Supplementary Information for

*In situ* investigation of scCO$_2$ assisted impregnation of drug into polymer by high pressure FTIR micro-spectroscopy

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**Supporting information a**

Adjustments performed to optimize the infrared spectra:

1) The cell was centered under the beam

2) The opening of the diaphragm was adapted to the sample to meet two requirements: it had to be not too large to avoid the detection of surrounding CO$_2$ but not too narrow to obtain a sufficient signal to noise ratio. A preliminary study was performed for each geometry and size of sample to determine the best size of the diaphragm opening.

3) The focusing of the beam was adjusted by changing the vertical position ($z$ axe in Figure 1) of the cell. Depending on how the focusing was performed, we found that the baseline changed (increased either in the near infrared region or in the far infrared region) and that...
 could entail a saturation of some of the characteristic peaks we have chosen for our investigation. Thus, after preliminary tests, we found that the best adjustment was to modify the vertical position to obtain the maximal value of the interferogram which corresponds to the maximal intensity of the reflected signal.

4) The background was recorded just nearby the polymeric sample on the mirror, without changing the adjustments performed at previous steps 2 and 3.

5) The horizontal position (x axe in Figure 1) of the cell was adjusted in order to focus on the middle of the sample.

**Supporting information b**

In order to evaluate the evolution of the pathlength during the impregnation process, we have done the following procedure. First of all, the initial pathlength $l_0$ is determined by measuring the thickness of the polymer before fitting it into the cell. Then, two impregnation experiments have been carried out, following the same experimental protocol described previously: one aimed at investigating the impregnation of the polymeric system where the IR beam was focused in the polymer sample and the other experiment dealt with the measurement of the pathlength $l$ by recording a spectrum of the surrounding CO$_2$ right next to the polymeric sample. In this last case, the absorbance $A_{CO2}$ of the characteristic peak of CO$_2$ is measured on the IR spectra and the concentration of CO$_2$ $C_{CO2}$ under these conditions is known from the literature (NIST $^1$). Thanks to the Beer-Lambert law, the pathlength can be determined:

$$l = \frac{A_{CO2}}{\varepsilon \cdot C_{CO2}}$$ (1)

The characteristic peaks of CO$_2$ centered at 3715 and 4950 cm$^{-1}$ were used for the absorbance measurements. Thus, the study of the impregnation required focusing the IR beam
in the polymer sample whereas the pathlength was measured by focusing the IR beam in the surrounding CO$_2$. Thus, recording the evolution of both the impregnation process and the variation of the pathlength simultaneously would have required displacing the high pressure cell under the microscope frequently. Because the microscope is equipped with a manual stage, it would have been inconvenient and imprecise to perform such displacements, yet it was necessary to focus at the same place in PEO to follow the kinetic of impregnation. It is doubtless that a simultaneous study of the impregnation process and the pathlength should be possible and more precise by using a microscope equipped with a motorized microscope stage and a programmed displacement of the stage to record successively a spectrum into PEO and one in surrounding CO$_2$.

**Supporting information c**

In order to determine the molar extinction coefficient of specific modes of aspirin and ketoprofen, we have performed Infrared absorption measurements on both APIs diluted in supercritical carbon dioxide at T=40°C and various pressures from 5 up to 30 MPa.

*a. Experimental set-up*

The infrared absorption measurements were performed on a ThermoOptek Nicolet 6700 FTIR spectrometer equipped with a globar as the infrared source, a KBr/Ge beamsplitter and a DTGS (Deuterated TriGlycine Sulphate) detector in order to investigate the spectral range 600-4000 cm$^{-1}$. Single beam spectra recorded with a 2 cm$^{-1}$ resolution were obtained after the Fourier transformation of 50 accumulated interferograms.

The infrared absorption experiments were performed using an in-house built stainless steel cell equipped with four cylindrical windows, two germanium windows with a variable path length (between 0.412 and 2.395 cm) for the infrared absorption measurements and two other sapphire windows for direct observation of the solution. The seal was obtained using the unsupported area principle. The windows were positioned on the surface of a stainless steel plug with a 100 μm Kapton® foil placed between the window and the plug to compensate for any imperfections between the two surfaces. Teflon® O-rings were used to ensure the seal
between the plug and the cell body. The cell was heated using cartridge heaters disposed in the periphery of the body of the cell. Two thermocouples were used, the first one located close to a cartridge heater for the temperature regulation and the second one close to the sample area to measure the temperature of the sample with an accuracy of about 2°C. The cell was connected via a stainless steel capillary tube to a hydraulic pressurizing system which allows the pressure to be raised up to 50 MPa with an absolute uncertainty of ± 0.1 MPa and a relative error of ± 0.3%. The stabilization of the operating conditions was controlled by recording several consecutive spectra.

**Figure c.1:** Picture and schematic diagram of the high-pressure optical cell

**b. Experimental Procedure**

First of all, the powder was placed in the bottom of the cell, in a cork that was then mounted on the cell (see in figure c.1). The powder was well below the incoming infrared beam such that the CO$_2$ phase can be analyzed. Then, the cell was heated up to the required temperature. Then the CO$_2$ was slowly added up to the desired pressure in order to avoid any spread of the drug powder inside the cell. The system was kept under isobaric and isothermal conditions for a period between 10 and 30 min. During the stabilization of the operating conditions, consecutive spectra were recorded every 5 min. The equilibrium was considered to be achieved when no changes of the spectral bands were noticed. Once the equilibrium was reached, the pressure was raised to a higher value. The mixture was constantly homogenized
during the experiment using a magnetically driven stirrer disposed into the cork (the stirring speed was optimized to avoid the powder to be spread into the cell).

For solubility measurements, a large amount of drug was placed into the cell (~30mg) i.e. in excess in order to allow saturation of scCO$_2$ with drug in all the studied conditions. On the contrary, a small and known amount of drug (2.00mg) was placed in the cell to determine the molar extinction coefficient of the characteristic peaks of the drug
c. Infrared spectra

CO$_2$ spectrum

The FTIR spectrum of raw scCO$_2$ in supercritical conditions is presented in figure c.2. The previously described vibrations modes appear on this spectrum as well as combination modes. One can detect four peaks at 3590, 3695 and 4950 cm$^{-1}$ which are assigned to the combination modes $2
\nu_2+\nu_3$, $\nu_1+\nu_3$, and $\nu_1+2\nu_2+\nu_3$ of the CO$_2$ molecule respectively $^3$. Since a large pathlength is used, the spectral regions between 600-800cm$^{-1}$; 2000-2500cm$^{-1}$ and 3500-4000cm$^{-1}$ are saturated. Thus, if the drug displays characteristic peaks in these wavenumber ranges, it cannot be observed. The band at 1400cm$^{-1}$ that corresponds to the symmetric stretching vibration $\nu_1$ is theoretically forbidden but can be observed on this spectrum. The band centered at 1100cm$^{-1}$ is due to the silicium window whereas the broad band around 3000cm$^{-1}$ is due to impurities.

![FTIR spectrum of scCO$_2$ at T=40°C P=90bar](image)

**Figure c.2:** FTIR spectrum of scCO$_2$ at T=40°C P=90bar
Ketoprofen spectra

The ATR-IR spectrum of ketoprofen powder is presented in figure c.3. The characteristic peaks of ketoprofen in the spectral range 1600-1800 cm\(^{-1}\) can be attributed. The peak centered at 1695 cm\(^{-1}\) corresponds to the \(\nu_{\text{C}=\text{O}}\) stretching mode of the carboxylic acid group of ketoprofen molecules organized in a crystalline structure, two ketoprofen molecules forming cyclic dimers by hydrogen bonding. The peak at 1655 cm\(^{-1}\) is assigned to the \(\nu_{\text{C}=\text{O}}\) stretching mode of the ketone group and the peaks between 1560-1610 cm\(^{-1}\) are characteristic to the \(\nu_{\text{C}=\text{C}}\) stretching mode of the phenyl group. The broad peak observed in the range 2200-3400 cm\(^{-1}\) is assigned to the \(\nu_{\text{O-H}}\) stretching mode. \(\text{CO}_2\) does not have any bands in these ranges (1600-1800 cm\(^{-1}\) and 2200-3400 cm\(^{-1}\)).

![ATR spectrum of ketoprofen powder](image)

Figure c.3: ATR spectrum of ketoprofen powder i.e. in crystallized form

Once ketoprofen is solubilized into scCO\(_2\), some changes can be noticed in the selected wavenumber range (see figure c.4). The bands at 1716 and 1672 cm\(^{-1}\) correspond to the \(\nu_{\text{C}=\text{O}}\) stretching mode of the carboxylic acid group of ketoprofen in its cyclic dimeric form and to the \(\nu_{\text{C}=\text{O}}\) stretching mode of the ketone group, they are shifted to higher wavenumber compared to the bands at 1695 and 1655 cm\(^{-1}\) in crystallized ketoprofen. Moreover, a new band appears at 1763 cm\(^{-1}\) which is assigned to \(\nu_{\text{C}=\text{O}}\) of the carbonyl group in its monomer form\(^{4,5}\).
Since the ν\text{C=O} bands at 1763 and 1695 cm\(^{-1}\) are dependent on the speciation of ketoprofen, they could not be used to determine the solubility since the speciation is likely to evolve with the experimental conditions (P, T). Hence the selection of the bands centered at 1672 cm\(^{-1}\) (ν\text{C=O}); 1605 cm\(^{-1}\) (ν\text{C=C}) and 1585 cm\(^{-1}\) (ν\text{C=C}) to measure the solubility of ketoprofen, depending on their saturation or not.

**Figure c.4:** Comparison of the IR spectra of (a) ketoprofen solubilized in scCO\(_2\) and (b) ketoprofen powder.

**Aspirin spectra**

Figure c.5 shows the comparison of IR spectra of aspirin solubilized into scCO\(_2\) (a) and crystallized aspirin (b). Similarly as previously done for ketoprofen, the characteristic peaks of aspirin have been assigned. The peak centered at 1780 cm\(^{-1}\) corresponds to ν\text{C=O} stretching vibrations of the ester group, the peaks at 1750 and 1706 cm\(^{-1}\) are respectively attributed to the aspirin ν\text{C=O} carboxyl group in its monomer and dimer form, the ones at 1608 and 1584 cm\(^{-1}\) are both assigned to ν\text{C=C} of the phenyl group.

The bands centered at 1780 cm\(^{-1}\); 1608 and 1584 cm\(^{-1}\) were chosen for the further study.
Figure c.5: Comparison of the IR spectra of (a) aspirin solubilized in scCO$_2$ and (b) aspirin powder.

d. Determination of the molar extinction coefficients

Applying the Beer-Lambert law, the molar extinction coefficient $\varepsilon_i$ was calculated for each peak $i$ using equation 1:

$$
\varepsilon_i = \frac{A_i}{l \times C_{drug}} \quad (1)
$$

where $A_i$ is the absorbance of the peak determined by measuring the height of the pic, and $C_{drug}$ (mol.L$^{-1}$) is calculated as the mole of drug introduced divided by the volume of the high-pressure cell (9mL in these experiments), and $l$ is the path length (2.395 cm).

For each peak, the molar extinction coefficient was calculated as the average of the values obtained for various pressures once the total amount of drug was solubilized (i.e. once the absorbance does not increase anymore). The molar extinction coefficients were considered to be independent on the CO$_2$ density.
Ketoprofen

The values of the molar extinction coefficient of the peaks of ketoprofen centered at 1672, 1605 and 1585 cm\(^{-1}\) are reported in table c.1.

<table>
<thead>
<tr>
<th>Characteristic band</th>
<th>Molar extinction coefficient value (mol.L(^{-1}).cm(^{-1}))</th>
<th>Ecart type s</th>
</tr>
</thead>
<tbody>
<tr>
<td>(\varepsilon_{1672})</td>
<td>380.5</td>
<td>9.4</td>
</tr>
<tr>
<td>(\varepsilon_{1605})</td>
<td>95</td>
<td>5.2</td>
</tr>
<tr>
<td>(\varepsilon_{1585})</td>
<td>49.7</td>
<td>3.5</td>
</tr>
</tbody>
</table>

*Table c.1: molar extinction coefficients of characteristic peaks of ketoprofen*

Aspirin

The values of the molar extinction coefficient of the peaks of aspirin centered at 1780, 1608 and 1584 cm\(^{-1}\) are reported in table c.2.

<table>
<thead>
<tr>
<th>Characteristic band</th>
<th>Molar extinction coefficient value (mol.L(^{-1}).cm(^{-1}))</th>
<th>Ecart type s</th>
</tr>
</thead>
<tbody>
<tr>
<td>(\varepsilon_{1780})</td>
<td>315.3</td>
<td>2.5</td>
</tr>
<tr>
<td>(\varepsilon_{1608})</td>
<td>147.2</td>
<td>1.45</td>
</tr>
<tr>
<td>(\varepsilon_{1584})</td>
<td>24.3</td>
<td>0.44</td>
</tr>
</tbody>
</table>

*Table c.2: molar extinction coefficients of characteristic peaks of aspirin*

Supporting information d

**Calculation of the molar extinction coefficient for the peaks at 3200 cm\(^{-1}\)** \(\varepsilon_{3200}\)

The calculation of the molar extinction coefficient of the peaks at 3200 cm\(^{-1}\) is presented for aspirin. For each characteristic peak, the Beer-Lambert law can be applied:

\[
A_{1608} = \varepsilon_{1608}L C_{\text{Aspirin}} \quad (1) \quad A_{3200} = \varepsilon_{3200}L C_{\text{Aspirin}} \quad (2)
\]
From equations 1 and 2, we can calculate the molar extinction coefficient $\varepsilon_{3200}$ of the peak at 3200 cm$^{-1}$ as follow:

$$\varepsilon_{3200} = \varepsilon_{1608} \frac{A_{3200}}{A_{1608}}$$ (3)

We have reported the evolution with time of the absorbance of the two peaks of aspirin at 1608 and 3200 cm$^{-1}$ in figure d.1 when the PEO film was subjected to a mixture of \{CO$_2$+Aspirin\} under 5 MPa, 10 MPa and 15 MPa successively during 2h, 5h and 3h respectively.

**Figure d.1:** Evolution with pressure and time of the absorbance of two characteristic peaks of aspirin during impregnation into PEO at $T=40^\circ$C

Up to 200min, the two peaks increase similarly. Beyond 200min, the peak at 3200 cm$^{-1}$ keeps increasing with time up to the end of the experiment whereas the peak centered at 1608 cm$^{-1}$ increases until reaching a maximum about 0.35 (u.a.) at 280min. This maximal absorbance is due to the saturation of the peak as we can see in figure d.2.
Figure d.2: IR spectra of a PEO film subjected to CO$_2$ and aspirin at $T=40^\circ$C and 0.1, 5, 10 and 15 MPa – saturation of the peaks in the range of 1600-1800 cm$^{-1}$

The equation 3 has been applied for spectra recorded between 115 and 185 min as the peak at 1608 cm$^{-1}$ was not saturating. The molar extinction coefficient $\epsilon_{3200}$ of the characteristic peak of aspirin at 3200 cm$^{-1}$ was determined to be:

$$\epsilon_{3200} = 103 \text{ L.mol}^{-1}.\text{cm}^{-1}$$

The same method has been applied to determine the molar extinction coefficient $\epsilon_{3200}$ of the characteristic peak of ketoprofen. It has been found to be equal to 162 L.mol$^{-1}$.cm$^{-1}$.

Supporting information e

The molar extinction coefficient of the CO$_2$ peak centered at 4950 cm$^{-1}$ was known from the literature to be $\epsilon_{4950}\text{cm}^{-1}= 0.25 \text{ L.mol}^{-1}.\text{cm}^{-1}$ for the neat CO$_2$ considering the peak height. As Buback and al. have shown that the molar extinction coefficients are independent on the CO$_2$ density for combination bands, we used this value for the CO$_2$ sorbed into the polymers.
We emphasize that using the CO$_2$ peak centered at 4950 cm$^{-1}$ in reference 6 for the determination of the mutual solubility of epoxide with CO$_2$, a satisfactory agreement was found with literature data.

The IR spectra of every polymer subjected to CO$_2$ were recorded at 40°C and for pressures up to 15 MPa. Then, we have selected the spectra on which the peaks at 3695 and 3590 cm$^{-1}$ did not saturate and the peak at 4950 cm$^{-1}$ was high enough to determine its intensity with a good accuracy. From these spectra, we have applied the Beer-Lambert law to the three peaks ($i$ being 3695 or 3590cm$^{-1}$):

\[ A_{4950} = \varepsilon_{4950} l C \quad (1) \]

\[ A_i = \varepsilon_i l C \quad (2) \]

After combination of equations 1 and 2, the molar extinction coefficients of the peaks centered at 3695 and 3590cm$^{-1}$ were calculated using the following equation:

\[ \varepsilon_i = \varepsilon_{4950} \frac{A_i}{A_{4950}} \quad (3) \]

Thus, the molar extinction coefficient considering the peaks height were estimated to be:

\[ \varepsilon_{3695\text{cm}^{-1}} = 8.28 \pm 1.2 \text{ L.mol}^{-1}.\text{cm}^{-1}; \]

\[ \varepsilon_{3590\text{cm}^{-1}} = 3.17 \pm 0.4 \text{ L.mol}^{-1}.\text{cm}^{-1} \]
References