

High Performance Liquid Chromatography (HPLC)

Motivation

- High Performance Liquid Chromatography (HPLC) is a chromatographic method to separate a great variety of organic and inorganic substances.
- HPLC methods are capable to qualify and quantify substances, using standards.
- Both, volatile and non-volatile substances can be analysed
- Fast method with a good accuracy

Experimental setup

- Standard HPLC device using 2 solvents as mobile phase
- The Diode Array Detector (DAD) offers detection of substances over a range of UV wavelength (200 to 900 nm)
- Complete data evaluation with PC

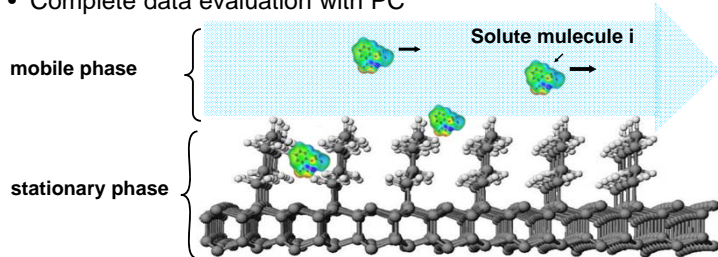


Fig. 1. Schematic of HPLC functional principle: Solute molecule partitions between two functional HPLC phases

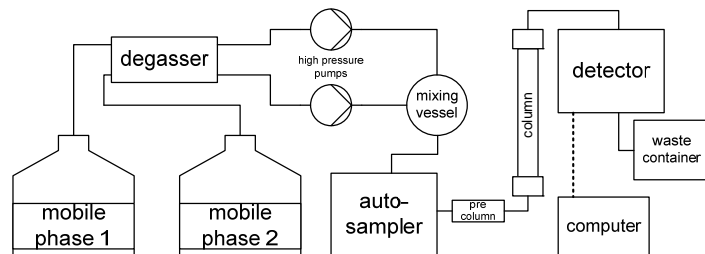


Fig. 2. Basic flow chart of a HPLC system setup

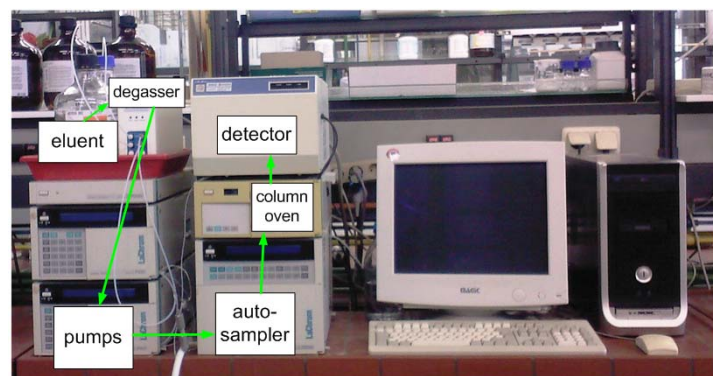


Fig. 3. HPLC system setup

Data reduction

- Peak areas are proportional to the amount of a component in the mobile phase
- Determination of LLE at finite and infinite concentrations
- Determination of selectivity α of two solutes in various solvents

$$\alpha_{21} = \frac{t_{R,2} - t_{0,ext}}{t_{R,1} - t_{0,ext}} = \frac{K_2}{K_1}$$

Example

Caffeine analysis

Caffeine analysis is used amongst others for practical training of chemical engineering students to give them an insight into established physico-chemical analysis methods.

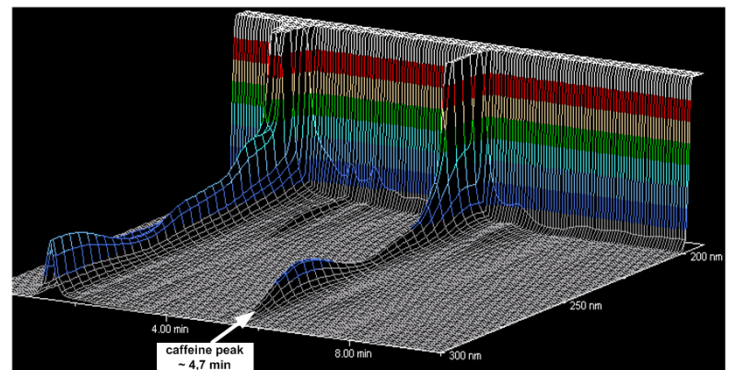


Fig. 4. 3D-Chromatogram of Coca Cola analysis on a C-18 column using a methanol /water mixture as mobile phase

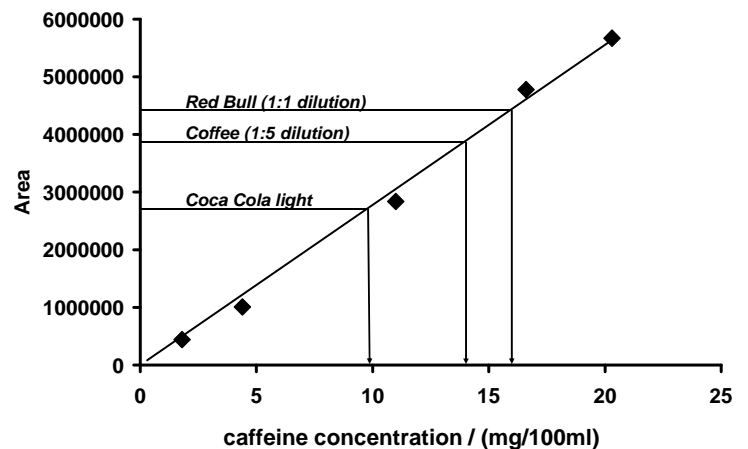


Fig. 5. Diagram showing data points of caffeine standard calibration as well as measured data for coffee, tee, and Coca Cola samples.