# REAL-TIME DYNAMICS OF SINGL-BIOMOLECULES BY TOTAL INTERNAL REFLECTION FLUORESCENCE MICROSCOPY : BIOCHIP APPLICATION

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The conformational dynamics and adsorption/desorption behavior of several individual bio-molecules such as DNA and protein were simultaneously observed and imaged within the evanescent-field layer using a total internal reflection fluorescence microscopy geometry at the fused-silica/water interface and various surfaces. The individual DNA molecular conformations and adsorption behaviors were found to depend both on pH and on buffer composition. The shape of the individual DNA molecules changed with decreasing pH as follows: (1) fluttering extended; (2) combed; (3) compact super-coiled; and, (4) precipitated. A histogram of individual -DNA molecule velocities (8-27 cm/min) produced by hydrodynamically flowing molecules along the fused silica-water interface exhibited nearly identical asymmetry to the corresponding elution peaks found in capillary liquid chromatography (CLC) and capillary electrophoresis (CE). The velocity was proportional to the capillary surface area-to-volume ratio ($2/r$), which was explained with capacity factor and relative adsorption factor. Hydrophobic interaction rather than electrostatic attraction was the major driving force for adsorption of individual DNA molecules. However, in the case of protein such as R-phycoerthrin, electrostatic attraction was the dominant factor causing adsorption of molecules onto the fused-silica surface. This information is valuable to studies of rate theory and the peak broadening in CE and LC. This technique can be also applied to study biochips at the range of micro and nanometer.