TPM-on-a-chip

Highthroughput analysis of DNA conformational changes by Tethered Particle Motion on single molecule chip

PROGRAMME NANOTECHNOLOGIES ET NANOSYSTEMES
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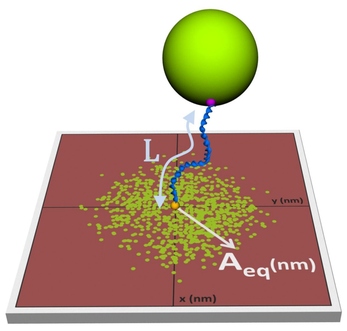
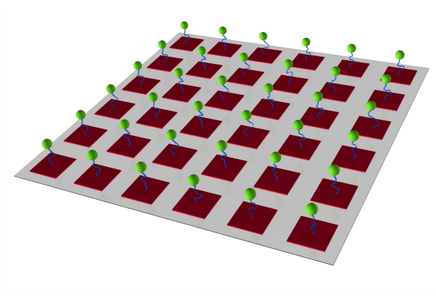
Context

Single-molecule techniques have demonstrated their powerful capabilities to dissect various complex biological systems, such as mechanisms of DNA machineries, DNA-protein interactions, …

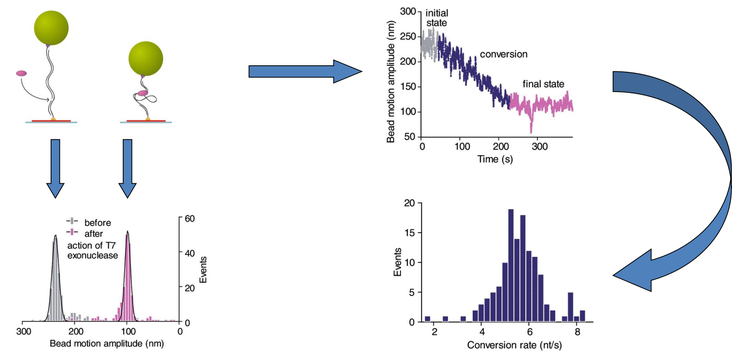
We have recently developed and protected (patent filed) such a biochip integrating the Tethered Particle Motion (TPM) technique. Based on a very simple principle, the TPM technique reveals the conformational dynamics of the DNA molecule.



The motion of the tethered particle (A_{eq}) is related to the effective length of the DNA (L).



The parallelization of the TPM allows high throughput data collection of single DNA molecules.

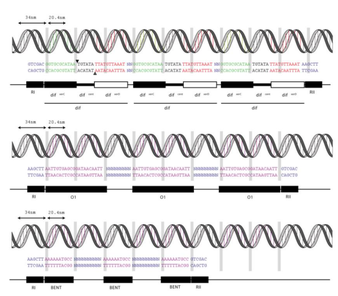


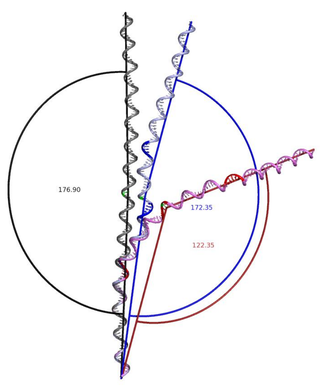
Our high throughput single molecule observations shed new light on the T7 exonuclease behaviour that had only been examined in bulk assays previously and, more specifically, on its processivity.

Objectives

- To explore the capabilities of the biochip to analyze a variety of biological and biophysical fundamental processes or effects of variations in the environmental conditions.
- To demonstrate the informative potential of the biochip and promote the oncoming commercial system.

1/ by screening extensively the performances of the biochip output through the analysis of a panel of synthetic model DNAs with distinct conformations induced by structural defects (bends, nicks).





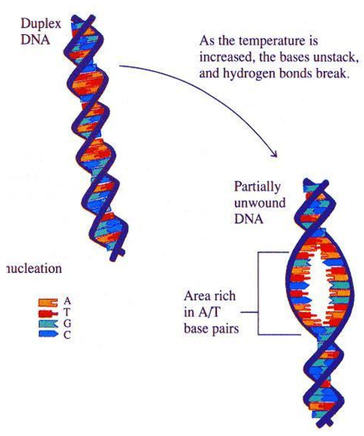
DNA fragments have been synthetized, and their analysis in under progress.

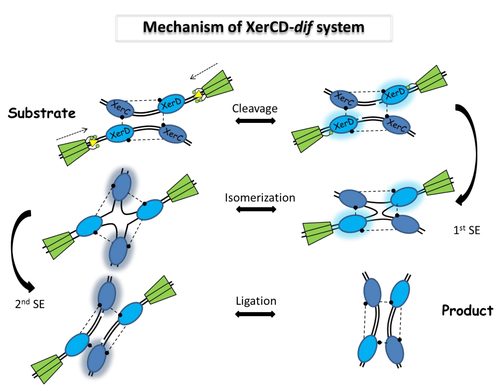
2/ by investigating specific biophysical and biological questions:

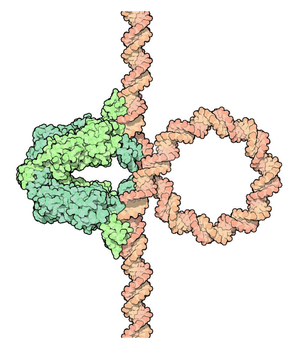
formation and life-time of denaturation bubbles

role of DNA bending in XerCD-dif recombination

DNA looping in the transcription regulation by Lac repressor







Scientific productions

The invention is protected by patent n° FR 1057031, filed on September 3, 2010, entitled: **“Biopuces pour l’analyse de la dynamique de molécules d’acide nucléique” ;** PCT/FR2011/052004 filed on September 1, 2011.

Software for images acquisition and analysis, entitled **“Nanomultiplex”,** is registered. N° APP : IDDN.FR.001.430005.000.S.P.2011.000.31235, on October 25, 2011, developed with the Magellium company.

Plénat & al., *High-throughput single-molecule analysis of DNA–protein interactions by tethered particle motion*, **Nucleic Acids Research**, 2012, 40 (12).

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