Claus Czeslik, Theodore L Hazlett, Enrico Gratton, Matthias Ballauff, and Catherine A Royer.
Variable protein binding to polyelectrolyte brushes.
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Abstract

We used two-photon excitation fluorescence fluctuation spectroscopy with photon counting histogram (PCH) analysis as a new tool to study the binding of globular proteins to colloidal particles in situ. The binding of two proteins, SNase and BSA, to spherical polyelectrolyte brushes (SPB) was measured as a function of protein concentration and ionic strength of the solution at pH-values where SNase and BSA are positively and negatively charged, respectively. It has been found that SNase and BSA strongly bind to the SPB. When the ionic strength of the solution is raised to 100 mM, the SPB become resistant to both proteins. These findings provide further evidence for a binding mechanism where the proteins are mainly driven to the SPB by the "counterion evaporation" force, while Coulomb interactions play a minor role. The changes in the secondary and tertiary structure of BSA induced by the interaction with the SPB has also been investigated using static fluorescence and circular dichroism (CD) spectroscopy. In the adsorbed state, the fluorescence spectrum of dansyl-labeled BSA indicates a distortion of the tertiary structure of BSA. Fluorescence and CD spectroscopic analysis of desorbed BSA shows that the adsorption induced conformational changes of BSA are largely reversible. The results of these studies characterize the potential of SPB as a new class of carrier particles for proteins whose use in biotechnological applications appears to be rewarding.