INNOVATING CROSS-DOMAIN SOLUTIONS TO DETECT EMERGING BIOLOGICAL THREATS

Giant Quantum Dots (gQDs) As Fluorescent Probes For Improved Antibody-antigen Interaction Studies Through 3D Quantum-enhanced Single Particle Tracking

Maria Nevarez Martinez Los Alamos National Laboratory **Demosthenes Morales** Los Alamos National Laboratory **Eric Bowes** Los Alamos National Laboratory **Duncan Ryan** Los Alamos National Laboratory **Jennifer Hollingsworth** Los Alamos National Laboratory **James Werner** Los Alamos National Laboratory

Traditional methods for the detection of pathogens, mainly rely on the assessment of antibody-antigen interactions through lateral flow or sandwich immunoassays. Despite the wide application of those technologies, they require immobilization of antibodies on a solid surface, what may involve conformational changes, denaturation, and steric hindrance that could result in loss of affinity 1. We propose to address those drawbacks by 3D quantum-enhanced single-particle tracking to acquire binding kinetics and analyze the dynamics of the interactions between pathogens and immobilized antibodies at the single particle level. The 3D quantum tracking of immunocapture pathogens (3D Q-TIP) that we present, depends on the development of fluorophores to tag specific pathogens. For this, our efforts have focused on the development of giant quantum dots (gQDs) which have proven to be efficient fluorescent probes offering non-blinking 2 and photon antibunching to enhance spatial resolution 3 even when imaging live cells 4,5. Herein, we have synthetized non-blinking gQDs with tunable IR-emission and photon-antibunching properties. The probes were ligand exchanged for aqueous compatibility and successfully attached to Burkholderia thailandensis. The effective bioconjugation and pathogen-labeling were assessed by cryo-EM. Close-future implementation of the 3D Q-TIP technology with the developed gQDs-tagged pathogens will provide in-depth understanding of epitope binding kinetics, and the collected data will allow optimization of existing diagnostic technologies, increasing their sensitivity and robustness. Additionally, this research approaches the nation's biosecurity concern and can be extended to different antigens/pathogens/diseases leading the advancement of diagnostic tools. (LA-UR-24-24181)

Figure 1. Cartoon of an immunoassay with immobilized capture antibodies and pathogens labeled with non-blinking giant QDs. Bacteria are represented as red rods and the gQDs as yellow circles. The Figure was partially generated with Servier Medical Art, licensed under a Creative Commons Attribution 3.0 Unported License

References:

1 Jung, Y., Jeong, J. Y. & Chung, B. H. Recent advances in immobilization methods of antibodies on solid supports. Analyst 133, 697-701 (2008). https://doi.org/10.1039/b800014j

2 Chen, Y. et al. "Giant" Multishell CdSe Nanocrystal Quantum Dots with Suppressed Blinking. Journal of the American Chemical Society 130, 5026-5027 (2008). https://doi.org/10.1021/ja711379k

3 Mangum, B. D. et al. Influence of the core size on biexciton quantum yield of giant CdSe/CdS nanocrystals. Nanoscale 6, 3712-3720 (2014). https://doi.org/10.1039/c3nr06558h

4 Keller, A. M. et al. 3-Dimensional Tracking of Non-blinking 'Giant' Quantum Dots in Live Cells. Adv Funct Mater24, 4796-4803 (2014). https://doi.org/10.1002/adfm.201400349

5 Lane, L. A., Smith, A. M., Lian, T. & Nie, S. Compact and blinking-suppressed quantum dots for single-particle tracking in live cells. J Phys Chem B 118, 14140-14147 (2014). https://doi.org/10.1021/jp5064325