LYCOPENE EXTRACTION FROM PROCESSED TOMATOES USING SUPERCRITICAL CO_2

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This work investigates the effects of the transformation processes of tomatoes especially using supercritical CO_2 on bioavailability and extractability of lycopene.
The experimental extraction curves measured on dried vegetable samples, obtained from different transformation processes of tomatoes, have been correlated with an equation of the type of a Langmuir gas adsorption isotherm and with a mathematical model based on differential mass balances written on a section of the extraction bed. In the second case it has been verified, both from literature and experimental data, that only the external mass transfer resistance controls the extraction process and under this hypothesis the model was developed by using a constant value for the mass transfer coefficient.

INTRODUCTION

Lycopene is a carotenoid mainly contained in the fresh tomatoes as red pigment. The importance of this compound is due to its antioxidant activity, which is fundamental in the treatment of cardiovascular diseases, aging degenerations and some cancer forms [1, 2].
Lycopene is very stable inside the vegetable cells of the deep red tomatoes but unfortunately in this situation its bioavailability is very low. On the opposite the assimilation of the carotenoid is really easy from cooked foods especially in presence of oils and fats because of the high solubility of lycopene in non polar solvents. Processing and packaging the fresh tomatoes lead to the degradation of the pigment by oxidation and isomerization.
Consequently it is important to define the relations between the concentration of lycopene and the variables (as air, light and heat) influencing the preservation of the active principle during the transformation processes of tomatoes [3, 4].
Thermal processing has been widely employed for concentrating and increasing the bioavailability of lycopene from ripe tomatoes: it has been demonstrated, by different authors, that this kind of treatment doesn't produce relevant losses of active principle [5].
A recent process employs high hydrostatic pressures to preserve active principles contained in vegetables and fruits. In this case principle concentrations and antioxidant activity don't change but the treatment induces structural modifications in the vegetable tissues, which are responsible for a different extractability of the compounds held inside the cells.

I - MATERIALS AND EQUIPMENT

Materials

Fourteen different dried vegetable matrices obtained from transformation processes of fresh tomatoes, which lead to the production of juices, puree, concentrates and 18°Brix concentrates, have been supplied by Stazione Sperimentale per l'Industria delle Conserve Alimentari (SSICA) - Parma (I).

Carbon dioxide from SIAD – Trieste (I), with purity higher than 99,98%, has been used as supercritical solvent.
Methanol (MeOH) and Tetrahydrofurane (THF) have been purchased by Fluka and employed to recover the extracted material and to determine the yield of the process.

Equipment for extraction

In figure 1 a schematic diagram of the experimental apparatus for the lycopene extraction with supercritical CO_2 (SF-CO_2) is shown.

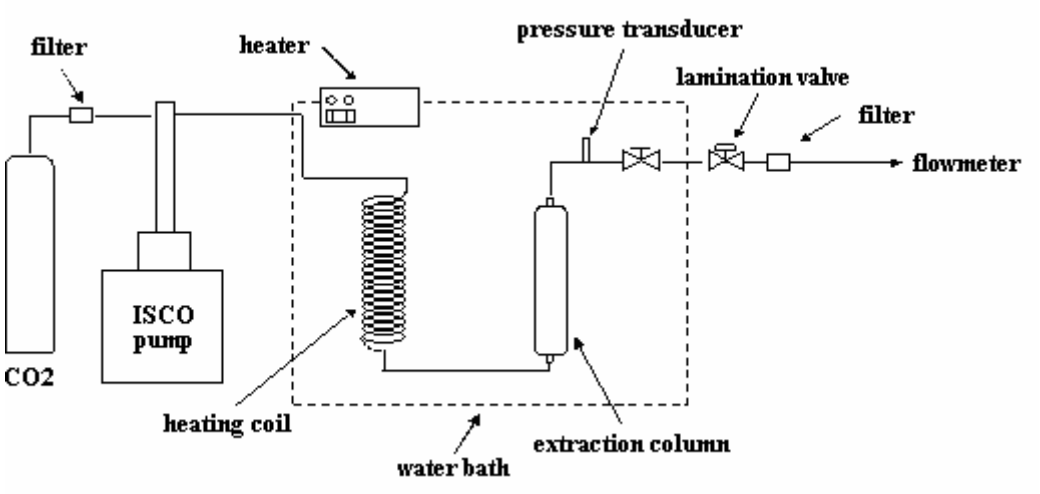

Figure 1: Experimental apparatus for extraction with SF-CO_2.

The CO_2 is supplied by a high pressure bomb and is pumped through the plant by an ISCO syringe pump (260D model) working at constant pressure. The cylinder of the pump is cooled to 0°C by a cooling unit (Haake F3) in order to keep the CO_2 in liquid phase during compression.
The CO_2 flows first through a heating coil and then through a stainless steel column (length 100 mm, inner diameter 10 mm) containing the vegetable matrix. This stuff is placed inside a water bath and the temperature is kept constant by a heater Haake DC3 (±0.1°C). The pressure is monitored with a pressure transducer Druck DPI 260 (±0.1%).
The total volume of CO_2 employed during each extraction test is measured with a flowmeter Brokhorst S200D (± 0.001 l).
At the outlet of the water bath the lycopene extracted by SF-CO_2 precipitates in the lamination valve and in the following filter (0.5 μm), due to the sudden expansion of the solvent from the process to the ambient conditions.
Lycopene is collected from the lamination valve and from the filter by using a known volume of THF/MeOH solution (1:1 by weight). The recovered solution is analysed with a UV spectrophotometer Unicam Heλios α to determine the lycopene concentration related to the maximum absorbance peak of the substance (λ_{max}= 472nm). The absorbance of a solution of 1g/100ml ($E^{1\%}_{1cm}$) at 472nm is equal to 3450 (Merck manual [6]) and this information gives the possibility to determine the amount of lycopene extracted after each extraction run.

II - RESULTS AND DISCUSSION

Preliminary studies on the tomatoes matrices have been performed in order to determine the treatments that preserve and concentrate better lycopene. Fourteen different samples were treated with THF/MeOH (1:1 by weight) and the solutions were analysed with the UV

spectrophotometer,after one day, in order to determine the active principle quantity after the complete release of lycopene from the vegetable matrix.
Afterwards, the extraction with SF-CO_2 has been carried out on the samples with higher concentrations of carotenoids, since the more concentrated samples can be effective in biological applications.
Six samples have been selected to perform the extraction of lycopene (see table 1).

Sample (related treatment)	**[mg] lycopene/ [g] matrix**
D (UHP of 18°Brix concentrate)	1.66
E (pasteurization of concentrate)	1.46
M (18°Brix concentrate)	1.65
N (UHP juice)	1.53
O (fresh tomatoes)	1.84
S (cold juice)	1.48

Table 1: Lycopene concentrations in the tomato matrixes (by UV technique).

The experimental conditions for extraction with supercritical CO_2, 300bar, 60°C and 450 ml/min for CO_2 flow rate, have been selected considering the operative limits of the laboratory plant and the literature suggestions [7, 8, 9, 10].
The experimental results obtained for each sample have been reported as the extraction curves representing yield expressed as [mg of lycopene]/[g of vegetable matrix] vs. time [minutes].
Initially an empirical model based on a function of the Langmuir gas adsorption isotherm type has been employed to fit the experimental extraction yield Y [11]:

$$Y = \frac{Y_\infty \cdot t}{B + t} \tag{1}$$

where Y_∞ is the yield after an infinite extraction time, t is the extraction time and B is a constant.
A model based on differential mass balances has also been used in order to have an instrument which gives a better understanding of what is the physical insight of the process [12, 13, 14].
With this aim the experimental curves have been correlated by using differential material balances written on a section of the extraction column and by considering a constant mass transfer coefficient:

$$\frac{\partial c}{\partial t} + \frac{u}{e} \cdot \frac{\partial c}{\partial z} + \frac{1-e}{e} \cdot \frac{r_s}{r} \cdot \frac{\partial q}{\partial t} = 0 \tag{2}$$

$$\frac{\partial q}{\partial t} = -k_e \cdot a \cdot (q / K - c) \tag{3}$$

where c is the fluid phase concentration (g/g), q the solid phase concentration (g/g), t the time (min), z the axial coordinate of the column (cm), u the superficial velocity (cm/min), ε the void fraction, ρ the fluid phase density (g/cm^3), ρ_s the solid phase density (g/cm^3), a the particle's surface.
The process has been modelled under the following hypothesis:

- constant u/ε;
- negligible axial dispersion;
- phase equilibrium expressed with a linear relation $q = K \cdot c$ (K is the equilibrium constant);
- only the fluid phase mass transfer controls the extraction process (k_e is the external mass transfer coefficient obtained by fitting the experimental data with the Langmuir isotherm);
- initial conditions: $t = 0,\ \ q = q_0,\ \ c = 0$;
- boundary condition: $z = 0,\ \ c = 0$ (c_0 is the fluid phase concentration at equilibrium);

In the next pictures the experimental curves correlated using the two models are reported for samples D, M and E, which gave the higher extraction yields.

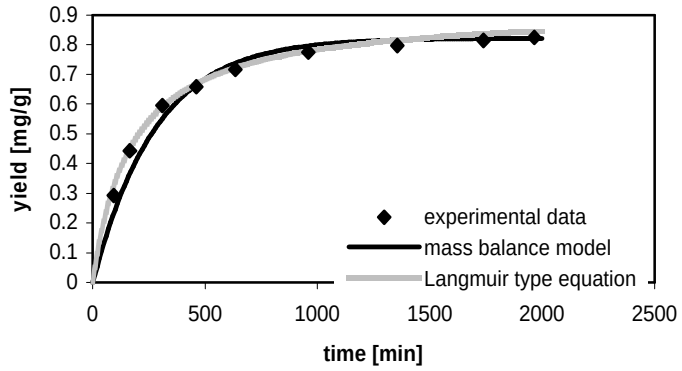

Figure 2: Extraction trends at 60°C and 300 bar for sample D.

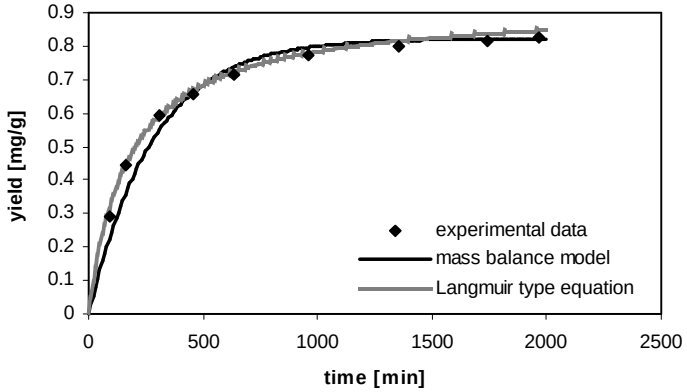

Figure 3: Extraction trends at 60°C and 300 bar for sample M.

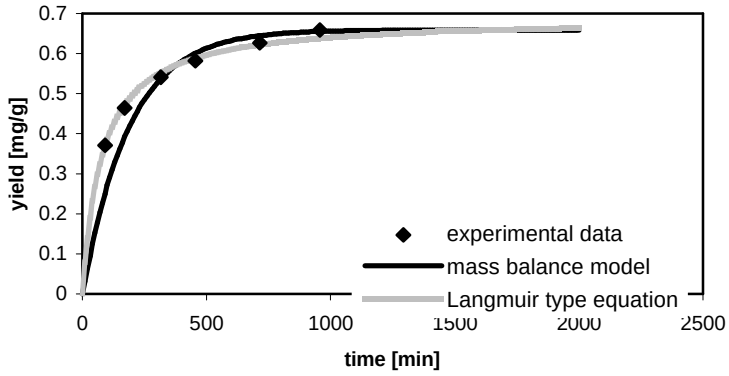

Figure 4: Extraction trends at 60°C and 300 bar for sample E.

A good agreement between the experimental data and the models can be evidenced. Furthermore it is necessary to notice the influence of the tomato treatments on the extractability of lycopene. Samples D, M and E, containing the higher percentages of lycopene, are produced from concentrated tomatoes: in particular samples D and M are obtained both from 18°Brix concentrated matrices. Sample D has been also subjected to a high pressure treatment and shows the best response towards the extraction with SF-CO_2.
The behaviour of the samples D and M has been investigated further on. The time dependency of lycopene release from solutions prepared by dissolving the matrices in the recovering solvent (mixture THF/MeOH (1:1 by weight)) was measured with the UV spectrophotometer. In the diagram of figure 5 the solvation profiles are shown.

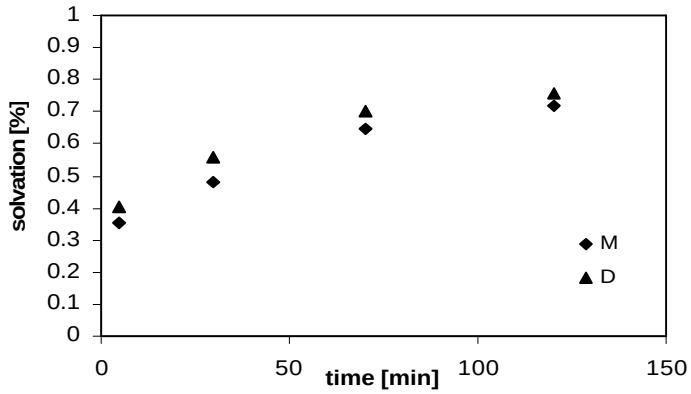

Figure 5. Release profiles of samples D and M from THF/MeOH solutions.

The UV analysis confirmed the good release of lycopene from the samples treated at high hydrostatic pressures.

CONCLUSIONS

The aim of the present work is the study of the effects of the transformation processes of tomatoes on the extractability of lycopene. It has been demonstrated by spectrophotometric tests that the concentration of the vegetable matrices leads to higher percentages of recovered lycopene after the extraction with supercritical CO_2.

The extraction curves obtained at 60°C, 300 bar and a CO_2 flow rate of 450 ml/min have been correlated first with an equation of the type of a Langmuir isotherm and then with a model based on differential mass balances written on a section of the extraction column.
The discussion of the experimental data revealed that the concentrated samples treated at high hydrostatic pressures release high percentages of lycopene after the extraction with supercritical CO_2 proving the good influence of this new technique on the recovering of the active principle.

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