Peptide separation by capillary electrophoresis with ultraviolet detection: Some simple approaches to enhance detection sensitivity and resolution [(pemisahan peptida oleh elektroforesis rerambut dengan pengesan ultralembayung: Pendekatan mudah untuk meningkatkan isyarat pengesanan dan resolusi)]

Abstract

Capillary electrophoresis (CE) is one of the leading separation technologies for analysis of water-soluble analytes. CE has many advantages over the more established methods such as liquid chromatography and gel electrophoresis particularly in rapid analysis, require very little sample, use less or no toxic organic solvent, high peak efficiency and ease of automation. Despite the many attractive advantages of CE, CE users continue to seek improvements particularly on detection sensitivity, resolution and selectivity. This paper presented several simple approaches to improve detection sensitivity using simple sample preconcentration called field-enhanced sample injection (FESI) and chromatographic-based ZipTip C 18 pre-concentrator. Also, some improvements in the resolution of complex peptides mixture when using two strategies namely, capillary coating and manipulation of the hydrophobicity of peptides using perfluorinated acids as background electrolyte (BGE), which have anionic conjugate base forms with hydrophobic character. As test compounds, standard peptide mixture and proteins digests were used for these studies. The results showed that FESI has significantly enhanced the detection signal of peptide standards and bovine serum albumin (BSA) tryptic digests. As for the use of ZipTip C 18 pre-concentrator, selective enhancement in detection signal was particularly notable on the late migrating peptides. Coating the capillary proved to have little changes on the CE of peptides when used in conjunction with acidic BGE. Electropherograms of BSA tryptic peptides in pentafluoropropionic acid (PFPA) and heptafluorobutyric acid (HFBA) showed interesting profile, with notable resolution improvement for peptides with close similarity in electrophoretic mobilities.