

**EXTRACTION OF BOTANICALS WITH SUPERCRITICAL CARBON DIOXIDE:
KINETICS OF LEMON BALM EXTRACTION
AT DIFFERENT OPERATIONAL CONDITIONS**

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Abstract

The supercritical extraction is considered as a “green” low-temperature process, using a solvent, which is non-toxic, safe, and easy-to-remove. This paper studies the extraction of Lemon Balm (Melissa officinalis L.) by treatment with carbon dioxide at supercritical conditions. The extract contains many bioactive compounds: flavonoids, phenolic acids, triterpene acid, etc. The main therapeutic properties of this medicinal plant are sedative, antispasmodic, antibacterial, anti-inflammatory, anti-oxidant, etc...

This work investigates the process kinetics at different operational conditions. The influence of some important regime parameters (solvent flow-rate, particle size, pressure, temperature) is experimentally determined. Besides the information for process mechanisms and process intensity at particular operational regimes, the results are useful for selection of favorable operational conditions for better extraction, i.e. for production of larger quantity of extracted substances from unit mass of raw material.

Key words: lemon balm, supercritical extraction, process kinetics

1. INTRODUCTION

The extraction with supercritical carbon dioxide (SC CO₂) is an attractive method for production of substances for application in pharmacology, medicine, perfumery and cosmetics, food industry etc., because this is a “green” low-temperature process, using non-toxic, safe, and easy-to-remove solvent. The physical properties of CO₂ near the critical point (smaller viscosity, higher diffusivity,...) are favorable to better penetration in the solid matrix, thus ensuring better extraction and intensifying the interphase mass transfer. Near the critical point the solubility is sensible to temperature and pressure variations resulting in possibility for tuning the selectivity and for production of substances of high purity. In general, the operational temperature is not high, which prevents from thermo-destructive processes and preserves the product qualities.

This paper deals with the extraction of Lemon Balm (*Melissa officinalis* L.) by treatment with SC CO₂. Lemon balm is an aromatic perennial herb, about 1 m high. It grows in Mediterranean region, Western Asia, South-western Siberia, and Northern Africa. [1]. Its main therapeutic properties are sedative, carminative, antispasmodic, antibacterial, anti-inflammatory, anti-oxidative [2]. It is used in the traditional medicine also for nervous disorders, lower abdominal disorders, gastric complaints, toothache, earache, headache, rheumatism, high blood pressure etc. [2-4]

Lemon balm contains many compounds: flavonoids, phenolic acids, triterpene acid, etc. [2,5,6]. The treatment with SC CO₂ produces a total extract rich in bioactive components, to mention among them essential oil, characterized by three chemical substances: citral, citronellal and β -caryophyllene [2,4,5, 7-9].

The aim of this work is to study experimentally the influence of main operational parameters (solvent flow rate, particle size, pressure and temperature) on the process kinetics in order to make an appropriate selection of favorable operational conditions for better extraction.

2. MATERIALS AND METHODS

2.1. Raw material

Dried aerial parts (leaves and stems) of the wild Lemon balm have been used in this study. It originates from sub-Balkan region of Bulgaria (Karlovo/Kazanlak). The preliminary treatment consists in milling and sieve separation and classification in a number of fractions of different particle size, stored in dark. Mainly three fractions with mean particle size of 0.94, 1.33 and 1.70 mm have been used.

We have determined the real geometry of raw material particles under study by scanning electron microscope (SEM). Fig. 1 visualizes these results.

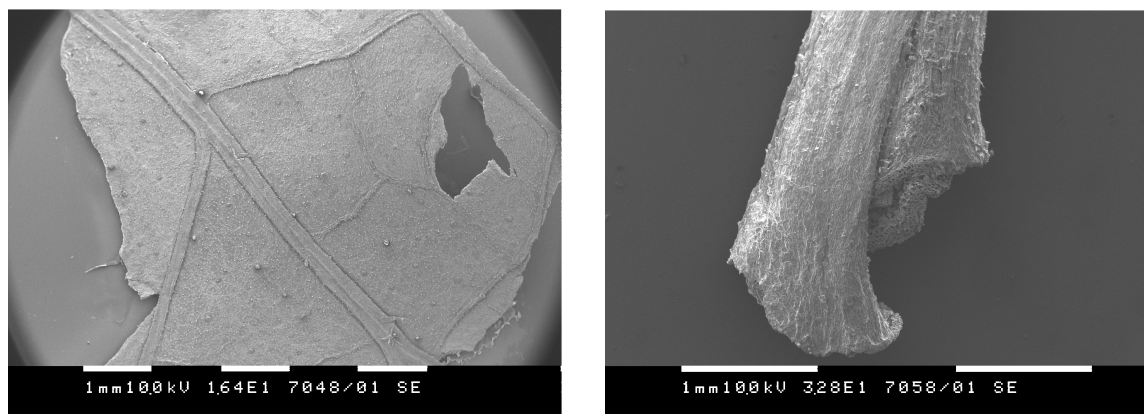


Fig. 1. Scanning Electron Microscope pictures of milled raw material particles

a) plate shape

b) cylinder shape

Two particle shapes are to be recognized: plate ones, corresponding to leaves and cylindrical particles originated from stems. These of plate shape have mean (average) thickness of 30 μm , and the other type has a mean (average) diameter of 626 μm . The proportion of two forms is about 90 % plates and 10 % cylinders. This information about the particular geometry is useful for the process modeling, because it becomes possible to insert real geometry parameters in the mathematical models.

2.2. Apparatus description

The extraction has been performed with a pilot plant from Separex Chimie Fine, France (serie 3417 type SF 200), having an extraction capacity of 200 ml and maximum CO₂ flow-rate of 5 kg/h. A scheme of the installation is shown in Fig 2.

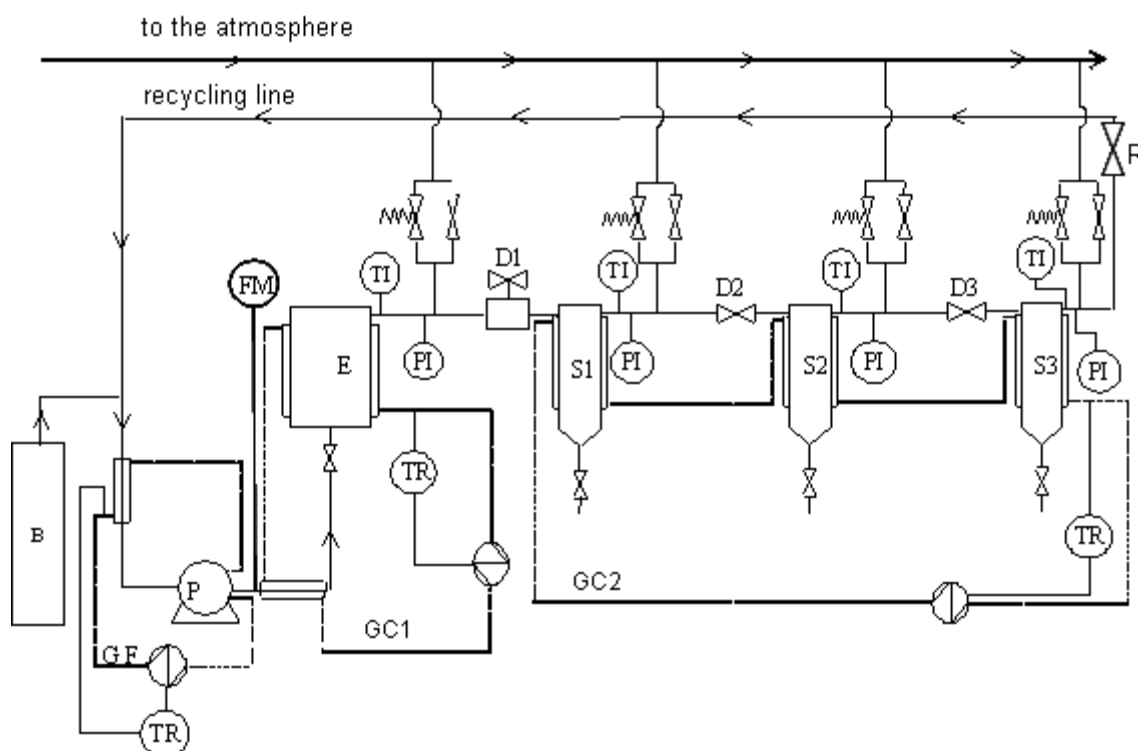


Fig. 2. Scheme of the experimental installation

Its main parts are extractor E and three separators S1-S3. They are jacketed in order to be maintained at a constant temperature by means of a temperature control system TR including heating devices GC1 and GC2. Carbon dioxide from a 50 bar-pressurized bottle B is turned to liquid by means of a cold group GF and pumped into the extractor E by the pump P with a prescribed flow-rate measured by flow-meter FM. The extractor pressure is kept at a given value by a control valve D1. After the extractor, the solvent passes through separators S1, S2 and S3, their pressure being maintained constant by the respective control valves. The temperature and pressure in the vessels are monitored by devices marked as TI and PI. There are options to operate with recycle of the CO₂ (through the “recycling line”) or to connect it to the atmosphere (through “to the atmosphere” line with recycling valve R closed).

2.3. Experimental protocol

The raw material samples are capsulated in the extraction thimble with inner diameter of 28 mm, weighed and placed in the extractor cylinder. The cylinder is then introduced into the temperature-controlled chamber. Care is taken to ensure that the air is purged before extraction is started. The SC CO₂ is pumped at constant flow-rate to the extractor’s bottom in up-flow configuration. The supercritical phase leaving the extractor is passed via three separators in series at a pressure throttled gradually by the corresponding valves. The mass of extraction thimble has been measured at 20 minutes intervals to register the negative mass changes. Then kinetics curves have been constructed based on the lost mass of the raw material.

After passage through the system, CO₂ is cooled and recycled.

The experiments are carried out with fractions of three different sizes obtained by sieving (0.94, 1.33 and 1.70 mm), at extractor temperatures being 40 and 60°C and separators temperature kept at 30°C. The pressure implemented is 200, 250 and 280 bar, and SCCO₂ flow-rate is varied from 5 to 30 g/min.

3. EXPERIMENTAL RESULTS AND DISCUSSION

Fig. 3 represents typical experimental results visualizing the variation of extraction rate versus the amount of used CO₂ for a kilogram of raw material (kg CO₂/kg raw mat.).

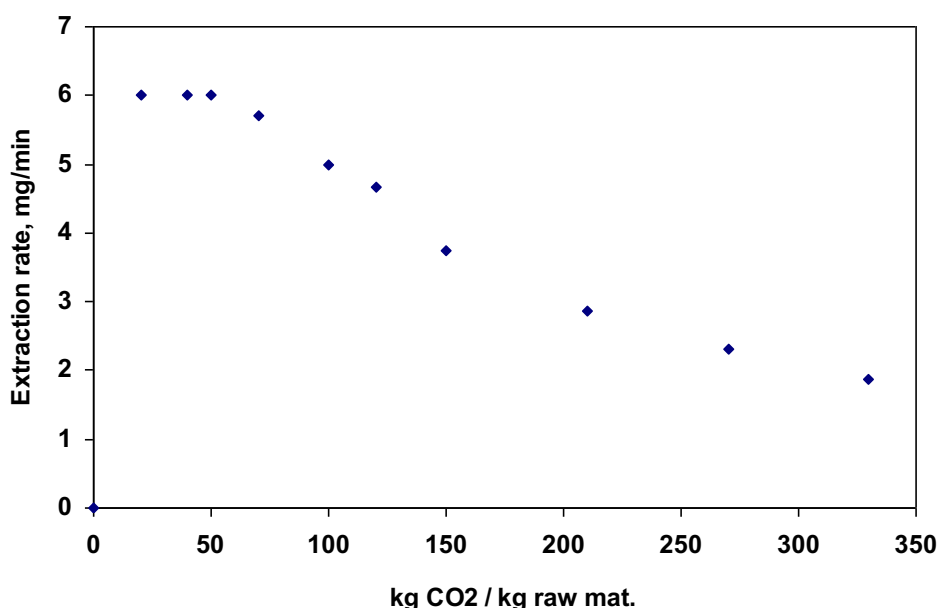


Fig. 3. Average extraction rate plotted versus the amount of used CO₂ per kg raw material, T= 40 °C, P=200 bar, average particle size 0.94 mm, CO₂ flow-rate 30 g/min.

Three periods can be distinguished. At the beginning, a relatively short period is seen corresponding to the extraction of free solute from the particle surface. A solubility controlled process can be suggested for this period. During the second (transition) period, the extraction rate drops rapidly. It corresponds to the extraction from both surface and internal cells. Consequently, the process is controlled by both external and internal resistances. The third period is characterized by a lower extraction rate, which is further faded, but rather slowly. It takes place when the solute in the surface cells is exhausted, and the process is controlled by the transfer from internal cells (internal resistance control). So, the system under study corresponds to the processes with standard type kinetics [10].

In this part the influence of various operational parameters on the process kinetics is studied experimentally. The results are presented as extracted mass from a unity of raw material (g extract/kg raw material) plotted versus the amount of used CO₂ per unity of raw material (kg CO₂/kg raw material).

3.1. Effect of temperature

Figures 4 a, b, c represent extraction isotherms at constant pressure and different temperatures. In all three figures, the initial period of linear rise corresponds to a process controlled by the solubility. From the line slope one can obtain approximately the apparent solubility of the extracted substance in the supercritical fluid [10].

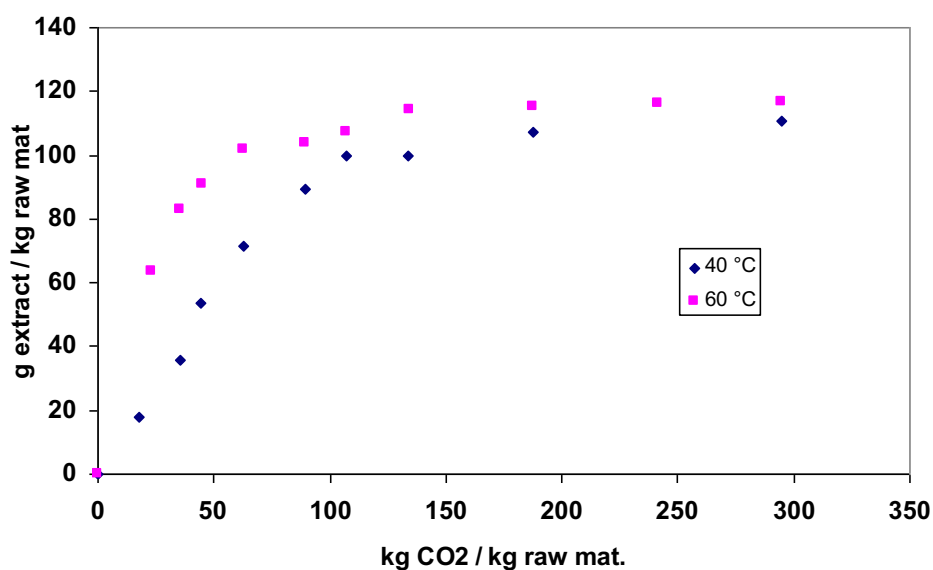


Fig. 4 a). Kinetic curves at different temperatures.

Particle size 0.94 mm, CO₂ flow rate 5 g/min, P = 200 bar

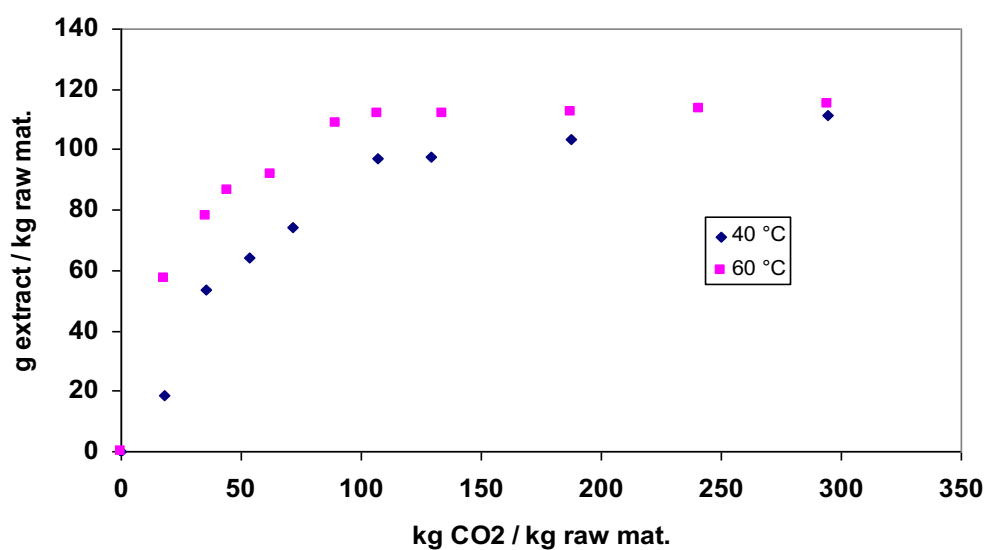


Fig. 4 b). Kinetic curves at same conditions as above, P = 250 bar

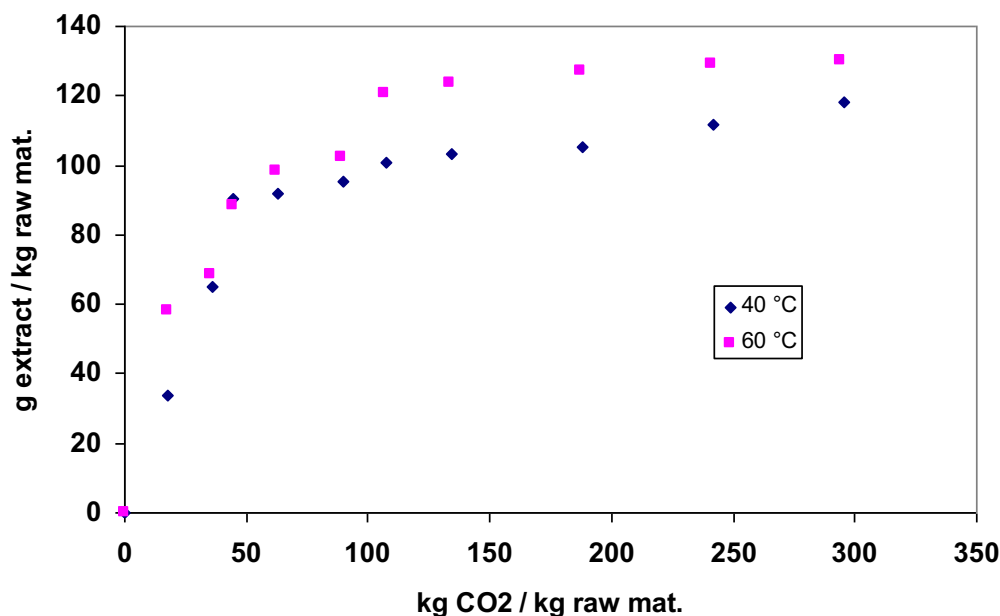


Fig 4 c). Kinetic curves at same conditions as above, $P = 280$ bar

As it is seen, for all pressure conditions the solubility resistance is more significant at lower temperature, which can be surely attributed to the increased solubility at higher temperature. As it is known [11, 12], the temperature dependence of the solubility in CO₂ is affected by two competitive factors: the solute's vapour pressure and the solvating effect of SC CO₂, which increases with its density. Initially, at lower temperatures the solubility decreases when temperature rises due to prevailing influence of decreased fluid density over solute's vapour pressure, then the latter becomes more pronounced, resulting in a dependence passing through a minimum followed by solubility rise at further temperature increase. The temperature at minimum solubility takes higher values at lower pressure. Evidently the range of pressure conditions of our experiments corresponds to a state after this minimum, and the temperature influence on solubility is predominated by the solute vapour pressure, resulting in the observed solubility increase with the temperature.

Comparing Figs 4 a, b, c it is to stress that the temperature influence during the first linear period becomes less pronounced at higher pressure. It might be attributed to the higher solvent density at higher pressure, which results in better solubility, respectively lower solubility resistance.

It is also seen that while at lower pressure the internal resistance (plateau at Fig. 4 a, b) is not very strongly influenced by the temperature, at higher pressure (Fig. 4 c) it becomes significantly higher at lower temperature, a possible reason being the increased viscosity at lower temperature.

From the above results it can be concluded that at all pressure regimes the extraction process under study is more efficient at higher temperature. We have suggested 60°C as a reasonable value, combining intensive kinetic regime and preventing from eventual thermal destruction of the extract.

3.2. Effect of the pressure

The effect of pressure on the extraction process at different particle sizes is shown on Fig. 5 a, b.

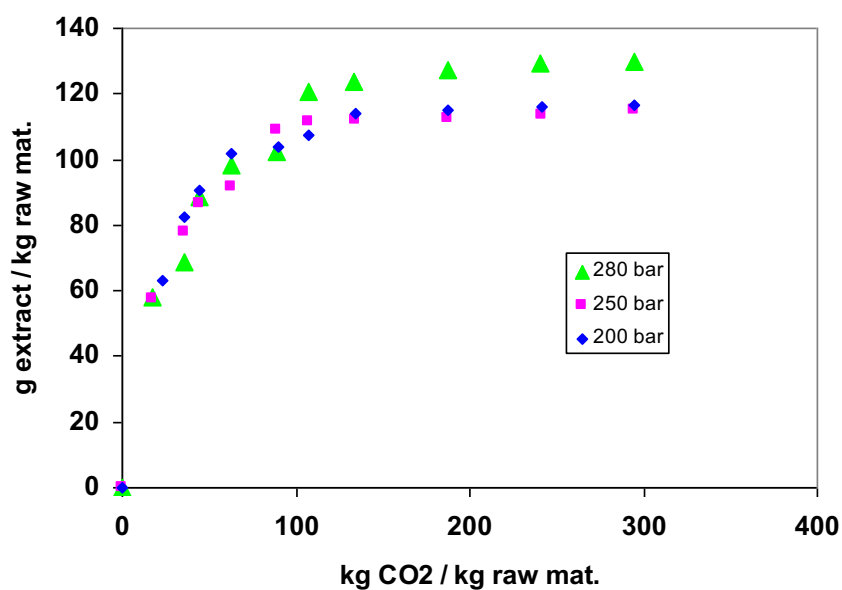


Fig. 5a)

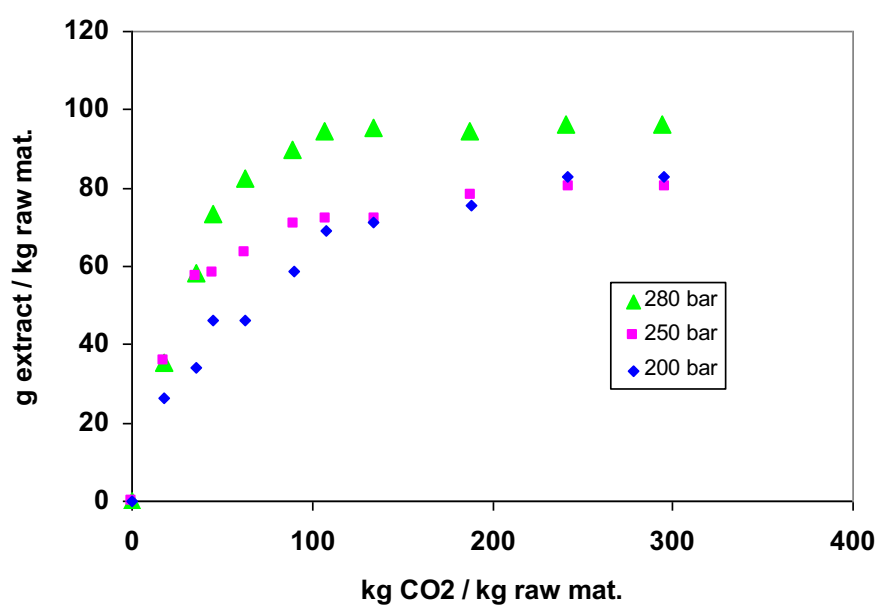


Fig. 5 b)

Fig 5. Extracted quantity versus quantity of solvent used at various pressure conditions,

T = 60 °C, CO₂ flow-rate 5 g/min,

a) Mean particle size 0.94 mm,

b) Mean particle size 1.33 mm

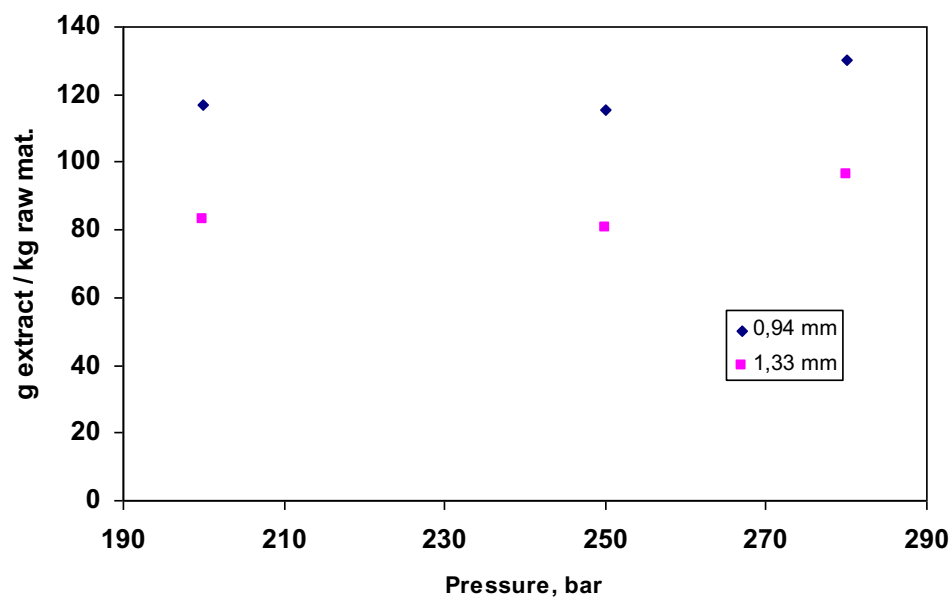


Fig. 6. Extracted quantity at different pressure and different particle size,
T = 60 °C, CO₂ flow-rate 5 g/min

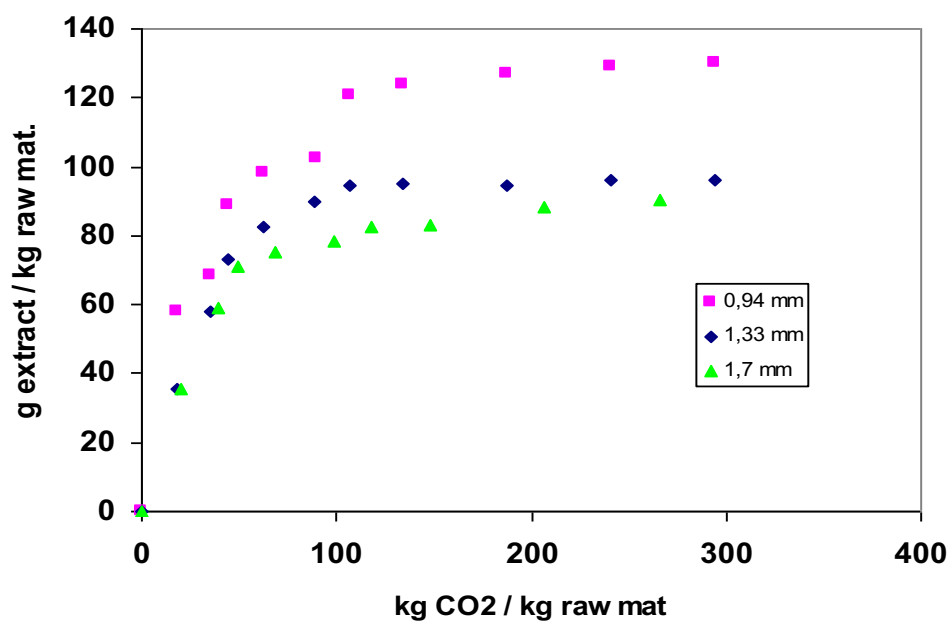


Fig. 7. Kinetic curves at different particle size, T = 60 °C, P = 280 bar, CO₂ flow-rate 5 g/min

3.3. Effect of particle size

For small sized fraction (Fig. 5 a), the initial and transition periods are nearly identical and seem as being not dependent on pressure. The entire extraction process in the pressure interval 200 - 250 bar is rather similar, while it is more significantly ameliorated at 280 bar. It can be considered as an indication for selection of appropriate pressure conditions. The impact of the pressure can be logically attributed to the increased solvent quantity due to increasing of its density at higher pressure.

Concerning larger particles (Fig 5 b), different kinetics of the initial and transition periods are registered at 200 and 250 bar, while the curves further become similar as in previous case of smaller particles. This behaviour seems to reflect the influence of increased internal resistance, which is more important in case of larger particles.

Fig. 6 shows clearly a stagnant extraction up to 250 bar followed by an increase of the extracted quantity at higher pressure. This behaviour is similar at different particle sizes. Consequently, it is to recommend operation at more than 250 bar in order to obtain better performance.

Fig. 7 illustrates the kinetics of extraction in case of particles of different size. Better results are registered with smaller particles. They have larger surface per unity of volume, i.e. larger fluid-solid contact area. A larger surface corresponds to increased quantity of open cells which makes the solutes more accessible and decreases the internal resistance. As it is seen, higher internal resistance is associated to larger particles.

3.4. Effect of solvent flow-rate

In order to cover a larger field of operational conditions, two rather different values of the solvent flow-rate have been chosen – 5 g/min and 30 g/min. The results are illustrated in Fig. 8.

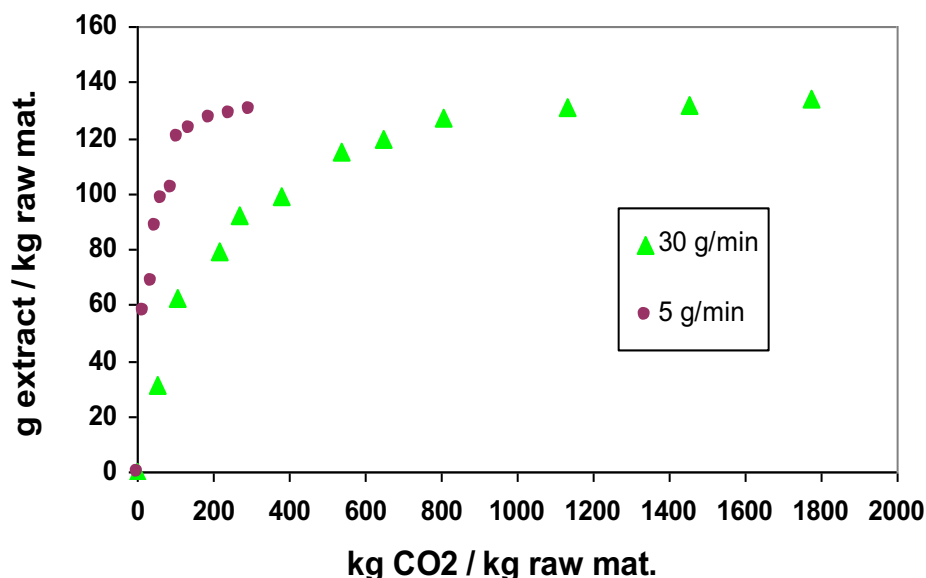


Fig. 8. Influence of flow-rate on the process kinetics, $P = 280$ bar, $T = 60^{\circ}\text{C}$, $D_p = 0,94$ mm

At both flow-rates, three standard extraction periods can be recognized – solubility controlled, mixed solubility – internal control (transition period) and dominant internal mass transfer control. In case of higher solvent flow-rate all of them are significantly longer although the total quantity extracted is

equal. At increased flow-rate the solvent residence time becomes smaller, which increases both resistances: solubility and internal transfer [13 - 15] resulting in less intensive mass transfer process. During the first period of solubility control, the solvent residence time is too short to allow dissolving of the available quantity of solutes. Consequently, this period becomes longer at higher flow-rates. The same reason is valid for the second period of mixed solubility-internal control, which also becomes longer because of lesser residence time. Thus, a lower flow-rate is more appropriate operational parameter ensuring more intensive process kinetics.

4. CONCLUSIONS

The process of extraction of Lemon Balm with carbon dioxide at supercritical conditions is experimentally studied. The influence of main operational parameters (pressure, temperature, particle size, solvent flowrate) is studied in order to determine appropriate conditions for better extraction. It is found that the extraction process under study is more efficient at higher temperature never mind the pressure regime. 60°C might be considered as a reasonable temperature, at which eventual thermal destruction of the extract is prevented, ensuring at the same time intensive process kinetics.

The pressure influence on the extraction yield is not very significant up to 250 bar, after this value the pressure intensifies significantly the process.

Better extraction yield is registered with smaller particles due to larger fluid-solid contact area, decreased internal mass transfer resistance and increased quantity of open cells with more accessible solutes.

A lower flow-rate of the solvent is more appropriate operational parameter ensuring larger residence time, resp. longer contact time and more intensive process kinetics.

Based on these results, appropriate conditions for better extraction can be selected and used in a practical application in order to produce larger quantity of extracted substances from unit mass of raw material.

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