SP3* Ultra-high sensitivity diagnostic assays in blood: Sergey Y. Tetin, M.D./Ph.D., Abbott Laboratories
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Significance: Abbott laboratory is one of major American health care companies. Abbott Diagnostic Division is the world leader in blood supply screening, core lab and point-of-care diagnostics with the net annual sales exceeding $4.6 billion in 2015. Within this Division, Dr. Tetin is the Head of the Molecular Binding Characterization group with annual internal funding of of ~$1.5M. His group participates in Abbott contracts with DOD on developing blood test for head injury (~$19.5M) and USAID on Zika infection (~$3.4M). His group pioneers in the development of new ultra-sensitive techniques for detection of molecular interactions and provides scientific guidance in reagent selection for diagnostic immunoassays. Dr. Tetin is a Senior Research Fellow in the Volwiler Society, which recognizes the highest level of achievements at Abbott. Abbott's strength in immunoassay products lies in their ability to develop some of the most specific and high affinity antibodies. Today, high sensitivity assay, such as ones for troponin, has sensitivity on the pg/ml level. These assays are routinely used for diagnosing cardiac ischemia. While existing assays have the required sensitivity for disease diagnosis, they often do not have the requisite sensitivity to detect the much lower concentration of analytes in the blood of healthy people. Abbott scientists hypothesize that if more sensitivity assays are available, such as ones that can robustly measure troponin level in a normal person, healthy people can routinely monitor their heart health. If a panel of ultrasensitive and robust assays can be developed for the healthy population, this advances can potentially be a major breakthrough in preventive medicine.

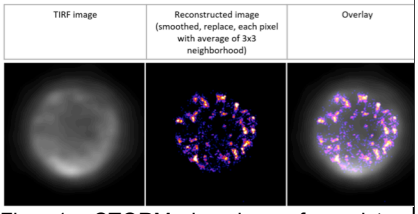

Fig. 1: STORM imaging of analyte binding on microp-articles commonly used in immunoassays. (Abbott data).

Approach: Towards better immunoassays, Dr. Tetin's group implements biophysical methods such as X-ray and NMR analysis, fluorescence fluctuation spectroscopy, advanced fluorescence microscopy to perform structure-function and binding characterizations of the assay components (Fig. 1). Measuring pertinent equilibrium and kinetic coefficients of antibodies and related target molecules reveals the inherent limits in using specific reagent combinations imposed by the detection concentration range and chosen incubation times. The apparent kinetic rates at each assay step are used to compute the assay calibration plot. By assaying good agreement between experimental assays with the computed data, they lay solid foundation for immunoassay design and optimization (1-14). In Abbott laboratories, they have often demonstrated exquisite assay sensitivity even on the single molecule level. However, providing a robust assay for blood analyte detection in the field is much more challenging. One of the two major limitations that they have identified is the slow reaction kinetics inherently associated with low analyte concentration in the blood. In order to compensate for the low concentration, it is required that they significantly increase the concentration of antibody coated microparticles associated with increasing reagent cost and assay time. The other limiting factor is the background of fluorescent immunoassay. In the case of low analyte concentration, while fluorescent probes, like phycoerythrin, are fairly bright, they are challenged by the ubiquitous autofluorescent background inherent to many assay components such as the residual fluorescence of the microparticles themselves. For these challenges, LBRC may have promising solutions that can help Abbott scientists towards developing more sensitive and robust assays. Addressing the problem of reducing fluorescence background, the short-wave infrared (SWIR) quantum dots (QDs) developed in TRD1 (and TRD4 in the next cycle) may be applicable for conjugation with high affinity antibodies by performing detection in a spectral region with almost no auto-fluorescent background. Very high sensitivity detection of analyte binding may become possible using high sensitivity SWIR microscope systems available in LBRC. While LBRC has no expertise in tackling analyte binding kinetics issues, LBRC has a long standing collaboration with Dr. Jay Han, a microfluidic expert in MIT. Dr. Han has invented microfluidic devices that Dr. Tetin may be deployed to isolate and to concentrate analytes in the blood prior to performing SWIR imaging based immunoassay.
Center Offering: First, LBRC provides bright, photobleaching resistant, low background SWIR quantum dots with flexible protein conjugation chemistry that Dr. Tetin can be optimized using Abbott's proprietary high affinity antibodies. Second, LBRC provide high sensitivity microscope system optimized for SWIR imaging and detection. By assaying in background-free SWIR spectral region, improvement in sensitivity may be achievable. Third, LBRC is able to offer Dr. Tetin novel microfluidic devices developed by the Han lab to isolate and pre-concentrate blood analyte prior to immunofluorescent assay to substantially improve binding rate that has the benefit of enhancing assay sensitivity and reducing reaction time. It is important to note that LBRC serves the community not only as an originator of photonic solutions, LBRC is also an important hub of biomedical technology development in general and serves as a matchmaker for our diverse group of collaborators.

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