Protein and Nucleic Acid Analysis with an Electro-Switchable DNA Chip – switchSENSE

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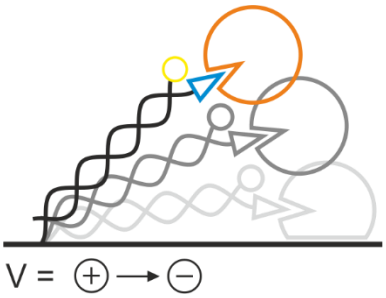

Measurements in stationary or mobile phases are fundamental principles in protein analysis. Although the immobilization of molecules on solid supports allows for the parallel analysis of interactions, properties like size or shape are usually inferred from the molecular mobility under the influence of external forces. However, as these principles are mutually exclusive, a comprehensive characterization of proteins usually involves a tedious multi-step workflow.
Here we show how these measurement modalities can be reconciled by tethering proteins to a surface via dynamically actuated nanolevers.
Short DNA strands, which are switched by alternating electric fields, are employed as capture probes to bind target proteins. By swaying the proteins over nanometer amplitudes and comparing their motional dynamics to a theoretical model, the protein diameter can be quantified with Angstrom accuracy [1], [2]. Alterations in the tertiary protein structure (folding) and conformational changes are readily detected, and even post-translational modifications are revealed by time-resolved molecular dynamics measurements.
Moreover, we demonstrate the analysis of protein interactions with exceptional sensitivity, i.e. the quantification of dissociation constants in the femtomolar concentration regime. Real-time measurements yield association and dissociation rate constants, and we discuss the artefact free characterization of high-affinity interactions like the binding of monoclonal antibodies to antigens. Since the capture probe surface density can be adjusted through an electrical desorption process, the interaction of multivalent analytes with the sensor surface can be evaluated which directly discloses the occurrence of avidity effects [3]. The thermal stability (denaturation) of proteins in the presence or absence of co-factors can be monitored in parallel and melting temperatures are quantified.
Finally, we discuss the possibilities and limitations of utilizing electro-switchable DNA nanolevers in the format of a 24x parallel microelectrode chip for the high-content analysis of molecular interactions.

[1] Nature Communications 4:2099 (2013)
[2] J. Phys. Chem. B. (2014) DOI: 10.1021/jp410640z
[3] J. Am. Chem. Soc. 134, 15225 (2012)