Gating-membranes based on functional DNA sandwich structures

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Introduction

DNA molecules consist of one or more biopolymer strands which can be as short as some nanometers or as long as a couple of meters. Aptamers are functional DNA capable of specifically interacting with a molecular target. The use of DNA-aptamers in stimulus-responsive membranes provides a degree of freedom that is not achieved with bulk polymer membranes because a modular system is obtained which can be easily adapted to different targets by simply exchanging the recognizing element (DNA-aptamer) but without changing the overall physico-chemical properties of the system, as frequently occurs in bulk polymer membranes.

It has been reported previously that single DNA-aptamers can be used as so-called "gate keepers" in nanoporous membranes where a molecular recognition event results in a conformational change that in turn modulates membrane permeability. The gating function of the aptamer was shown to be specific, concentration dependent, and reversible [1]. However, such systems depended on a fine-tuning between pore size and DNA-aptamer dimensions in order to be efficient.

To overcome this limitation, we studied the possibility of creating a DNA-aptamer sandwich structure (Figure 1) which is scalable and to a certain extent independent of the pore size such that the flexibility of the system is significantly increased. In absence of a molecular target, the sandwich structure is maintained; upon exposure to the target, the structure disintegrates as the recognition of the target is energetically more favourable than the hybridization between individual DNA segments.

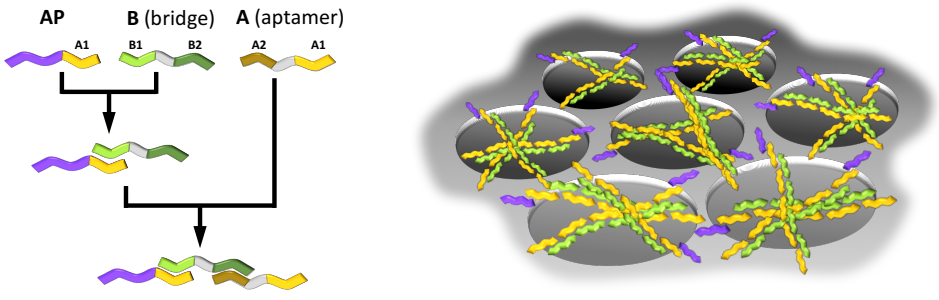


Figure 1. *Left*: Scheme of the systematic build-up of the DNA-aptamer sandwich structure: AP denotes the anchor probe attached to the surface, followed by a bridge segment which then links to an aptamer, and so on. *Right:* Illustration of the stimuli-responsive DNA-aptamer sandwich closing a porous membrane structure.

Material and Methods

The build-up of the DNA-aptamer sandwich was monitored in real-time by quartz crystal microbalance with dissipation monitoring (QCM-D) and multi-parameter surface plasmon resonance (MP-SPR). The DNA-aptamer was specifically recognizing adenosine monophosphate (AMP). The permeability of the DNA-sandwich modified AAO membranes was measured spectrophotometrically using a side-by-side diffusion cell and erioglaucine disodium salt as a tracer dye.

Results and Discussion

QCM-D and MP-SPR revealed that the systematic and controlled build-up of the DNA-sandwich resulted in using less DNA material and a far lower polydispersity than a one-pot self-assembly proposed in previous studies [2]. The sandwich build-up was highly reproducible and DNA-sandwich lengths of up to 30 nm were obtained. Addition of the target AMP resulted in the disintegration of the whole structure down to the anchor probe covalently attached to the surface (Figure 2, left). However, it was also found that this disintegration was heavily dependent on the liquid composition and the function of the DNA-sandwich could not be directly inferred from single DNA kinetics.

Membranes that were systematically modified with a DNA-aptamer sandwich indeed maintained an effective pore closing and opening function (Figure 2, right). This corroborated the hypothesis of using DNA-aptamers as functional units for constructing a matrix which responds to a molecular stimulus.

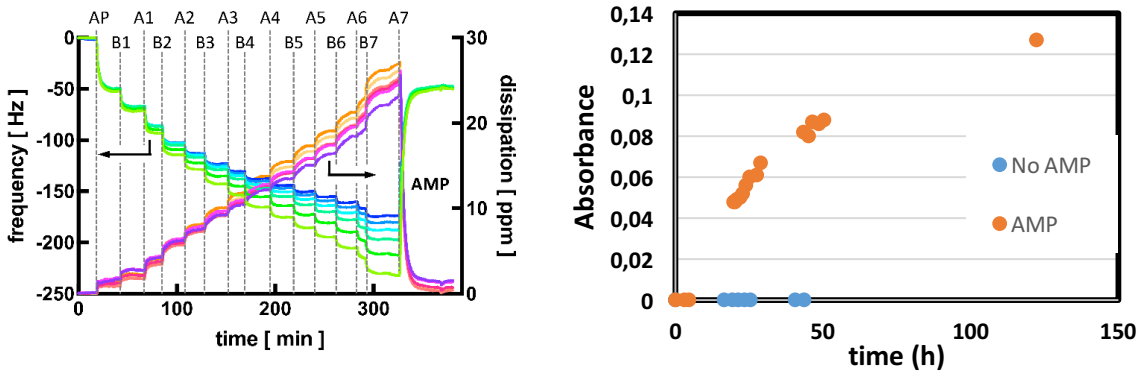


Figure 2. *Left*: Systematic build-up of the DNA-aptamer sandwich structure in QCM-D and total disintegration upon exposure to the target (AMP). First, the anchor probe (AP) was attached, followed by alternating seven times bridge (B) and aptamer (A) probes. *Right:* Permeation experiment of the DNA-sandwich gating membrane. The tracer dye was blocked in absence of the target AMP. Upon recognition of AMP, the sandwich structure (in this case consisting of 10 segments) opened the pore.

References

(1) Schäfer T., Özalp VC., *Chem Commun*. 25 (2015) 5471-5474.

(2) Olave, B., Rafaniello, I., Schäfer, T., (2018) *in preparation*