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Microencapsulation of pulp and ultrafiltered cactus pear (*Opuntia ficus-indica*) extracts and betanin stability during storage

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ABSTRACT

Pulp (CP) and ultrafiltered (UF) cactus pear extracts were encapsulated with Capsul (C) by applying a central composite design (CP–C and UF–C systems) by spray-drying. To evaluate the effect of the extract, microparticles obtained under optimal conditions were characterised and stored at 60 °C. Betacyanin and betaxanthin encapsulation efficiency reached values above 98% for both systems studied. This efficiency was attributed to strong interactions between betalains and the polymer. Betalain degradation in CP–C and UF–C microparticles followed pseudo-first order kinetics. The betacyanin degradation rate constant was significantly higher for CP–C than for UF–C. These results suggested that the mucilage or higher sugar content of CP increased the hygroscopicity of the CP–C microparticles, leading to the degradation of betalain. The hydrolysis pathway was the main mechanism of betanin degradation during microparticle storage. These results demonstrate the potential utility of both CP–C and UF–C microparticles as natural colourants for healthy foods.

1. Introduction

The food industry uses thousands of tons of colourant in a year because colour is one of the most important attributes of food appearance and is considered to be an indicator of quality and acceptability (Azeredo, 2008). Synthetic red colorants used as additives in food have been restricted by official regulations of the EU and the USA due to their possible adverse effects on human health (Tsuda, Murakarni, Kano, Taniguchi, & Sasaki, 2001). Therefore, there is a growing interest in the search for new, red, natural pigment sources and potential applications in foods.

The betalains are natural water-soluble pigments composed of two structural groups: betacyanins (reddish-purple) and betaxanthins (orange–yellow). The red beet (*Beta vulgaris* L.) is a commercial source of betanin, which is classified as additive E-162 (EU) and 73.40 (FDA, USA). The red beet is used mainly to color food, such as dairy products, confectionery, ice cream, desserts, beverages and sausages (Obón, Castellar, Alacid, & Fernández-López, 2009). However, red beets have some disadvantages, including the earth-like flavour imparted by geosmin and 3-s-butyl-2 methoxypyrazine and high nitrate levels. Therefore, the purple cactus pear (*Opuntia ficus-indica*) is an interesting alternative as a source of betanin for the production of food colouring (Sáenz, Tapia, Chavez, & Robert, 2009; Castellar, Obón, Alacid & Fernández-López, 2003; Delgado-Vargas, Jiménez, & Paredes-López, 2000).

The betalain pigment is usually extracted from fruit pulp or pieces of other raw material (e.g., red beet) with a solvent (water, ethanol or methanol), with or without heat treatment or acidification, to improve the pigment yield (Delgado-Vargas et al., 2000). Ultrafiltration (UF) is an alternative method to obtain betalain extract at low-temperatures. Low temperature extraction avoids the problem of pigment degradation during the separation and concentration process (Bayindirli, Yildiz, & Özilgen, 1988). UF processes have previously been reported to concentrate red beet juice (Bayindirli et al., 1988) and to clarify yellow and red cactus pear pulp (Cassano, Conidi, & Drioli, 2010; Cassano, Conidi, Timpone, D'Avella, & Drioli, 2007).

In addition, some studies have reported that purple cactus pear pulp provides protection against diseases related to oxidative stress (Stintzing et al., 2005; Tesoriere, Butera, Pintaudi, Allegra, & Livrea, 2004). Therefore, purple cactus pear extract could have a double application in healthy foods design as a natural colorant, and as an antioxidant.

The stability of betanin has been shown to be affected by pH, water activity, exposure to light, oxygen, enzymatic activities,

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and temperature, which is the most important factor (Azeredo, 2008; Herbach, Stintzing, & Carle, 2006, 2004; Castellar et al., 2003). Therefore, the stabilisation of betalains could be improved by using microencapsulation technologies, such as spray-drying.

There are few studies on encapsulation of betacyanins (Pitalua, Jimenez, Vernon-Carter, & Beristain, 2010; Azeredo, Santos, Souza, Mendes, & Andrade, 2007; Ravichandran et al., 2012; Cai & Corke, 2000; Sáenz et al., 2009; Serris & Biliaderis, 2001; Gandía-Herrero, Cabanes, Escribano, García-Carmona, & Jiménez-Atienzar, et al., 2013), spray-drying being the most used encapsulation technology (Pitalua et al., 2010; Azeredo et al., 2007; Ravichandran et al., 2012; Cai & Corke, 2000; Sáenz et al., 2009; Gandía-Herrero et al., 2013). Encapsulation of betacyanins from red beet, *Lampranthus productus* flowers, cactus pear and amaranth has been reported, but among them only one study was conducted on betacyanins from cactus pear by spray-drying (Sáenz et al., 2009). In these studies, encapsulating agents such as gum arabic, starch and maltodextrin, with different dextrose equivalents, inulin, and pullulan, have been used. These studies established that the nature and properties of the biopolymer used as an encapsulating agent (film capacity, oxygen permeability, and hygroscopicity, among others) play an important role in the stability of the betalain microparticles (Cai & Corke, 2000; Serris & Biliaderis, 2001). Capsul®, a chemically modified corn starch by incorporation of a lipophilic component (octenyl succinate groups), has not been previously reported as an encapsulating agent of cactus pear extracts; this is the polymer assessed in this study. The composition of the extracts, such as the sugars and mucilage of the cactus pear pulp, can influence the encapsulating process and the stability of the encapsulated pigments. Therefore, encapsulation efficiency and pigment stability were evaluated in this study.

In summary, the aim of this work was to evaluate the effect of the type of extract (cactus pear pulp (CP) and ultrafiltration (UF)) on betalain encapsulation efficiency and on the stability of the microparticles.

2. Materials and methods

2.1. Materials

Purple cactus pear fruits (*Opuntia ficus-indica*) were obtained from a plantation located in the Antumapu Experimental Station that belongs to the University of Chile, Santiago, Chile. The encapsulating agent, Capsul® (C), was obtained from the National Starch & Chemical, S.A., Chile.

2.2. Methods

2.2.1. Preparation of the pulp (CP) and ultrafiltered (UF) cactus pear extracts

The fruits were washed, manually peeled, and pulped in an Alexanderwerk screw press with a 2 mm screen. The cactus pear pulp (CP) was packed in polypropylene bags and frozen at −20 °C.

Cactus pear pulp (CP:water (1:1)) was clarified by microfiltration with a ceramic multi-tubular membrane module (cut-off 0.2 μm, effective membrane filtration area of 0.0886 m^2, microdyn-Nadir GmbH, Germany). The experimental conditions were as follows: transmembrane pressure, 0.64 bar and temperature, 20 ± 5 °C. A microfiltration permeate solution was used to feed the ultrafiltration system. The ultrafiltration process was performed in a ceramic multi-tubular membrane module (cut-off 1 kDa, effective membrane filtration area 0.013 m^2, Tami, France). The module was operated at a transmembrane pressure of 8.5 bar and a temperature of 31 ± 5 °C for 100 min. The permeate was collected separately in a non-stationary condition (batch).

2.2.2. Analysis of the pulp (CP) and ultrafiltrated (UF) cactus pear extracts

Moisture, soluble solids (°Brix), pH, and acidity were determined according to AOAC (1996) methods. Total sugars were determined by the Antrona method (Osborne & Voogt, 1986) in a UNICAM UV3 UV/Vis spectrometer (U.S.A.). The turbidity was determined using a turbidity meter (HI 93703, Hanna Instruments, U.S.A.).

Betacyanin and betaxanthin analyses were performed spectrophotometrically according to the method of Stintzing et al. (2005). The total phenolic content was determined according to the Folin–Ciocalteu method (Singleton & Rossi, 1965). The results were expressed as microgrammes of gallic acid equivalent per ml of extract according to a calibration curve (204–714 μg/ml; $r^2 = 0.9983$). All of the analyses were carried out in triplicate.

2.2.3. Preparation of the microparticles

Pulp (CP) and ultrafiltered (UF) cactus pear extract microparticles were prepared with Capsul (C) (CP–C and UF–C) as follows: C (5.5–51.72 g) was dissolved in water (64.5–18.28 g), and heated to 82 °C, then cooled to 30 °C and mixed with CP or UF (30 g) with constant stirring. The resulting solutions were homogenised at 11,000 rpm for 3 min with a Polytron PT 2100 (Kinematica A.G, Switzerland) and fed into a B-290 mini spray-dryer (Büchi, Switzerland). The spray-dryer was operated at an air inlet temperature ranging from 133 to 219 °C. The air flow, rate of feeding and atomisation pressure were 600 l/h, 3 ml/min (5%) and 5 bar, respectively, for both systems. The obtained powders were stored in the dark and were kept at −20 °C for subsequent analysis.

2.2.4. Experimental design

The microencapsulation experiments for the CP–C and UF–C systems were performed using a central composite design (CCD) with 12 runs: 4 experimental points, 4 axial points, and 4 central points (used to determine the experimental error) (Bezerra, Santelli, Oliveira, Villar & Escaleira, 2008). The air inlet temperature (133–219 °C) and the (CP or UF)/C ratio (0.58:1–5.42:1) were evaluated as independent variables according to encapsulation efficiency (EE), recovery (R) and yield of betacyanins and betaxanthins. In this study, response surface methodology (RSM) was used to determine the optimal conditions for each of the systems studied by applying a multiple response optimisation using the desirability function (DF) to determine the optimal conditions, considering all of the response variables (Bezerra et al., 2008).

The data were fitted to a second-order regression model according to Eq. (1). All of the experiments were conducted randomly to avoid systematic bias.

$$Y = b_0 + \sum_{i=1}^{2} b_i X_i + \sum_{i=1}^{2} b_{ii} X_i^2 + \sum_{i=1}^{1} \sum_{j=i+1}^{2} b_{ij} X_i X_j \quad (1)$$

where Y was the response; subscripts i and j ranged from 1 to the number of variables ($n = 2$); b_0 was the intercept term; b_i values were the linear coefficients; b_{ij} values were the quadratic coefficients; and X_i and X_j were the levels of independent variables.

2.2.5. Characterisation of the microparticles

2.2.5.1. General. Water activity (a_w) (AquaLAB, China) at 20 ± 0.3 °C and moisture content were determined according to AOAC methods (1996). Colour parameters (L, $a*$, $b*$, $C*$, and $h°$) were determined with a HunterLab Spectrophotometer UltraScan PRO (USA). Hygroscopicity was determined according to the procedure described by Cai and Corke (2000).

2.2.5.2. Surface betacyanin and betaxanthin determination. Microparticles (100 mg) were dispersed in a 2 ml ethanol:methanol

(1:1) solution, stirred on a vortex mixer for 1 min and centrifuged at 112,000*g* for 5 min. The betalain content was spectrophotometrically quantified according to the Stintzing et al. (2005) method.

2.2.5.3. Total betacyanin and betaxanthin determination. The coating material structure of the microparticles was completely destructed by the following procedure: microparticles (200 mg) were dispersed in 2 ml of methanol:acetic acid:water (50:8:42 v/v/v), were stirred using a vortex mixer for 1 min, were ultrasonicated twice for 50 min, and were then centrifuged at 112,000*g* for 5 min. The betalain content was spectrophotometrically quantified by the Stintzing et al. (2005) method.

The encapsulation efficiency (EE) was calculated according to Eq(2).

$$EE(\%) = \frac{\text{experimental total betalains} - \text{superficial betalains}}{\text{experimental total betalains}} \times 100 \quad (2)$$

2.2.5.4. Morphology and particle size of the microparticles. The outer structures of the microparticles obtained under optimal conditions were studied by scanning electron microscopy (SEM). Microparticles (CP–C and UF-C) were coated with gold/palladium, using a Varian Vacuum Evaporator PS 10E and analysed using a LEO 142 OVP (LEO Electron Microscopy Ltd., Cambridge, UK) operated at 20 kV. The scanned images were collected digitally using EDS 7424 software (Oxford Instruments, Oxford, UK).

Particle size was determined by laser light scattering using a Mastersizer X (Malvern Instruments, Worcestershire, UK) with a lens of 300 mm. Microparticles were dispersed in propylene glycol prior to analysis.

2.2.6. Accelerated storage stability test of the microparticle powders

2.2.6.1. General. Microparticle powders (CP–C and UF–C) (100 mg) obtained under optimal conditions were transferred to clear glass vials (16 × 100 mm) and stored at 60 ± 1 °C in a forced-air oven (Memmert model UFE 500, Germany) with controlled temperature and in the absence of light. For the spectrometric characterisation of betacyanins and betaxanthins, triplicate vials were removed every 7 days during the first 5 weeks, and then every 15 days until the study was completed (25 weeks).

2.2.6.2. Analysis of the degradation products of the microparticle powders. To determine the betanin degradation mechanism (oxidation or hydrolysis) during microparticle storage at 60 °C, an HPLC study was carried out to check for the presence of cyclodopa-D-glucoside and betalamic acid (degradation products). The HPLC equipment was composed of a Merck Hitachi L6200 pump, a Waters 996 photodiode-array detector, and a C18 column (5 μm × 4.6 mm i.d. ×25 cm, Symmetry, Waters, Ireland). The mobile phases used were solvent A (1% aqueous acetic acid) and solvent B (1% acetic acid in acetonitrile) according to the programme described by Fernández-López and Almela (2001). Betanin, cyclodopa-D-glucoside and betalamic acid were identified at 535, 280 and 410 nm, respectively, by comparison of the UV–visible spectra with those of the literature (Fernández-López & Almela, 2001; Herbach et al., 2004).

2.3. Statistical analysis

Linear regression (95% confidence limit) was used to determine the reaction order and the rate constants. A one-way analysis of variance was performed to test for differences between the parameters and the betacyanin and betaxanthin rate constants of the microparticle systems obtained under optimal conditions. Statistically significant terms in the model were found using ANOVA for each response variable. The statistical analyses were performed using Statgraphics Centurion XV (StatPoint Inc., 2011) and MODDE 8 software (Umetrics AB, 2008).

3. Results and discussion

3.1. Characterisation of the pulp (CP) and ultrafiltered (UF) cactus pear extracts

Table 1 shows the chemical and physical characteristics of the CP and UF cactus pear extracts. The amount of total sugar, soluble solids, betalains and total phenolic content of the pulp (CP) were similar to those reported in other studies of the same cactus species (Sáenz et al., 2009; Stintzing et al., 2005).

Ultrafiltration of the cactus pear pulp was carried out to obtain an extract with different physical and chemical characteristics, which would affect to some extent the stability of betalains in the microparticles. The UF extract was a clarified solution (0 NTU) because the mucilage of the pulp remained in the retentate during the membrane process. The total soluble solids and total sugar content of the UF extract were significantly ($p < 0.05$) lower than in the CP extract. Moreover, there was no difference in the betacyanin and betaxanthin content between the UF and CP extracts ($p \geqslant 0.05$), whereas the total polyphenol content was significantly higher ($p \leqslant 0.05$) in the UF extract than in the CP extract.

3.2. Optimisation of multiple variables using response surface methodology

A central composite design (CCD) for each system studied (CP–C and UF–C) was applied to evaluate the effect of the process (air inlet temperature) and the formulation ((CP or UF)/C ratio) on variables. The following five response variables were considered: betacyanin and betaxanthin encapsulation efficiency (EE), betacyanin and betaxanthin recovery (R) and yield.

The betacyanin and betaxanthin EEs ranged from 95.5% to 99.7% and from 94.8% to 97.9%, respectively, for CP–C, and from 92.9% to 100% and from 91.4% to 97.9%, respectively, for UF–C. The betacyanin and betaxanthin Rs ranged from 25.2% to 65.4% and from 50.1% to 77.5%, respectively, for CP–C, and from 44% to 67.5% and from 73.1% to 100%, respectively, for UF–C. Yields ranged between 15.5% and 71.5% for CP–C and between 27.2% and 73.9% for UF–C.

The response surface methodology was applied to optimise the response variables considering the linear, quadratic and cross-product forms of the independent variables studied (air inlet temperature and (CP or UF)/C ratio) at $p \leqslant 0.05$ for each system.

In general, the quadratic form of the (CP or UF)/C ratio was significant for all of the response variables and in both systems studied, while the quadratic form of the air inlet temperature was not

Table 1
Physical and chemical characteristics of cactus pear pulp (CP), and ultrafiltrated (UF) cactus pear extract.

Parameters	CP	UF
Moisture (%)	86.1 ± 0.2[b]	91.9 ± 0.5[a]
Total soluble solids (°Brix a 20 °C)	14.0 ± 0.1[a]	8.1 ± 0.0[b]
Total sugars (%)	13.2 ± 0.0[a]	9.2 ± 0.3[b]
Turbidity (NTU)	2453 ± 64.2[a]	0.0 ± 0.0[b]
Betacyanins (mg BE/100 g)	25.4 ± 0.2[a]	24.8 ± 4.3[a]
Betaxanthins (mg IE/100 g)	8.5 ± 0.1[a]	8.8 ± 1.2[a]
Total phenolics compounds (mg GAE/L)	535 ± 4.4[b]	660 ± 25.0[a]

CP: cactus pear pulp; UF: ultrafiltrated extract; BE: betanin equivalent; IE: indicaxanthin equivalent; GAE: gallic acid equivalent. Different letters show significantly different between systems ($p \leqslant 0.005$).

[a] Corresponds to highest values.

[b] Corresponds to lowest values

significant for any of the response variable in either of the systems studied; therefore, this form was not included in the models. The cross-product form was significant for the EE of betacyanin in the CP–C system and for the EE of betaxanthin, and the R of both betacyanin and betaxanthin in the UF–C system. When the quadratic and/or cross-product forms were significant, the linear form was considered in the model, although the linear form was not significant (i.e., temperature) (Romero & Zuñica, 2005). Therefore, as we expected, both the (CP and UF)/C ratio and the air inlet temperature were significant ($p \leqslant 0.05$) for the CP–C and UF–C systems.

For the multiple optimisation (Desirability Function) of the CP–C and UF–C microparticle systems, the response variables with a high value of coefficient of determination (R^2adj) adjusted by degrees of freedom (d.f.) and residual below 6.0 (Romero, 2005) were considered. However, the lack-of-fit test (non-significant) indicated that the mathematical model fitted well with the experimental data (Bezerra et al., 2008).

Fig. 1(A and B) shows the surface response graphics of the CCD for the CP–C and UF–C systems, respectively. As observed, the response variables were maximised at medium values of the (CP or UF)/C ratios and at low air inlet temperatures within the range studied for each system.

3.3. Characterisation of the microparticles obtained under optimal conditions

The optimal inlet air temperature was 133 °C for both the CP–C and the UF–C systems, independently of extract type. In contrast, the optimal ratios were influenced by the extract type; indicating a lower incorporation of the encapsulating agent in the CP (3:1) than the UF (2.5:1) systems. This difference in ratios suggests that the mucilage of the pulp could act as an encapsulating agent, as was reported by Medina-Torres et al. (2013) and Sáenz et al. (2009). These results demonstrate that the optimal conditions in spray-drying are characteristic of each system studied.

High betacyanin and betaxanthin EEs (99.1–99.3% and 98.6–98.7%, respectively) were found in this study for the CP–C and the UF–C systems, without significant differences between the systems. These results agree with studies where encapsulating agents of different natures have been used (maltodextrin with different dextrose equivalent, inulin, pullulan, corn starch and gum arabic) (Sáenz et al., 2009; Gandía-Herrero, Jiménez-Atienzar, Cabanes, García-Carmona, & Escribano, 2010; Pitalua et al., 2010; Azeredo et al., 2007). These results could be explained by a high betalain-polymer interaction due to electrostatic interactions or hydrogen bonding (Kanner, Harel, & Granit, 2001; Cai & Corke, 2000).

The recovery of betacyanins in spray-drying has been reported to be mainly influenced by the following properties: encapsulating agent properties (i.e., viscosity and solubility) (Ravichandran et al., 2012), the betacyanin/encapsulating agent ratio (Cai & Corke, 2000; Azeredo et al., 2007), and the temperature (Cai & Corke, 2000). The temperature is one of the most important factors in betalain degradation (Herbach et al., 2006), emphasising the relevance of the air temperature during the drying process (Gharsallaoui, Roudat, Chambin, Voilley, & Saurel, 2007). In this study, despite the high drying temperatures, the recovery of betacyanins and betaxanthins were high (70.9–72.4% and 88.5–96.8%, respectively) for the CP–C and the UF–C systems, leading to significant differences only for betaxanthins recovery. Betalains recovery agree with the results of beetroot microparticles (88.2% and 90%) (Pitalua et al., 2010; Azeredo et al., 2007). High recovery of betalains could be attributed to short drying times or to the rapid formation of a dry crust, which allows for water diffusion but retains the bioactive

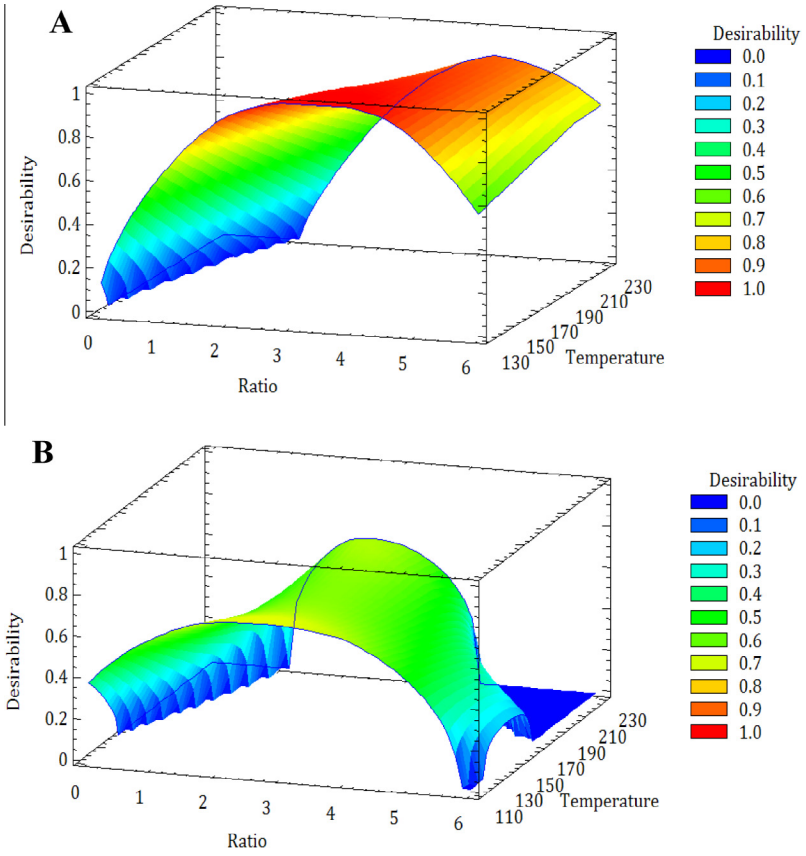

Fig. 1. Desirabilities function overlay surfaces plots: CP–C (A) and UF–C (B).

Table 2
Chemical and physical characteristics of the CP–C and UF–C microparticles obtained under optimal conditions.

Parameters	CP–C	UF–C
Moisture (%)	3.7 ± 0.3[b]	5.1 ± 0.04[a]
a_w	0.23 ± 0.01[b]	0.30 ± 0.03[a]
Hygroscopicity (g/100 g)	49.7 ± 2.4[a]	40.4 ± 0.3[b]
Colour parameters		
L (lightness–darkness)	69.9 ± 0.0[a]	68.1 ± 0.6[a]
a∗ (redness–greenness)	17.2 ± 0.1[a]	16.9 ± 0.3[a]
b∗ (blueness–yellowness)	−7.2 ± 0.0[a]	−6.6 ± 0.1[a]
h° (hue)	337.3 ± 0.1[a]	338.6 ± 0.1[a]
C∗ (chroma)	18.7 ± 0.1[a]	18.2 ± 0.3[a]
Betacyanins (mg BE/g)	0.36 ± 0.03[a]	0.36 ± 0.01[a]
Betaxanthins (mg IE/g)	0.16 ± 0.04[a]	0.16 ± 0.01[a]
Particle size D (3.2) (μm)	4.7	4.6

CP: Cactus pear pulp; UF: Ultrafiltrated extract; C: Capsul; BE: betanin equivalent; IE: indicaxanthin equivalent; GAE: gallic acid equivalent. Different letters show significantly different between systems ($p \leqslant 0.005$).
[a] Corresponds to highest values.
[b] Corresponds to lowest values

compounds (Gharsallaoui et al., 2007). Furthermore, betaxanthin recovery was slightly greater than betacyanin recovery, in accordance with the better temperature stability of betaxanthin over betacyanin (Gandía-Herrero et al., 2010; Azeredo et al., 2007). The yield ranged from 62.6% to 65.3% for UF–C and CP–C, respectively, without differences between systems.

Table 2 shows the chemical and physical characteristics of the CP–C and UF–C microparticles obtained under optimal conditions; there were no significant differences in the betacyanin, betaxanthin and polyphenol contents between the CP–C and the UF–C microparticle systems. Moreover, moisture, a_w and the particle size of the CP–C and the UF–C microparticles obtained under optimal conditions were within the range described for microparticles obtained by spray-drying (Gharsallaoui et al., 2007; Cai & Corke, 2000). The particle size (D 3,2) ranged between 4.4 and 4.7 μm, with a unimodal distribution. The colour parameters showed a slight tendency towards a purple colour. The hygroscopicity of the CP–C system was significantly higher ($p \leqslant 0.05$) than the UF–C system.

Fig. 2(A and B) shows the SEM microphotographs of microparticles obtained under optimal conditions for CP–C and UF–C, respectively. All microparticles were spherical in shape with indented surfaces and fused particles. The formation of indented surfaces observed in the SEM microphotographs could be attributed to the shrinkage of particles during the drying process (Alamilla-Beltrán, Chanona-Pérez, Jimenez-Aparicio, & Gutierrez-López, 2005), which can occur at low or high air inlet temperatures. At low air inlet temperatures, there is less water diffusion and the particles have more time to shrink. At high air inlet temperatures, the rapid evaporation and high pressure inside the particles also produce shrinkage.

3.4. Evaluation of storage stability

It is known that the encapsulation of betalains greatly improves their stability (Gandía-Herrero et al., 2010). The effect of the cactus pear extract (CP or UF) on the stability of the encapsulated betacyanins and betaxanthins at 60 °C was studied. Betacyanin and betaxanthin degradation in CP–C and UF-C microparticles, obtained under optimal conditions, followed pseudo-first order kinetics. The same order was reported for the degradation of betacyanin extracted from beetroot (Serris & Biliaderis, 2001), cactus pear microparticles (Sáenz et al., 2009), and *Amaranthus* betacyanin microparticles (Cai & Corke, 2000).

Storage conditions of beetroot microparticles, such as light, temperature and relative humidity or water activity, have been

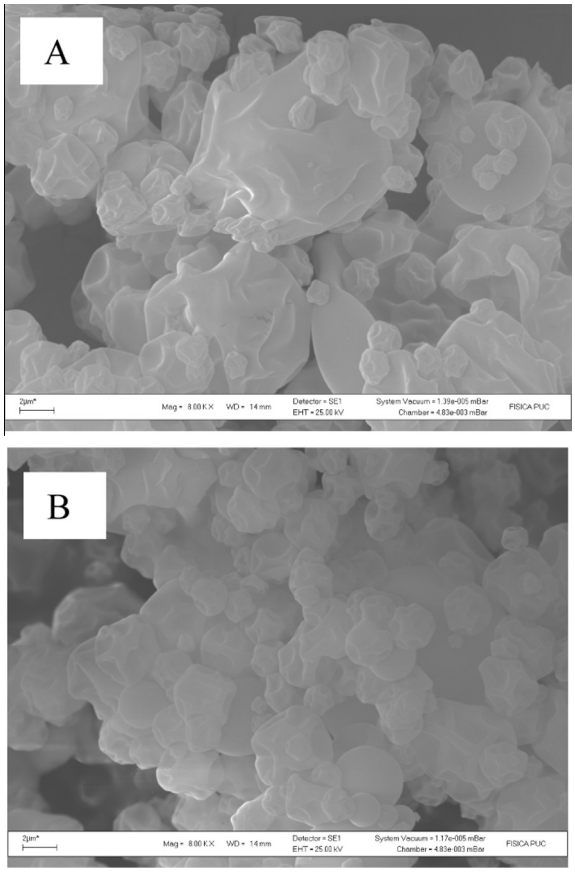

Fig. 2. Scanning electron microscopic photographs for CP–C (A) and UF–C (B).

reported to affect the stability of betanin (Serris & Biliaderis, 2001; Pitalua et al., 2010). In the present study, differences between the components of the cactus pear extracts (CP and UF), such as mucilage and/or sugars, influenced the betacyanin and betaxanthin stability during storage at 60 °C.

Table 3 shows the betacyanin and betaxanthin degradation rate constants (k_{obs}) for the CP–C and the UF–C microparticles obtained under optimal conditions, which were obtained from the slope of a plot of the natural log of the betalain retention percentage vs. time. Thus, the betacyanin and the betaxanthin degradation rate constants were significantly lower ($p \leqslant 0.05$) for UF–C than for CP–C (Table 3). The higher betanin degradation in the CP–C system could be attributed to mucilage (3.8%) and/or higher sugar content of CP (Matsuhiro, Lillo, Sáenz, Urzúa, & Zárate, 2006). Mucilage has high water absorption capacity and sugar could be solubilised, thus both increase water available in the microparticles for reactions. This is in agreement with the high hygroscopicity of the CP–C system

Table 3
Betacyanins and betaxanthins degradation rate constant at 60 °C of CP–C and UF–C microparticles obtained under optimal conditions.

System	$10^3\ k_{(obs)} \pm$ DS (days^{-1})	r^2
Betacyanins		
CP–C	8.2 ± 0.0003[a]	0.98
UF–C	7.2 ± 0.0002[b]	0.99
Betaxanthins		
CP–C	4.3 ± 0.0002[a]	0.95
UF–C	3.3 ± 0.0002[b]	0.97

Different letters show significantly different between systems ($p \leqslant 0.005$).
[a] Corresponds to highest values.
[b] Corresponds to lowest values

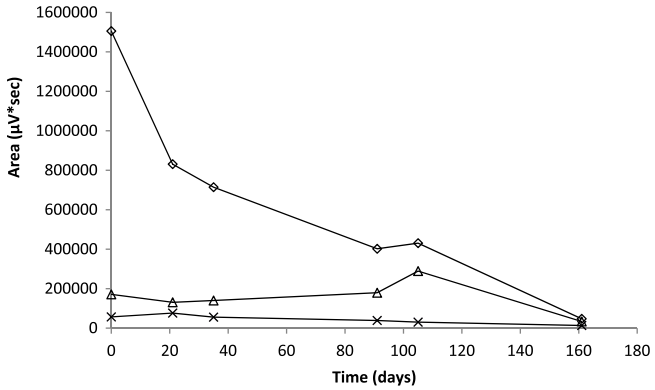

Fig. 3. Evolution of betanin and hydrolysis products formation for UF–C microparticles stored at 60 °C, (◇) betanin, (Δ) cyclodopa-D-glucoside and (x) betalamic acid.

(Table 2), which could lead to the hydrolysis of the aldimine bond of betanin. This hydrolysis reaction produces cyclodopa-D-glucoside (colourless) and betalamic acid (yellow) (Herbach et al., 2006). Although extracts with low molecular weight polymers, such as saccharides, have been reported to cause permeability and allow oxygen into the microparticle (Serris & Biliaderis, 2001), this mechanism is negligible with regard to hydrolysis. In addition, betanin degradation by hydrolysis was demonstrated in this study by HPLC, where cyclodopa-D-glucoside (280 nm) and betalamic acid (410 nm) were observed as reaction products. Fig. 3 shows the evolution of betanin and the hydrolysis products for the UF–C microparticles stored at 60 °C. The findings of the present study suggest that the degradation of betanin in microparticles occurs mainly through a hydrolysis pathway (Herbach et al., 2006), and that hygroscopicity is a critical factor of stability in microparticle storage.

4. Conclusions

In summary, the betalain encapsulation efficiency was high for both types of cactus pear extracts (CP and UF), suggesting a strong betalain-polymer interaction. However, encapsulation of the UF extract improved the stability of betalain over that of the CP extract during storage at 60 °C, as shown by lower degradation rate constants. This behaviour was attributed to the composition of the UF extract (lower sugar content than the CP extract and no mucilage). Hydrolysis was the main mechanism of betalain degradation during storage, with the critical condition being the hygroscopicity of the microparticles.

The cactus pear pulp and ultrafiltered extract microparticles described in this study represent a potential natural red colorant for use in healthy foods.

5. Declaration of interest

The authors report no conflict of interest. The authors alone are responsible for the content and writing of this article. The authors acknowledge the FONDECYT project Grant No. 1110126 (CONICYT-Chile) the CONICYT/CONACYT project Grant No. PCCI 12015 and the CONICYT scholarship Nos. 21090694 and 24110060.

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