Abstract

This thesis deals with finding optimal chromatographic conditions for a separation of p-aminophenol, paracetamol, caffeine and propyphenazon by HPLC. These analytes have different polarity and acidobazic properties. The application of traditional octadecylsilane stationary phases for their simultaneous determination is problematical. The PEG column have proved to be suitable for an efficient separation of the analytes with different polarities, i.e. with different chromatographic behavior. The resulting method brings shortening of analysis time, which is important for analysis of big series.

Within the optimization of chromatographic conditions:

• a wide range of mobile phases has been tested (various organic solvents, various ratio of organic components to water phase components)
• a various level of acidity of the mobile phase has been tested ranging from 4 to 7 pH units
• various buffers have been tested
• behavior of the analytes has been tested under an enhanced temperature
• two PEG columns with different lengths have been tested

Optimal chromatographic conditions for a separation of the analytes mentioned above, as well as the fulfillment of requirements for simplicity, analysis time and high degree of reliability have been found.

The final chromatographic conditions:

Stationary phase: Supelco Discovery HS PEG column (15 x 4 mm, 3 µm)

Mobile phase: acetic buffer pH 4.5, 0.04% of triethylamine

Flow-rate: 1 ml/min

Detection: UV detection at 254 nm

Isocratic flow

Temperature: laboratory

Analysis time: 7 minutes