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# Rapid Affinity Reagent Development for Emerging Pathogen Detection
158820

**Year 2 of 2**
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## Project Purpose:

We face constantly emerging and naturally occurring global biological threats, from pandemic influenza to novel pathogens or engineered bioagents. In the event of disease outbreak or biothreat release, fast and sensitive diagnostic tools deployable to multiple point-of-care (POC) locations are crucial for effective biosurveillance and crisis management. Rapid antigen-based tests, which are the most amenable approaches to POC diagnostics, typically employ antibody affinity capture schemes for pathogen detection. However, antibody production relies on hybridoma technology which requires several months for full scale production, making antibodies inadequate for emerging pathogen outbreak scenarios. Moreover, antibodies are not very stable to storage, especially in resource-poor areas lacking refrigeration. Polymerase chain reaction (PCR) approaches require extensive sample preparation, are very expensive because of thermal cycling needs, and often fail to detect newly emerged or divergent pathogen strains. Thus there remains a critical need for versatile POC diagnostic approaches that are rapidly reconfigurable and deployable in emerging pathogen outbreak scenarios.

Here we propose to remove this bottleneck in biological emergency responsiveness by employing Sandia's virus-like particles (VLPs) as rapidly-deployable affinity reagents. VLPs have shown efficacy in cell targeting and drug delivery applications; this project will pioneer their use as diagnostic affinity reagents. VLPs require 5-12 days for development with affinities for target analytes that rival commercial monoclonal antibodies. VLPs are less-likely to denature and hence have much longer shelf-life than antibodies. VLPs can also be loaded with 'cargo' such as multiple quantum dots (QDs) facilitating a novel sedimentation assay directly targeting infectious agents using Sandia's centrifugal microfluidics platform. The proposed assay scheme will exceed the stringent limits of detection for emerging pathogens (10^3 PFU/mL), and may enable single particle resolution. This project merges two of Sandia's proven technologies to tackle the pressing challenge of emerging pathogen detection; the final product will vastly outperform state-of-the-art techniques and directly address this glaring national and global security need.

## Summary of Accomplishments:

We designed methods of utilizing virus-like particles with specific affinity ligands on the surface of the constructs to bind to pathogens and subsequently detected these complexes on device. We demonstrated sensitive detection using these methods for model systems in a laboratory setting.

Furthermore, we concurrently designed non-amplified nucleic acid assay methods which were demonstrated to be highly sensitive and specific, comparing favorable with FDA-approved gold standard methods. These methods are further enhanced by deployment on the SpinDx platform.

## Significance:

By developing methods of rapidly producing affinity reagents for pathogens which are

poorly characterized or understood, we have enhanced the ability to respond to threats both naturally occuring and man-made. Additionally, these strategies are incorporated into Sandia's proprietary point-of-care medical devices allowing for rapid deployment of these solutions to low-resource settings.

**Refereed Communications:**