# New Advances in Supercritical Fluid Extraction for the Quality Control/Quality Assurance Laboratory for Fat Analysis

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## Abstract

Supercritical fluid extraction has been gaining acceptance in quality control/quality assurance laboratories as a replacement technology for the traditional “organic solvent intensive” fat analysis methods. Conventional methods of fat analysis for baking dough, milk, and chocolate products are time and labor intensive, and require large amounts of hazardous organic solvents. Supercritical fluid extraction using $CO_2$ as a solvent is an alternative method for extraction and isolation of fat content from these products. SFE for baking dough, liquid milk, and chocolate will be explored in this poster using the SFT-100 Extraction Unit and results compared to the traditional solvent intensive methods.


## Supercritical Fluid Extraction – SFT-100 SFE Unit

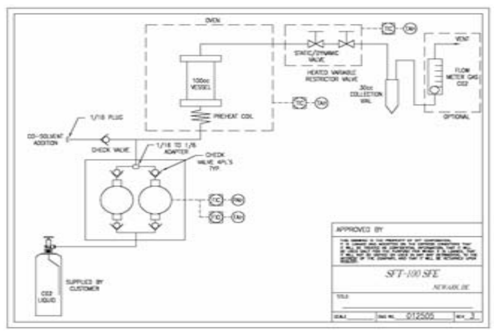
The SFT-100 Supercritical Fluid Extractor (SFE) possesses many features typically found in more costly SFE equipment. The Unit may be employed for a wide variety of applications from routine analytical QC/QA work to detailed process development. The SFT-100 SFE was designed for ease of use for scientists who want to investigate the feasibility of applying supercritical fluid techniques to a wide variety of analytical and processing problems.

The SFT-100 SFE can accommodate 10 ml to 100 ml sample extraction vessels and may be operated at pressures up to 10,000 psi (68.9 MPa) and at temperatures ranging from ambient to 200°C. The wide range of vessel volumes available makes the unit well suited for both analytical scale QC/QA SFE applications and basic process development work. For example, with a 100 ml sample vessel, the SFT-100 can extract very low levels of key components from materials and process larger amounts of bulk material than would be possible with smaller, analytical scale SFE equipment. Inside the SFT-100's oven, a preheater ensures that the temperature of the fluid reaching the extractions vessel is controlled precisely. This is essential to obtaining accurate, repeatable extraction results. The SFT-100 incorporates a high performance, dual piston pump which produces the high pressures required for supercritical fluid work. The system has built-in safety precautions to prevent accidental over-temperature or overpressure conditions. As an additional safety backup, a rupture disc assembly provides mechanical protection against accidental over pressurization of the system. Manually operated valves ensure long term, maintenance free operation. An integrated program logic controller monitors and adjusts fluid pressure inside the extraction vessel to achieve and maintain a desired set point. A PID temperature controller monitors and maintains the precise fluid temperature inside the high pressure vessel. A robust, variable restrictor valve (back pressure regulator) provides precise control over flow rates, which is essential in obtaining highly reproducible results from run to run. Flow rates can range from 0.1 to 24 ml/min. (0.08 to 18 grams/min.) of liquid CO2 under typical operating conditions. While carbon dioxide is the most commonly used solvent, the SFT-100, with some modification, allows the user flexibility to work with a variety of supercritical fluids. An optional co-solvent addition module is available for the SFT-100 which can allow the extraction of more polar analytes of interest. Extract collection options include: solid phase extraction (SPE) cartridges, fractional cyclone separators, and EPA type sample vials.

## Typical Supercritical Fluid Extraction Unit

A typical SFE Extractor is comprised of a high pressure carbon dioxide pump, fluid preheater, co-solvent addition module, extraction sample cell, heated micrometering restrictor valve, extract collection/separator vial and flow meter. The sample to be analyzed is placed in the extraction vessel and supercritical carbon dioxide flows through the preheat assembly into sample vessel. Analytes of interest are solublized in the supercritical fluid and then collected the collection vial. The micrometering restrictor valve depressurizes the sample from the supercritical fluid state (to the gas state) and the analyte of interest precipitates in the collection vial for further analysis. Typically in fat analysis the extract or the remaining sample is weighed to determine the overall fat content in the original sample.


SFT-100 SFE Unit




SFT-100 SFE Flow Diagram

## Case Study #1: Fat Analysis of Pie Crust Dough Using SFE:

The traditional fat analysis method for determination of fat content in pie dough crust is very labor and organic solvent intensive. SFT developed a simple SFE/Ethanol extraction Method to determine the total fat content in pie crust dough.

**Typical Procedure:** Approximately 3.5 grams of pie dough was placed into the SFT-100 fitted with a 25 ml Extraction Vessel (5 micron frit assemblies), sealed, the Static/Dynamic Valve was moved to the closed position, 1 ml of Ethanol was metered into the sample chamber using the Co-Solvent addition module. The Pressure and Temperature in the sample vessel was brought to 9000 psi and 115 degrees Celsius. The sample was allowed to soak at these conditions for 15 minutes. The Static/Dynamic valve was opened and the restrictor valve was set to allow steady flow of 5 ml of neat CO2 liquid to flow through the sample vessel for 5 minutes. The carbon dioxide pump was turned off and the system allowed to vent and cool to atmospheric conditions.

**Results:** Three trials yielded total fat content of 23.10%, 22.96%, and 23.04% compared to the traditional solvent method analysis of 23.05% for the same sample lot of dough. Total sample processing time was 22 minutes compared to 1.0 hour by the traditional method.

## Case Study #2: Gravimetric Fat Analysis of Milk Using SFE:

The traditional fat analysis method for determination of fat content in milk is very labor and organic solvent intensive. A simple SFE/Ethanol extraction method was employed to determine the fat content in liquid milk samples.

**Typical Procedure:** Approximately 2.0 grams of milk was mixed with 5.0 grams of solid phase extraction matrix and mixed thoroughly. The sample mixture was then poured into a 10 ml sample extraction vessel (fitted with 5 micron frit assemblies), sealed, the Static/Dynamic Valve was moved to the closed position, and 0.5 ml of Ethanol was metered into the sample chamber using the Co-Solvent addition module. The Pressure and Temperature in the sample vessel was brought to 9000 psi and 100 degrees Celsius. The Static/Dynamic valve was opened and dynamic flow was set to 6 ml of carbon dioxide liquid for 25 minutes using the restrictor valve. Collection of the milk fat was into a pre weighed collection vial.

**Results:** Three trials yielded fat content of 2.10%, 2.06%, and 2.04% compared to the traditional solvent method analysis of 2.05% for the same milk sample. Total sample processing time was 28 minutes compared to longer with the traditional Mojonnier method.

## Case Study #3: Fat Analysis of Chocolate Samples Using SFE:

Traditional Gravimetric fat analysis of chocolate samples are typically performed using a standard soxhlet apparatus with petroleum ether as the solvent. The soxhlet method from start to a result being obtained is approximately 8 hours. Supercritical extraction of fats from chocolate products eliminates the organic solvent and significantly on the processing time.

**Typical Procedure:** Approximately 3.0 grams of milk was added to 5.0 grams of solid phase extraction matrix and mixed thoroughly. The sample mixture was then poured into a 10 ml sample extraction vessel (fitted with 5 micron frit assemblies), sealed, the Static/Dynamic Valve was moved to the closed position. The Pressure and Temperature in the sample vessel was brought to 9000 psi and 80 degrees Celsius. The sample was allowed to soak statically for 10 minutes and then the Static/Dynamic valve was opened and dynamic flow was set to 6 ml of carbon dioxide liquid for 15 minutes using the restrictor valve. Collection of the chocolate fat was into a pre weighed collection vial.

**Results:** 4 trials yielded a % fat of 37.05% in comparison to the traditional soxhlet method of 37.10%. Sample preparation and processing time was reduced significantly using SFE from a typical 8 hour experimental run with soxhlet to 25 minutes as a replacement for standard soxhlet or Foss-Let techniques.

## Conclusions

In each of the cases the fat analysis using supercritical fluid extraction of the pie dough, liquid milk, and chocolate closely repeated the results found by the more traditional time, solvent, and labor intensive sample preparation and analysis methods.