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Supercritical CO_2 extraction of *trans*-lycopene from Portuguese tomato industrial waste

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ABSTRACT

Supercritical CO_2 extraction of *trans*-lycopene from Portuguese tomato industrial wastes (skins and seeds) was carried out in a flow apparatus. The effects of moisture content, feed initial composition, particle size, solvent flow-rate, pressure and temperature on the extraction yield and recovery were evaluated.

The recovery of *trans*-lycopene depended on the content of the compound in the starting material and increased with increases in pressure and solvent flow-rate, and with a decrease in the particle size. The effects of temperature and feed moisture content were more complex. When temperature rose from 40 to 60 °C the recovery increased, but a further rise of the temperature to 80 °C led to a decrease in the *trans*-lycopene recovery, although the total lycopene (*cis* + *trans*) remained the same as that obtained at 60 °C. On the other hand, an increase in the moisture content of the samples, from 4.6% to 22.8% led to a rise in the extraction yield and to a decrease in the recovery of *trans*-lycopene. At higher moisture contents (58.1%), both yield and recovery decreased. Moreover, the highest *trans*-lycopene recovery, 93%, was obtained at 60 °C, 300 bar, solvent flow-rate of 0.59 g/min, particle size of 0.36 mm and feed moisture content of 4.6%.

1. Introduction

Lycopene is a well-known carotenoid, responsible for the red colour of fresh tomatoes. The importance of this natural carotenoid as a colouring and antioxidant agent in the food industry has increased in recent years (Anonymous, 2003). Lycopene can also be used as nutraceutical, due to its high antioxidant activity, reducing the risk of atherosclerosis and coronary heart disease. Moreover, epidemiological studies have related the intake of lycopene with a lower risk of the incidence of certain types of cancers (Shi & Le Maguer, 2000).

The major sources of lycopene in the human diet are tomato and tomato products (Schwartz, Hadley, Miller, & Clinton, 2002), although watermelon, apricot, papaya, pink grapefruit and rosehip are also dietary sources, but with lower contents (Bohm, Frohlich, & Bitsch, 2003; Perkins-Veazie & Collins, 2004; Pol, Hyitylainen, Ranta-Aho, & Riekkola, 2004). Lycopene can represent about 80–90% of the total carotenoids in tomato (Sharma & Le Maguer, 1996; Shi & Le Maguer, 2000). Its content in this vegetable source can vary widely, from 10 to 200 mg/kg (wet basis), with typical values of 40–90 mg/kg (wet basis) (Brandt, Pék, Barna, Lugasi, & Helyes, 2006; Dumas, Dadomo, Di Lucca, & Grolier 2003; Shi & Le Maguer, 2000), depending on many parameters, such as genetic factors (tomato variety), environmental factors (temperature and light) and agricultural techniques (water, mineral nutrients, etc.) (Brandt et al., 2006; Dumas et al., 2003). Most of the compound (70–90%) is located in the insoluble fraction and in the seeds, which can contain about five times more lycopene than tomato pulp (Sharma & Le Maguer, 1996; Shi & Le Maguer, 2000; Toor & Savage, 2005). Al-Wandawi, Abdul-Rahman, and Al-Shaikhly (1985) quantified the lycopene in tomato skins and obtained a value of 120 mg/kg (wet basis). This value was close to that obtained by Toor and Savage (2005). Both authors verified that the seeds had much lower lycopene content. This fact was also checked by Sábio et al. (2003), who found that the lycopene extraction yield was higher in samples consisting only of skins than in those containing skins and seeds. Also, it was verified that lycopene from processed tomato (submitted to heating and trituration) had more bioavailability than that of raw tomatoes (Britton, Gambelli, Dunphy, Pudney, & Gidley, 2002; Schwartz et al., 2002; Shi & Le Maguer, 2000).

The tomato processing industry produces large amounts of solid waste. About 10–40% of the total tomato processed in the facility are as skins and seeds (Al-Wandawi et al., 1985; Topal, Sasaki, Goto, & Hayakawa, 2006), which are usually used for animal feed (Knoblich, Anderson, & Latshaw, 2005). This waste is obtained after

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the trituration and pre-heating (65–70 °C) of the processed tomatoes. The fact that skins are rich in lycopene and have been submitted to such treatment makes tomato industrial waste a suitable source of this carotenoid and its extraction a good alternative use for this by-product (Schieber, Stintzing, & Carle, 2001; Shi, Mittal, Kim, & Xue, 2007).

Extraction of carotenoids from vegetal sources is usually carried out using organic solvents, e.g., *n*-hexane, acetone, chloroform, ethanol, etc. These hazardous solvents possess many disadvantages, such as toxicity, disposal, difficult separation from final product (presence of solvent traces) and the need to work at high temperatures. Supercritical fluid extraction using CO_2 is a suitable alternative to the conventional extraction techniques of biological products, since this solvent allows working at moderate temperatures, is non-toxic and easily separated from the extract.

Some studies concerning the supercritical CO_2 extraction of lycopene from tomato by-products have already been published, such as those of Baysal, Ersus, and Starmans (2000), Rozzi, Singh, Vierling, and Watkins (2002); Favati, Pietrafessa, & Galgano, 2003; Sábio et al. (2003); Pol et al. (2004); Topal et al. (2006) and Vagi et al. (2007). However, there was a large variation in their results. However, in most of these studies the effect of the content of lycopene in the initial feed and pre-treatment of the samples is not considered. Also, most of them deal with the extraction of both *cis* and *trans* isomers of lycopene, not distinguishing the compounds. The lycopene in fresh tomato occurs predominantly in the *all-trans* form (Schierle et al., 1997), which is the most thermodynamically stable isomer, and so this isomer is more suitable to manipulate and incorporate in functional foods and nutraceuticals than the *cis* form. On the other hand, some studies attribute to the *cis* isomer a higher bioavailability (Boileau, Boileau, & Erdman, 2002; Schieber & Carle, 2005; Schierle et al., 1997), but the mechanisms related to the *trans–cis* isomerisation process are not yet clear and some studies indicate that this process can occur in the human digestive system (Boileau et al., 2002), which would favour the intake of *trans*-lycopene.

The objective of this work was to investigate the effect of moisture and *trans*-lycopene content of the starting material, particle size, solvent flow-rate, pressure and temperature on the extraction of this isomer from tomato industrial wastes obtained from a Portuguese tomato processing unit.

2. Materials and methods

2.1. Materials

Carbon dioxide (99.998% purity) was purchased from Air Liquide (Portugal). Tomato industrial waste (a mixture of skins and seeds) were supplied by a local tomato processing company: FIT – Fomento da Industria do Tomate, S. A. Standard of all-*trans*-lycopene (90–95% purity) was obtained from Sigma–Aldrich (St. Louis, MO). Acetone (p.a.), methanol (HPLC grade), acetonitrile (HPLC grade), and *n*-hexane (p.a) were purchased from Merck (Darmstadt, Germany).

2.2. Sample preparation

Three samples of Portuguese tomato industrial waste (mixture of skins and seeds) were used: **M1**, **M2** and **M3**. Samples **M1** and **M2** were collected at the same time and sample **M3** was obtained on a different occasion.

Sample **M1**, with a moisture content of 82.9%, was dried, immediately after being collected, in an oven at 40 °C. Different times of drying were used and so three batches, with different moisture contents, were obtained: 4.6%, 22.8% and 58.1%. The dried tomato wastes were then packed under nitrogen and stored at −20 °C. These samples were used for experiments concerning the effect of moisture content, particle size and solvent flow-rate.

Samples **M2** and **M3** were also dried in an oven at 40 °C and a moisture content of 4.6% was achieved, but whilst sample **M3** was dried immediately after being collected, sample **M2** was stored (as fresh, under nitrogen at −20 °C) for 4 months and then dried. The dried tomato wastes were then packed under nitrogen and stored at −20 °C for further use. Sample **M2** was used for experiments concerning the effect of temperature and pressure, whilst sample **M3** was used to study the effect of the feed composition.

All samples were ground prior to the supercritical fluid extraction measurements using a cutting mill. For experiments concerning the effect of particle size, three samples of tomato paste with moisture content of 4.6% and mean particle size of 0.15, 0.36 and 0.72 mm were prepared. For the experiments concerning the effect of the flow-rate, samples of tomato paste with a moisture content of 4.6% and an average particle size of 0.48 mm were used. Furthermore, for the experiments concerning the effect of moisture content, four samples were studied: 58.1% moisture content (mean particle size = 0.70 mm), 22.8% moisture content (mean particle size = 0.76 mm) and 4.6% moisture content (mean particle size = 0.48 mm). For the remaining experiments an average particle size of 0.36 mm was used.

Lycopene (*cis* + *trans*) and lipids of the Portuguese tomato industrial wastes were determined by Soxhlet extraction with acetone:hexane (1:1) for 6 h. The amounts of *trans*-lycopene and *cis*-lycopene were quantified by HPLC (see below). The amount of lipids (Soxhlet extract) was determined gravimetrically. The obtained extracts were concentrated in a vacuum evaporator and the residue was weighed. *trans*-Lycopene, *cis*-lycopene and lipid contents of the three samples, and treatments prepared from these starting materials are shown in Table 1.

2.3. Experimental procedure

The supercritical studies were carried out in a flow-type apparatus described in a previous paper (Mendes et al., 1995). The liquid solvent was pumped from the cylinder to the extraction vessel through a metering pump, the pressure controlled by a back-pressure regulator. In order to guarantee that the fluid reaches the extraction vessel at the desired temperature, it passes through a heat exchanger, which involves a coil and a temperature-controlled water bath. After flowing through the extraction vessel, the fluid is expanded to atmospheric pressure using a three-way valve and the extract precipitates inside a cooled glass U-tube, which contains glass wool. Gas flow-rate is monitored by a rotameter and total volume of gas is measured with a wet test meter.

The extraction vessel consisted of a 5-cm^3 pressure cell, with an internal diameter of 7.9 mm, which was filled with about 1.5 g of tomato industrial waste, packed between two layers of glass wool.

Fractions of 5–10 l of expanded gas were collected over time. Three solvent flow-rates were tested: 0.26 g/min (surface velocity of 0.63 cm/min), 0.59 g/min (surface velocity of 1.44 cm/min) and 1.18 g/min (surface velocity of 2.58 cm/min).

The extracts were collected by washing the glass wool, the U-tube, the three-way valve and the expansion tubing with acetone. The collected solutions were analysed by HPLC, in order to quantify the amount of *trans*-lycopene and *cis*-lycopene extracted. The system consisted of a liquid chromatograph, Hewlett Packard 1100 series, with UV/Vis detector adjusted to 470 nm. A mobile phase of methanol:acetonitrile (90:10 v/v) was used at 1 ml/min with a reversed phase column (250 × 4.6 mm, Vydac 201 TP54; Grace, Deerfield, IL). *trans*-Lycopene was identified by comparing the

Table 1
trans-Lycopene, *cis*-lycopene and lipid content of the samples (Soxhlet extraction).

Sample	Mean particle size (mm)	Moisture content (%)	*trans*-Lycopene ($\mu g/g_{oil\text{-}free\ dry\ matter}$)	*cis*-Lycopene ($\mu g/g_{oil\text{-}free\ dry\ matter}$)	Lipids ($mg/g_{oil\text{-}free\ dry\ matter}$)
M1a	–	82.9	691	63.9	201
M1b	0.70	58.1	560	–	299
M1c	0.76	22.8	578	–	211
M1d	0.72	4.6	52.1	14.7	107
M1e	0.48	4.6	245	–	313
M2	0.36	4.6	154	34.8	202
M3	0.36	4.6	254	76.3	199

retention time of the carotenoid with that of the standard compound, which was also used to obtain calibration curves, in order to determine its total amount. The *cis*-lycopene content was determined by comparing the chromatographic areas of the *cis* and *trans* isomers and considering a response factor of 1. The amount of lipids in the supercritical extracts was measured gravimetrically, concentrating each fraction collected and weighing the obtained residue.

3. Results and discussion

3.1. Effect of moisture content

To our knowledge, all the studies dealing with supercritical fluid extraction of lycopene from tomato or tomato industrial waste only used fresh, or well-dried samples (moisture content of 2.3–10%). It has been shown that the supercritical extraction of compounds from fresh tomatoes or fresh tomato industrial wastes resulted in a low extraction recovery of lycopene (Favati et al., 2003; Vasapollo, Longo, Rescio, & Ciurlia, 2004). Also, it has been reported that the lycopene content of dried samples was lower than that of fresh tomato waste samples (Favati et al., 2003).

In this work, fresh tomato industrial wastes (mixture of skins and seeds), with moisture content of 82.9%, were dried to several moisture contents: 58.1%, 22.8% and 4.6%. Soxhlet extraction of these samples (Table 1) showed that the *trans*-lycopene content of tomato waste dried to moisture contents of 58.1% and 22.8% was only slightly lower than that of the fresh (82.9%) material. On the other hand, the *trans*-lycopene content decreased to half of the initial value when tomato waste was dried to 4.6% of moisture content. This reduction in the *trans*-lycopene content of the dried sample was unlikely to be due to degradation of this compound during the drying of the sample, because HPLC at 470 and 450 nm of the Soxhlet organic extracts did not detect any degradation product. Most probably, the decrease in *trans*-lycopene content of the dried sample could be due to physical changes in the structure of the skins (Brunner, 1994); namely, lipid pillars of a plant cell elementary membrane change with the water content and, if there is not enough water in the system the pillar closes the membrane making it impermeable (Brunner, 1994).

Fresh tomato industrial waste (82.9% moisture content) was submitted to supercritical CO_2 extraction at 60 °C, 300 bar and solvent flow-rate of 0.59 g/min, but there was no extraction of *trans*-lycopene except in trace amounts. Supercritical CO_2 extraction of samples with different moisture contents (4.6%, 22.8% and 58.1%) was carried out at 60 °C, 300 bar and solvent flow-rate of 0.59 g/min. In Fig. 1A and B is shown the *trans*-lycopene yield of extraction (mass of extracted lycopene/mass of oil-free dry matter) and its percentage recovery (mass of extracted lycopene by supercritical CO_2/mass of extracted lycopene by Soxhlet), respectively, for the extraction curves of the samples with different moisture contents. For the purpose of comparison it is shown in these figures the results obtained for two samples with 4.6% moisture content, one sample with a mean particle size of 0.72 mm (value near those used for the samples with different moisture contents) and the other sample with a mean particle size of 0.48 mm.

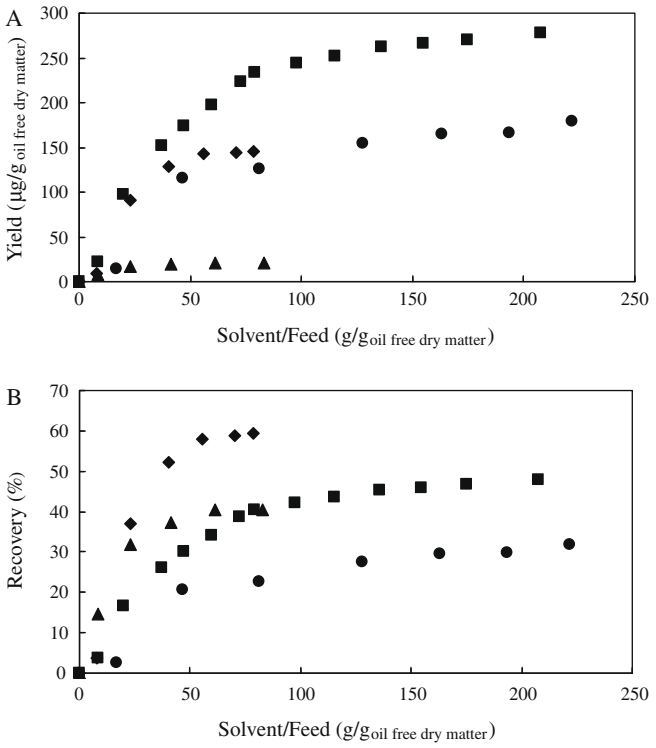

Fig. 1. (A) Yield and (B) recovery of *trans*-lycopene as function of CO_2 amount, at 60 °C, 300 bar, solvent flow-rate of 0.59 g/min (solvent surface velocity of 1.44 cm/min). (♦) 4.6% moisture content, 0.48 mm mean particle size; (▲) 4.6% Moisture content, 0.72 mm mean particle size; (■) 22.8% moisture content, 0.76 mm mean particle size; (●) 58.1% moisture content, 0.70 mm mean particle size.

From Fig. 1A it was verified that the extraction yield of *trans*-lycopene rose when the moisture content of the sample increased from 4.6% to 22.8%, possibly due to the modifications in the skin structure mentioned before, implying less available compounds in tomato waste with lower moisture content. However, for higher moisture contents the yield decreased, in this case, probably due to the water preventing contact between CO_2 and tomato particles (Ge et al., 2002). Moreover, water has higher solubility in CO_2 than *trans*-lycopene (de la Fuente, Oyarzún, Quezada, & del Valle, 2006; Goméz-Prieto, Caja, & Santa-Maria, 2002; Sabirzyanov, Il'in, Akhunov, & Gumerov, 2002; Topal et al., 2006), and possibly some competition with the extraction of carotenoids can occur.

It is also shown (Fig. 1A) that although the yield of extraction decreased due to the dryness of the sample at low moisture, the reduction in the particle size can increase the yield of extraction, allowing yield values near those obtained for the samples with higher moisture content (58.1%) and at the same time consuming less solvent. In effect, the samples with higher moisture contents (22.8% and 58.1%) consumed a higher amount of solvent because

the lycopene content was also higher. On the other hand, the recovery percentage of *trans*-lycopene tends to increase when the moisture content of the samples decreased (Fig. 1B). The sample with the lowest moisture content (4.6%), and with a mean particle size of 0.48 mm, presented the highest recovery of *trans*-lycopene. Comparing the results obtained for this sample with those obtained for the sample with the same moisture content (4.6%), but with a higher mean particle size (0.72 mm) it can be seen that reduction of particle size improved the recovery of *trans*-lycopene. Also, from Fig. 1B, comparing the recovery of *trans*-lycopene for the sample with 4.6% moisture content and 0.72 mm mean particle size, with that obtained for the sample with 22.8% moisture content, it can be seen that the recovery is almost the same for both samples. Increasing the moisture content to 58.1% led to a lower recovery of *trans*-lycopene.

3.2. Effect of feed initial composition

Two samples of tomato industrial waste, **M2** and **M3**, with different *trans*-lycopene contents (Table 1), moisture content of 4.6% and particle size of 0.36 mm, were submitted to supercritical CO_2 extraction at 60 °C, pressure of 300 bar and solvent flow-rate of 0.59 g/min. The analysis of the starting material (Table 1) showed that the sample of tomato waste **M3** had a higher content of *trans*-lycopene.

It can be seen (Fig. 2) that when the starting material had a lower content of *trans*-lycopene the recovery was faster and a higher value was achieved (93%). On the other hand, when the starting material had a higher content of *trans*-lycopene the solvent/feed ratio was much higher (220 g/g) with a consequently much slower extraction. As the amount of CO_2 needed for the extraction of **M3** was also much higher than that necessary for the extraction of sample **M2**, for the same flow-rate, the extraction time increased having as a possible consequence some degradation of the *trans*-lycopene in the extraction cell, lowering the recovery of *trans*-lycopene from the sample **M3**.

3.3. Effect of feed particle size

The effect of the particle size on the supercritical extraction behaviour was studied more intensely, submitting three samples of tomato industrial waste, with different mean particle sizes (0.15, 0.36 and 0.72 mm) and moisture content of 4.6%, to supercritical CO_2 extraction at 60 °C, 300 bar and solvent flow-rate of 0.59 g/min. In Fig. 3 is shown the recovery percentages of *trans*-lycopene for the three samples studied. As it can be seen, the

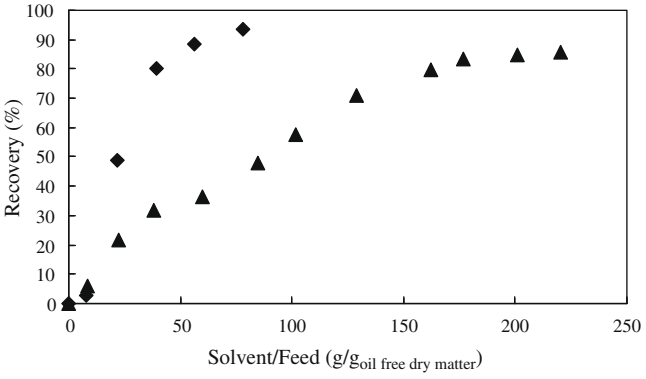

Fig. 2. Recovery of *trans*-lycopene as a function of CO_2 amount, at 60 °C, 300 bar, solvent flow-rate of 0.59 g/min (solvent surface velocity of 1.44 cm/min). Samples with 4.6% moisture content and mean particle size of 0.36 mm. (♦) Sample **M2** (128.1 μg/g, *trans*-lycopene); (▲) sample **M3** (213.5 μg/g, *trans*-lycopene).

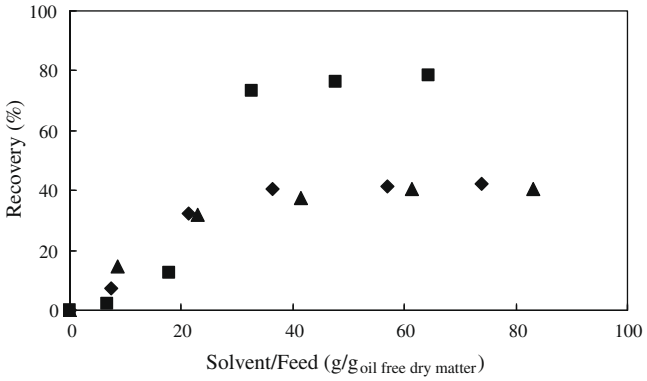

Fig. 3. Recovery of *trans*-lycopene as a function of CO_2 amount, at 60 °C, 300 bar and solvent flow-rate of 0.59 g/min (solvent surface velocity of 1.44 cm/min), sample **M1**, moisture content of 4.6%. (■) Mean particle size of 0.15 mm; (♦) mean particle size of 0.36 mm; (▲) mean particle size of 0.72 mm.

recovery of *trans*-lycopene was almost the same for the samples with mean particle sizes of 0.72 and 0.36 mm, although the recovery value was slightly higher for the sample with the lower mean particle size (0.36 mm), showing a trend for an increase in the recovery of *trans*-lycopene with a decrease in the particle size. When the particle size decreased to 0.15 mm the recovery of *trans*-lycopene increased drastically, reaching a value of 78%. The obtained results are expected, since the particle size reduction of plant material increases the lipids recovery, because it may cause ruptures of the cell walls and therefore increase the surface area of contact with CO_2 (Eggers, 1996).

On the other hand, Fig. 3 shows also that in the initial part of the extraction the recovery of *trans*-lycopene was slower for the sample with the lower particle size and faster for the sample with the higher particle size. The particle size reduction process can increase the solute accessible for the solvent, as was verified by the Soxhlet extraction of the three samples, which showed that the lipid content highly increased for the sample with lower particle size (lipid contents of 22.1, 15.3 and 9.7 g/100 g dry matter were obtained for the samples with mean particle sizes of 0.15, 0.36 and 0.72 mm, respectively). So, it is possible that, for the sample with lower particle size, the possible effect of competition between carotenoids and lipids for the supercritical solvent (the extraction of carotenoids occurs only after the major part of the lipids, namely the triglycerides which, have higher solubility in CO_2, have been extracted) was more accentuated. Therefore, the rate of *trans*-lycopene extraction was lower at the beginning of the extraction.

Nevertheless, the use of very small particle sizes can have some problem, since smaller particles can cause packing of the extraction bed, which can result in channelling effects (Brunner, 1994). Sábio et al. (2003) verified that the extraction of lycopene from tomato wastes decreased at very small particle sizes (0.08 mm). These authors attributed this fact to the channelling of the extraction bed. On the other hand, Reverchon and De Marco (2006) suggest, for supercritical extraction processes, optimal particle diameters between 0.25 and 2.00 mm.

3.4. Effect of solvent flow-rate

To examine the effect of flow-rate, supercritical CO_2 extraction experiments were carried out at 60 °C, pressure of 300 bar, using sample **M1** with mean particle size of 0.48 mm, and moisture content of 4.6%. Three different flow-rates were tested: 0.26, 0.59 and 1.05 g/min (surface velocities of 0.63, 1.44 and 2.58 cm/min, respectively). In Fig. 4 A and B are shown the results for the recovery of *trans*-lycopene, as a function of the extraction time and of the solvent/feed ratio, respectively. It was verified that the

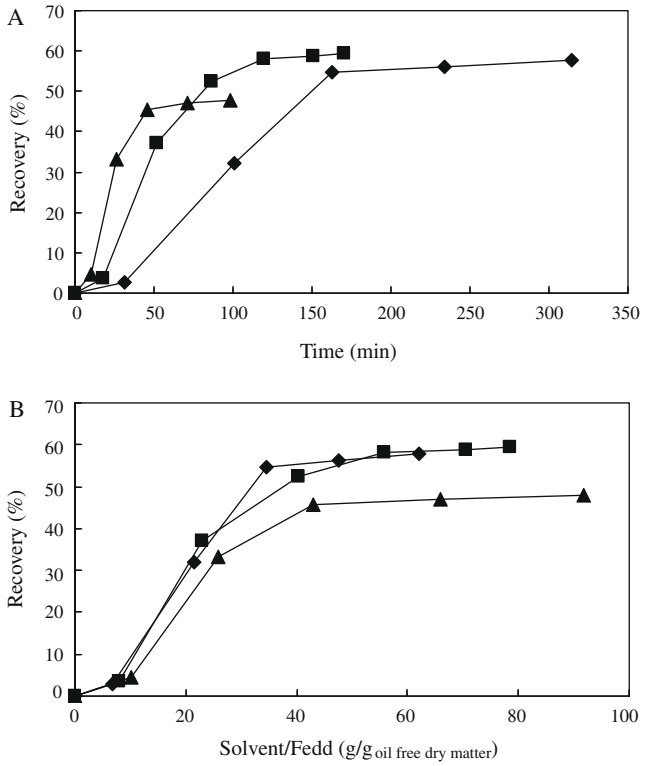

Fig. 4. Recovery of *trans*-lycopene at 60 °C, 300 bar, sample **M1**, moisture content of 4.6% and mean particle size of 0.48 mm, for several solvent flow-rates (solvent superficial velocity), as a function of (A) time, (B) CO_2 amount. (♦) 0.26 g/min (0.63 cm/min); (■) 0.59 g/min (1.44 cm/min); (▲) 1.05 g/min (2.58 cm/min).

recovery of *trans*-lycopene increased when the flow-rate decreased from 1.05 to 0.59 g/min, but remained practically the same for the lowest flow-rates (0.26 g/min). From Fig. 4A it can be seen that although the recovery of *trans*-lycopene, at the beginning of the extraction, was faster at higher flow-rates, at the end of extraction the recovery is almost the same at the lowest flow-rates (0.26 and 0.59 g/min) and is higher than that obtained for the highest flow-rate (1.05 g/min). However, Fig. 4B shows that the extraction curves agree at the lowest flow-rates (0.26 and 0.59 g/min), but the extraction curve for the highest flow-rate is below those. These results could mean that the mass transfer process is controlled by the equilibrium between the solid and the fluid phase. The fact that the highest flow-rate presented the lowest recovery could possibly be due to channelling effects and the impossibility of reaching equilibrium at such a high flow-rate.

3.5. Effect of pressure

The effect of the pressure on the extraction recovery was also studied. In Fig. 5 is represented the recovery of *trans*-lycopene (from sample **M2**), at temperature of 60 °C, CO_2 surface velocity of 1.44 cm/min and pressures of 200 and 300 bar. This figure shows that when the pressure is lowered from 300 to 200 bar the recovery of *trans*-lycopene decreases drastically (from 93% to 30%) and the extraction is much slower. This could probably be due to the fact that *trans*-lycopene solubility, at 60 °C, is much lower at 200 bar than at 300 bar (de la Fuente et al., 2006; Goméz-Prieto et al., 2002) and also because, at a pressure of 200 bar, there could possibly exist a competition between carotenoids and the lipids (oil from crushed seeds) for the supercritical solvent, and so the extraction of *trans*-lycopene occurred only after the major part of the lipids, namely the triglycerides, which have higher solubility in CO_2, were extracted.

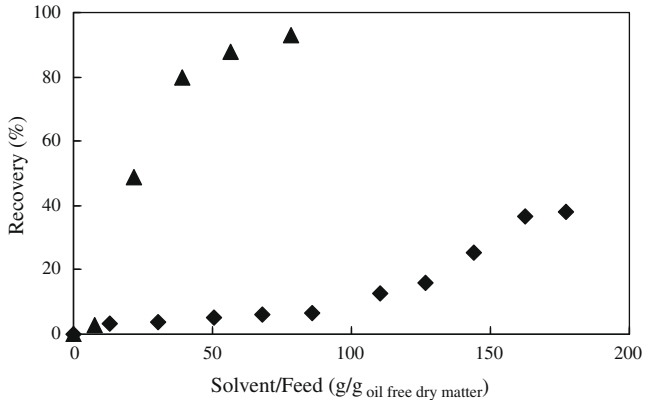

Fig. 5. Recovery of *trans*-lycopene as a function of CO_2 amount, at 60 °C, solvent surface velocity of 1.44 cm/min, sample **M2**, moisture content of 4.6% and mean particle size of 0.36 mm, for the two pressures studied. (♦) 200 bar; (▲) 300 bar.

3.6. Effect of temperature

The effect of temperature is represented in Fig. 6A and B for *trans*-lycopene and total lycopene (sum of *cis* and *trans* isomers), respectively. When the temperature increases from 40 to 60 °C the recovery of *trans*-lycopene rises from 40% to 93%, but if the extraction temperature is further increased to 80 °C there is a decrease in the recovery of this isomer. On the other hand, for total lycopene (*cis* and *trans*), the recovery increases when the temperature rises from 40 to 60 °C, and remains almost the same with further increase of temperature to 80 °C. This could possibly indicate that there could have been some isomerisation of *trans*-lycopene promoted by the higher temperature. Lee and Chen (2002)

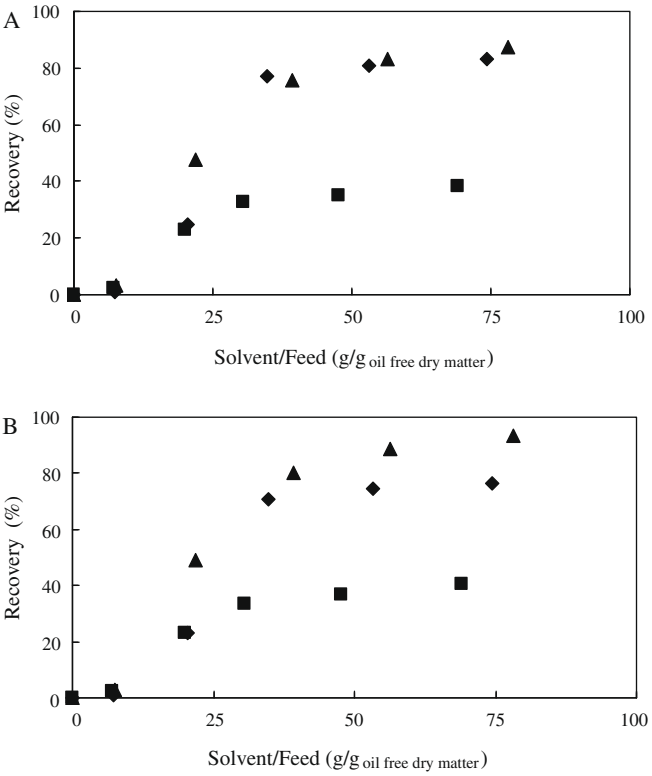

Fig. 6. Recovery of (A) *trans*-lycopene, (B) total lycopene (*cis* + *trans*), as a function of CO_2 amount, at 300 bar, solvent surface velocity of 1.44 cm/min, sample **M2**, moisture content of 4.6% and mean particle size of 0.36 mm, at several temperatures studied. (♦) 80 °C, (▲) 60 °C, (■) 40 °C.

checked the isomerisation of *trans*-lycopene to *cis*-lycopene at temperatures higher than 50 °C and Wang and Chen (2006) reported that at temperatures of 80 and 90 °C there was a maximum *trans–cis* isomerisation of lycopene in the supercritical CO_2 extraction of this carotenoid from tomatoes. Some degradation could also have occurred at 80 °C.

4. Conclusions

The supercritical CO_2 extraction of *trans*-lycopene from tomato industrial waste was carried out using a flow-type apparatus. It was shown that the moisture content of the feed is an important parameter to be optimised. The recovery of *trans*-lycopene is higher and faster for matrices with lower moisture content, although the drying of tomato waste to lower values of moisture (4.6%) led to a significant decrease in its *trans*-lycopene content.

Is was also shown that when two samples of tomato industrial waste, with different *trans*-lycopene content, were submitted to supercritical CO_2 extraction at 60 °C and 300 bar, the recovery of *trans*-lycopene was faster and higher (about 93%) for the sample with a lower *trans*-lycopene content.

The recovery of *trans*-lycopene increased with decrease of the particle size and rose significantly with increase of pressure from 200 to 300 bar. On the other hand, the rise in temperature, to values higher than 60 °C, can lead to isomerisation of this carotenoid. Moreover, it was verified that working with high surface velocities (2.58 cm/min) of supercritical fluid can lead to a lower recovery of lycopene.

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