

## **Abstract of the PhD thesis**

### ***From simple to complex systems - development of capillary electrophoresis methods for the analysis of medicinal and psychoactive substances***

For the analysis of multi-component mixtures, separation techniques, such as: gas chromatography, liquid chromatography, or capillary electrophoresis, are most often used. These techniques make it possible to separate and determine many components in one measurement. The technique of capillary electrophoresis (CE) is still an intensively developed analytical technique. In comparison to other separation techniques CE is characterized by high efficiency, requires smaller amount of the sample and consumes less reagents. Furthermore, it enables conducting reliable analysis both using commercial instruments and simple systems constructed with low costs in laboratory.

The topics covered in this doctoral thesis include the use of simple, as well as complex capillary electrophoresis systems. Both in terms of constructionally simple instruments and complex and automated systems. In addition, chemical systems (i.e. separation buffers) can also be considered as simple, when consisting of only two substances, where the behaviour of the compounds introduced into them can be described and simulated. However, some of the chemical systems used with the capillary electrophoresis technique are not easy to describe, and the interactions affecting the substances introduced in the sample segment are complex (such as in the presence of micelles and an organic solvent). The aim of this work was to elaborate the possibilities of the CE technique both in development of new methods and further advanced automation of the analytical procedure with the use of commercially available instrument as well as present possibilities offered by portable systems for forensic and pharmaceutical analyses.

In the first part the possibilities of the portable capillary electrophoresis system with the contactless conductivity detection are presented. The constructed in the laboratory system is miniaturized, transportable and much cheaper than the commercial ones. It is constructed of high voltage power supply, electrodes, capillary and detector, the sampling is performed manually and it does not have capillary thermostating. Two methods were developed for determination of tropane alkaloids (atropine and scopolamine) isolated from plant material. First one, uses the extraction with the methanol:water mixture (7:3, v/v) and capillary zone electrophoresis technique. While the second one, utilize menthol-based deep eutectic solvent

for the extraction, and the analysis is performed with the non-aqueous capillary electrophoresis technique. Both methods enable determination of selected analytes with acceptable accuracy and precision.

Automation of the analytical process enables reduction of the reagents' consumption, limits the influence of the operator on the results and may also shorten the time of the whole analytical procedure. The automated methods not only in case of the analysis but also of the sample pre-treatment are nowadays particularly desirable. Thus, in this work the procedure for the preparation of the calibration solutions with the CE instrument (without any modification into the system) is proposed. The base of the procedure is the transfer of the standard solution and the diluent via the capillary with the appropriate pressure and time to obtain the assumed fold dilution. The procedure was verified by comparing the results for a series of calibration solutions prepared using the developed method and in the traditional way (manual pipetting). Proposed procedure allows to reduce the reagents consumption, however it requires more time for preparation of solutions than manual pipetting. Nevertheless, such procedure can be used when working with expensive or very expensive biochemical reagents.

The last part covers the subject of the extraction and analysis of the neutral and hydrophobic compounds found in plants. Analysis of such substances is challenging both in case of selective isolation from the plant material, and analysis with the CE technique. Due to the hydrophobic character of the analysed compounds, it was necessary to exploit the microemulsion electrokinetic chromatography and to modify the background electrolyte with organic solvent and cyclodextrin, obtaining chemically complex system. For the extraction of the desired compounds micelle-mediated extraction was chosen, to ensure the compatibility of the extraction and separation techniques. Developed method was successfully used to analyse samples of nutmeg containing myristicin and elemicin.

The developed methods include the multi-faceted development of capillary electrophoresis methods, enabling its wider application in both analytical chemistry and pharmaceutical analysis. They also emphasize the wide possibilities of CE, resulting from the variety of techniques offered within its framework. The presented methods were also evaluated using the *RGB12* procedure, which allows to highlight the strengths and weaknesses of the compared methods or techniques, taking into account the aspects of greenness, analytical efficiency and practicality of operating the systems.