The sequestration mechanism: a practical approach to narrow the dynamic range of biosensors and bioassays

Alejandro Chamorro-Garcia
Claudio Parolo, Gabriel Ortega, Andrea Idili, Joshua Green, Francesco Ricci, Kevin W. Plaxco
University of Rome Tor Vergata. Via della Ricerca Scientifica 1, 00133 Rome, Italy.
chmlnd01@uniroma2.it

Abstract

Biosensors and bioassays have seen significant success largely due to the extraordinary versatility, affinity, and specificity of biomolecular recognition. Nevertheless, these receptors suffer from two inherent limitations: the single saturable binding site hyperbolic relationship (Langmuir isotherm) between target concentration and receptor occupancy, and the position of the linear response region with respect of the detection range of interest. That limits the sensing capabilities of technologies based in bioreceptors. To overcome this limitation and generate more responsive assays, researchers have previously explored the nature-inspired mechanism of sequestration [1], which improves steepness of the input/output curves and allows to shift the position of the linear response range of bioanalytical methods. This mechanism relies on the use of two biorecognitions elements: a higher-affinity, non-signalling "depletant", and a lower affinity, signal-generating "receptor" which generates an output signal upon target binding. The resulting enhancement in responsiveness improves our ability to measure small relative changes in target concentration [2]. In this talk I will present how by incorporating sequestration we can induce steeper transitions in dose response curves of an aptamer based electrochemical biosensor (EAB sensor), lateral flow immunoassay (LFIA), and ELISA. All three techniques adapted for the detection of neutrophil gelatinase-associated lipocalin (NGAL), a biomarker for kidney damage [3].

References

[1] Nicolar E. Buchler, Frederick R. Cross, **Mol. Syst. Bio.,** 5 (2009), 272.
[2] Francesco Ricci, Alexis Vallé-Bèelisle, Kevin W. Plaxco, **PLoS comput. Biol.**, 7 (2011), (10):e1002171.
[3] Claudio Parolo, Andrea Idili, Gabriel Ortega, Andrew Csordas, Alex Hsu, Netz Arroyo-Curras, Qin Yang, Brian S. Ferguson, Jinping Wang, and Kevin W. Plaxco, **ACS Sens.,** 5,7 (2020), pp. 1877 – 1881.

Figures

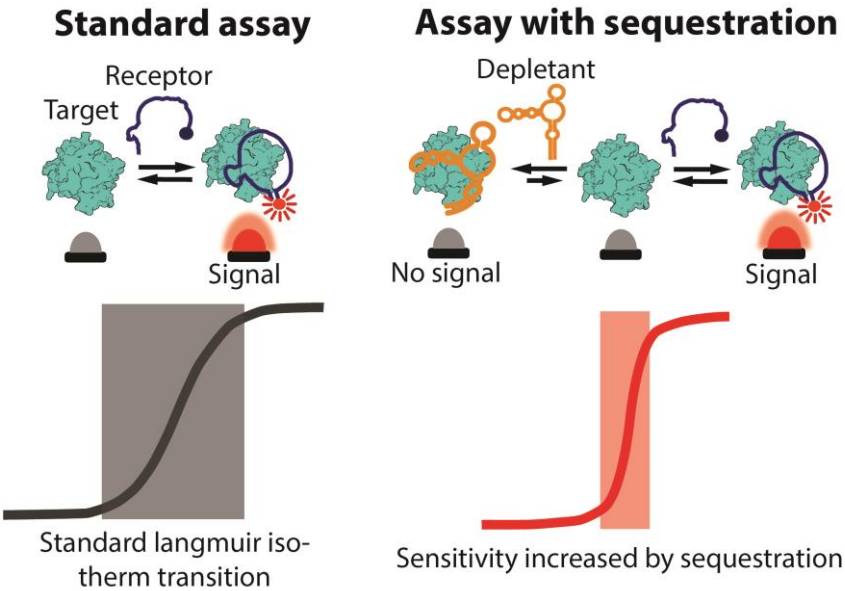

Figure 1: Sequestration mechanism employs a depletant: biomolecule that binds its target with hight affinity without producing any signal. The depletant acts as a "sink", ensuring the low concentration free target. When target concentration exceeds the depletant's, target's relative concentration increases dramatically. This, in turn, activates a second, lower affinity "receptor" that generates a signal change. Sequestration then (right) leads to a steeper binding curve, improving the response to small changes in target concentration.