Distance measurement along DNA molecules using fluorescent quantum dots

HELMUT STREY, Stony Brook University — To create and design better micro- and nanofluidic devices, we need to understand how macromolecules behave when squeezed by lateral barriers to create pseudo-two-dimensional confinement. We present experiments in which we visualize DNA molecules of varying sizes (2 kbp - 50 kbp) trapped in 10 micrometer wide slits, the slit height varying from the radius of gyration of the unconfined molecule (micrometer) down to 25 nm (half the persistence length of DNA). We present data on the diffusion coefficient and electrophoretic mobility (no electroosmotic flow) of SYBR-gold labeled DNA molecules as a function of slit height. Simultaneously, we have assessed the DNA conformation by examining molecules that are end-labeled with differently colored fluorescent quantum dots. By determining the distance between labels, we measure directly the end-to-end distance - a conformational measure much discussed but rarely measured. Using the same approach but turning the problem around, we determined if contour length can be estimated from visualization experiments. The answer to this question becomes important when the distance between specific binding sites on the DNA backbone must be measured. One such application, for example, is the determination of haplotypes (genetic variability due to blocks of single nucleotide polymorphisms (SNP)) in diploid individuals.

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