

AFM IMAGING AND FORCE-DISTANCE CURVES ANALYSIS OF SINGLE STRANDED DNA-BINDING (SSB) PROTEIN COMPLEX AND COMPARISON OF KINETIC WITH SURFACE PLASMON RESONANCE AS REFERENCE BIOSENSOR METHOD

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Abstract

Formation of specific non-covalent complexes among nucleic acids and proteins seems to be essential for regulation of cellular processes and biochemical pathways. On the molecular level, the interactions between DNA and proteins are playing crucial role in the control of DNA replication, transcription, translation, and also repair of damaged DNA. Thus, detailed characterization of DNA-protein interactions improves the knowledge about abnormal cells and provides a better understanding of tumor growth , its prevention and medical treatment.

The interaction between ssDNA and the ssDNA-binding protein SSB was analyzed using atomic force microscopy (NanoWizzard3) providing images of the formed complexes on mica. Furthermore, this was complemented by determination of binding forces using atomic force spectroscopy with SSB-modified cantilever tip. The interaction was also characterized using the surface plasmon resonance based real-time bioanalysis (Biacore) providing reference data on kinetics of the interaction. In all cases, the direct label-free approach was conveniently employed and the disturbing labelling was not required.

Keywords: AFM, SPR, DNA-protein complex, SSB protein

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