From Nanobody Forests to Drug-producing MOFs: Designer Proteins for Designer Materials

Raik Grünberg
Ainur Sharip, Somayah S. Qutub, Keying Guo, Escarlet Diaz-Galicia, Yuxiang Ren, Tianrui Chang, Anil Koklu, Shofarul Wustoni, Ondrej Strnad, Sahika Inal, Niveen M. Khashab, Stefan T. Arold
King Abdullah University of Science and Technology (KAUST), Thuwal, 23955-6900, Saudi Arabia
raik.grunberg@kaust.edu.sa

Proteins are found at the core of most biosensing platforms as well as in many recent nanotechnology devices or hybrid materials. However, too often this research relies on off-the-shelf, commercially available model proteins. The collaboration between protein engineers and material scientists can unlock applications that would be inaccessible to either side alone. In our work with the Inal team, we continue to improve a self-assembling nanobody architecture to build sensitive and rapid bioelectronic sensors that operate in unprocessed, complex samples [1]. The spyDirect biofunctionalization method creates ultra-high-density arrays of correctly oriented and fully binding competent nanobodies that are stably linked to a gold surface (Figure 1). SpyDirect biorecognition layers improve the long-term stability and lower the background noise of organic electrochemical transistors (OECTs) for the detection of SARS-CoV-2 in saliva or unprocessed waste water [2]. In our work with the Khashab team, we are combining their hierarchically engineered metal organic framework (MOF) nanoparticles with the *in vitro* reconstituted six-enzyme pathway for the biosynthesis of violacein [3]. Pathway-MOF nanoreactors produce violacein in amounts comparable to solute enzymes, but enable pathway reuse, lyophilisation and storage. MOF nanoreactors can deliver the entire multi-protein system into mammalian cells where it interfaces with the metabolic state of cancer cells leading to the enhanced production the cytotoxic violacein as an *in-situ* therapeutic (Figure 2). We believe that violacein nanoreactors may pave the way towards a novel class of intracellular protein systems therapies.

References

[1] Guo, Wustoni & Koklu *et al.* Nat Biomed Eng, **5** (2021) 666–677
[2] Guo *et al.* (2023) submitted
[3] Sharip & Qutub *et al.* (2023) submitted

Figures

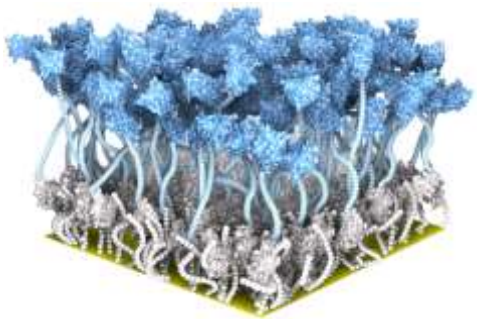

Figure 1: Rules-based model of a spyDirect nanobody-functionalized electrode surface.

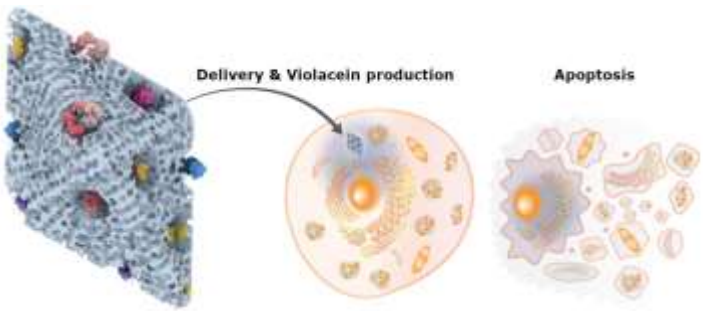

Figure 2: Illustration of eMIL - multi-enzyme nanoreactors and their delivery into mammalian cells.