SP-13

Quantitative Analysis of Surface-Immobilized Streptavidin with Different Orientations: The Critical Role of Conformation

김영필, 홍미영, 김진모*, 손현경*, 문대원*, 김학성, 이태걸*
한국과학기술원, *한국표준과학연구원

We describe a quantitative analysis of differently oriented surface-immobilized streptavidin (SA), for which time-of-flight secondary ion mass spectrometry (TOF-SIMS) and surface plasmon resonance spectroscopy (SPR) techniques were both used. A trehalose-induced conformational stability was sequentially examined for quantitative analysis while introducing immobilized SA both by random configuration (amine-coupling) and by orientedconfiguration (biotin-coupling). Due to the symmetric properties of the SA molecules, the characteristic secondary ion peaks from SA provided little information regarding its orientation on surfaces. However, with a combination of principal component analysis (PCA) onTOF-SIMS data and SPR analysis it was possible to use these characteristic peaks to quantify the surface amounts of SA. Compared to random configuration, the oriented configuration of SA showed a high range of surface density under the same deposit condition, showed a better linear correlation between the TOF-SIMS and SPR data, and was less dependent on the trehalose effect in the quantitative analysis. More importantly, the trehalose-protected SA showed a stronger linear correlation between the TOF-SIMS and SPR data in both random and oriented SA than did the trehalose-unprotected SA. This result indicates that conformational stability is critical when surface-immobilized SA is directly quantified with TOF-SIMS and principal component regression analysis. A TOF-SIMS-based quantitative approach has the potential to allow a direct, label-free SA detection with extreme sensitivity and selectivity.