehydration-induced gradual loss of shape of cell wall, disconnection between the cells and breakdown of the tissue. Ultrasound induced the formation of microscopic channels in the fruit structure but did not induce breakdown of the tissue. The changes observed on the structure of the fruit explain the effects of these two pre-treatments on the water diffusivity of the subsequent air-drying step. Osmotic dehydration when carried out for less than 30 min decreases the water diffusivity due to the incorporation of sugar, but increases water diffusivity when carried out for more than 1 h due to the breakdown of cells lowering the resistance to water diffusion. Ultrasound treatment increases water diffusivity due to the formation of microscopic channels which also offers lower resistance to water diffusion.

Keywords: *Curcumis melo* L.; Melon; Image analysis; Ultrasound; Osmotic dehydration; Drying

1. Introduction

Drying is the most common method of food preservation and is used to reduce post-harvest loss and to produce several dried fruits which can be directly consumed or used in processed foodstuffs. Conventional air-drying is energy intensive and consequently cost intensive because it is a simultaneous heat and mass transfer process accompanied by phase change. A pre-treatment can be used to reduce the initial water content of the fruit or can be used to modify the fruit tissue structure in a way that air-drying becomes faster.

Osmotic dehydration is the most reported pre-treatment used prior to air-drying. The technique consists in immersing the fruit in hypertonic solution to partially remove water from the fruit. The driving force for water removal is the difference in osmotic pressure between the fruit and the solution where the complex cellular structure of the fruit acts as a semi-permeable membrane (Corzo & Gómez, 2004; Fito, 1994; Prokharkar, Prasad, & Das, 1997; Raoult, Lafont, Rios, & Guilbert, 1989; Raoult-Wack, 1994; Rastogi, Eshtisghi, & Knorr, 1999; Rastogi & Niranjan, 1998; Rastogi & Raghavarao, 1997; Simal, Benedito, Sánchez, & Rosello, 1998; Torreggiani, 1993).

Power ultrasound is a novel technology in the food industry and its use is increasing as new uses are studied. Ultrasonic waves can cause a rapid series of alternative compressions and expansions, in a similar way to a sponge when it is squeezed and released repeatedly (sponge effect). The forces involved by this mechanical mechanism can be higher than surface tension which maintains the moisture inside the capillaries of the fruit creating microscopic channels which may ease moisture removal. In addition, ultrasound produces cavitation which may be helpful to remove strongly attached moisture. Deformation of porous solid materials, such as fruits, caused by ultrasonic waves is responsible for the creation of microscopic channels that

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reduce the diffusion boundary layer and increase the convective mass transfer in the fruit (Fuente-Blanco, Sarabia, Acosta-Aparicio, Blanco-Blanco, & Gallego-Juárez, 2006; Tarleton, 1992; Tarleton & Wakeman, 1998).

The aim of this study was to evaluate the effect of osmotic dehydration and ultrasound pre-treatments on melon tissue. The evolution of cellular structure was studied by light microscopy analysis and the structural modifications resulting from osmotic dehydration and ultrasonic waves were compared. The influence of both pre-treatments on water diffusivity in the air-drying step was analyzed.

2. Materials and methods

2.1. Preparation of samples

Melons were bought from the producer (Fortaleza, Brazil) being commercial melons harvested at 65 days. Melon samples were cut and sliced to obtain cubes of same dimensions (0.02 m average side). The moisture content was determined by heating in a drying oven (Marconi model MA-085) at 105 °C for 48 h according to AOAC method 934.06 (AOAC, 1990). The initial concentration of solute (Brix) was determined by refractometry.

2.2. Ultrasound pre-treatment

An experimental set consisting of three melon samples was immersed in distilled water and submitted to ultrasonic waves during 20 and 30 min. These pre-treatment times were chosen after the results of kinetics studies carried out beforehand. The results showed that the effects of ultrasound pre-treatment started to influence the drying process after 10 min (little effect) and after 30 min the changes inferred in the drying process became insignificant.

The experiments with ultrasound were carried out in separate 250 mL Erlenmeyer flasks to avoid interference between the samples and runs. The water to fruit ratio was maintained at 4:1 (weight basis). The experiments were carried out under ambient water temperature (30 °C) in an ultrasonic bath (Marconi model Unique USC) without mechanical agitation. The temperature was measured before and after the runs and the temperature increase during the runs did not exceed 2 °C. The ultrasound frequency was 25 kHz and the intensity was 4870 W/m^2. Each assay was made in triplicate. From each replicate, one random sample was used in microscopy image analysis studies and two samples were air dried.

At the end of the ultrasound pre-treatment, a sample of the liquid medium was taken to determine its sugar content using the DNS method (Miller, 1959) and its glucose content using the enzymatic method (Fleming & Pegler, 1963). This procedure was carried out to quantify the amount of sugar that the fruit loses, by mass transfer, to the liquid medium.

2.3. Osmotic dehydration

An experimental group consisting of three melon cubes was immersed in the osmotic solution for 0.5, 1 and 2 h. The osmotic dehydration was carried out in separate 250 mL Erlenmeyer flasks to avoid interference between the samples and runs. The osmotic solution used in each experiment was prepared mixing food grade sucrose with distilled water to give a concentration of 70 °Brix. The osmotic solution to fruit ratio was maintained at 4:1 (weight basis). The experiment was performed with constant mechanical agitation (150 rpm) in a rotary shaker (Tecnal model TE-420), applied to homogenize the osmotic solution avoiding formation of local concentration gradients. The temperature was monitored using the thermocouple of the rotary shaker heating plate and was set at 42.5 °C. This operating condition was chosen based on optimization carried out in previous studies and has presented the highest water removal rate and lower processing time (Rodrigues & Fernandes, 2006; Teles et al., 2006).

After removal from the solution, the dehydrated samples from each group were drained and blotted with absorbent paper to remove excess solution. Each assay was made in triplicate. From each replicate, one random sample was used in microscopy image analysis studies and two samples were air dried.

Weight and moisture content of the samples, and the sugar content of the fruit and liquid medium were used to calculate water loss (WL) and solid gain (SG), according to the following equations:

$$\mathrm{WL}\ (\%) = \frac{w_i X_i - w_f X_f}{w_i} \times 100, \quad (1)$$

$$\mathrm{SG}\ (\%) = \frac{w_f X_{sf} - w_i X_{si}}{w_i} \times 100, \quad (2)$$

where X_i is the initial fruit moisture on wet basis (g water/g), X_f is the final fruit moisture on wet basis (g water/g), X_{si} is the initial fruit soluble solid content (g solid/g), X_{sf} is the final fruit soluble solid content (g solid/g), w_i is the initial fruit mass (g), and w_f is the final fruit mass (g).

2.4. Air-drying

After removal from the osmotic solution and from the ultrasonic bath, the samples were drained, blotted with absorbent paper to remove excess solution and transferred to a forced circulating air-drying oven (Marconi model MA-085) set at 60 °C. The air moisture content was 18% and was determined by psychrometry (dry and wet bulb temperature). This operating condition was chosen based on optimization carried out in a previous study (Teles et al., 2006).

The samples were weighted every 30 min during 12 h and the data were used to calculate the water diffusivity of melons during air-drying according to Fick's Law of

diffusion. The equation used for the falling-rate period of the drying process was based on the simplification of Fick's second law considering long processing period (Perry & Green, 1999). The effective water diffusion parameter was adjusted using Eq. (3) with a parameter estimation procedure based on the minimization of the error sum of squares:

$$\frac{dH}{dt} = -\frac{2\pi}{\delta^2} D(H - H_{eq}), \quad (3)$$

where D is the effective water diffusivity (m^2/s), H is the moisture content, H_{eq} is the equilibrium moisture content, t is the time (s), and δ is the thickness of the fruit (m).

2.5. Light microscopic analysis

After the end of each pre-treatment, the cube samples were carefully cut into smaller cubes (0.005 m average side). The sample cubes were fixed with 4% solution of paraformaldehyde in 0.1 m phosphate buffer, pH 7.2 and 1% glutaraldehyde for 24 h at ambient temperature (Karnovisky, 1965). The material was then dehydrated in a graded ethanol series and embedded in Historesin embedding kit (Jung). The tissue blocks were sectioned at 8 μm on a Leica RM 2065 microtome. Histochemical reaction was carried out with Toluidine Blue (TB) at pH 4.0 as metachromatic stain to detect polyanionic pectins (Vidal, 1977).

Photomicrographs of the cell structure were taken using an Olympus BX51 light microscope with digital image capture system.

3. Results and discussion

The effects of both pre-treatments (osmotic dehydration and ultrasound) on water loss and sugar gain are presented in Table 1. The results show that significant differences exist between the two pre-treatments. During osmotic dehydration pre-treatment, the fruit lost water and gained sugar, while during ultrasonic treatment, the fruit gained water and lost sugar. Both treatments have also increased the diffusivity of water during the air-drying stage.

The results regarding water loss and sugar gain were expected and were due to the concentration gradient of water and sugar between the fruit and the liquid medium. The changes on water diffusivity during the air-drying stage were due to changes in the tissue structure of melons which will be discussed below.

The microscopic image analysis of the fresh fruit showed typical thin-walled cells with normal morphology and some intercellular spaces (Fig. 1). Very few cells had undulated walls.

After 30 min of osmotic treatment, several differences were observed due to water loss. The cell walls became distorted and smaller in all regions of the samples. In some regions, the junctions between adjacent cells were present and the intercellular spaces were reduced (Fig. 2A), while in other regions disruption of the cells were observed causing an increase of intercellular spaces which may be due to the solubilizing of chelator-soluble pectin of the middle lamella (Fig. 2B). Chelator-soluble pectin is the substance that most contributes to cell adhesion and firmness and according to the microscopic images may solubilize at the early stages of osmotic dehydration.

After 1 h of osmotic treatment, most intercellular spaces between the cells disappeared and the cell walls became more distorted (Fig. 3A). In some regions of the samples, cell wall breakdown started to appear (Fig. 3B) although it was still rare. At this point in the process, 21.7% of water was removed from the fruit and sugar gain was at 70.9% as

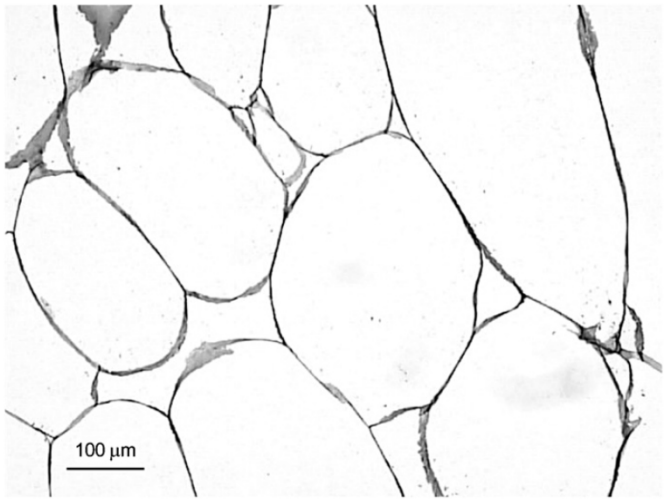

Fig. 1. Photomicrographs of melon cubes before processing (raw fruit). Magnification of 380 ×.

Table 1
Water loss and sugar gain after pre-treatment and water diffusivity during air-drying

Operating condition*	Water loss (%)	Sugar gain (%)	Water diffusivity (m^2/s)
Air-drying only	–	–	5.00×10^{-9} ($R^2 = 0.995$)
After 20 min of ultrasound treatment	−8.7 ± 0.6	−44.7 ± 2.0	6.42×10^{-9} ($R^2 = 0.996$)
After 30 min of ultrasound treatment	−7.7 ± 2.3	−52.2 ± 1.0	6.97×10^{-9} ($R^2 = 0.998$)
After 30 min of osmotic dehydration	+19.5 ± 1.0	+40.4 ± 2.0	4.43×10^{-9} ($R^2 = 0.994$)
After 60 min of osmotic dehydration	+21.7 ± 1.1	+70.9 ± 3.5	6.27×10^{-9} ($R^2 = 0.993$)
After 120 min of osmotic dehydration	+32.0 ± 1.6	+87.5 ± 3.5	8.55×10^{-9} ($R^2 = 0.996$)

*Initial sugar content = 8%; initial moisture content = 90%.

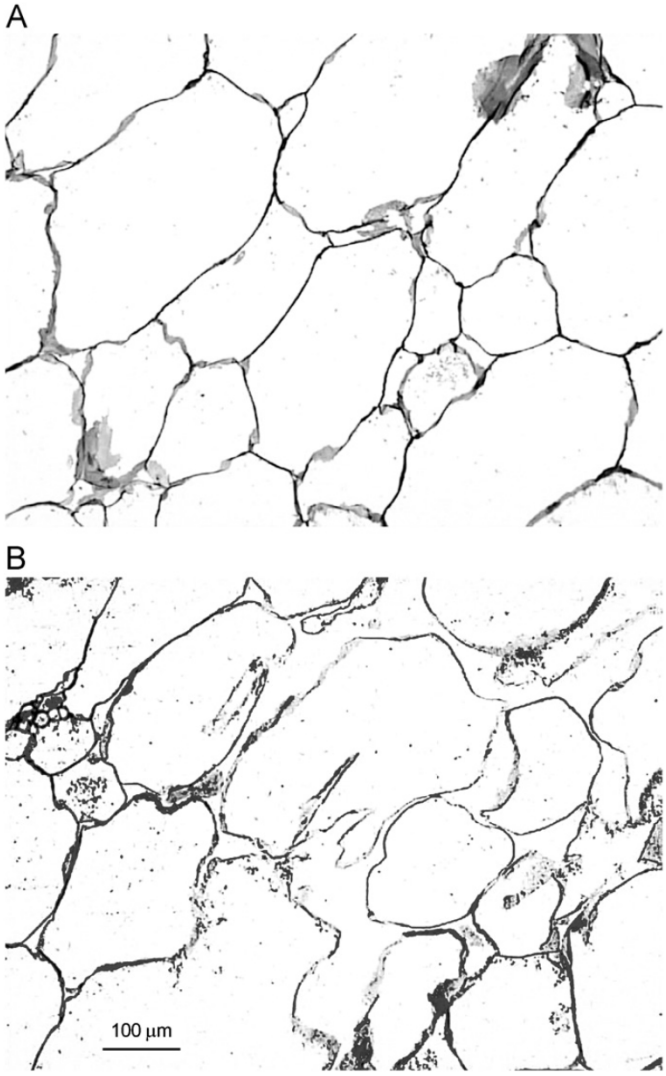

Fig. 2. Photomicrographs of melon cubes after 30 min of osmotic dehydration: (A) region with distorted cells and (B) region with disrupted cells. Magnification of 380 ×.

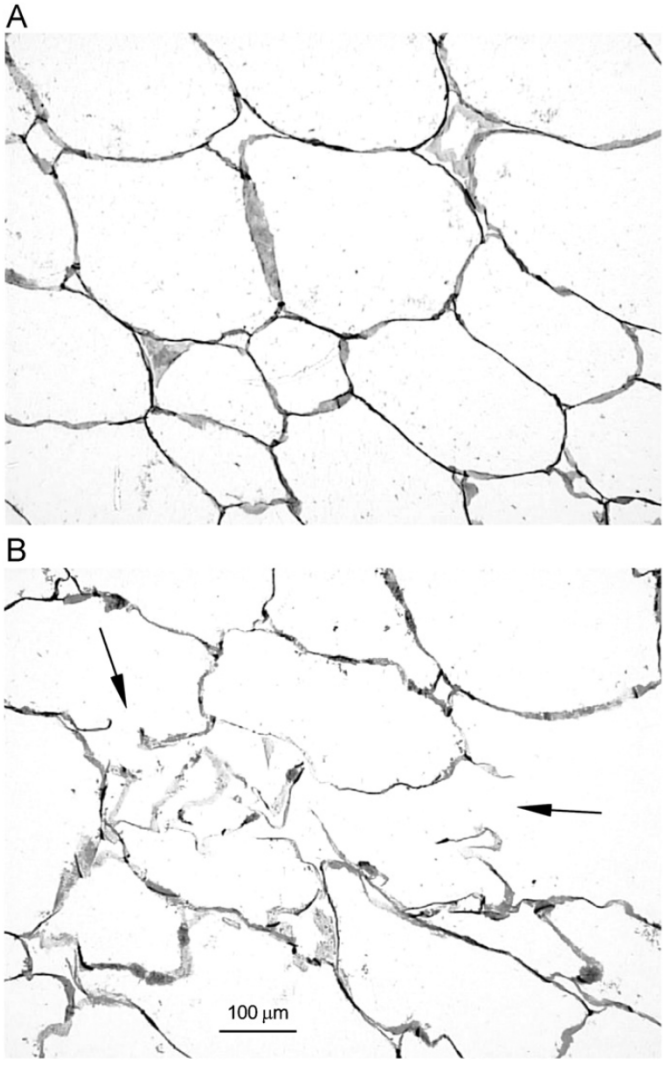

Fig. 3. Photomicrographs of melon cubes after 1 h of osmotic dehydration: (A) region with distorted cells and (B) region with cells in initial stage of cell breakdown. Magnification of 380 ×.

shown in Table 1. Diminished cell wall strength, due to solubilizing of pectin, combined with high osmotic pressure and flow of sugar molecules into the fruit may contribute to the breakdown of cell walls. Pectin solubilizing during osmotic dehydration was shown by Prinzivalli, Brambilla, Maffi, Scalzo, and Torreggiani (2006).

A greater change was observed after 2 h of osmotic treatment when most cell walls were broken down and the few remaining cells had severely distorted walls (Fig. 4). Solubilizing of pectin can be visually noted by decreasing cell wall strength. Fig. 5 shows the effect of knife cutting on the border of the fruit sample. After 30 min of osmotic treatment, the cell walls were more rigid and could be cut without breaking down the cells (Fig. 5A), whereas after 2 h of osmotic treatment, the cell walls were weaker and failed under stress (Fig. 5B).

Water diffusivity for the drying process was $3.00 \times 10^{-7}\,m^2/min$ for melons without any prior pre-treatment. In a previous study, Rodrigues and Fernandes (2006) showed that the use of the osmotic dehydration pre-treatment increased water diffusivity in the air-drying process when carried out for more than 1 h. The increase can be now explained by the collapse of cell wall membranes. Between 30 and 60 min of osmotic dehydration, the cell wall membranes started to break down and the phenomena increased after 60 h of osmotic dehydration. Water diffusion within the fruit became easier when the cell wall membranes broke down and the effective water diffusivity increased as shown in Table 1. After 2 h, most of the cells collapsed and the water diffusivity came to a maximum value. Rodrigues and Fernandes (2006) showed that water diffusivity decreased after 2 h in the osmotic treatment and this may be caused by incorporation of solids by the fruit increasing water diffusion resistance.

At the end of the ultrasonic pre-treatment, little change was observed in the fruit moisture content. The fruit incorporated water after submitted, during 20 min, to ultrasound increasing 8.7% its water content. Microscopic

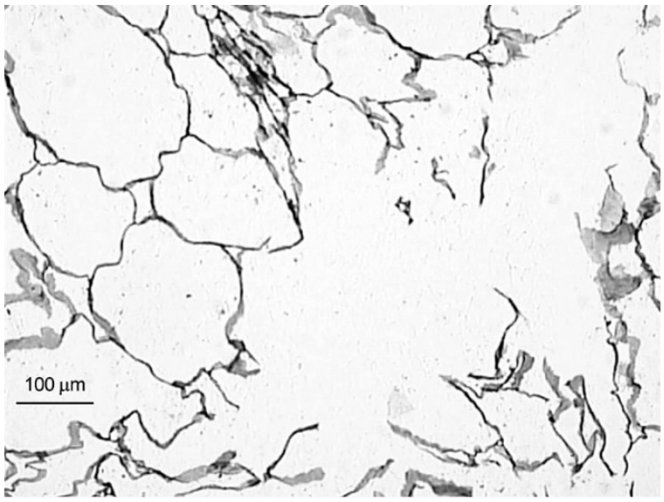

Fig. 4. Photomicrographs of melon cubes after 2 h of osmotic dehydration showing cells in advanced stage of cell breakdown. Magnification of 380 × .

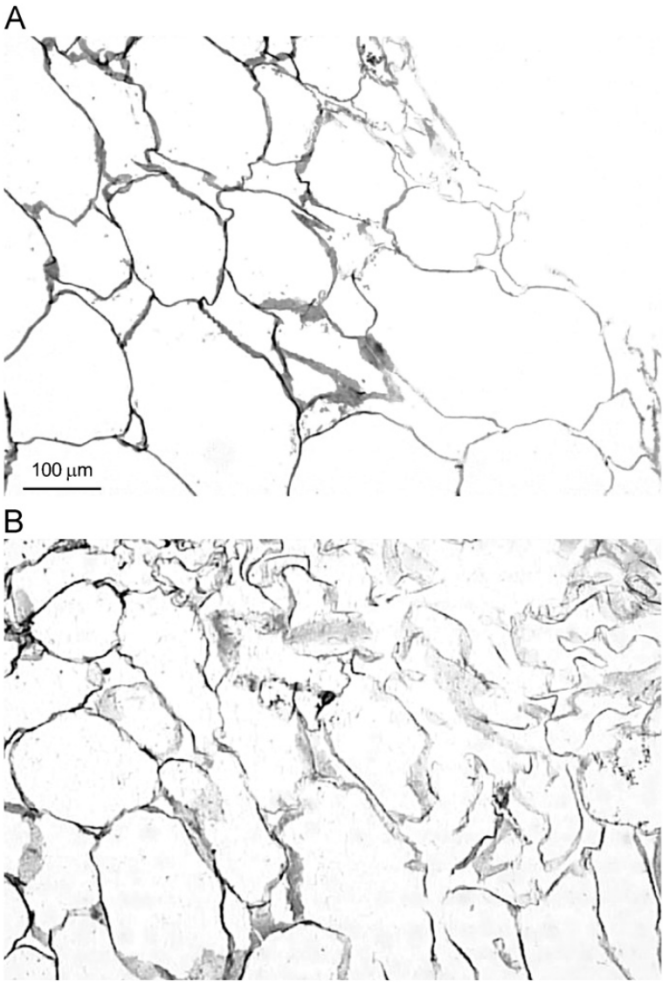

Fig. 5. Photomicrographs of the borders of melon cubes samples before processing (A) and after 2 h of osmotic dehydration (B). Magnification of 380 × .

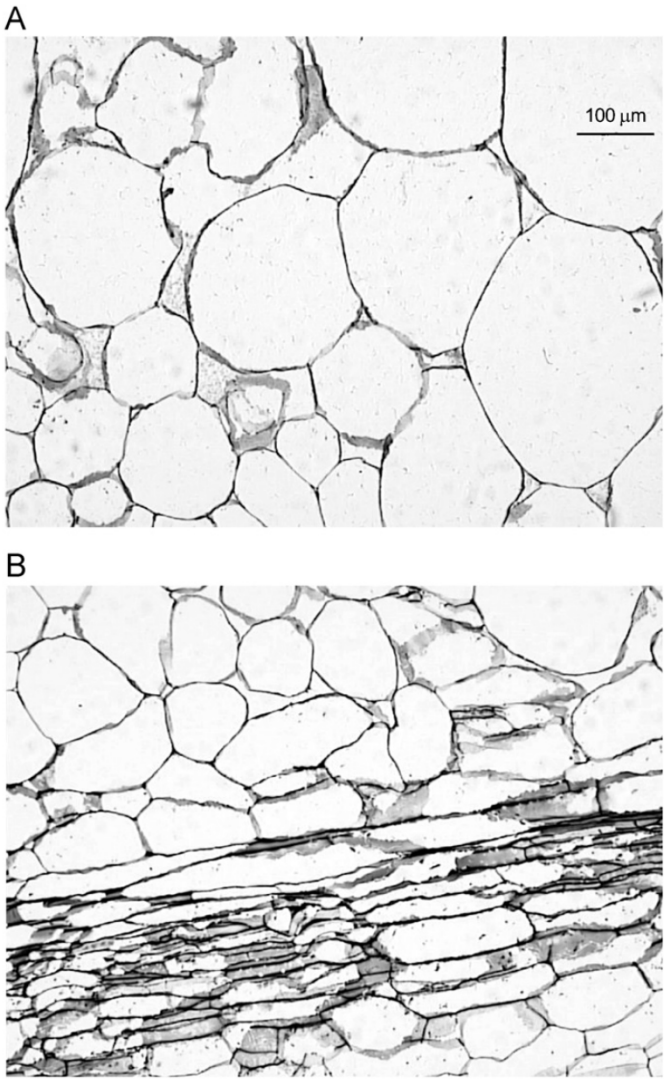

Fig. 6. Photomicrographs of melon cubes after 20 min of ultrasound pre-treatment: (A) region with bloated cells and (B) region with microscopic channels. Magnification of 380 × .

image analysis showed two distinct regions, one region where the cells became bloated as a result of water gain (Fig. 6A) and a second region where the cells became much smaller and needle shaped (Fig. 6B). These needle shaped cells were not observed in the fresh fruit or in the samples submitted to osmotic dehydration and were observed in all samples submitted to ultrasound pre-treatment. Some researchers have reported that ultrasonic waves may create microscopic channels (Fuente-Blanco et al., 2006; Tarleton, 1992; Tarleton & Wakeman, 1998), but still no microscopic image has been reported. Fig. 6B shows that the microscopic channels are formed by the elongation and flattening of cells in some regions of the sample.

The use of ultrasound increased 39.3% water diffusivity during the air-drying process (Table 1) reducing the time required for drying. This result confirms the observations of Fuente-Blanco et al. (2006) that the ultrasonic pre-treatment affects the fruit tissue making easier for the water

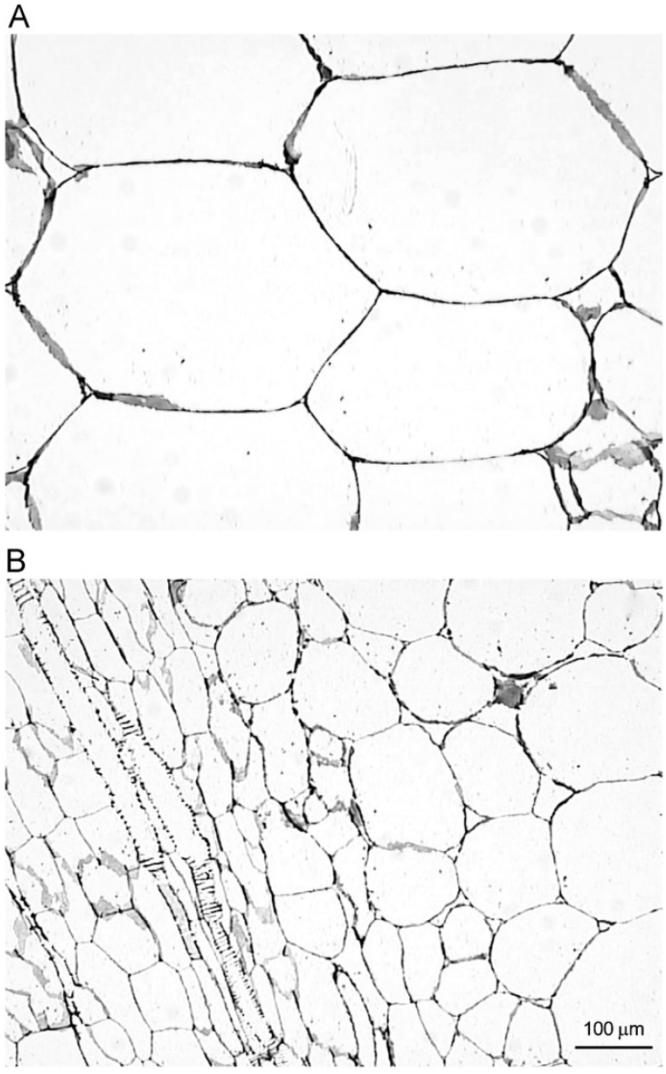

Fig. 7. Photomicrographs of melon cubes after 30 min of ultrasound pre-treatment: (A) region with bloated cells and (B) region with microscopic channels. Magnification of 380 × .

to diffuse during air-drying and showed that the microscopic channels may contribute with the higher water diffusivity.

After 30 min of ultrasonic treatment, the microscopic channels became broader (Fig. 7B) and some new smaller microscopic channels seamed to appear. The region with bloated cells was still observed (Fig. 7A). The broadening of the microscopic channels may explain further increase in water diffusivity of the fruit in the air-drying process (Table 1). There was no indication of loss of strength of the cell wall and during knife cutting the cells at the border did not fail under stress as happened when the fruit was submitted to osmotic dehydration. No cell breakdown was observed in the samples submitted to ultrasound.

The changes in the tissue structure had direct effect on the values of water diffusivity during the air-drying process as shown in Table 1. The effective water diffusivity in fruits is dependent on tissue structure since the cell walls act as a semi-permeable membrane and also on the fruit porosity. The pre-treatments studied herein have increased the effective water diffusivity of melons but through different effects. Ultrasonic waves have created microscopic channels in the fruit which increased the effective water diffusivity because water could use these microscopic channels as an easier pathway to diffuse towards the surface of the fruit. On the other hand, the osmotic dehydration increased the effective water diffusivity by breaking down part of the cell walls reducing the resistance for water to diffuse through the cells.

In a quantitative context, the changes in the value of the water diffusivity have great significance during the air-drying stage. For example, if melons are dried from an initial moisture content of 9.10 g of water/g of solids to a final moisture content of 0.25 g of water/g of solids, it will take 760 min to dry the melons if the water diffusivity is at $5.00 \times 10^{-9}\,m^2/s$ (fresh melons water diffusivity). Submitting the melons to 30 min of ultrasound treatment, the drying time will be reduced to 550 min due to the increase in water diffusivity to $6.97 \times 10^{-9}\,m^2/s$. Submitting the melons to 1 h of osmotic dehydration, the drying time will be reduced to 605 min due to the increase in water diffusivity to $6.27 \times 10^{-9}\,m^2/s$.

4. Conclusion

Osmotic dehydration induced significant changes on melon cell structure. After 30 min of osmotic dehydration, there was a gradual disruption of cells together with loss of shape of cell walls. From 1 h onwards, a significant breakdown of the tissue was observed. A good agreement was obtained between cellular structure changes and water diffusivity. Water diffusivity decreased due to the incorporation of sugar when the osmotic dehydration was carried out for less than 30 min, but increased when the osmotic dehydration was carried out for more than 1 h due to the breakdown of cells which lowered the resistance to water diffusion.

Ultrasound also induced changes on melon cell structure, but different from osmotic dehydration, no cell breakdown was observed. Microscopic channels appeared in the cell structure and may be responsible for the increase on water diffusivity because offered lower resistance to water diffusion.

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