From electrokinetic preconcentration to high resolution separation: a novel capillary electrophoresis approach to quantify amyloid peptides directly from cerebrospinal fluids

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The Amyloid Beta (Aβ) peptide 1-42 is an established Cerebrospinal Fluid (CSF) biomarker for the diagnosis of Alzheimer's disease (AD) as its level is lowered in the CSF of AD patients(1). Nevertheless, its quantification is not sufficient for a reliable molecular diagnosis of AD. To discriminate further AD from other neurodegenerative diseases, combinations of various Aβ isoforms have been proposed (2). Immunoassays are widely used to measure Aβ 1-42 levels. While antibodies specific for Aβ 1-42 and Aβ 1-40 are readily available, this is not the case for other Aβ peptides found in biological fluids. In addition, unsatisfactory inter-laboratory reproducibility of these ELISA is a major issue. Our aim is to explore the potential of Capillary Electrophoresis (CE) to propose a new approach of AD diagnosis overcoming current limitations.

The first challenge has been to develop high resolution separation of Aβ peptide family having close-related structures, differing sometimes only one or two amino acids. CE and microchip capillary electrophoresis were employed for this purpose with encouraging achievements(3,4). CE-based techniques can provide rapid and highly-efficient separations, however they suffer from poor sensitivities. We thus investigated two strategies to overcome this issue: fluorescent labelling of peptides and on-line preconcentration of the analytes prior to CE operation. Both allowed to increase the detection sensitivity to some extend (LOQ down to 10 nM)(5, 6). Very recently, we developed a novel electrokinetic preconcentration approach, combining fluorescent labelling and a new concept of multiple electrokinetic preconcentrations(7). For the first time, reliable quantification of Aβ 1-42, Aβ 1-40 and Aβ 1-38 down to sub nM in CSF was made possible without recourse to immunoassays or immunoprecipitations. Sensitivity enhancement factors up to 170 with LOQ better than 0.05 nM could be achieved with this antibody free' approach. Excellent agreement between our results and the gold standard ELISA was demonstrated for measurements of Aβ 1-42 in CSF, opening the route to new possibilities in AD diagnosis.

References: (1) Querfurth et al., N Engl J Med 2010, 362, 329-344; (2) Lewczuk P et al. JAD 2015, 43, 183-191 (3) Verpillot et al. J chromatogr. A 2008, 1214, 157-164. (4) Mesbah K.et al. Analyst 2014, 139, 6547-6555. (5) Oukacine et al., Anal Chem. 2014, 86, 3317-3322 (6) Mai TD et al. J Chromatogr A. 2016 Jul 1;1453:116-23. (7) Crosnier de Lassichère C et al. Anal Chem. 2018 ;90(4):2555-2563