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A highly sensitive biosensor for the detection of circulating Alzheimer's Disease biomarker pTau

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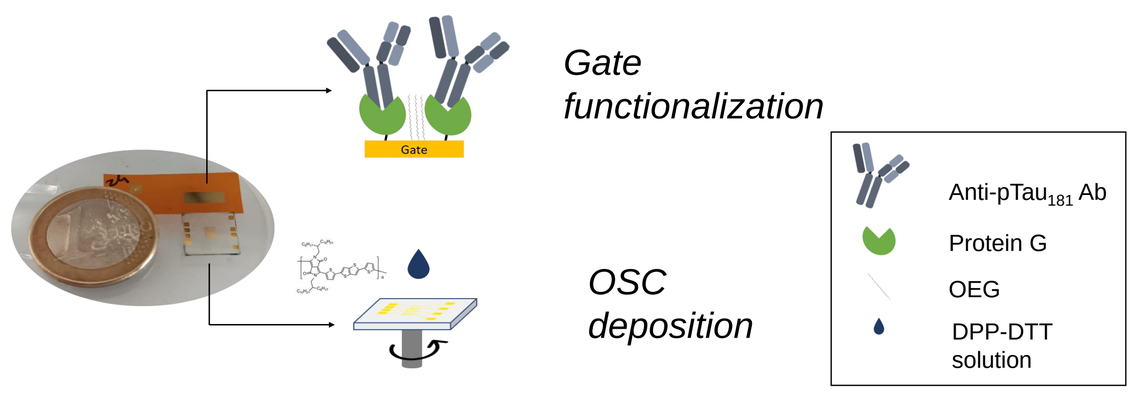
1. INTRODUCTION

Alzheimer's disease (AD) is a progressive neurodegenerative disorder that primarily affects cognitive function. Neuroimaging is currently the most accurate biomarker for AD prediction, but new biomarkers are needed for better drug development, personalized medicine, and pre-clinical diagnosis. Phosphorylated Tau protein (pTau) is a promising candidate for early diagnosis, with a pathological concentration level of 67.87±18.05 pg/mL.[1]

The technology we used as an alternative for the detection of p-Tau is called Electrolyte-Gated Organic Transistors (EGOTs), which have the ability to amplify small biological signals in biological samples, bridging the gap between the biological and technological worlds.[2]

2. EXPERIMENTAL SECTION

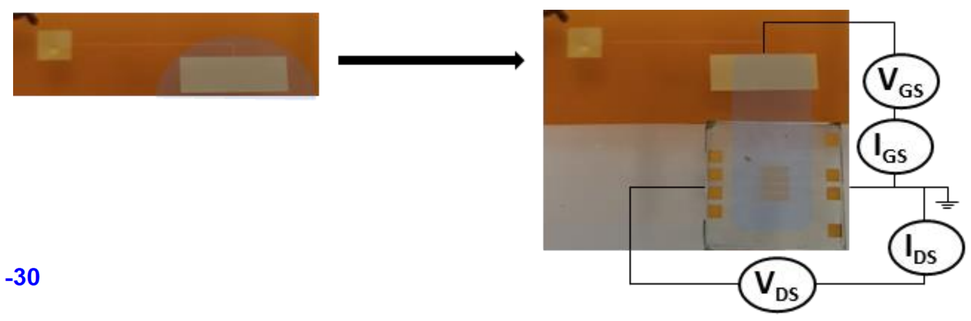
Device fabrication



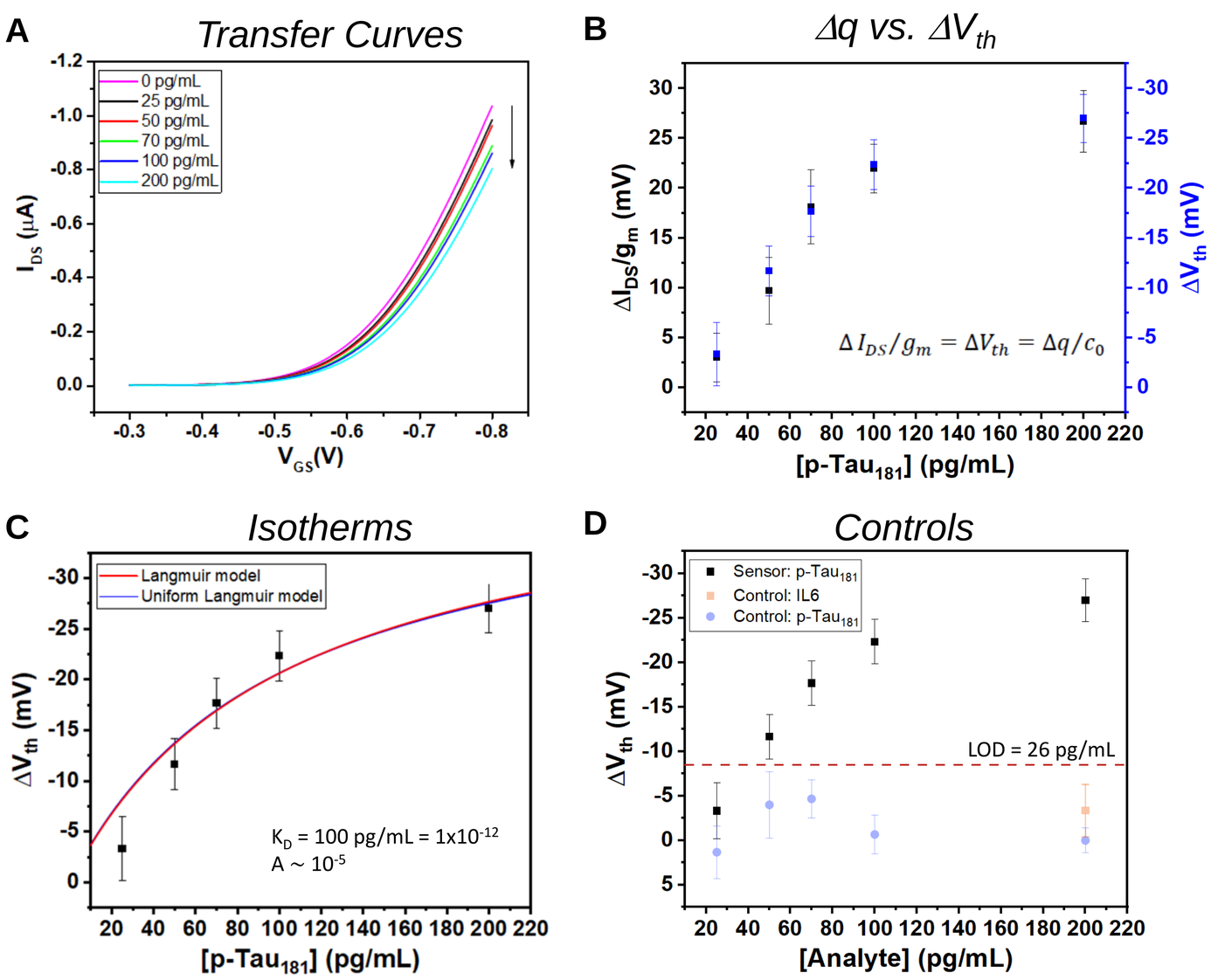
Electrical sensing

ex-situ incubation in pTau$_{181}$ in aCSF

electrical measurements in 50 mM phosphate buffer



3. RESULTS



A) Transfer characteristics of DPP-DTT EGOT-based biosensor monitoring the changes in drain current upon analyte binding at the functionalized gate. Transfer curves were recorded in 50 mM Phosphate Buffer solution, pH 7.4, at a fixed V_{DS} of -0.2 V. **B)** Black squares: dose curve in terms of $\Delta I_{DS}/g_{m[0]}$ *vs.* [p-Tau$_{181}$], change in current and g_m were calculated at V_{GS} = -0.63 V and V_{DS} = -0.2 V. Blue squares: shift in V_{th} *vs.* [p-Tau$_{181}$]. **C)** Dose curves in terms of ΔV_{th} *vs.* [p-Tau$_{181}$]. The classical Langmuir fitting is represented as a red line, while the uniform Langmuir fitting is represented by a blue line. **D)** Comparison of the dose curve (black squares, ΔV_{th} *vs.* [p-Tau]) compared with the sensor response to the control experiments. IL-6 control (pink square) is the developed p-Tau sensor exposed to a high (200 pg/mL) concentration of IL-6, and p-Tau$_{181}$ (blue square) is an EGOT-based biosensor with a different antibody not specific towards p-Tau$_{181}$, exposed to increasing concentration of p-Tau$_{181}$. The dashed line represents the LOD of the biosensor. Error bars represent SEM.

4. CONCLUSIONS

- The comparison between the drain current change normalized by the transconductance with the *Vth* shift shows very similar values, suggesting that the device response is dominated by electrostatic effects upon analyte binding.
- We investigated the thermodynamics of the binding between p-Tau$_{181}$ and its corresponding antibody by applying either the Langmuir or the Uniform Langmuir model:[3] the two isotherms yield very similar values, possibly hinting to the fact that the adsorption sites on the gate surface might have a narrow distribution of binding energies.

$$S = \frac{S_{max}}{2A} ln\left(\frac{1+K_{avg}e^{A_c}}{1+K_{avg}e^{-A_c}}\right)$$

- Our independent experiments showed good consistency and successfully detected p-Tau$_{181}$ at concentrations ranging from 50 pg/mL to 200 pg/mL in a complex medium such as artificial cerebrospinal fluid (aCSF), with a theoretical limit of detection (LOD) of 26 pg/mL. These results, together with the negligible non-specific response, corroborate the successful applicability of EGOT-based biosensors for the detection of p-Tau$_{181}$.

5. REFERENCES

1. E. B. Erhardt, J. C. Adair, J. E. Knoefel, A. Caprihan, J. Prestopnik, J. Thompson, S. Hobson, D. Siegel, G. A. Rosenberg, Front. Aging Neurosci. 2021, 13, 1.
2. D. T. Simon, E. O. Gabrielsson, K. Tybrandt, M. Berggren, Chem.
3. A. Paradisi, M. Berto, M. Di Giosia, S. Mazzali, M. Borsari, D. Marforio, F. Zerbetto, M. Calvaresi, A. Orieshyna, N. Amdursky, C. A. Bortolotti, F. Biscarini, *Chem. A Eur. J.* **2023**.

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