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Standard Guide for Supercritical Fluid Chromatography Terms and Relationships[1]

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1. Scope

1.1 This guide deals primarily with the terms and relationships used in supercritical fluid chromatography.

1.2 Since many of the basic terms and definitions also apply to gas chromatography and liquid chromatography, this guide is using, whenever possible, symbols identical to Practices E355 and E682.

1.3 The values stated in SI units are to be regarded as standard. No other units of measurement are included in this standard.

2. Referenced Documents

2.1 *ASTM Standards:*[2]

E355 Practice for Gas Chromatography Terms and Relationships

E682 Practice for Liquid Chromatography Terms and Relationships

3. Names of Techniques

3.1 *Supercritical Fluid Chromatography,* abbreviated as SFC, comprises all chromatographic methods in which both the mobile phase is supercritical under the conditions of analysis and where the solvating properties of the fluid have a measurable affect on the separation. Early work in the field was performed under a broader heading–*dense gas chromatography*. Related work in the field uses subcritical or near-critical conditions to affect separation.

3.2 Separation is achieved by differences in the distribution of the components of a sample between the mobile and stationary phases, causing them to move through the column at different rates (differential migration).

3.3 In supercritical fluid chromatography, the pressure may be constant or changing during a chromatographic separation.

3.3.1 *Isobaric* is a term used when the mobile phase is kept at constant pressure. This may be for a specified time interval or for the entire chromatographic separation.

3.3.2 *Programmed Pressure Supercritical Fluid Chromatography* is the version of the technique in which the column pressure is changed with time during the passage of the sample components through the separation column. Isobaric intervals may be included in the pressure program.

3.4 In supercritical fluid chromatography, the temperature may be constant, or changing during a chromatographic separation.

3.4.1 *Isothermal Supercritical Fluid Chromatography* is the version of the technique in which the column temperature is held constant during the passage of the sample components through the separation column.

3.4.2 *Programmed Temperature Supercritical Fluid Chromatography* is the version of the technique in which the column temperature is changed with time during the passage of the sample components through the separation column. Isothermal intervals may be included in the temperature program.

3.5 In supercritical fluid chromatography, the density may be constant or changing during the chromatographic separation.

3.5.1 *Isoconfertic* is a term used when the density of the mobile phase is kept constant for a specified time or for the entire chromatographic separation.

3.5.2 *Programmed Density Supercritical Fluid Chromatography* is the version of the technique in which the column density is changed with time during the passage of the sample components through the separation column. Isoconfertic intervals may be included in the density program.

3.5.3 *Flow Programming* is a technique where the mobile phase linear velocity is changed during the chromatographic procedure. However, with fixed orifice restrictors, flow programming is more complex requiring an increase in pressure to effect an increase in linear velocity.

3.6 In supercritical fluid chromatography, the composition of the mobile phase may be constant or changing during a chromatographic separation.

[1] This guide is under the jurisdiction of ASTM Committee E13 on Molecular Spectroscopy and Separation Science and is the direct responsibility of Subcommittee E13.19 on Separation Science.

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[2] For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

3.6.1 The term *Isocratic* is used when the composition of the mobile phase is kept constant during a chromatographic separation.

3.6.2 The term *Gradient Elution* is used to specify the technique when a deliberate change in the mobile phase composition is made during the chromatographic procedure. *Isocratic* intervals may be included in the gradient program.

4. Apparatus

4.1 *Pumps*—The function of the pumps is to deliver the mobile phase at a controlled flow rate to the chromatographic column.

4.1.1 *Syringe Pumps* have a piston that advances at a controlled rate within a smooth cylinder to displace the mobile phase.

4.1.2 *Reciprocating Pumps* have a single or dual chamber from which mobile phase is displaced by reciprocating piston(s) or diaphragm(s).

4.2 *Sample Inlet Systems* represent the means for introducing samples into the columns.

4.2.1 *Direct Injection* is a sample introduction technique whereby the entire volume of sample is swept onto the head of the analytical column. Its use is most prevalent in packed column SFC.

4.2.2 *Split-Flow Injection* introduces only a portion of the sample volume onto the analytical column so as to prevent overloading of the column in open tubular SFC. This is achieved by the use of a splitter tee or similar contrivance, such that the incoming slug of sample is divided between the analytical column and a flow restrictor vented to waste. The amount of sample deposited on the column is a function of the ratio of the flow to the column versus the flow through this restrictor. This ratio can thus be adjusted for different samples and column capacities.

4.2.3 *Timed-Split (Moving-Split) Injection* achieves the same end result as split-flow injection. The volume of sample introduced onto the column is governed by the rapid back-and-forth motion of an internal-loop sample rotor in a valve. The time interval between the two motions determines the volume of sample injected, with shorter times delivering smaller volumes.

4.2.4 *On-Line Supercritical Extraction* is a means of directly introducing a sample or portion of a sample into a supercritical fluid chromatograph. The sample is placed in an extraction cell and extracted with the supercritical fluid. The extraction effluent containing the solutes of interest are ultimately transferred to the column by the action of switching or sampling valves. This can be accomplished with or without solute focusing (that is, using a suitable trap such as a cryogenic trapping).

4.3 *Columns* consist of tubes that contain the stationary phase and through which the supercritical fluid mobile phase flows.

4.3.1 *Packed Column Supercritical Fluid Chromatography* uses an active solid or a liquid that is chemically bonded to a solid and packed into a column, generally stainless steel or fused silica; as the stationary phase.

4.3.2 *Wall-Coated Open-Tubular Supercritical Fluid Chromatography* uses a liquid that is chemically bonded to the wall of an open-tubular column as stationary phase. Fused silica tubing columns, internal diameter (i.d.) > 100 µm, may shatter at pressures employed in SFC. A high degree of crosslinking is desirable to reduce stationary phase solubility in the mobile phase.

4.4 *Restrictors* are devices employed to maintain the pressure in the chromatographic system. The pressure of the supercritical fluid is usually reduced to ambient after passage through the restrictor. The mobile phase flow rate is determined by the restrictor dimensions or operation. The restrictor is placed before some types of detectors (for example, flame ionization, mass spectrometer) and after other types of detectors (for example, UV).

4.4.1 A *Linear Restrictor* is a length of small i.d. tubing of uniform bore. Linear restrictors are made of polyimidecoated fused silica tubing, or stainless steel or other tubing of the appropriate diameter. The amount of restriction provided is dependent upon both the length and i.d. of the tubing.

4.4.2 A *Tapered Restrictor* is a length of small i.d. tubing where one end has been reduced by drawing in a flame in the case of fused silica tubing, or crimped in the case of metal tubing.

4.4.3 An *Integral Restrictor* (**1**)[3] consists of a length of fused silica tubing with one end closed by heating with a microtorch. This closed end is then ground until a hole with the desired initial linear velocity is obtained.

4.4.4 A *Converging-Diverging Restrictor* (**2**) has the wall of the tubing collapsed slightly near one end forming a constriction. This constriction is similar to a venturi in profile and the point of smallest diameter is located about 1 to 2 mm from the end of the tubing.

4.4.5 An *Orifice* is a type of restrictor which uses a metal disk or diaphragm with an appropriately sized opening. This type normally requires an adapter or holder specifically designed to couple the device to a detector.

4.4.6 A *Porous Frit Restrictor*[4] consists of a length of fused silica tubing containing a porous plug at one end.

4.4.7 A *Back Pressure Regulator* consists of a diaphragm valve which can be adjusted to control the pressure maintained on its inlet (instrument) side. The outlet discharge pressure is nominally one atmosphere.

4.5 *Detectors* are devices that respond to the presence of eluted solutes in the mobile phase emerging from the column. Ideally, the response should be proportional to the mass or concentration of solute in the mobile phase. Detectors may be divided either according to the type of measurement or the principle of detection.

4.5.1 *Differential Concentration Detectors* measure the proportion of eluted sample component(s) in the mobile phase passing through the detector. The peak area is inversely proportional to the mobile phase flow rate.

[3] The boldface numbers in parentheses refer to a list of references at the end of this standard.

[4] Cortez, H., Pfeiffer, C., Richter, B., and Stevens, T. U. S., Patent No. 4 793 920, 1988.

4.5.2 *Differential Mass Detectors* measure the instantaneous mass of a component within the detector per unit time (g/s). The area under the curve is independent of the mobile phase flow rate.

5. Reagents

5.1 *Supercritical Fluid* is a fluid state of a substance intermediate between a gas and a liquid. A supercritical fluid may be defined from the accompanying phase diagram (Fig. 1). The supercritical fluid region is defined by temperatures and pressures, both above the critical values. A subcritical fluid (or liquid) is a compound that would usually be a gas at ambient temperature but is held as a liquid by the application of pressure below its supercritical point.

5.1.1 The *Critical Temperature* is the temperature above which a substance cannot be liquefied or condensed no matter how great the applied pressure.

5.1.2 The *Critical Pressure* is the pressure that would just suffice to liquefy the fluid at its critical temperature.

5.1.3 The *Reduced Pressure* is the ratio of the working pressure to the critical pressure of the substance.

5.1.4 The *Reduced Temperature* is the ratio of the working temperature to the critical temperature of the substance.

5.1.5 The *Density* of a supercritical fluid (the weight per unit volume of the fluid) in chromatographic separations is calculated from an empirical equation of state.

5.2 A *Modifier or co-solvent* is a substance added to a supercritical fluid to enhance its solvent strength, usually by increasing the polarity of the mobile phase, or binding to active sites on a stationary phase.

5.3 The *Stationary Phase* is composed of the active immobile materials within the column that selectively retard the passage of sample components. Inert materials that merely provide physical support or occupy space within the columns are not part of the stationary phase.

NOTE 1—Extremely porous stationary phases may exhibit exclusion phenomenon in addition to adsorptive interactions.

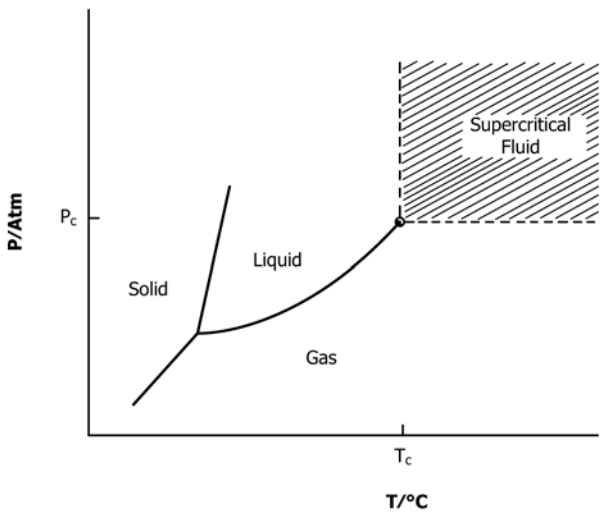

FIG. 1 Phase Diagram

5.3.1 An *Interactive Solid* is a stationary phase material with bulk homogeneity where the surface effects separation by adsorptive interactions. Examples are silica and alumina.

5.3.2 A *Bonded Phase* is a stationary phase that has been covalently attached to a solid support. The sample components partition between the stationary and mobile phases which results in separation. Octadecylsilyl groups bonded to silica gel particles and polydimethylsiloxane (or dimethyl polysiloxane) bonded to deactivated fused silica column wall represent examples for packed column and open tubular column phases, respectively.

5.4 The *Solid Support* is the inert material that holds the stationary phase in intimate contact with the mobile phase. It may consist of porous or impenetrable particles or granules or the interior wall of the column itself, or a combination of these.

5.5 The *Column Packing* consists of all the material used to fill packed columns, including the solid support and the bonded phase or the interactive solid.

5.6 *Solutes* are the sample components that are introduced into the chromatographic system and are transported by the mobile phase and elute through the column. Some solutes may be unretained.

6. Readout

6.1 A *Chromatogram* is a plot of detector response against time or effluent volume. Idealized chromatograms obtained with a differential detector for an unretained substance and one other component are shown in Fig. 2.

6.2 The definitions in 6.2.1 – 6.2.6 apply to chromatograms obtained directly by means of differential detectors or indirectly by differentiating the response of integral detectors.

6.2.1 A *Baseline* is that portion of a chromatogram where no detectable sample components emerge from the column.

6.2.2 A *Peak* is that portion of a chromatogram where a single detectable component, or two or more unresolved detectable components, elute from the column.

6.2.3 The *Peak Base*, CD in Fig. 2, is the interpolation of the baseline between the extremities of a peak.

6.2.4 The *Peak Area*, CHFEGJD in Fig. 2, is the area enclosed between the peak and the peak base.

6.2.5 *Peak Height*, EB in Fig. 2, is the perpendicular distance measured in the direction of detector response, from the peak base to peak maximum.

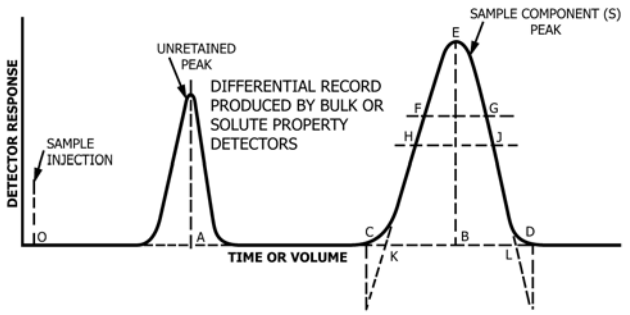

FIG. 2 Typical Chromatogram