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The dynamic properties of an intramolecular transition from DNA duplex to cytosine–thymine motif triplex

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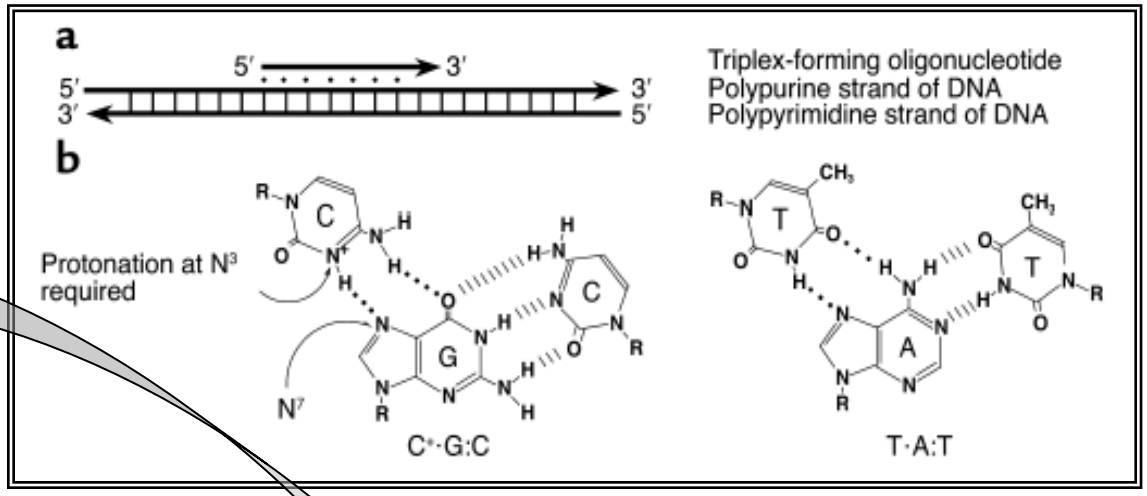
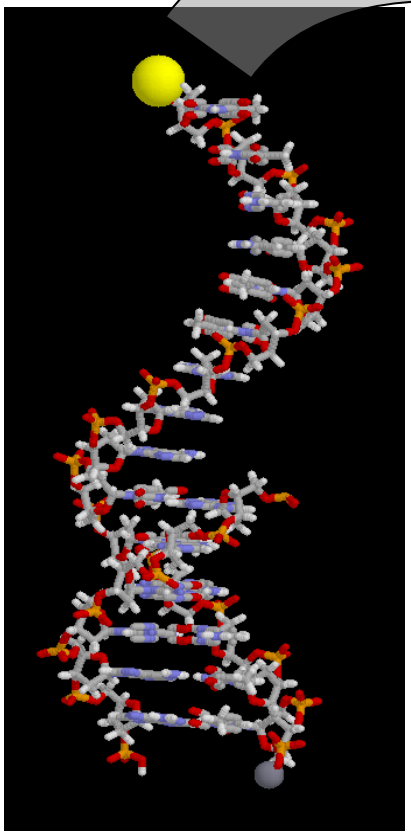
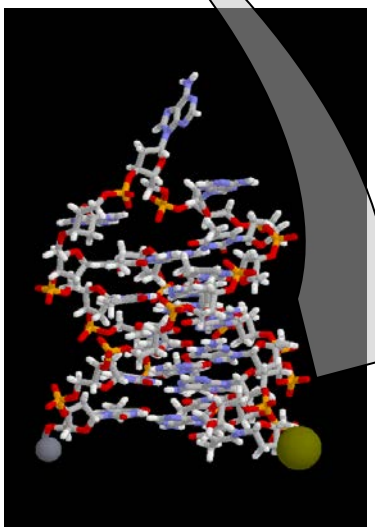
Progettazione di un nanomotore basato sulla triplex







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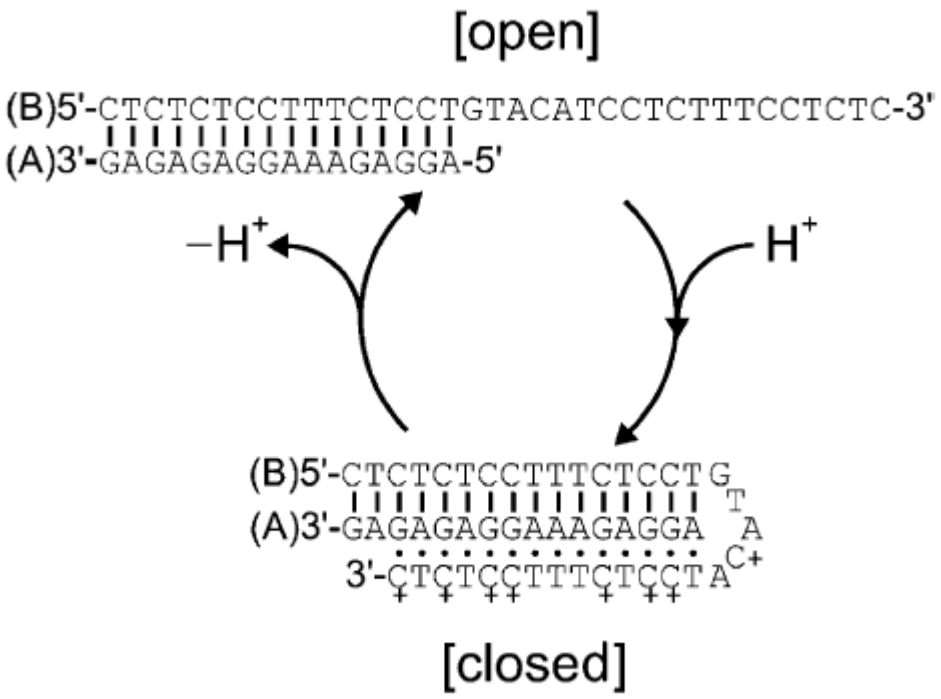
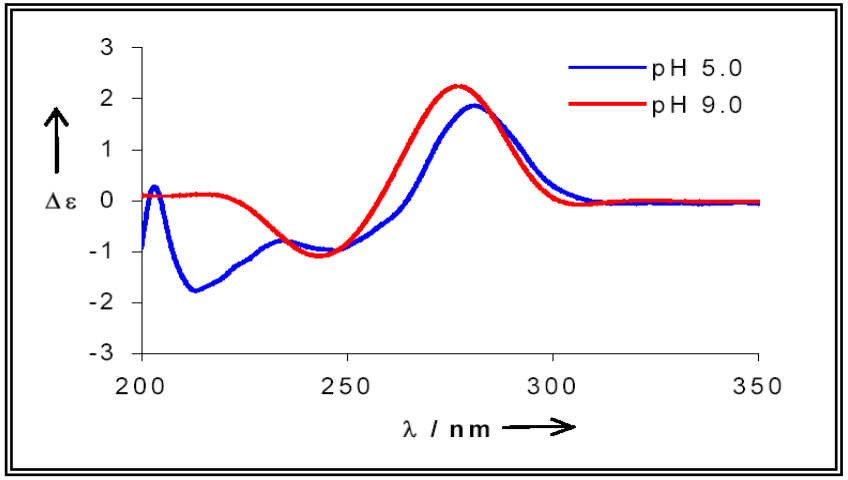
Design and definition of base sequence

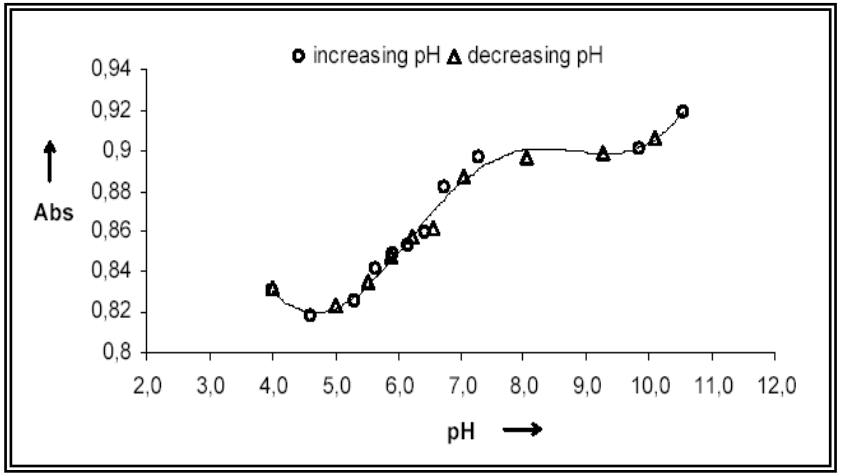


Scheme 1 Oligonucleotide sequences and conformations.

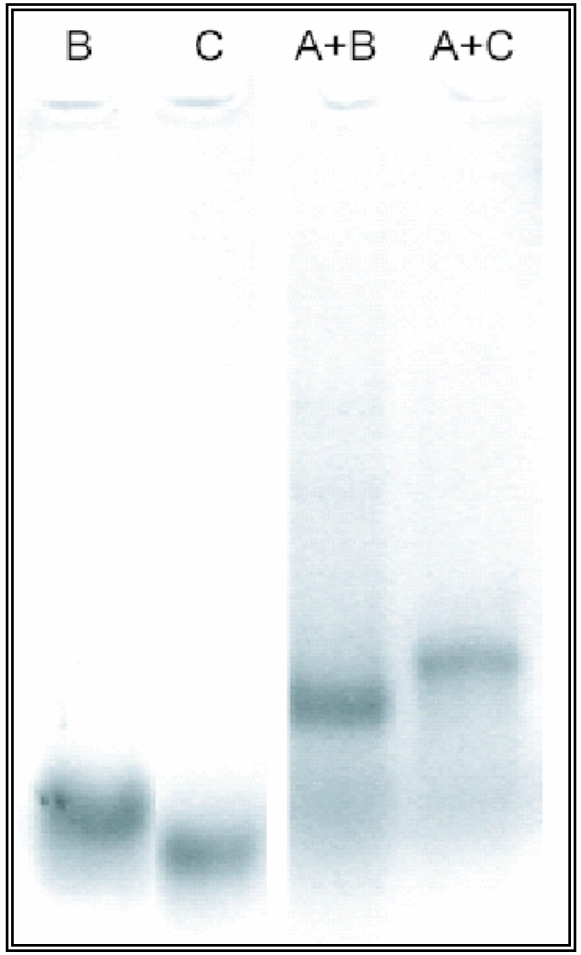
Caratterizzazioni statiche: una macchina a 2 stati



Spettro CD del costrutto a differenti pH.

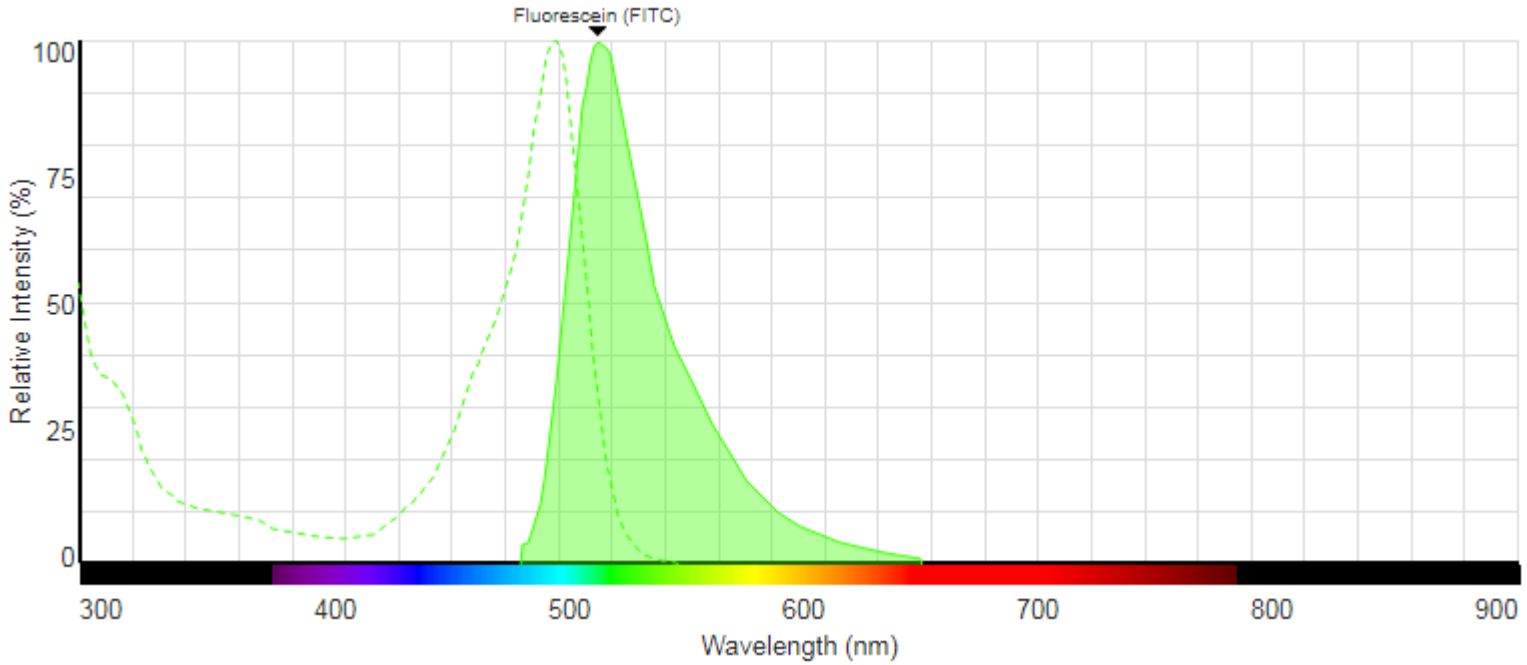


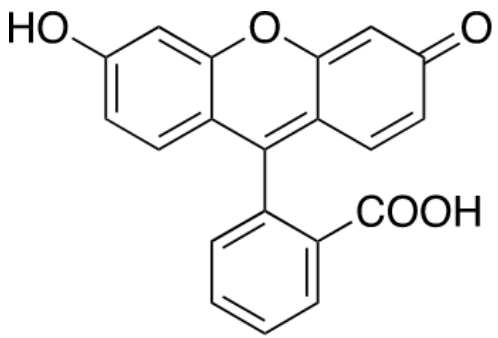
Assorbanza a 260 nm a differenti pH.



Mobilità elettroforetica del costrutto con TFO e di un analogo senza TFO (oligo C non può ripiegare)

Uno spettro di fluorescenza per un comune fluoroforo organico



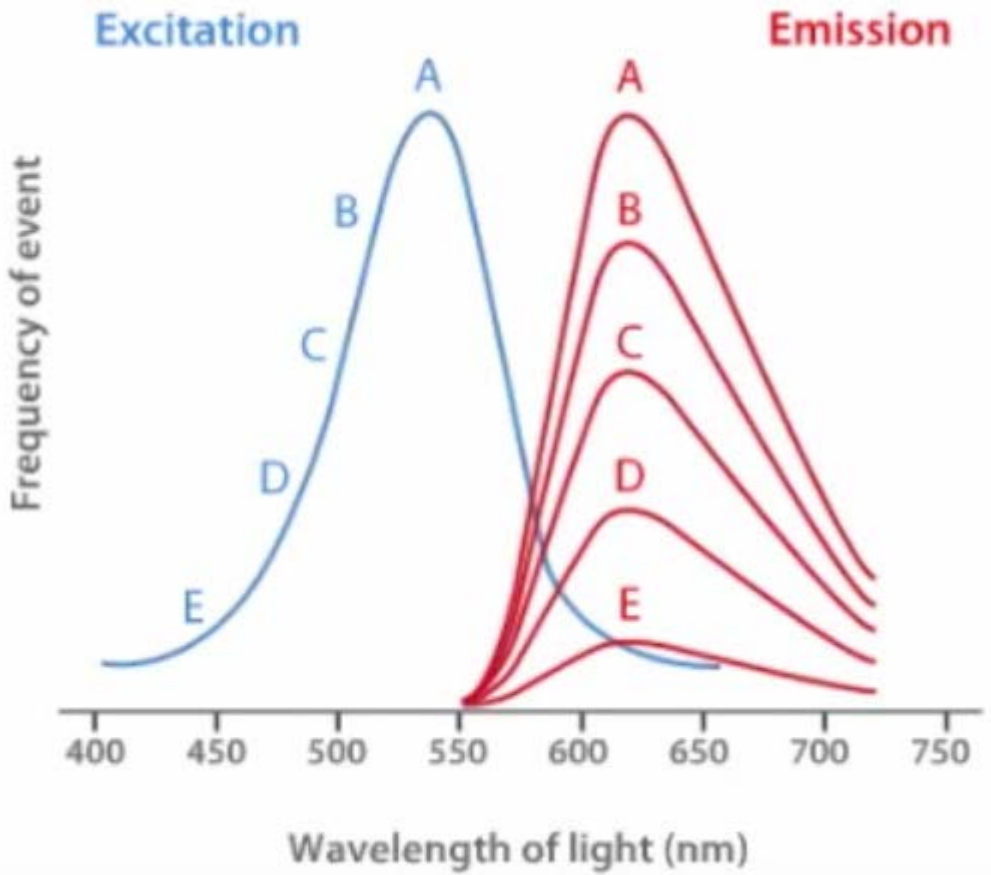


Instruments types for reading fluorescence

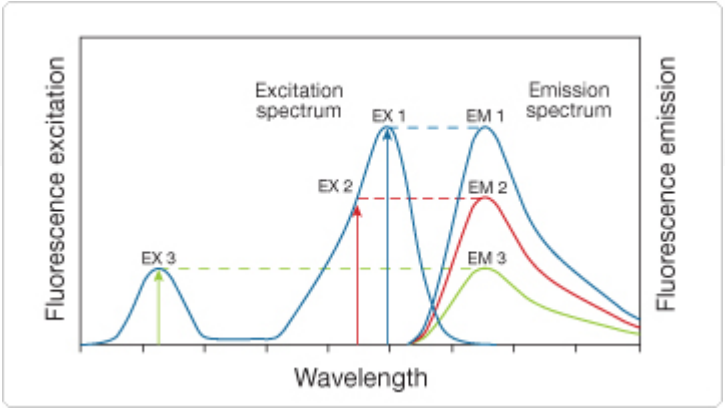
•Spectrofluorometers and microplate readers measure the *average* properties of bulk (µL to mL) samples.
•Fluorescence microscopes resolve fluorescence as a function of spatial coordinates in two or three dimensions for microscopic objects (less than ~0.1 mm diameter).
•Fluorescence scanners, including microarray readers, resolve fluorescence as a function of spatial coordinates in two dimensions for macroscopic objects such as electrophoresis gels, blots and chromatograms.
•Flow cytometers measure fluorescence per cell in a flowing stream, allowing subpopulations within a large sample to be identified and quantitated

Other types of instrumentation that use fluorescence detection include capillary electrophoresis apparatus, DNA sequencers and microfluidic devices. Each type of instrument produces different measurement artifacts and makes different demands on the fluorescent probe. For example, although photobleaching is often a significant problem in fluorescence microscopy, it is not a major impediment in flow cytometry because the dwell time of individual cells in the excitation beam is short.

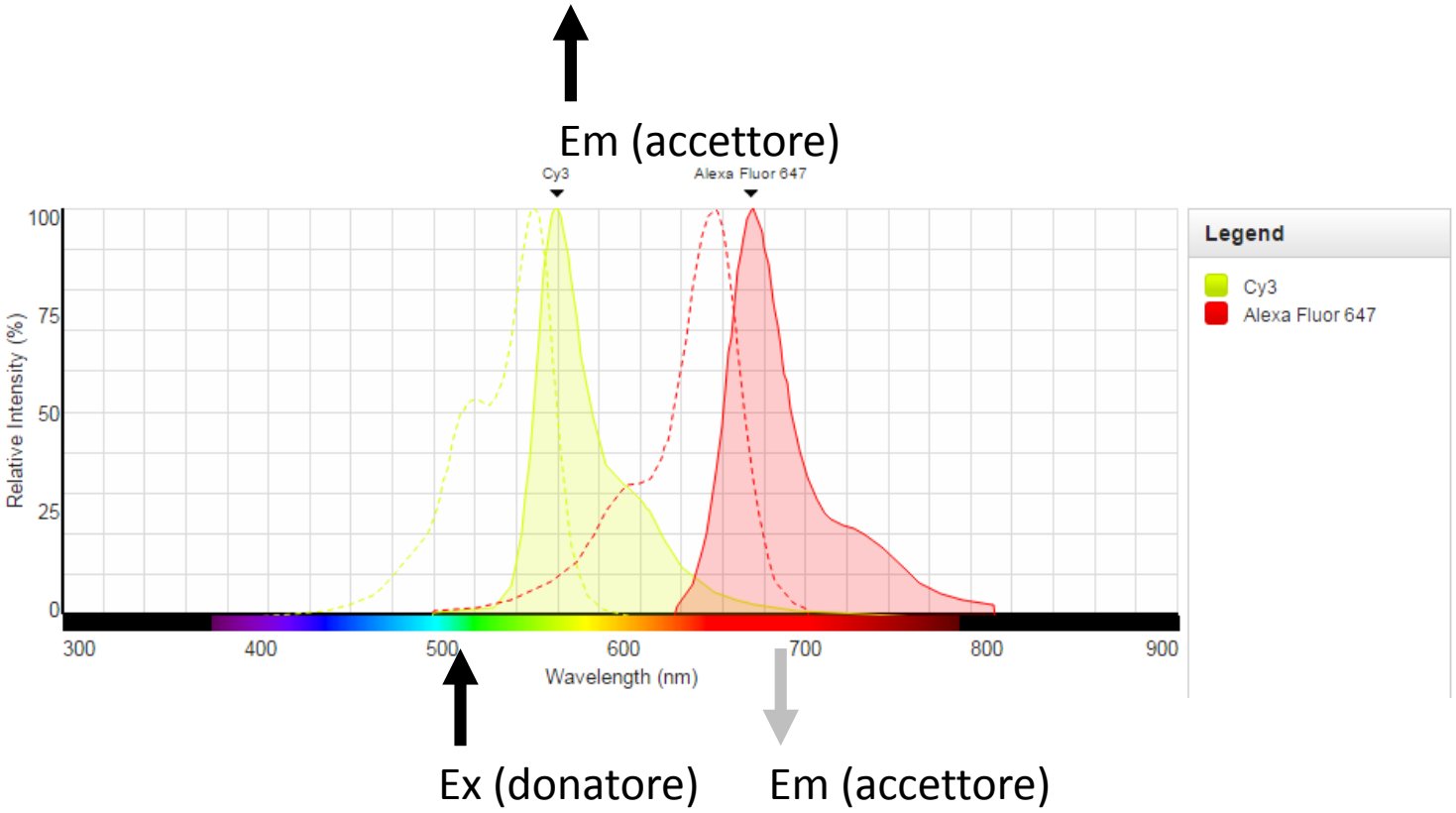
Dove eccitare la fluorescenza di un fluoroforo?



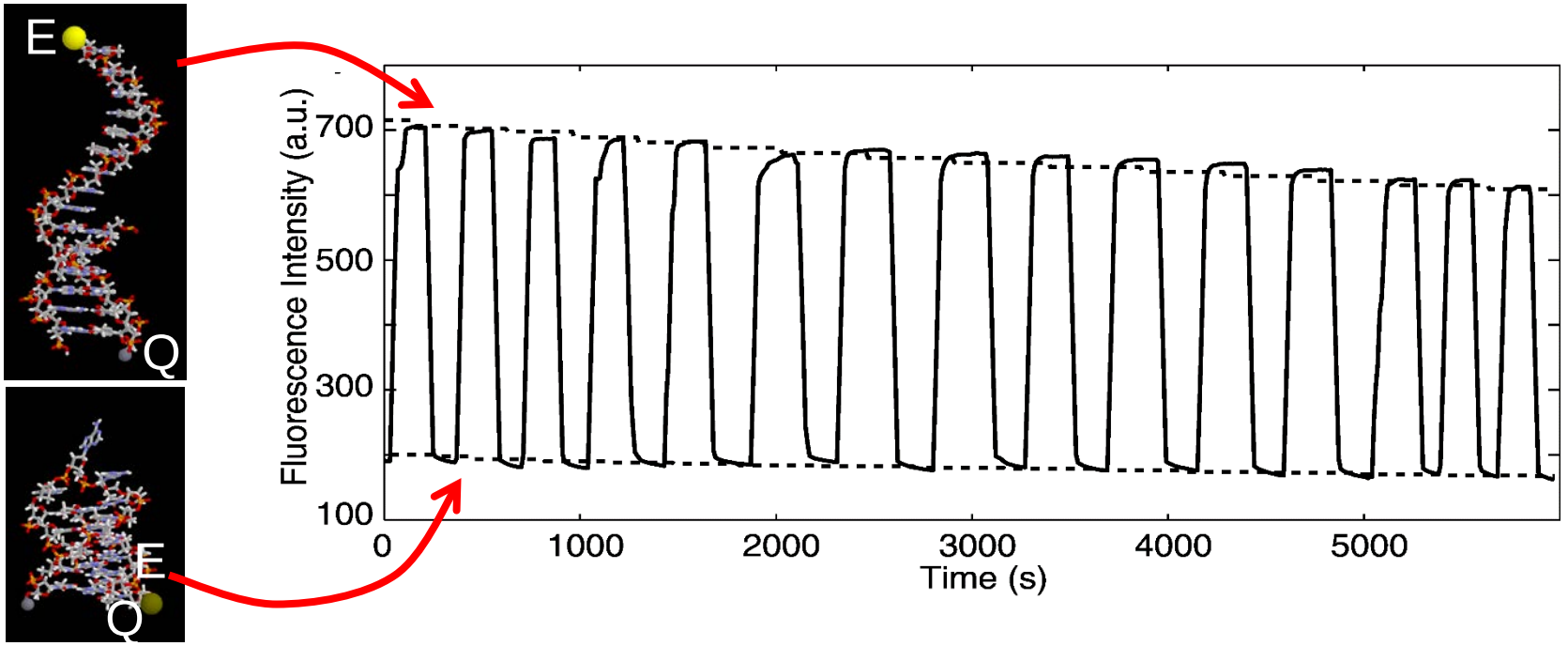
Illumination at lower or higher wavelengths affects only the intensity of the emitted light



FRET: fluorescence resonance energy transfer



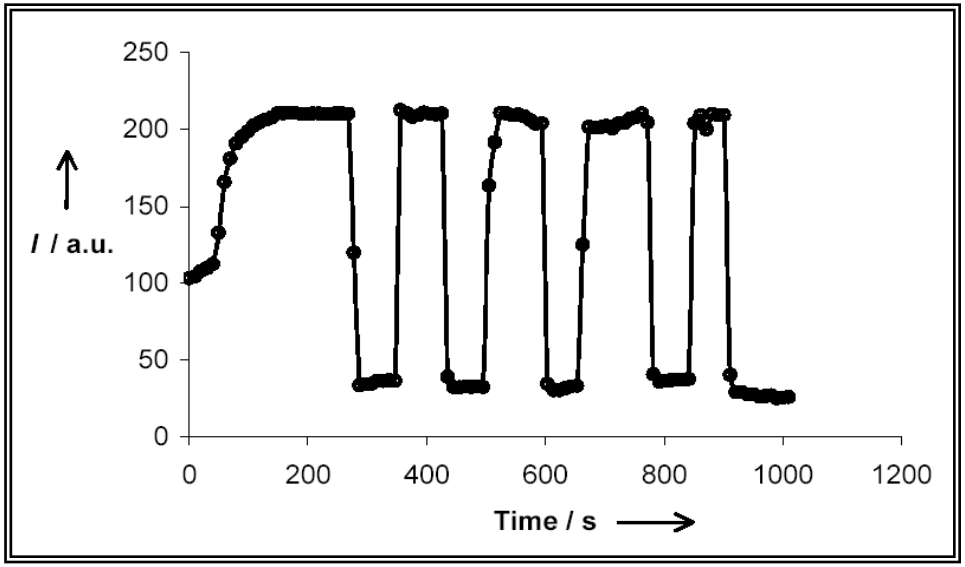
Caratterizzazioni dinamiche / 1



Emissione di fluorescenza di A+B* con pH alternante tra 5 e 9

- Intensità dipendente esclusivamente dalla separazione della coppia E-Q
- Diminuzione dell'intensità = lineare (pendenza = diluizione) → nessuna diminuzione di rendimento dovuto all'accumulo di rifiuto

Caratterizzazioni dinamiche / 2



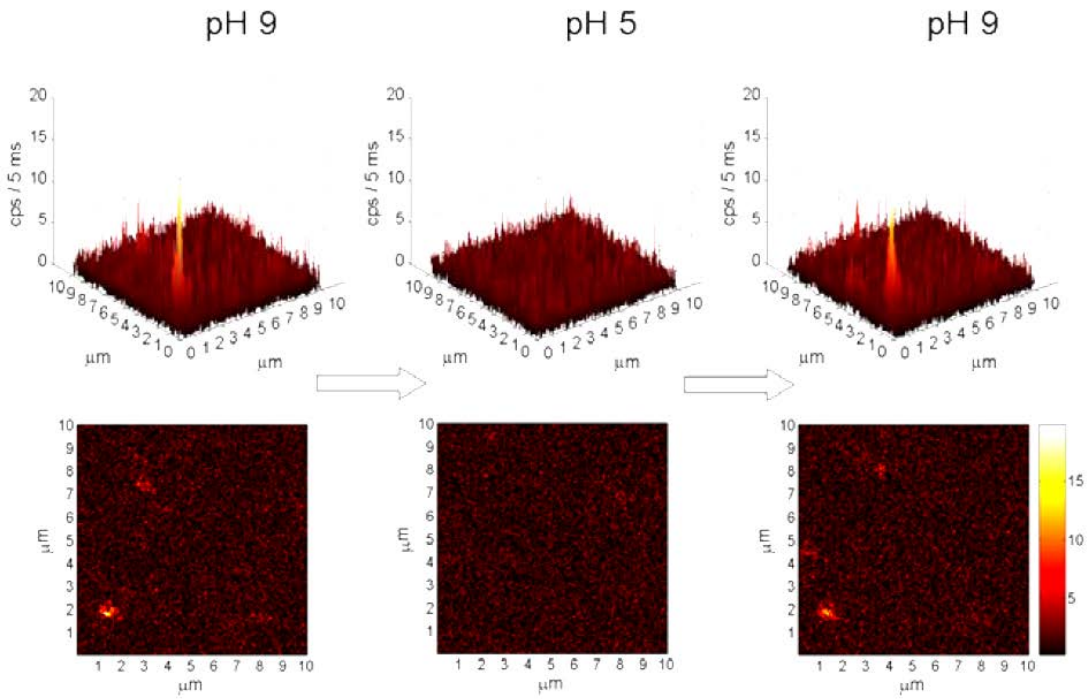
Ripetizione dell’esperimento ad alta diluizione

- Si evidenzia il medesimo comportamento.

Cycling at the single-molecule level

We demonstrated, at the single-molecule level, that the device functions also when confined on a surface.

It was one of the first times that the functioning of a DNA nanodevice was proved at the single-molecule level.



Such devices can decorate larger self-assembled nanostructures and move nano-objects in space

[B. Kolaric et al., Photochem. Photobiol. Sci. 2007]