GREEN SUPERCRITICAL FLUID NANOTECHNOLOGY

Can Quan[1], Gunilla Jacobson-Andrews[2], Charlotta Turner[3,] *
[1]*Division of Chemistry, National Institute of Metrology (NIM), Beijing, China*
[2]*Department of Chemistry, Stanford University, CA, USA*
[3] *Department of Physical & Analytical Chemistry, Uppsala University, Sweden*
** Charlotta.turner@kemi.uu.se Tel: 46 18 471 3677*

ABSTRACT

It is argued that nano-engineered materials at the nano-scale have mechanical, optical, chemical, and electrical properties quite different from the bulk material and even potential safety issues to human health or environment. The safest possible route for advancing nanotechnology in a sustainable world can be reached using green technology. Green Nanotechnology has two goals: firstly, to produce nano-materials and products without harming the environment or human health; and secondly, to produce nano-products that provide solutions to environmental challenges without compromising the function of the product.

Supercritical carbon dioxide (SC-CO_2), with properties such as low viscosity, high solvent strength and in particular, zero surface tension, presents as a natural choice in environmental friendly nanotechnology processes, and plays a more and more critical role in many advanced technology applications. The use of SC-CO_2 in various particle formation processes in which plant-derived molecules and materials are used as raw-materials potentially result in the production of solvent-free nano/micro-sized natural product materials designed for various high-value applications.

In this presentation, a selected list of examples studied in our group concerning green SC-CO_2 nanotechnology will be presented, which include microencapsulation of quercetin with biodegradable polymers by supercritical antisolvent process (SAS) and astaxanthin nanoparticle formation by rapid expansion of supercritical solutions (RESS).

Keywords: Supercritical Antisolvent, Rapid Expansion of Supercritical Solution, Quercetin, Astaxanthin

INTRODUCTION

The production of ultra-fine particles using a supercritical fluid as antisolvent has several advantages over other precipitation methods. The mixing between the supercritical antisolvent and the liquid is much faster than in conventional liquid antisolvent processes, thus leading to higher supersaturations and smaller particle diameters. Moreover, it is possible to control the particle size distribution (PSD) with changes in the operating conditions. The supercritical antisolvent can be easily removed from the final product by reducing pressure; in contrast with the complex purification processes often required when organic antisolvents are used. And with a proper selection of the antisolvent, it is possible to carry out the process at near ambient temperatures, avoiding the thermal degradation of the product. For these reasons, supercritical antisolvent processes have been studied during the last years, for applications which include explosives, polymers, pigments, pharmaceuticals and natural compounds [1-2].

Rapid expansion of supercritical solutions (RESS) [3-4] is used when the compound has some degree of solubility in supercritical fluids. The compound is dissolved in a supercritical fluid and this high-pressure solution is rapidly depressurized through an orifice to lead to compound precipitation at a low pressure. Supercritical antisolvent process (SAS) [5-6] is similar to the aerosol solvent extraction system (ASES) and precipitation with a compressed antisolvent (PCA) process. In these techniques, a supercritical fluid acts as an antisolvent for solutions as in the GAS (gas antisolvent) process.

Astaxanthin is one of the main pigments included in crustacean shrimp, and other farmed fish feeds, that belongs to the family of the xanthophylls, the oxygenated derivatives of carotenoids whose synthesis in plants derives from lycopene. Its main role is to provide the desirable reddish-orange color

in these organisms as they do not have access to natural sources of carotenoids. The use of astaxanthin in the aquaculture industry is important from the standpoint of pigmentation and consumer appeal but also as an essential nutritional component for adequate growth and reproduction [7-8]. Quercetin is a potent antioxidant that reduces the risk of cancer and cardiovascular diseases, and possibly also neurodegenerative diseases such as Alzheimer [9]. Both chemical structures are shown in Figure 1.

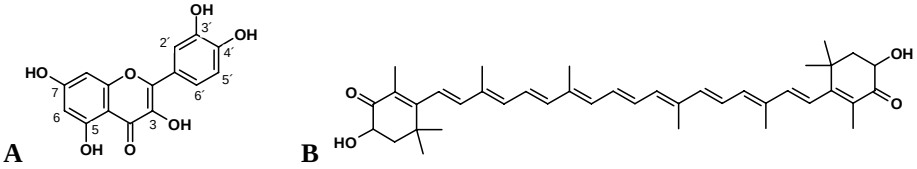

Figure 1. Chemical structures of quercetin (**A**) and astaxanthin (**B**)

In this paper, some preliminary data related to particle formation using supercritical fluid technologies are described, including making quercetin particles by SAS and astaxanthin particles by RESS. This report is the preliminary results in the study of the co-precipitation of quercetin and astaxanthin with bio-polymers, with the objective of obtaining stable formulations suitable for the food industry.

EXPERIMENTAL

Materials

Quercetin and astaxanthin with purity of 98% were purchased from Sigma (Steinheim, Germany). Solvents-- ethyl acetate and hexane (Merck, Darmstadt, Germany) were all of HPLC grade and used without further purification. Liquid carbon dioxide purchased from Air Liquide Gas AB (Eskilstuna, Sweden) was used in both the SAS and RESS process.

Experimental apparatus and procedures

A schematic diagram of the pilot plant, Bench Scale unit, is given in Figure 2. , the RESS system is designed to operate at pressures and temperatures up to 400 bar and 200°C, respectively.

Solvent (CO_2), from the cylinder, is delivered through a pipe to the condenser. Liquid CO_2 reaches the inlet of the high pressure pump. Compressed fluid is fed to the heater prior to entering the RESS vessel. When the desired operating conditions (temperature, pressure and flow rate) are achieved and supersaturated after remaining stable for certain time (0.5-1 h), the compound can be depressurized through a nozzle and collected in a chamber.

A schematic diagram of the pilot plant used for the supercritical antisolvent precipitation of quercetin is shown in Figure. 3. The equipment used are two diaphragm pumps, one for the CO_2 and the other for the solution; an isolated and jacketed stainless steel precipitator with 0.5 L volume and with a porous metallic frit at the exit; an external stainless steel filter from headline filters.

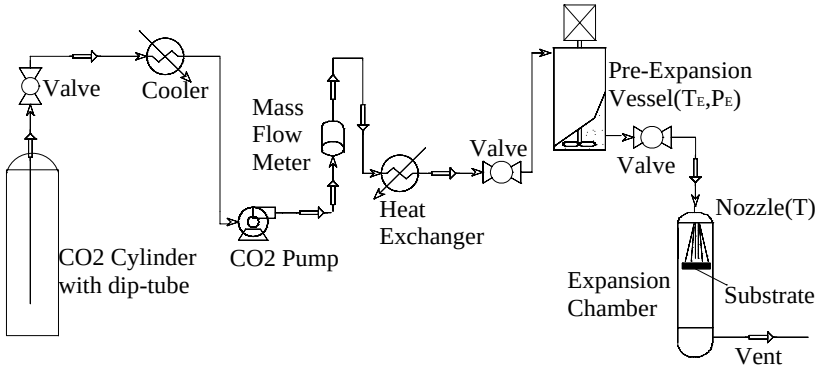

Figure 2. Schematic diagram of the RESS apparatus

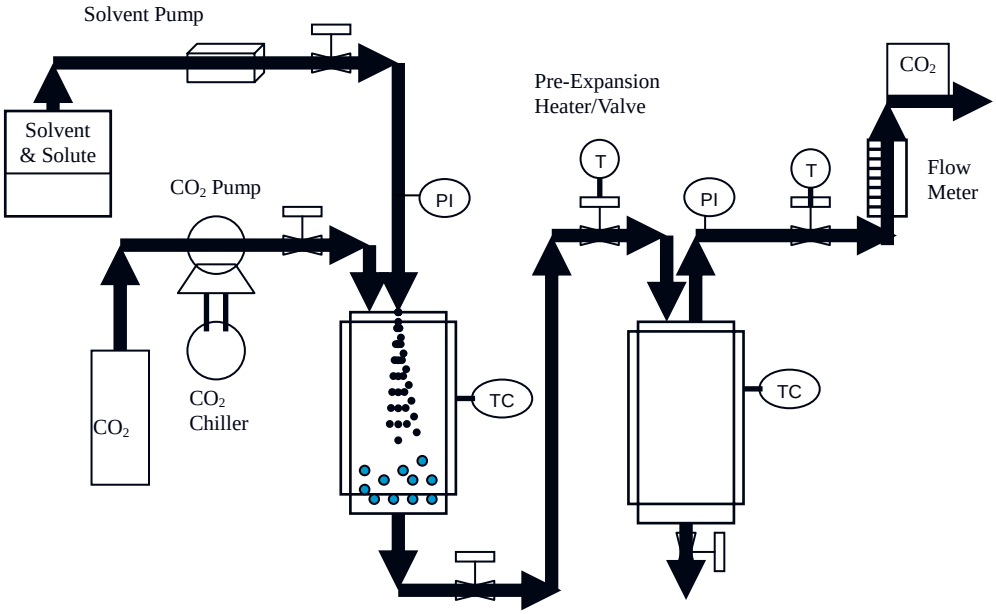

Figure 3. SAS system used in this work

A standard experiment for SAS is as follows: the experiment starts by pumping pure CO_2 into the precipitator. When the desired operating conditions (temperature, pressure and flow rate) are achieved and remain stable, the solution is fed to the precipitator. When the desired amount of solution has been injected, the liquid pump is stopped and only pure CO_2 is pumped. The flow of CO_2 is maintained during a period long enough for the complete removal of solvent from the precipitator. The minimum amount of CO_2 required for this step was determined experimentally, after the decompression, a sample of the particles retained in the frit is collected for the scanning electron microscopy (SEM) analysis.

Characterization

An emission scanning electron microscope (SEM) was used in this study for morphological observations. Specimens were sputter-coated with palladium (SPI Sputter) for 20 s to make the surface conductive without compromising fine surface microstructure. A nonconductive surface would produce a severe surface charge problem under the high intensity electron beam and accumulated surface charge would cause abnormal contrast, image deformation and distortion.

RESULTS AND DICSUSSIONS

Quercetin particles produced by SAS

Figure 4a shows the morphology and size of the unprocessed quercetin powder and Figure 4b- 4c show the quercetin particles made by SAS with methanol and IPA as cosolvent, respectively. As can be observed, the quercetin particles exhibit needle structure.

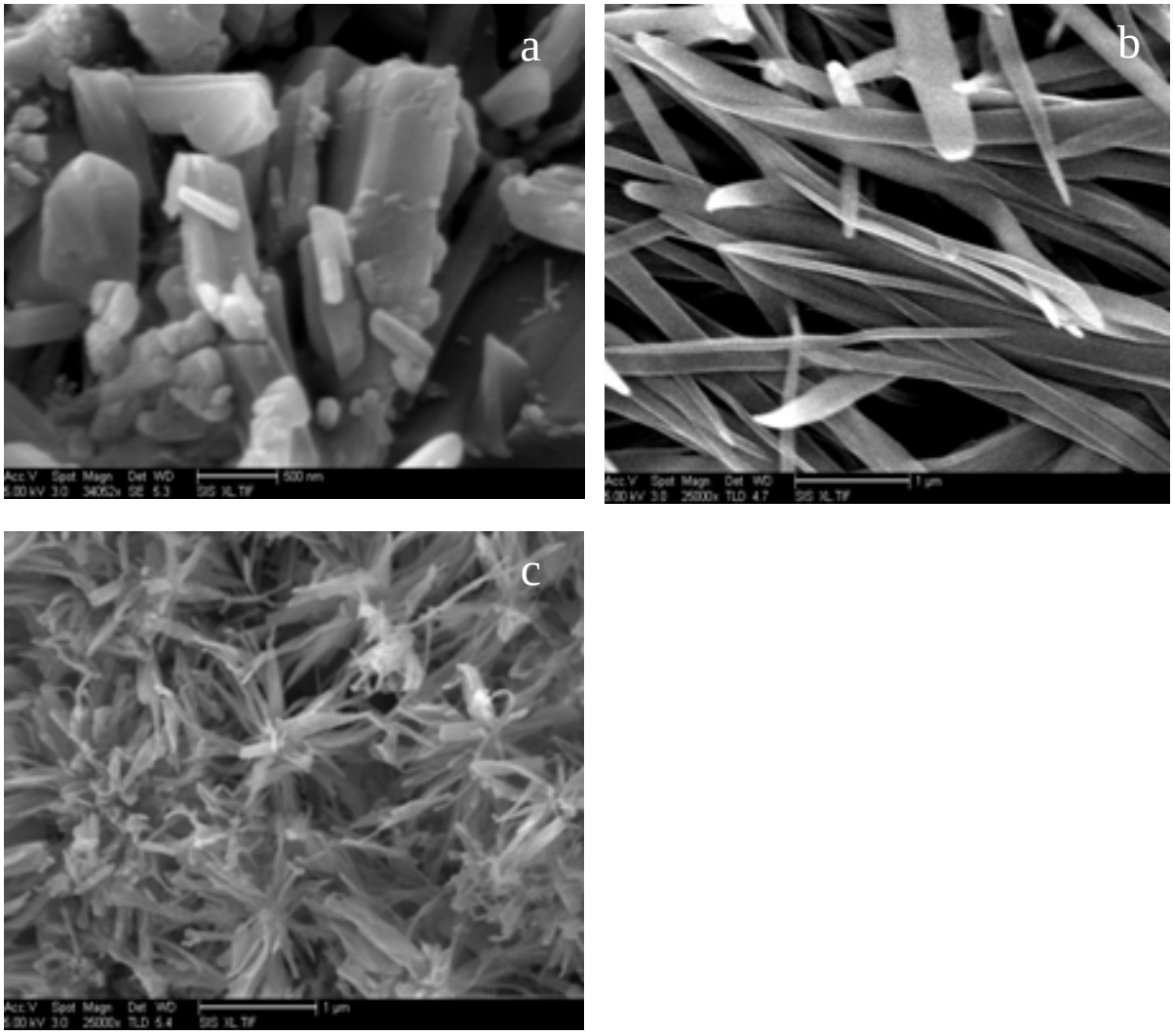

Figure 4. SEM image of Quercetin particles A: unprocessed quercetin; B: SAS with methanol as cosolvent; C： SAS with IPA as cosolvent. T = 40 ºC, P = 300 bar, CO_2 flow = 80 g/min, Inj. flow = 1 ml/min

Astaxanthin particle precipitation via RESS

Binary solution RESS experiments were performed in order to study the final particle morphology as a function of processing variables. The results of the binary solution RESS study were used to establish favorable conditions for the ternary solution (with polymer) RESS experiments.

The SEM micrographs of astaxanthin particles obtained at different experimental conditions, and their particle size histograms with estimations of the mean particle size, a (μm), and standard deviation, d (μm), in normal distribution mode N(a, d), was calculated by Matlab and shown in Figure 5, which giving sphere-like particles with average particle size of around 0.5 μm. Compared to quercetin particles produced by SAS, the astaxanthin particles formed by RESS are of completely different morphology.

The effect of the pre-expansion pressure on the sizes of particles was tested at two different pressures (150 and 300 bar) and the result is reported in Figure 5: the astaxanthin mean particle size decreases when increasing the pressure from 150 bar to 300 bar by comparing Figure 5c and 5d to Figure 5a and 5b at 40°C and 60°C, respectively. This could be that increasing pre-expansion pressure is expected to decrease critical nucleus size and thus produce smaller particles.

The effect of pre-expansion temperature was investigated at 40 and 60°C, and two different pre-expansion pressures, 150 bar and 300 bar. SEM images in figure 5 show that smaller particles are

produced at higher pre-expansion temperature, when comparing figure 5a and 5c to figure 5b and 5d, respectively. This might be attributed to that lower temperature results in the earlier solute nucleation and higher nuclei concentration during spraying.

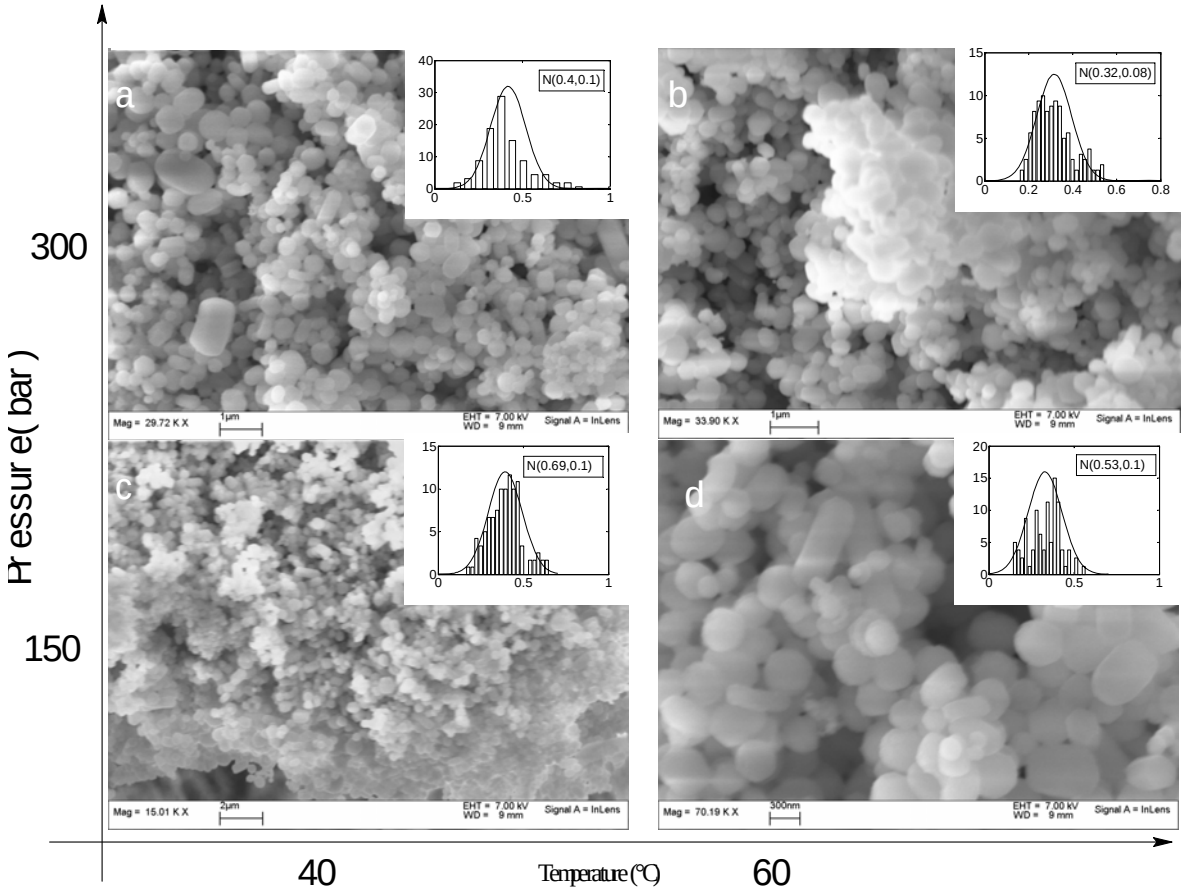

Figure 5. SEM micrographs of astaxanthin particles collected at different conditions: (a) 40°C, 300 bar; (b) 60°C, 300 bar; (c) 40°C, 150 bar; and (d) 60°C, 150 bar. Calculated particle size distributions are shown for each SEM image, also giving the mean particle size (a, in μm) and the standard deviation (d, in μm) in normal distribution mode N(a, d).

CONCLUDING REMARKS

Factors including co-solvent selection, concentration of drug in co-solvent, temperature, pressure and CO2 flow rate were investigated for quercetin particle formation by SAS. Preliminary results show that changing the cosolvent had a significant effect on particle size of queretin.

For the astaxanthin formation process by RESS, micronized particles with mean particle size of 0.3-0.8 μm were also made successfully. The effects of RESS operating parameters on the astaxanthin particle size has been studied: higher pre-expansion pressure and pre-expansion temperature resulted in a reducing effect on the average particle size of the collected astaxanthin.

REFERENCES

[1] J. Jung, M. Perrut, Particle design using supercritical fluids: literature and patent survey, J. Supercrit. Fluids 20 (2001) 179.
[2] T. Gamse, F. Miguel, M.J. Cocero, Supercritical fluid techniques for production of micron and submicron particles, in: Proceedings of the 16th Congress of Chemical and Process Engineering (CHISA),
Prague, Czech Republic, 2004.
[3] Sang-Do Yeo, Erdogan Kiran, Formation of polymer particles with supercritical fluids: A review, J. of Supercritical Fluids 34 (2005) 287–308
[4] Y. Chernyak, F. Henon, R.B. Harris, Formation of perfluoropolyether coatings by the rapid

expansion of supercritical solutions (RESS) process. Part 1. Experimental results, Ind. Eng. Chem.Res. 40 (2001) 6118.
[5] E. Carretier, E. Badens, ,Hydrodynamics of supercritical antisolvent precipitation: characterization and influence on particle morphology, Ind. Eng. Chem. Res. 42 (2003) 331.
[6] N. Elvassore, T. Parton, A. Bertucco, V. Di Noto, Kinetics of particle formation in the gas antisolvent precipitation process, AIChE J. 49 (2003) 859.
[7]I. Higuera-Ciapara, L. Felix-Valenzuela, Astaxanthin: A Review of its Chemistry and Applications, Critical Reviews in Food Science and Nutrition, 46(2006)185-196
[8] Lies Van Nieuwerburgh, Ingrid Wa¨nstrand, Jianguo Liu, Pauli Snoeijs, Astaxanthin production in marine pelagic copepods grazing on two different phytoplankton diets, Journal of Sea Research 53 (2005) 147-160
[9] K. Ono; Y. Yoshiike; A. Takashima; K. Hasegawa; H. Naiki; M. Yamada, "Potent anti-amyloidogenic and fibril-destabilizing effects of polyphenols in vitro: implications for the prevention and therapeutics of Alzheimer's disease", J. Neurochem., 87, pp. 172-181, 2003