Capillary electrophoresis in analysis of biomarkers

Pavol Ďurč[1,2], Júlia Lačná[1,2], František Foret[1,3], Petr Kubáň[1,3]

[1]*Department of Bioanalytical Instrumentation, CEITEC Masaryk University, Veveří 97, 602 00 Brno, Czech Republic*
[2]*Department of Chemistry, Masaryk University, Kotlářská 2, 611 37 Brno, Czech Republic*
[3]*Department of Bioanalytical Instrumentation, Institute of Analytical Chemistry of the Czech Academy of Sciences, v.v.i., Veveří 97, 602 00 Brno, Czech Republic*

I. A new non-invasive method for diagnosis of cystic fibrosis based on ion ratios determined by CE-C4D

A new approach for diagnosis of cystic fibrosis (CF) particularly in newborn CF screening is presented. The current CF screening plan consists of IRT analysis, DNA genetic analysis and sweat test. It has significantly improved the detection rate and lead to improved survival rate due to the early treatment initiation.

Currently, the sweat test is considered a golden standard for diagnosis of CF. The sweat test is based on measuring elevated chloride concentration in sweat of CF patients (above 60 mmol/L) in comparison to healthy individuals. However, the sweat test is time-consuming, may be uncomfortable and sometimes lead to inconclusive diagnosis, when the chloride concentration falls within 30-60 mmol/L.

Here, we present fast sampling technique and capillary electrophoresis method for analysis of ionic content in sweat. The novel sampling technique consists of wiping the skin of the forearm and extraction of the obtained skin-wipe sweat sample. The analysis of skin-wipe sweat sample is performed by capillary electrophoresis with contactless conductivity detection (CE-C4D), that allows to analyze not only Cl^- but also other ions, that can improve diagnostics of CF. We present here the analytical parameters of the method, such as sensitivity, specificity and repeatability based on a statistically significant group of patients and healthy individuals. The correlation of chloride concentration in sweat measured by our CE method and standard coulometric analysis method used in routine clinical practice is also shown.

Our developed sampling technique and CE-C4D method for multi-ion analysis can be a suitable surrogate to standard sweat test in CF diagnostics that is simpler, cheaper and completely non-invasive. The method has also a potential to be a novel tool for improvement of the classification of CF patients.

II. Fast blood plasma separation device for point-of-care applications

In this work, a simple device for extremely fast separation of blood plasma from diluted whole blood was developed. The device accommodates an asymmetric polysulfone membrane/supporting membrane sandwich that allows collection of 10 µL blood plasma into a narrow glass capillary in less than 10 s. The composition of diluent solution was optimized in order to achieve maximum recoveries for selected metabolites of alcohol intoxication. 5% solution of [tris(hydroxymethyl)methylamino] propanesulfonic acid provided recoveries of formate, oxalate and glycolate close to 100% and only moderate erythrocyte lysis. Both charged and uncharged compounds from the whole blood samples can be analyzed in the separated blood plasma by capillary electrophoresis with contactless conductometric detection and spectrophotometry,

respectively. The developed device might find wide application in on-site testing and point-of-care analysis, when only microliter volumes of whole blood are available.