Detection of short nucleotides with DNA nanopores

Juan Elezgaray[1],
Luyan Yang[1], Chirstophe Cullin[2]

[1] CRPP, CNRS, UMR5130, Av. Schweitzer, Pessac, France
[2]CBMN, CNRS, U. Bordeaux, Allée St Hiliare, Pessac, France

juan.elezgaray@u-bordeaux.fr

MiroRNAs (miRNA) are single strand, non-coding RNAs that play a role in the regulation of the genetic expression, through their capacity to hybridize with the 3'UTR of specific target mRNA (messenger RNA). miRNAs are known to be associated with the normal development and function of the organism, but are also involved in diseases such as cancer [1]. The attractiveness of extracellular miRNA as cancer biomarkers relies on their stability and their dysregulation in the diseased cells. However, because of their short sequence and low concentration, miRNA detection is intrinsically difficult. This paper shows how DNA nanotechnology [2,3] provides with an efficient way to detect single miRNAs in a single step process, where no amplification steps (as PCR based methods) are required. This method is based on DNA nanopores, nanometric cylindrical structures (Figure 1) that are able to perforate lipid bilayers. The geometry (width [4], length [5]) of DNA nanopores can be regulated by the interaction with short nucleotides such as miRNAs. Detection of nanopores relies on electric measurements of membrane conductance inspired from electrophysiology. We show that the presence of single nanopores with well defined conductance can be detected. Because of the close relation between geometry and conductance, this is equivalent to the detection of single miRNAs. We also describe how the same approach can be extended by the use of aptamers to the detection of small organic molecules.

References

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Figures

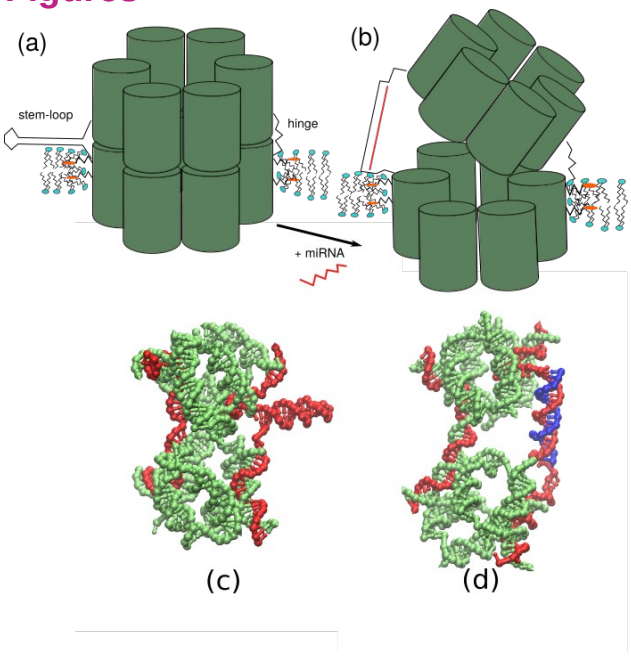

Figure 1. (a) and (b): schematic representation of the opening mechanism. Each cylinder represents a double helix. DNA nanopore is inserted into a lipid bilayer thanks to cholesterol modifications (orange ellipses). (a) Closed state: the stem loop imposes a short distance between two of the helices. (b) Open state: upon addition of miRNA, the stem loop unfolds giving rise to a mixed single and double stranded linker which pushes the two halves apart. (c) and (d) are oxDNA [6] simulations of closed and open conformations, respectively. Strands that form the hinge or the stem-loop locking mechanism are in red. The input signal is in blue.