



## Supercritical Fluid Extraction-A Green Paradigm in the Area of Separation Science

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### ABSTRACT

Supercritical fluids are increasingly replacing the organic solvents that are used in industrial purification and recrystallization operations because of regulatory and environmental pressures on hydrocarbon and ozone-depleting emissions. With increasing scrutiny of solvent residues in pharmaceuticals, medical products, and nutraceuticals, and with stricter regulations the use of supercritical fluids is rapidly proliferating in all industrial sectors. One such major application is the use of supercritical fluids for extraction which is an environment-friendly process. Extracts obtained using this technology is superior in quality with higher yield and no residual solvent. In all the extraction processes generally carbon dioxide is used as solvent, which is regarded as safe for extraction of natural products. The extraction processes are being commercialized in the polymers, pharmaceuticals, specialty lubricants and fine chemicals industries and are advantageously applied to increasing product performance to levels that cannot be achieved by traditional technologies. Such applications for supercritical fluids offer the potential for both technical and economic success.

### 1. INTRODUCTION:

A supercritical fluid (SCF) is any compound at a temperature and pressure above the critical point. Above the critical temperature ( $T_c$ ) of a compound, the pure, gaseous component cannot be liquefied regardless of the pressure applied. The critical pressure ( $P_c$ ) is the vapor pressure of the gas at the critical temperature. In the supercritical environment only one phase exists. The fluid, as it is termed, is neither a gas nor a liquid. This phase retains solvent power approximating liquids as well as the transport properties common to gases. By increasing the pressure of the gas above the critical point it is possible to give liquid-like densities and solvating strengths. Near the critical point, the density of the gas will increase rapidly with increasing pressure. Here, the solubility of many compounds is several orders of magnitude greater than predicted from the classical thermodynamics of ideal gases. As the average distance between molecules decreases, non-ideal gas behavior will begin to govern the interactions between the solvent and the sample accounting for a tremendous enhancement in solubility. In supercritical region, solvating strength is a direct function of density, which in turn is dependent on system pressure

(at constant temperature). Solvating strengths can be fine-tuned by adjusting the pressure and/or temperature, using the solvent anywhere in the range of ideal gas to nearly pure liquid. Because of the non-compressibility of liquids, this phenomenon is unique to supercritical fluids. When a fluid approaches the critical conditions, its density gets closer and closer to that of the liquid state.<sup>2</sup>

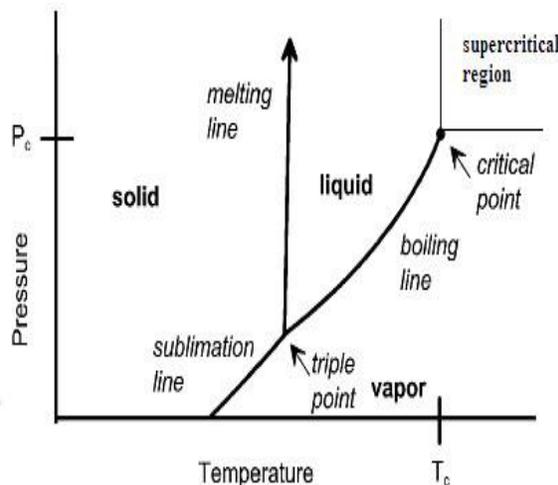


Figure 1 : Phase diagram

As shown in Figure 1, phase diagram describing physical state of substances has three curves as sublimation curve, melting curve, and boiling curve. The curve defines the regions corresponding to the gas, liquid and solid state. The critical point marks the end of the vapor liquid coexistence curve. Above the critical temperature there is no phase transition in that the fluid cannot undergo a transition to liquid phase, regardless of the applied pressure. In the supercritical environment only one phase exists and as it is termed is neither a gas nor a liquid and characterized by physical and thermal properties that are between those of the pure liquid and gas.<sup>3,4</sup>

Table 1 below lists examples of compounds along with their critical temperature and pressure to be used as SCF

Compound	Critical temperature (°C)	Critical pressure (atm)
Carbon dioxide	31.3	72.9
Ammonia	132.4	112.5
Water	374.15	218.3
Nitrous oxide	36.5	71.7
Xenon	16.6	57.6
Krypton	-63.8	54.3
Methane	-82.1	45.8
Ethane	32.28	48.1
Ethylene	9.21	49.7
Propane	96.67	41.9
Pentane	196.6	33.3
Methanol	240.5	78.9

**Table 1: Critical points for typical solvents**

### Supercritical fluid extraction

Extraction can be defined as the removal of soluble material from an insoluble residue, either liquid or solid, by treatment with a liquid solvent. It is therefore, a solution process and depends on the mass transfer phenomena. The controlling factor in the rate of extraction is normally the rate of diffusion of the solute through the liquid boundary layer at the interface.

Soxhlet extraction is a classical technique especially for extraction of oils. Ideally, extraction procedures should be environment friendly and should not create additional pollution. Soxhlet extraction does not meet these criteria because it generates large volumes of contaminated, hazardous solvents and emits toxic fumes, costly and high-purity solvents are required and it is a time consuming procedure.

Liquid-Liquid extraction is also a classical approach for extraction; however the technique usually requires several hours or more to achieve satisfactory recoveries of analytes and even sometimes fail to do. The solvent costs are often high. The solutions of the recovered analytes are often so dilute so that a concentration step must follow the extraction. Analyte degradation or loss as well as atmospheric pollution may accompany this concentration step. Due to these pitfalls, the need for easier and quicker analyses has prompted the development of new

techniques. Amongst them, the most outstanding technique is supercritical fluids extraction (SFE).

In recent decades, SFE has gained much more widespread acceptance in recent years as a promising alternative to conventional solvent extraction for extracting oils and other materials as it offers a number of advantages, including a lack of solvent residue and better retention of aromatic compounds. Moreover, conventional methods are usually carried out at high temperatures, which can be responsible for the destruction of valuable substances.<sup>8</sup>

### Advantages of SFE:

1. SCFs have relatively low viscosity and high diffusivity (the diffusivity of SCFs is  $10^4 \text{ cm}^2 \text{ s}^{-1}$  while that of liquid solvents is  $10^5 \text{ cm}^2 \text{ s}^{-1}$ ). Therefore, they can penetrate into porous solid materials more effectively than liquid solvents, and may render much faster mass transfer resulting in faster extractions. For in-stance, with comparable or better recoveries, the extraction time could be reduced from hours or days in a liquid–solid extraction (L–S) to a few tens of minutes in SFE

2. In SFE, a fresh fluid is continuously forced to flow through the samples; therefore, it can provide quantitative or complete extraction

3. In SFE, the solvation power of the fluid can be manipulated by changing pressure and/or temperature; therefore, it may achieve a remarkably high selectivity. This tunable solvation power of SCFs is particularly useful for the extraction of complex samples such as plant materials. One good example is the selective extraction of a vindoline component from among more than 100 alkaloid compounds from the leaves of *Catharanthus roseus*

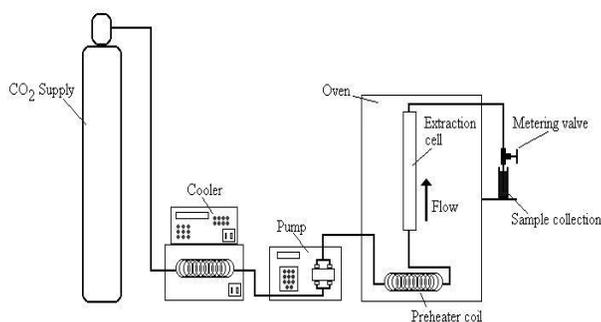
4. Solute dissolved in supercritical fluid can be easily separated by depressurization. Therefore, SFE can eliminate the sample concentration process, which is usually time-consuming and often results in loss of volatile matter.

5. SFE is performed at low temperatures, so it may be an ideal technique to study thermally labile compounds and may lead to the discovery of new natural compounds. For example, use of SFE to extract the chemical actives of ginger can effectively prevent many undesirable reactions such as hydrolysis, oxidation, degradation and rearrangement.

6. Compared with the 20–100 g of sample typically required in classical extraction methods, as little as 0.5–1.5 g of sample is needed in SFE methods. It has been reported that from only 1.5 g of fresh plant samples, more than 100 volatile and semi-volatile compounds could be extracted and detected by gas chromatography –mass spectroscopy (GC-MS), of which more than 80 compounds were in sufficient quantity for accurate quantifications

- 7. SFE uses no or significantly less environmentally hostile organic solvents. A while classical extraction methods require tens to hundreds of milliliters
- 8. SFE may allow direct coupling with a chromatographic method, which can be a useful means to extract and directly quantify highly volatile compounds
- 9. In large scale SFE processes, the fluid, usually CO<sub>2</sub> can be recycled or re-used thus minimizing waste generation.
- 10. SFE can be applied to systems of different scales, for instance, from analytical scale (less than a gram to a few grams of sample), to preparative scale (several hundred grams of sample), to pilot plant scale (kilograms of sample) and up to large industrial scale (tons of raw materials, such as SFE of coffee beans).<sup>8</sup>

**INSTRUMENTATION OF A SUPERCRITICAL FLUID EXTRACTOR**



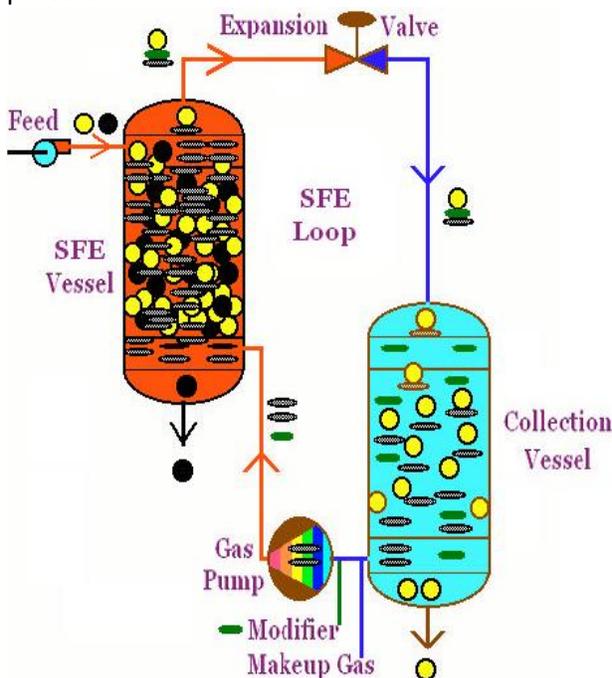
**Figure 2: Schematic diagram of supercritical fluid extractor**

As shown in Figure 2, the system contains a pump for the supercritical fluid, a pressure cell to contain the sample, a means of maintaining pressure in the system and a collecting vessel. The liquid is pumped to a heating zone, where it is heated to supercritical conditions. It then passes into the extraction vessel, where it rapidly diffuses into the solid matrix and dissolves the material to be extracted. The dissolved material is swept from the extraction cell into a separator at lower pressure, and the extracted material settles out. The supercritical fluid can then be cooled, re-compressed and recycled.

**Principle of extraction:**

SFE is a two step process which uses a dense gas e.g carbon-dioxide, as a solvent for extraction, above its critical temperature (31<sup>o</sup>) and critical pressure (74 bar). The feed, generally ground solid, is charged into the extraction vessel. Carbon dioxide is fed to the extraction vessel through a high-pressure pump (100 to 500 bars). The extract rich carbon dioxide is then sent to a separation vessel, which maintained at reduced pressure and temperature conditions, and the extract precipitates out in the separation vessel. The extract free carbon dioxide stream, leaving the separation vessel is then recycled back to the extraction vessel till the end of the batch.<sup>4</sup>

Figure 3 Depicts a schematic representation of the SFE process.



**Fig 3: Supercritical fluid extraction process**

**Modes of SFE:**

SFE allows for two extraction modes – dynamic (continuous) and static (batch). In the dynamic mode (Figure 4), the extraction vessel is constantly supplied with fresh supercritical fluid, while the recovery container is continually supplied with the solute. In the static mode the outlet of the extraction vessel is closed (Figure 5) and extraction takes place without the regeneration of supercritical fluid. On completion of extraction, the supercritical fluid rapidly rinses the vessel in order to permit the recuperation of the solute. Static extraction is preferred in situation dealing with low concentration of active constituents strongly bound to matrix.

There are three ways of recuperating the analyte after depressurization of the supercritical fluid viz. thermal trapping, sorbent trapping and solvent trapping. Thermal trapping is the simple technique because the supercritical fluid is simply depressurized in a cooled recovery container. Unfortunately, this technique is limited to non-volatile components as high gas flow lead to the loss of relatively volatile compounds. Slightly volatile compounds can be led by aerosol formation. The other two techniques offer increased potential for the quantitative extraction of highly or moderately volatile compounds. Sorbent trapping provides favorable results and is relatively easy to apply. The supercritical fluid is depressurized and then adsorbed on a solid support. Once the supercritical phase extraction has terminated, the analyte is recovered by elution with a small volume of solvent liquid. This method has the advantage of being able to increase the selectivity of the extraction by way of

Area of application	Sample name	Active constituent	Supercritical fluid	Application	Ref. No.
Forensic	Opiates from hair	Methadone, Morphine, Codeine, Ethyl morphine	CO <sub>2</sub> modified with a polar mixture: MeOH/TEA/H <sub>2</sub> O at a volume ratio of 85:6:3	Extraction & separation of drugs	15
	Post-mortem & vitrous humor	Methadone, Morphine	CO <sub>2</sub> with 15% MeOH/Et modifier		
	Human Hair	Cocaine	CO <sub>2</sub> modified with triethyl amine & water		
	Biological matrixes e.g. blood, urine	Benzodiazepine & derivatives	CO <sub>2</sub> modified with 2% (v/v) MeOH		
	Spiked human hair sample	Amphetamine	CO <sub>2</sub>		
Pharmaceutical	Propolis tincture	Flavonoids	CO <sub>2</sub>	Cosmetics & health food preparation for antimicrobial property	21
	Marihuana	Tetrahydrocarbinol	CO <sub>2</sub>	illicit drug in Hungary	13
	Pyrethrum	Pyrethrins	CO <sub>2</sub>	Strong action against insect & highly biodegradable pesticides	21
Food	Neem seed	Azadirachtins (tetranotriterpenoid), Nimbin	CO <sub>2</sub>	pesticides	21
	Guava seed	Linoleic acid	CO <sub>2</sub> with ethanol as co solvent	Nutritional cosmetic and medicinal industry	16
	Grape seed	Grape seed oil	CO <sub>2</sub>	Extraction of oil	19
	Almond seed	Almond oil	CO <sub>2</sub>		
	Pennyroyal leaves & flowers	Pennyroyal oil	CO <sub>2</sub>		
	Aqueous solution	Ethanol	CO <sub>2</sub>	Flooding point measurement	18
	Spiked solid	t-resveratol & phenolic compound	CO <sub>2</sub> with methanol as modifier	Source of antioxidant	20
	Eucalyptus bark	Triterpenoid	CO <sub>2</sub> with 8% (wt) of ethanol	Extraction & separation of terpenoid	17
	Rice bran oil	Tocopherol & Tocotrienol	CO <sub>2</sub>	Extraction & separation of Tocopherol & Tocotrienol	23
	Peppermint leaves	Peppermint oil	CO <sub>2</sub>	flavouring confectionery, liquors, cosmetics, toothpastes and many other products	14
	Soyabean oil	Lecithin	CO <sub>2</sub>	Extraction of oil	17
	Tobacco	Protein extraction from tobacco	Ethanol extraction	Reduce the nicotine & tar content of the burning tobacco	21
Nutraceutical	Chamoli flower	sesquiterpene, lactone, matricin	CO <sub>2</sub>	anti-inflammatory and antispasmodic	11
	Valarian	valepotriates	CO <sub>2</sub>	sedative preparation	
	Chitin & chitosan	Astaxanthin	CO <sub>2</sub>	Used in wound healing preparation	
	Wormwood extract	Toxic b thjuone	CO <sub>2</sub>	carminative, cholagogue and stomachic	

**Table 2 summarizes the use of SFE in various fields along with the type and nature of SCF.**

the elution solvent. The simplest technique, however, procedure does not offer the selectivity seen in sorbent remains that of trapping in a liquid solvent. While this trapping, it is undoubtedly simpler and quicker.<sup>3</sup>

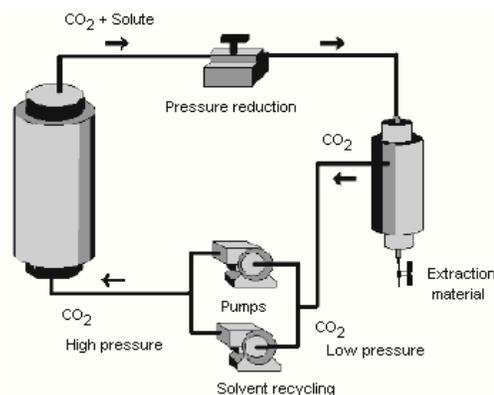


Figure 4: Schematic diagram of SFE in dynamic (continuous) mode

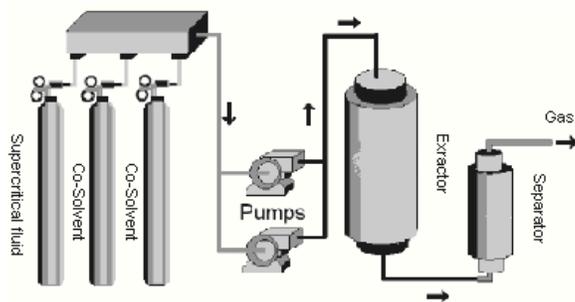


Figure 5: Schematic diagram of SFE in static (batch) mode

SFE comprises two integrated parts: extraction of the analytes from the sample matrix and subsequent collection (trapping) of the analytes. The collection can be achieved either on-line into a chromatographic instrument such as GC or SFC, or off-line by depressurizing the supercritical fluid (SF) into a collection device. The collection device can be an empty vessel, a vessel containing a small volume of organic solvent, a solid-phase trap, or a cryogenically cooled capillary. Hence, there are many possibilities for achieving collection in SFE. Consequently, for analytical applications, SFE is usually used in conjunction with chromatographic techniques, to improve the overall selectivity of the process in isolating specific solutes. SFE combined with chromatography can be either “off-line” or “on-line”

#### Advantages of on-line SFE:

1. Direct coupling of the analyte containing supercritical fluid to a chromatographic separation system with appropriate detection.
2. Eliminating sample handling after loading in the extraction<sup>10</sup>

SFE-based methods have a promising future in analytical lipid chemistry. Also they can be employed to considerable advantage as an aid in optimizing and testing the feasibility for non-analytical applications, i.e., to investigate the potential for scaling up SFE for industrial application. Using automated analytical SFE instrumentation for such purposes allows the rapid assessment as to whether a SFE will work at minimal expense and time. The commercial SFE instrumentation discussed herewith can be applied to solving a wide

variety of problems that encompass extractions, fractionations, and even reactions in critical fluid media. Such demonstrated utility further justifies the cost of the instrumentation and provides a synergism between analytical research and process development which utilize supercritical fluids.<sup>24</sup>

General awareness and understanding of supercritical and liquefied gas processing technology is increasing, as is understanding of where this technology best fits for processing of a wide range of emerging biological products. In particular, the use of microbiological systems for the primary production of chemicals has much promise for high volume sustainable production of dietary, medical and biochemical products. Associated with this is the need for development of efficient processes for separation of organic compounds from dilute aqueous systems and for the isolation and concentration of target compounds. Also of increasing importance is for products to be formulated for efficacy and stability. In all of these areas supercritical fluid processing need to be investigated, along with other processing alternatives, to determine the most favorable overall integrated process.<sup>25</sup>

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