

Introduction

The Joint USFK (United States Forces Korea) Portal and Integrated Threat Recognition Advanced Technology Demonstration (JUPITR ATD), led by JPEO-CBD, will provide unique biological detection capabilities to address the demand for stronger bio-surveillance capabilities on the Korean Peninsula. The JUPITR ATD has four legs: #1: Biosurveillance Portal (BSP); #2: Biological Identification Capability Sets (BICS); #3: Assessment of Environmental Detectors (AED); #4: Early Warning. During 2013-2015, ECBC's Aerosol Sciences Branch supported the AED leg, culminating in the assessment of 10 different biological detectors/identifiers from August – November, 2014 in our Ambient Breeze Tunnel (ABT). The objective of the demonstration was to provide information on the Limits of Detection (LoD) and Limits of Identifications (LoID) for each of the systems in a “real-world” environment using four Agent Like Organisms (ALO).

Preliminary Characterization



The ABT in E5884 is 14' x 14' x 200' tunnel capable of wind speeds from 0-6 mph. It is equipped with HEPA filter banks that enable aerosol experimentation up to BSL-2. This facility brings in outdoor air, which allows for testing in an environment that more closely resembles the “real world” while maintaining a controlled experiment. To ensure a fair and unbiased assessment, the ABT was extensively characterized to ensure uniformity of aerosol concentration across the test section. SUDs in the 10' x 20' test section (right) were staggered to mitigate perception of favoritism.

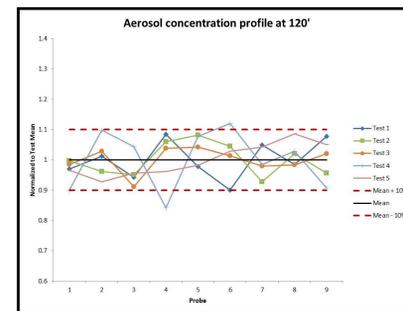
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Preliminary Characterization (cont)

In addition to the ABT characterization, QA/QC procedures were implemented in the microbiology labs to ensure the validity of referee sample analysis. Five different PCR extraction kits were evaluated for reproducibility and sensitivity, referee filter extraction protocols were verified, and each of the ALOs were characterized upon receipt of materials to corroborate the reported concentrations. Additionally, extensive characterization of the aerosolized ALOs were conducted to enumerate the number of organisms per particle. Finally, the aerosol generation techniques were validated in the ABT to establish the required parameters to achieve reproducible concentrations in terms of organisms per liter of air in the test section.



Aerosol Uniformity characterization setup (above)
Uniformity data (right) shows concentration is roughly $\pm 10\%$



Execution of Trial Matrix

Four Agent Like Organisms (ALOs) were used to establish LoDs and LoIDs for the Systems Under Demonstration (SUD): Live *Ba* Sterne to represent spore forming bacteria; gamma irradiated *Yp* to represent vegetative bacteria; gamma irradiated *Vaccinia* Lister to represent viruses; and inactivated *C. Botulinum* Toxoid to represent toxins. Dilutions of each material were made to produce 3 micron particles (mass mean aerodynamic diameter), which is an optimal size for most aerosol collector/detectors. The SUDs were challenged at concentrations ranging from 50 – 1000 Agent Containing Particles per Liter of Air (ACPLA). Twenty-six trials were conducted for each ALO at the various concentrations. Three Aerodynamic Particle Sizers were used to measure the size and concentration of the challenges in real time. Additionally, three reference filters were used to quantify the concentrations of each challenge. The filters were extracted and analyzed. *Ba*, *Yp*, and *Vaccinia* were analyzed using RT Q-PCR on an ABI 7500 Fast DX with assays from the CRP, and Bot samples were analyzed using ELISA kits from Tetracore, Inc. and read with a Molecular Devices Vmax kinetic microplate reader.

Layout in the ABT



View from the open end of the ABT (above). A large fan at the aft end brought in outside air at a wind speed of 2mph. Aerosols were generated using a SonoTek aerosol generator at the open end of the ABT and traveled down the tunnel to the test section 100 feet away. Fans were used to create a turbulent flow to mix the aerosol and achieve a uniform concentration in the test section.

Acknowledgements

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