NEXT-GENERATION BIOAEROSOL DETECTION & IDENTIFICATION

Development Of Micro-fluidic Capability For Rapid Environmental Bioreconnaissance Point Sensor Targeting Biological Threats

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The COVID-19 pandemic has demonstrated the role of bioaerosols with regards to the spread and dissemination of infectious agents. Bioaerosols are one of the primary ways infectious and toxic agents are spread and dispersed, as the small size of the particles are readily dispersed and can remain suspended in the air for hours to days. As a result, aerosolized agents are more likely to be inhaled, which increases their toxicity and infectivity due to the semi-permeable nature of the mucosal membranes in the respiratory tract. In spite of this, few rapid, deployable methods exist to collect, detect, and identify bioaerosols.

Biological threats are often detected using methodologies based off of immunoassays including enzyme-linked immune specific assays (ELISA), but those are often both labor and time intensive; they are thus insufficient for implementation in pseudo real-time environmental monitoring and field deployment. Centrifugal microfluidics (CMs) provides one solution to the problem for sample handling, as microfluidic gating techniques that utilize centrifugal force can simultaneously control fluidic motion as well as mixing of liquids. This technique has also substantially reduced the time of some ELISAs from ~four hours to 25-60 minutes. However, additional, non-linear steps in assay development are needed to further reduce the timescale. Additionally, reagents must be identified and incorporated into the system that do not require cold-chain storage and are shelf-stable for extended periods of time.

Utilizing the expertise of our multidisciplinary team we have developed methods to collect and detect biotoxins in under 15 minutes using a centrifugal microfluidic device (CMFD). Notably, our team has demonstrated the following achievements. First, protein bioaerosols, including those containing the simulant human serum albumin (HSA), can be collected, concentrated, and re-suspended to detectable concentrations in a matter of minutes. Notably, the reconstituted protein antigens are compatible with ELISAs, demonstrating the efficacy of the technique. Second, shelf-stable bio-recognition elements (BREs) were discovered through high-throughput screening processes that 1) have high selectivity, 2) bind to orthogonal epitopes, and 3) have a strong affinity for the target toxin. Notably, the identified toxin-specific BREs are suitable for sandwich-based assays. Third, our team has explored and demonstrated electrochemical and sandwich assays for rapid detection of toxins at concentrations as low as 25 ng/mL under 10 min. Notably, using synthetic BREs, we demonstrate that multiple steps in traditional assays can be skipped or combined to simplify assay protocol and shorten the run-time to reach the final YES/NO answer. Additionally, we also demonstrate that time is a critical parameter to explore when executing sandwich assays using synthetic BREs due to the more rapid binding kinetics (both on-rate and off-rate). Finally, our team has designed and produced CM devices that can execute these assays, leveraging both antibody and synthetic BREs immobilized on beads. The accomplishments described above provide a fundamental platform and methods towards future development of rapid, cost-effective, and minimal-maintenance autonomous assays that are deployable towards defending civilians and military from bioaerosol threats.

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