On-chip Digestion and Identification of Captured Analyte Proteins using MALDI-TOF Mass Spectrometry

Hak-Joon Seok, Young-Ja Kim, Mi-Young Hong, Jung-Hwa Lee[1], Jong-Shin Yoo[1], Hak-Sung Kim*
Department of Biological Sciences, Korea Advanced Institue of Science and Technology
373-1, Kusung-dong, Yusung-ku, Taejon 305-701, Korea
[1]Korea Basic Science Institute (KBSI), Taejon 305-333, Korea
TEL: 82-42-869-2656, FAX: 82-42-869-2610

Abstract

We demonstrated the strategy of selection and identification of analyte proteins of interest by combining a chip-based biosensor with a matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS). For this, the affinity surface of a biosensor chip was constructed over a dendrimer monolayer on gold by employing the interaction between bovine serum albumin (BSA) and its antibody. After a trypsin digestion of captured BSA antigens under an optimized condition, a digested product was collected and analyzed through a peptide mass fingerprinting with MALDI-TOF MS. The detection limit in this approach reached to a low femtomolar level. Comparison of a direct on-chip digestion with an in-solution digestion after an elution of captured proteins reveals that an exposed region of BSA molecule, affinity-captured on an antibody layer, is likely to be trypsin digested. Efficacy of a used chip platform to an on-chip digestion was also tested in other antibody layers. In real applications of treating a protein mixture and reconstituted serum onto the prepared antibody layer, BSA antigen can be detected down to a level of 1 ng/ml. These results represent that combined use of a chip-based technique and MALDI-TOF MS is capable of identifying an analyte of very low concentration in complex biological media.

Reference

1. Dobrin Nedelkov, Randall W. Nelson (2003), Surface plasmon resonance mass spectrometry : recent progress and outlook trends in biotechnology, **21**(7), 301-305.