# HPLC Terminology (Short Guide)

Absorption:
The process where a chemical entity enter the bulk of a liquid, solid or gas phase. In chromatography the term usually signifies the process by which a solute partitions into a liquid-like stationary phase.

Additive:
A compound added to the mobile phase to improve the chromatographic analysis.

Adsorbent:
The packing used in adsorption chromatography.

Adsorption:
The process where a chemical entity is accumulated on a surface.

Adsorption Chromatography
Separation based on differences in adsorption of the components to the stationary phase surface.

Affinity Chromatography
Chromatographic separation based on a specific interaction between the analyte and a ligand bound to the stationary phase surface.

Anion exchange chromatography:
The chromatographic process that is used to separate anions by using an ionized positively charged stationary phase. Tetraalkylammonium ions are often used as anion exchange functional groups.

Bonded phase
A stationary phase which is covalently bonded to the support particles or the inside wall of a tube.

Capillary column:
Columns with an inner diameter less than 0.5 mm.

Capillary LC:
Liquid chromatography performed by using a capillary column.

Column
The tube and the stationary phase through which the mobile phase flow.

Counterion:
When the term is used in ion exchange chromatography it means the ions added to the mobile phase with charge opposite to the ions bonded to the stationary phase.

Eluent:
Another word for the mobile phase.

Gradient elution:
The chromatographic technique by which a mobil phase gradient is used to modulate the retention times. Usually the mobile phase composition changes so that its strength increases with time.

Guard column:
A small column that protects the analytical column from contamination, it is placed between the injector and the analytical column.

High performance liquid chromatography (HPLC):
A term coined for the modern and instrumentally developed form of column liquid chromatography. It is characterised by high flow rates and high back column pressure.

Hold-up Volume
The volume ( or corresponding time ) of mobile phase required to elute a component that does not interact with the stationary phase. I.e. the component is not retained by the stationary phase.

Indirect detection:
A detection technique where the solute is indirectly detected by measuring the change in mobile phase composition at column outlet. A prerequisite for this technique is that the adsorption isotherm of a component in the mobile phase depends on the concentration of the solute. E.g. a non-UV absorbing solute is indirectly detected with an UV-detector by adding an UV absorbing component to the mobile phase. If the adsorption isotherm of this component depends on the concentration of the solute, its variation in concentration at the column outlet, caused by the elution of the solute, can be detected with an UV-detector.

Ion chromatography (IC):
A technique in which low concentrations of ionic solutes are determined by using ion exchangers of low exchange capacity and mobile phase with low ionic strength.

Ion exclusion:
The exclusion of co-ions from the surface layer. In chromatography the ion exclusion effect implicates that co-ions migrates faster through the column than a neutral molecule.

Ion pair chromatography:
A form of reversed phase chromatography in which a charged organic molecule, the ion pair reagent, is added to the mobile phase. The ion pair reagent adsorbs to the stationary phase surface and creates a charged surface layer. Ions of opposite charge are attracted to the charged surface layer and ions of the same charge are repelled . The retention of ions is modulated by changing the concentration of ion pair reagent in the mobile phase.

Partition Chromatography
Separation based mainly on differences in solubilities between the mobile and stationary phase.

Peak
The part of the chromatogram where the detector response is caused by a solute.

Peak Area
The area of the peak as registered by the detector.

Peak-Width
The width of the peak registrered by the detector. It may be represented in the dimension time or volume. For a Gaussian formed peak, the peak-width is related to the standard deviation (s) of the peak. The peak width can be estimated by several different methods.

Plate Height ( H)
The column length ( L ) divided by the plate number:
$H = L / N$

Plate Number ( N )
A dimensionless number that is a measure of the effectivity of a column.
$N = ( V_R / s)2$

Resolution ( Rs )
A measure how well two peaks are separated. It is defined as:
$R_s = 2(t_{R2} - t_{R1}) / (w_{b1} + w_{b2})$
$t_R$ = Retention time, $w_b$ = peak width at base

Retention Factor (k)
The ratio of the adjusted retention volume ( or time ) and the hold-up volume ( or time );
$k = V'_R / V_M = t'_R / t_M$
The retention factor has for many years also been called the capacity factor, k´. This usage is not recommended by IUPAC.

Retention Volume ( $V_R$ ) ( or Time ( $t_R$ )
The volume ( or corresponding time ) of mobile phase that passes through the column between sample injection and the emergence of the peak maximum. Note the difference between this and the adjusted retention volume ( or time ).

Separation Factor ( a )
The relative retention values for two adjacent peaks;
$a = V'_{R2}/V'_{R1} = k_2 / k_1$
$V'_{R2}$ is chosen to be the larger value so that the separation factor becomes larger than unity.

Size Exclusion Chromatography
Separation based mainly on differences in molecular size. Differences in shape and/or charge may also contribute to the separation.

Stationary phase
One of the two phases in a chromatographic system. In a chromatographic system the analyte is distributed between the mobile phase and the stationary phase.

Void Volume
The volume in the column that is filled with the mobile phase. In the ideal case it is equal to the mobile phase hold up volumne.