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BIOSENSORS FOR PANDEMICS

Electrical Detection of Amyloid ß Aggregates Using Nanoporous Membrane Integrated Microfluidic Organic Electrochemical Transistor

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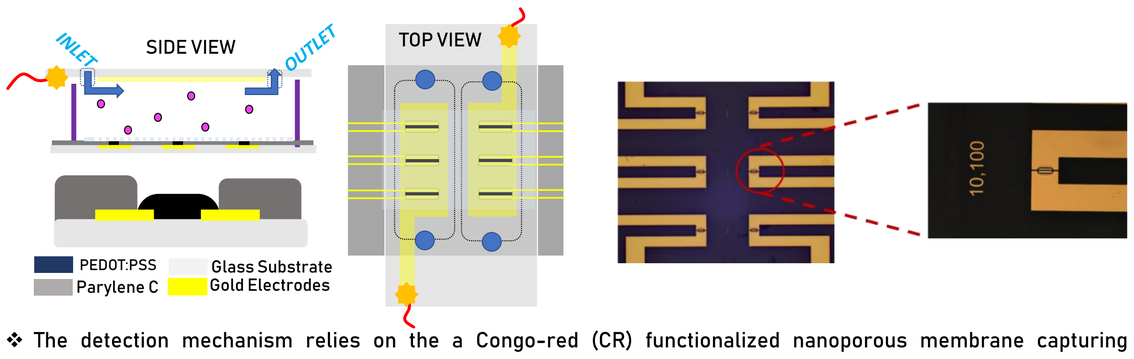
Objectives

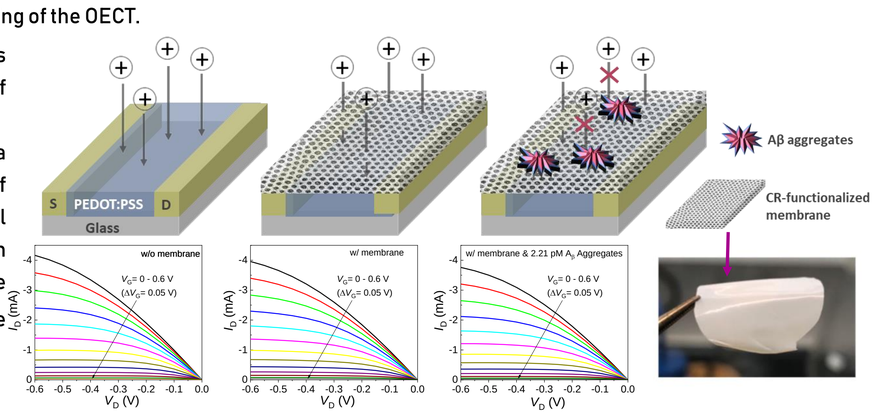
- Alzheimer's disease (AD) is a progressive neurodegenerative disease associated with severe memory loss and impaired cognitive skills. A common pathological change found in AD-affected brains is the accumulation of a peptide named amyloid-β (Aβ) that can form plaques.
- In this work, we design an organic electrochemical transistor (OECT) based chip for in vitro detection of Aβ aggregates in physiologically relevant media. This method incorporates the structural advantage of nanoporous membrane functionalized with receptors and the confined detection unit owing to the microfluidic integration.

Working Principle of Biosensor

- The schematic of nanoporous membrane integrated microfluidic OECT and organic channels microscopy images.



- The detection mechanism relies on the a Congo-red (CR) functionalized nanoporous membrane capturing Aβ aggregates larger than the size of its pores and thus blocking the penetration of electrolyte ions into the channel underneath, suppressing the gating of the OECT.
- The OECT signal thus varies depending on the concentration of Aβ aggregates in the solution.
- Consequently, we measure a decrease in the total number of cations that can enter the channel which results in changing drain current (I_D), , transconductance (g_M), and slower response time (τ).



Electrical Characterization

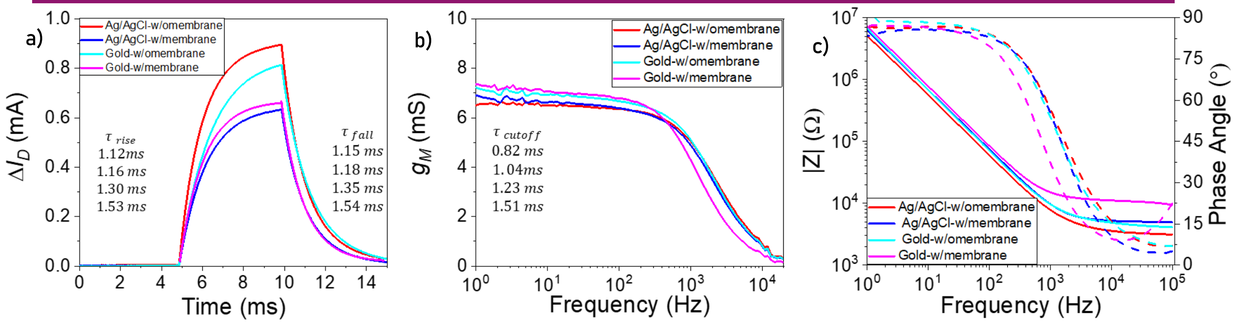
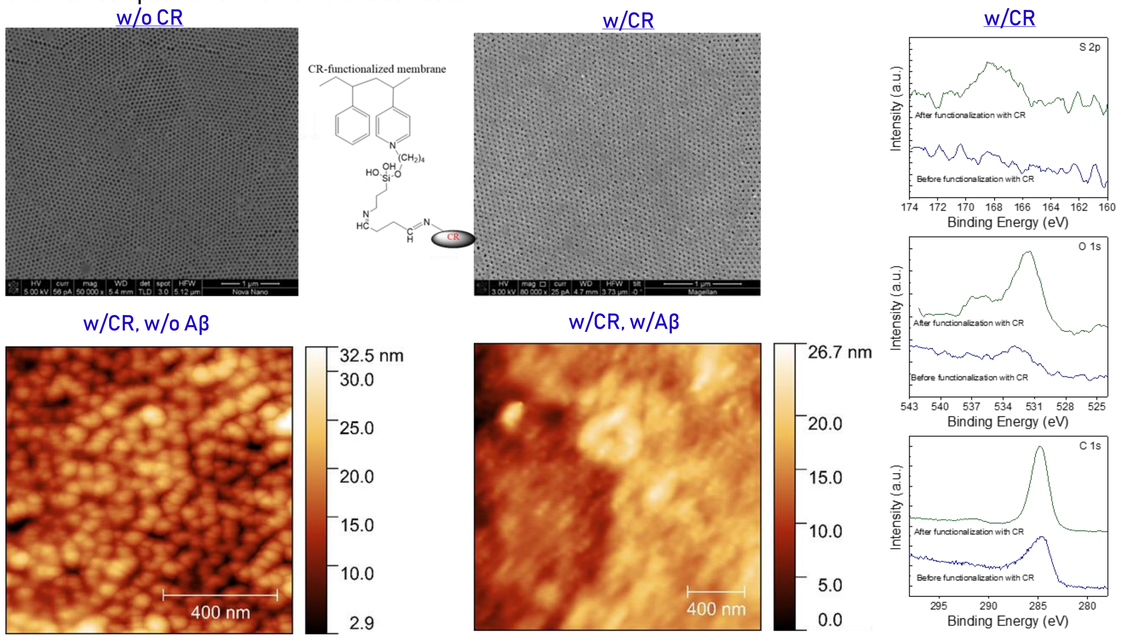


Figure. a) Time response, b) bandwidth, and c) impedance characteristics of nanoporous membrane integrated microfluidic OECTs.

- The response times which gives the time for how fast the channel will be de-doped (doped) by injected (extracted) cations were measured. The presence of the membrane on top of the channel decreases the current and increases response time slightly.
- The impedance of the system was also increased after the integration of CR conjugated nanoporous membrane on to the OECT.

Surface Characterization

- The nanoporous membrane has a pore size smaller than 50 nm and a surface functionalized with CR, i.e., a ligand with a strong affinity to a cross-β structure of Aβ aggregates
- To verify the presence of CR on the membrane surface and analyze the surface after each modification step, we carried out SEM, and XPS measurements. The pore size slightly decreased after CR functionalization.The high-resolution of S2p, O1s and C1s spectra of the surface after APTES modification and upon CR immobilization reveal significant differences in the chemical composition of the membrane surfaces.

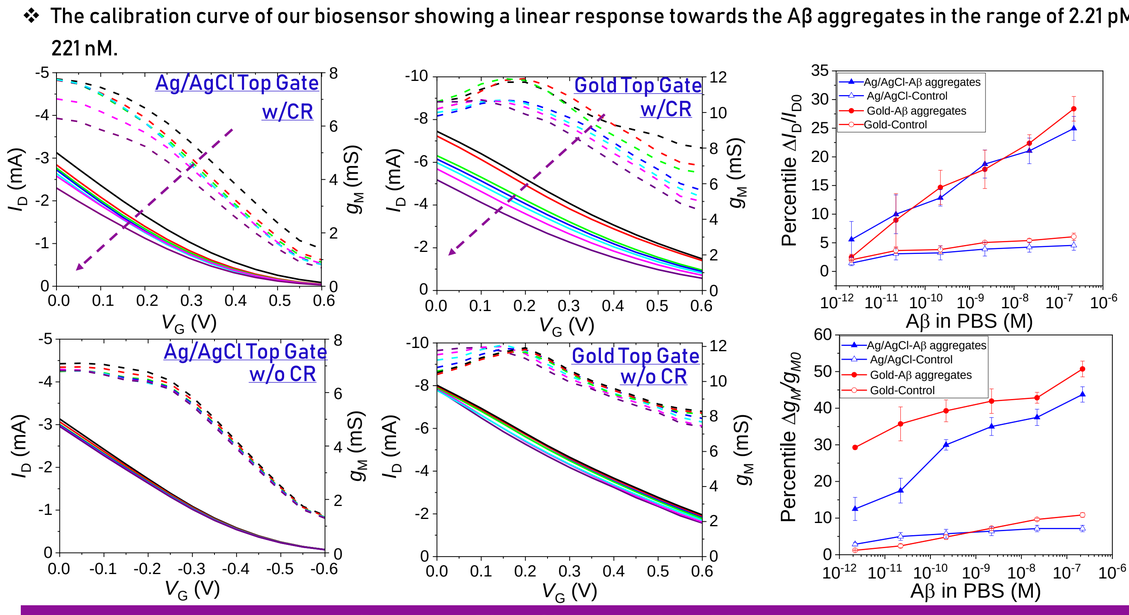


- The changes in the AFM images of the membrane upon interactions with Aβ evidence that CR units capture the protein aggregates which then adsorb on the membrane surface

Results

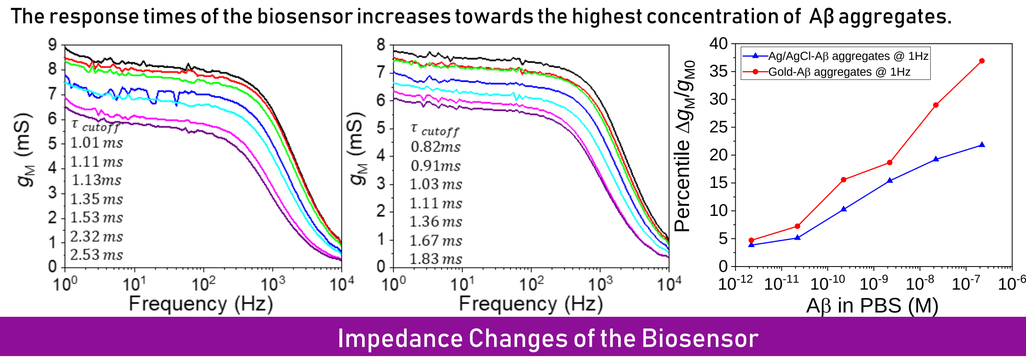
Sensitivity of the Biosensor

- The transfer curve of the CR functionalized membrane integrated microfluidic OECT shows that a continuous decrease in the drain current and its transconductance with an increase in the Aβ aggregates concentration.
- The CR-free, bare membrane, on the other hand, has no specific interactions with Aβ aggregates, leading to a device response independent of protein concentration
- The calibration curve of our biosensor showing a linear response towards the Aβ aggregates in the range of 2.21 pM–221 nM.



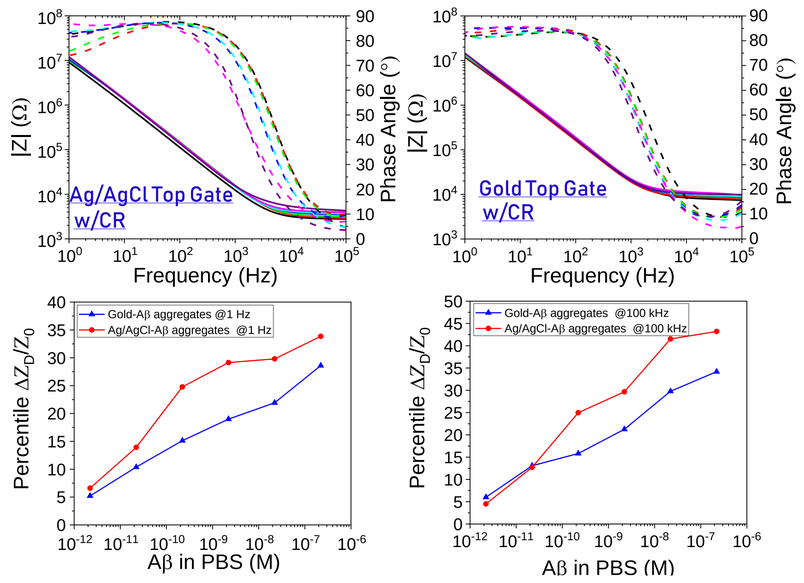
Response Time Changes of the Biosensor

- The response times of the biosensor increases towards the highest concentration of Aβ aggregates.



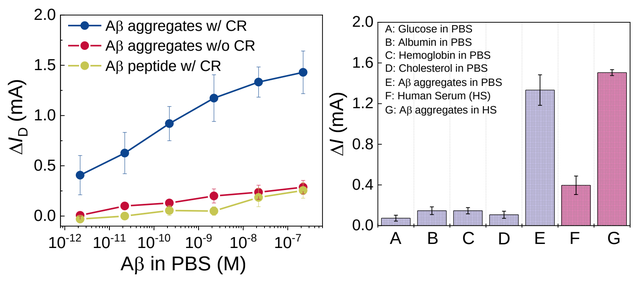
Impedance Changes of the Biosensor

- We further performed electrochemical impedance spectroscopy measurements.
- The results show that after incubation with Aβ aggregates, the impedance increase drastically, indicating that cations transport towards the channel is hindered.
- In addition, the channel capacitance decreases accompanied with an increase in charge transfer and electrolyte resistance due to a drop in the ion diffusion ability.
- The sensitivity of the microfluidic chip increased at high frequencies at which the effect of Aβ aggregates is more pronounced.



Selectivity and Specificity of the Biosensor

- To rule out the possibility of the CR-functionalized membrane also interacting with the Aβ monomers, we monitored the device response to a broad range of peptide concentrations.
- The negligible change in channel conductance with the peptide confirms the selectivity of the sensor to the aggregate form of the protein.
- The device is also not responsive to molecules which can clog the membrane pores because of their size, further evidencing that the specific interactions of Aβ aggregates with CR is essential for their detection.



Conclusions

- The microfluidic OECT integrated with a Congo-red functionalized nanoporous membrane shows a strong affinity for Aβ aggregates.
- Combining the high transconductance of the OECT with the precise porosity and selectivity of the membrane, the device detects the presence of Aβ aggregates in physiologically relevant media with an excellent sensitivity
- This robust, low-power, non-invasive, and miniaturized sensor aids in the development of point-of care tools for early diagnosis of AD
- Sensor performance will be tested with recombinant SARS-CoV-2 RBS (receptor binding domain of the spike protein) fusion proteins as described in the literature.

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REFERENCES

1) Wustoni, S., Wang, S., Alvarez, J. R., Hidalgo, T. C., Nunes, S. P., & Inal, S. (2019). An organic electrochemical transistor integrated with a molecularly selective isoporous membrane for amyloid-β detection. Biosensors and Bioelectronics, 143, 111561.

2) Madhavan, P., Hong, P. Y., Sougrat, R., & Nunes, S. P. (2014). Silver-enhanced block copolymer membranes with biocidal activity. *ACS applied materials & interfaces*, *6*(21), 18497-18501.

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