

Astra and the Nanoparticles

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Oh! Hello and welcome to this poster! My name is Astra and I'm here to tell you a story!

I'm gonna start with the bad news: many people die from neurological diseases that we still don't understand well [1]

...but some parts, like synapses and dendritic spines, are way too small to study with electrodes!

Luckily, scientists recently created special proteins, called Genetically Encoded Voltage Indicators (GEVIs) [2,3]...

We suspect that in many of these diseases the electrical signals in neurons don't manifest properly...

...which show a fluorescence that is directly dependent on the membrane potential! In other words, they let us see the electrical signal!

Amazing, right? Well, it might be too good to be true...

Where are the spines here?

Their dim signal is covered by the bright signal of the rest of the cell! [4]

This is the part of the story where I come into play!

Thanks to the plasmonic resonance, I can locally enhance the fluorescence! So we get more light, and we know where it comes from! [5]

Why a nanostar and not a simple sphere?

Like humans, nanoparticles are all diverse and unique! Some have more or less spikes... ..some longer or shorter tips... ..some are just different from the average...

That's because nanospheres are not resonant with the protein's emission, but carefully designed nanostars are [6]

Fortunately, simulations show that all this diversity is not a problem! Each one of us can help in this important task! [7]

Normalized extinction cross sections [a.u.] vs Wavelength [nm] graph showing Absorption (dashed lines) and Emission (solid lines) for Nanostar (green) and Nanosphere (blue).

Avg. field enhancement $|E|/|E_0|^2$ (a.u.) vs Tips density (a.u.) and Randomness factor (a.u.) plots.

Time to go to the lab! This is how we're made... [8]

...and this is how we look like when you're close enough! [9]

I am ready! We only need a final proof of principle: the signal enhancement of a coupled GEVI!

Chemical structure: CH3O-Si(OCH3)-OCH3 with HS and SMTMS groups.

We plate HEK293T cells expressing the GEVI on an optical dish covered in a layer of nanostars...

Control

Nanostars

...and the result proves a higher signal! It really works! [10]

Absolute fluorescence signal [a.u.] vs Control and NSs box plot showing a significant increase for NSs (****).

Now that we have a working technique, the challenge is to bind nanostars to certain parts of the neuron... ..but this is a story for another time!

Notes:

- [1] Feigin, V. L. et al. "Global, regional, and national burden of neurological disorders, 1990–2016: a systematic analysis for the Global Burden of Disease Study 2016". The Lancet Neurology 2019, 18.
- [2] Kralj, J. M. et al. "Electrical spiking in Escherichia coli probed with a fluorescent voltage-indicating protein". Science 2011, 333.
- [3] Yang, H. H. et al. "Genetically encoded voltage indicators: Opportunities and challenges". Journal of Neuroscience 2016, 36.
- [4] Hochbaum, D. R. et al. "All-optical electrophysiology in mammalian neurons using engineered microbial rhodopsins". Nature Methods 2014, 11.
- [5] Kühn, S. et al. "Enhancement of single-molecule fluorescence using a gold nanoparticle as an optical nanoantenna". Physical Review Letters 2006, 97.
- [6] FDTD simulations confirm that colloidal gold nanostars are resonant with the emission of Arch(D95N). Nanospheres, on the contrary, could enhance the absorption of Arch(D95N), but the nanoscopic localization due to the tips of the nanostar would be lost.
- [7] A series of FDTD simulations highlighted how the average field enhancement at the resonance wavelength (680nm) does not change drastically when some tips are missing, or when the tips have different lengths from the ideal case. This result motivates our approach based on colloidal synthesis of nanostars, which generates a population of different shapes and sizes.
- [8] Barbosa, S. et al. "Tuning size and sensing properties in colloidal gold nanostars". Langmuir 2010, 26.
- [9] SEM Micrograph of colloidal gold nanostars deposited on silicon substrate, coated with MPTMS.
- [10] The automated screening approach shows a clear 31% enhancement of QuasAr6a fluorescence. The null hypothesis of the one-sided Mann-Whitney test is that the median absolute brightness of the control is greater or equal to the median of the nanostars sample. At the 0.0001 level, the median of the nanostars is significantly higher than the control. Patch clamp analysis (not depicted) shows that the voltage sensitivity is unaffected by the plasmonic effect.

