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ORIGINAL PAPER

High Pressure Micronization of Tristearate

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Abstract The effect of the pre-expansion temperature, pre-expansion pressure and storage time was studied in order to investigate the crystal form, degree of crystallinity, particle size, particle size distribution and morphology of the tristearate. The experiments were performed at pressures between 115 and 215 bar and at temperatures of 54, 60 and 70 °C with the substance being already melted. Samples were characterized immediately after the experiments and later after 3 months of storage at 5 and 20 °C. Results showed an increase in the amorphous phase. During the storage, the micronized samples re-crystallized. With time, the agglomeration was more emphasized for particles produced at higher temperatures and pressures and stored at a higher temperature. The results of X-ray analysis did not show a change in crystal form of tristearate (β' form) before and after micronization, however, for samples stored at 20 °C slow transformation from the β' (less stable) form to the β (more stable) form occurred. In addition, the morphology of micronized samples showed an irregular and porous shape. The re-crystallization was more evident with time for samples stored at 20 °C where needle-like crystals appeared on the surface.

Keywords $SCCO_2$ · Tristearate · PGSS^{TM} · Crystal form · Degree of crystallinity · Particle size · Morphology

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Introduction

High-pressure spray processes, using supercritical fluids, have made a big impact in many industrial fields over the past 15 years. These processes, reported by Jung and Perrut [1] and Weidner et al. [2], which are designated by acronyms such as PGSS^{TM} (particles from gas saturated solution) and others, allow the production of powders with properties that are difficult or even impossible to achieve by classical methods. Products processed in these fields have to satisfy some requirements: suitable morphology, proper diameter, narrow particle size distribution, high reproducibility and high purity [3]. The use of supercritical fluids (SCF) shows advantages over conventional processes like milling, grinding and chemical precipitation, where product damage is often caused. Mechanical treatment often results in product damage or degradation which can be caused by particle traumatization, frictional heat or a broad particle size distribution [4, 5]. High pressure spray processes can overcome these limitations. They offer the possibility of controlling the particle dimensions by adjusting the operating temperature and pressure and enabling one to obtain completely solvent-free products [6].

The work of Himawan et al. [7] and Kellens and Reynaers [8] proved that tristearate forms three polymorphic crystalline structures, which are α, β' and β, and are widely used in many fields of food and other consumer products. Each of these three crystal forms has typical morphological and physical properties such as melting point and heat of fusion. Polymorphism of fats, reported by Narine and Marangoni [9] and Kalnin et al. [10], shows an important phenomenon where the stability of an individual phase crucially influenced the processing of fats in food products, the final presentation of the product (hardness and texture) and longer term storage (re-crystallization process and changes in the microstructure).

The experiments were focused on producing the micro particles of tristearate at different operating temperatures and pressures with a batch PGSS process aimed at investigating the influence of process parameters like pre-expansion pressure, temperature and aging on the crystallization behavior, particle size, particle size distribution and morphology of micronized tristearate. Micronizations were performed within a pressure range from 115 to 215 bar and at three different melting point temperatures: 54, 60 and 70 °C. The degree of crystallinity and crystal form, particle size and morphology of micronized samples were determined immediately after micronization and later after 3 months of storage at 5 and 20 °C.

Experimental

Materials

Tristearate (CAS N. 555-43-1) was purchased from Chemos, Germany. According to DSC and X-ray, tristearate was in the β' form and was used in experiments in the same form as received. CO_2 (purity 99.5 vol%) was purchased from Messer, Slovenia.

Methods

Micronization

The scheme of the apparatus can be seen in the literature [11]. The autoclave was filled with the substance (10–12 g) and CO_2 was introduced by a high pressure pump to the desired pressure, depending on pre-expansion parameters. The autoclave was then heated to the temperature near the melting point of the substance. After reaching equilibrium (2 h of stirring by a mechanical oscillating device) the gas-saturated solution was expanded through a nozzle with a diameter 0.4 mm in a spray tower. Spraying took approximately 10 s and, during this step, the pressure in the autoclave was not kept constant (it was lowered to atmospheric pressure). Micronizations of tristearate were performed in pre-expansion pressure range from 115 to 215 bar at 54, 60 and 70 °C with the substance already being melted. Each experiment was performed twice.

Characterization of Powdered Samples

The particle size and particle size distribution of micronized and stored samples were measured with a granulometer

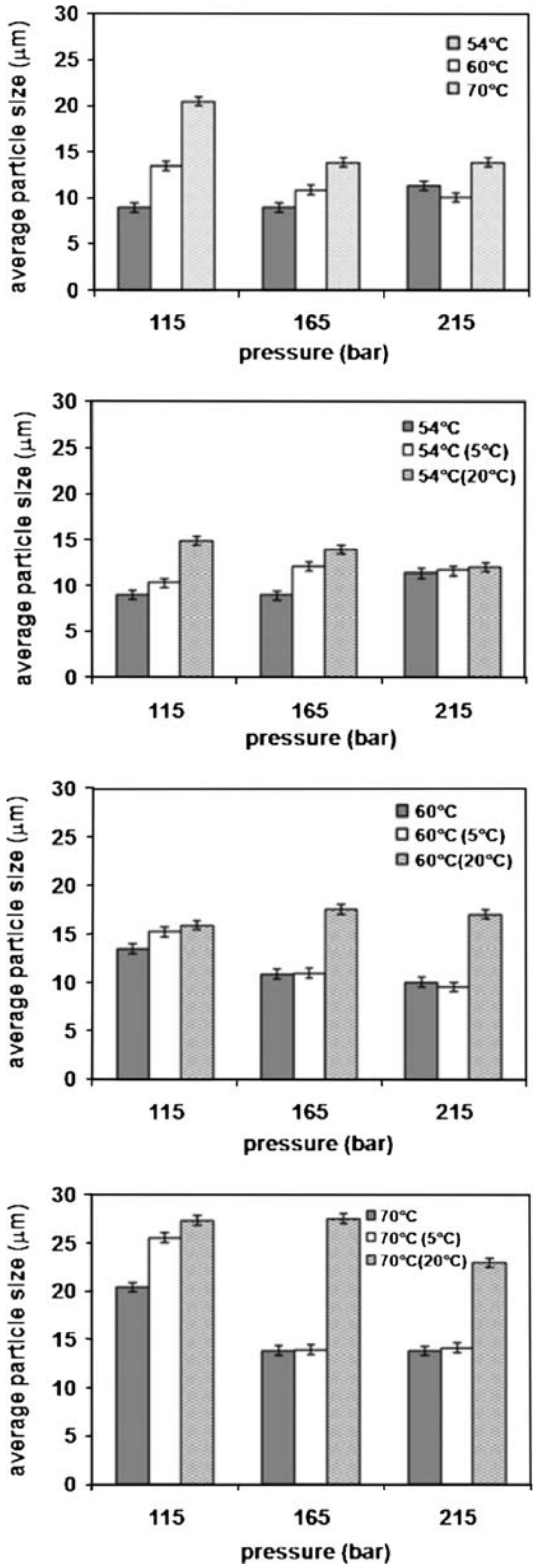


Fig. 1 Comparison of average tristearate particle size of against the operating temperatures (54, 60 and 70 °C), pressures (115, 165 and 215 bar) and storage temperature (5 and 20 °C)

(Fritsch analysette 22 compact, Germany), which operated on the principle of laser diffraction spectroscopy by a wet method using water as the solvent. Every measurement was repeated three times.

To determine the degree of crystallinity and crystal forms based on the melting points (MP) and change in enthalpy of fusion and crystallization (ΔH) a METTLER DSC 20 apparatus was used. DSC is the most widely used thermo-analytical technique in fat research. DSC offers a sensitive, rapid and reproducible method for the identification of fats and their polymorphism. Calibration of energy and temperature was performed with an indium sample. All samples were placed in an aluminum pan with a weight of about 3 mg against a reference empty pan. Micronized samples and the starting substance received from the supplier were heated from 25 to 80 °C at a heating rate of 2.5 K/min and cooled down from 80 to 25 °C with a cooling rate of 0.5 K/min. Comparison of the crystallinity degree between micronized sample and substance before micronization was calculated using Eq. 1:

$$\mathrm{cr}(\%) = \frac{\Delta H_{mx}^{\mathrm{fus}}}{\Delta H_{m0}^{\mathrm{fus}}} \times 100, \qquad (1)$$

where $\Delta H_{mx}^{\mathrm{fus}}$ is enthalpy of fusion of micronized sample, $\Delta H_{m0}^{\mathrm{fus}}$ is enthalpy of fusion for the sample before micronization.

To determine the crystal form of powders, X-ray diffraction measurements were performed on an AXS-Brüker/Siemens/D5005 diffractometer using Cu–Kα radiation ($\lambda = 0.1542$ nm). Before each measurement the diffractometer was calibrated with a SiO_2 holder. The diffraction measurements started at 10° and ended at 80° (2θ) with steps of 0.036 (2θ) and a step time of 1 s at 25 °C.

The particle shape and surface were observed by a scanning electron microscope under high vacuum (Quanta 200 3D, SEM).

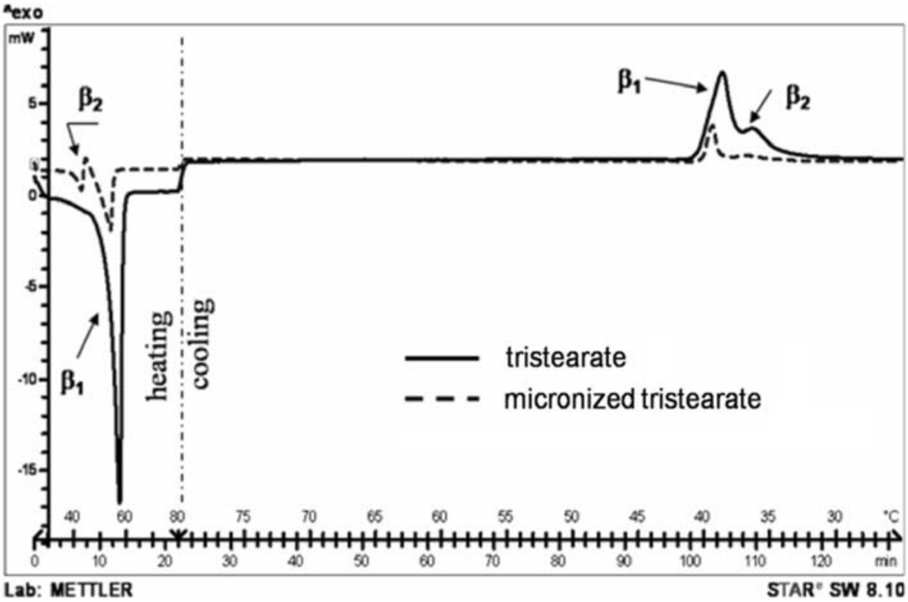
Results and Discussion

Tristearate was received in a form of flakes. After micronization the produced powder had a monomodal particle size distribution with an average particle size in a range from 7 to 28 µm (±0.5 µm). Figure 1 presents the comparison of average particle size of powders obtained at different pre-expansion temperatures, pre-expansion pressures and storage temperatures. The average particle size of powders with regard to operating temperatures showed two different trends; at 54 °C the size of particles increased with increasing pre-expansion pressure, while at 60 and 70 °C, it decreased with increasing pressure. At all investigated pressures the largest particles were obtained at 70 °C and the difference in the size of particles obtained at various operating temperatures decreased with increasing the operating pressure.

For the powders produced at 54 °C and stored at 20 °C the agglomeration process is more evident with time when produced at lower pressures, while for powders stored at 5 °C, the increase in particle size is small and it may be rather due to the change of shape of the crystalline cold stored particles. When the micronization is performed at higher working temperatures like 60 and 70 °C and pressures above 115 bar, samples must be stored at 5 °C to avoid the agglomeration process.

In order to meet more practical requirements, thermo-analytical techniques were used for fat characterization to

Fig. 2 DSC melting and crystallizing curves of tristearate before processing (11 mg) and micronized tristearate (115 bar, 60 °C, 3 mg). The curves were recorded 1 h after preparation



determine melting and crystallization profiles, enthalpies of fusion and crystallization, and solid fat content.

Results of examinations by DSC are shown in Fig. 2. Samples were first heated and cooled at a rate of 10 K/min and only one peak was observed. While the scanning rate plays an important role in thermal transition, every measurement was repeated with slower heating and cooling rates as given above. The existence of two β' forms was found and had already been reported by Elisabettini et al. [12] and Matovic and van Miltenberg [13]. The DSC curve for purchased tristearate revealed one endothermic peak at 54.7 °C with the enthalpy of 157 J/g (β'_1 form) and two exothermic peaks at 39.8 °C with the enthalpy of 61.8 J/g

Table 1 Content of β'_2 and β'_1 (%) polymorphic structure of micronized samples of tristearate

Sample		Melting		Crystallization	
(°C)	(bar)	β_2 (%)	β_1 (%)	β_2 (%)	β_1 (%)
After micronization					
54	115	19.2	80.8	16.6	83.4
	165	24.8	75.2	20.6	79.4
	215	15.6	84.4	21.7	78.3
60	115	16.5	83.5	8.7	91.3
	165	11.2	88.8	30.7	69.3
	215	–	100.0	11.5	88.8
70	115	19.8	80.2	20.9	79.1
	165	12.1	87.9	31.7	68.3
	215	14.7	85.3	21.1	70.9
Stored at 5 °C, 3 months					
54	115	17.0	83.0	23.9	76.1
	165	19.2	80.8	17.7	82.3
	215	22.4	77.6	21.7	78.3
60	115	21.5	78.5	18.9	81.1
	165	20.1	79.9	28.1	71.9
	215	21.9	78.1	20.8	79.2
70	115	20.5	79.5	37.9	62.1
	165	22.2	77.8	21.6	78.4
	215	24.8	75.2	24.3	75.7
Stored at 20 °C, 3 months					
54	115	–	100.0	18.4	81.6
	165	–	100.0	19.9	80.1
	215	–	100.0	22.2	77.8
60	115	7.4	92.6	13.0	87.0
	165	–	100.0	22.0	78.0
	215	–	100.0	13.5	86.5
70	115	5.8	94.2	23.2	76.8
	165	17.3	82.7	16.5	83.5
	215	4.3	95.7	15.0	85.0

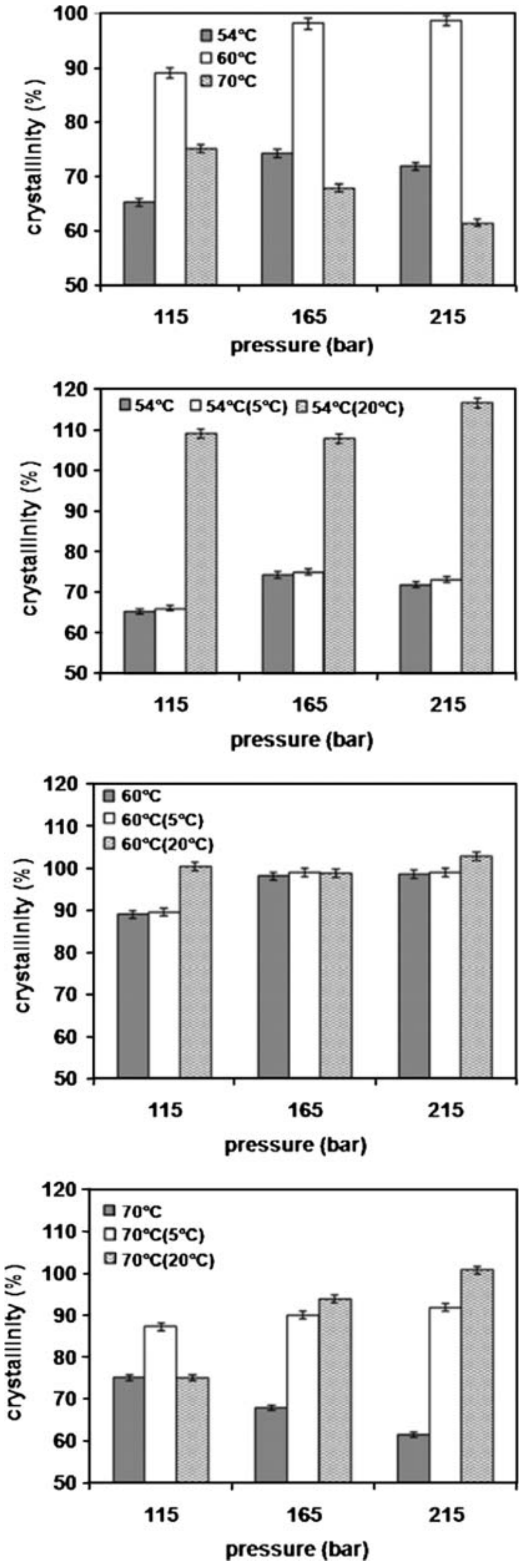


Fig. 3 Comparison of tristearate crystallinity degree against the operating temperatures (54, 60 and 70 °C), pressures (115, 165 and 215 bar) and temperature of storage (5 and 20 °C)

(β'_1 form) and at 36.5 °C with 8.5 J/g (β'_2 form). For tristerate micronized at 165 bar and 60 °C the DSC curve revealed two endothermic (43 °C and 32 J/g for β'_2 form; 53.7 °C and 163 J/g for β'_1 form) and two exothermic peaks (40 °C and 69 J/g for β'_1 form; 37 °C and 6.6 J/g for β'_2 form). Generally, enthalpies of crystallization were lower than enthalpy of fusion for about 60%.

The percentages of β'_2 and β'_1 forms in micronized samples were calculated based on peak areas indicating fusion and crystallization of β'_2 and β'_1 polymorphs obtained by DSC analyses. The percentage of polymorph can be expressed by Eq. 2. Results are presented in Table 1.

$$\beta_1^{'}(\%) = \frac{\Delta H_{\beta_1'}}{\Delta H_{\beta_1'} + \Delta H_{\beta_2'}} \times 100, \qquad (2)$$

where ΔH is enthalpy calculated from the peak area corresponding to the melting and crystallization of β'_2 and β'_1 polymorphs and weight of sample.

DSC measurements of micronized samples showed two melting peaks in some cases while for the purchased material a single peak was obtained. The results showed that in micronized samples the quantity of β'_1 crystals was much higher, on average around 80%. After 3 months of storage at 5 °C this quantity remains approximately the same, while for the samples stored at 20 °C mostly β'_1 crystals are present. Immediately after heating to 80 °C the samples were cooled to 25 °C. Again, every sample showed two peaks corresponding to β'_2 and β'_1 crystals, with mainly the β'_1 form present (around 80%). Even the samples stored at 20 °C presented around 20% of the β'_2 crystals.

The degree of crystallinity (±1%) against operating pressure and operating temperatures is shown in Fig. 3. It was calculated by Eq. 1 based on enthalpies of fusion, where the melting enthalpy of fresh material was taken as 100%.

It is clear that the degree of crystallinity of micronized samples is lower than the one of the samples before processing and that it depends on the operating temperature and pressure. At temperatures of 54 and 60 °C the degree of crystallinity slowly increases, while at 70 °C it markedly decreases with increasing pressure. As the re-crystallization process is an undesired result, it is necessary to establish optimal terms of aging. For unprocessed tristearate no change in crystallinity was observed after storage of samples for 3 months at 5 and 20 °C, while processed samples showed a different storage behavior. Samples micronized at 54 °C and stored at 5 °C did not show re-crystallization, while the same samples stored at 20 °C strongly re-crystallized after 3 months. The degree of crystallinity for those samples is even higher than the one for starting material. The results of samples processed at 60 °C showed advantages of storing at 20 °C, where the unwanted phenomena is strongly reduced. However, these samples already had a degree of crystallinity much higher than the samples processed at 54 and 70 °C. The lowest degree of crystallinity (60%) was obtained for the samples processed at the highest temperature (70 °C) and the highest pressure (215 bar). However, when the samples were stored at 5 and 20 °C for 3 months, the re-crystallization rate was high. The crystallinity increased to 90 and 100%, respectively.

The use of X-ray diffraction shows the most important advancement in understanding the structure of fats. Over a long period, many researchers used the X-ray investigation to prove that fats crystallized in different polymorphic forms. The three typical polymorphic crystal structures, α, β′ and β, are precisely described by Ghotra et al. [14].

Figure 4 presents the X-ray patterns of tristearate before micronization, tristearate micronized at 115 bar and 60 °C and micronized samples after the 3 months of storage at 5 and 20 °C.

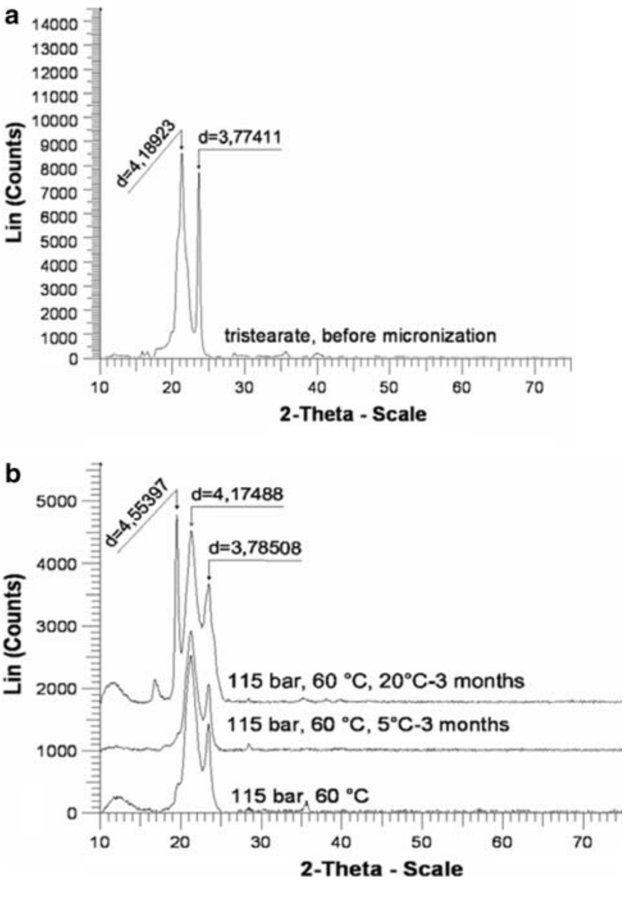


Fig. 4 X-ray patterns of tristearate before (**a**) and after micronization (powder produced at 115 bar, 60 °C and stored at 5 and 20 °C) (**b**)

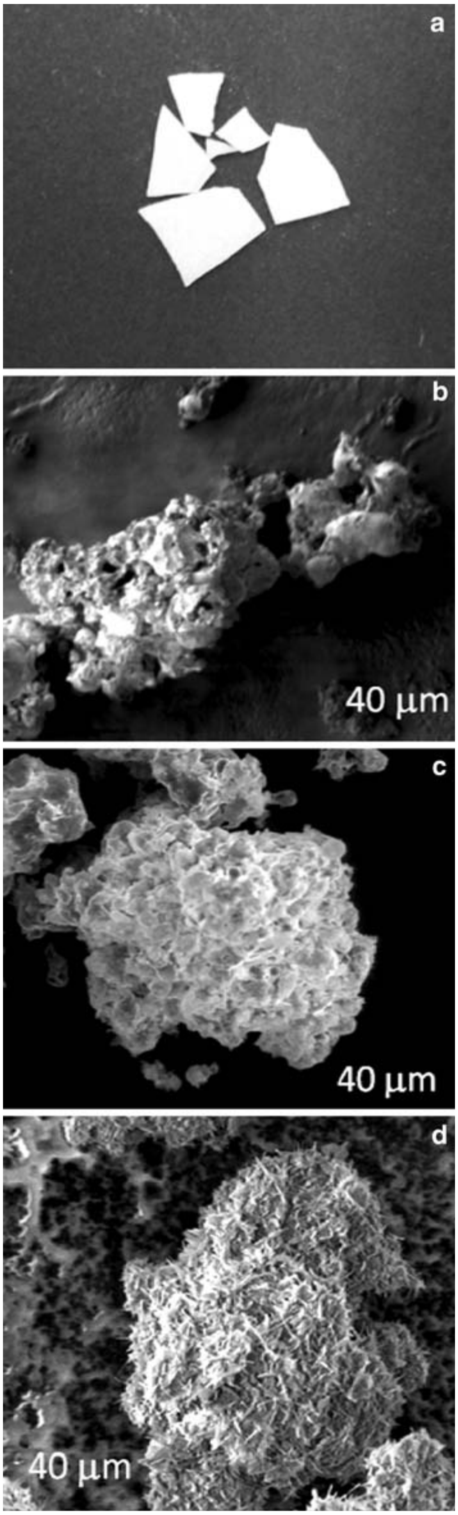


Fig. 5 Morphology of tristearate **a** before micronization (taken with a digital camera), **b** particles micronized at 115 bar and 60 °C (*SEM* Scanning Electron Microscope), **c** stored at 5 °C (SEM), **d** stored at 20 °C (SEM)

Sample of tristearate before processing showed two intensive peaks at 2θ of about 20 °C indicating the β' form: 4.18 and 3.77 Å. Peaks of the tristearate sample after micronization (115 bar, 60 °C) remained in the same place but intensities were lower showing a lower degree of crystallinity. The same sample stored at 5 °C kept the β' form and the peaks are still in the positions 4.17 and 3.78 Å, while the same micronized sample, stored at 20 °C for 3 months, resulted in a third intensive peak at 4.55 Å showing transformation from the β' to the β form.

In general, it was observed that micronization process parameters of pre-expansion temperatures and pressures did not have an effect on the crystal form of tristearate. Samples stored at 5 °C for 3 months also kept the β' polymorphic form, while micronized tristearate samples stored at 20 °C after 3 months showed transformation to the more stable β form. Tristearate samples micronized at higher pressure and temperature exhibited lower intensity of the third peak, which confirmed the more stable form, and for these cases the transformation was slower as for samples micronized at lower temperature and pressure, where the intensity of the third peak was much higher.

The morphology of the tristearate particles observed by SEM is presented in Fig. 5b–d on a scale of 40 μm. Particles before micronization were in the shape of flakes. Figure 5a presents the flakes of tristearate (approximate size 4 mm) taken with a digital camera. After the process, the particles became porous with an irregular shape (Fig. 5b). A change in particle morphology is connected to the evaporation of CO_2 from the melt during expansion and solidification of the melt caused by the cooling when the compressed gas expands. Figure 5c shows particles stored at 5 °C after 3 months. The surface changed due to the shape of the crystalline cold stored particles. The recrystallization process is more evident for particle stored at 20 °C (Fig. 5d), where needle-like crystals appeared.

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