



NIDS[®] Multiplexed Assays for Biothreat Agent Detection by Civilian First Responders

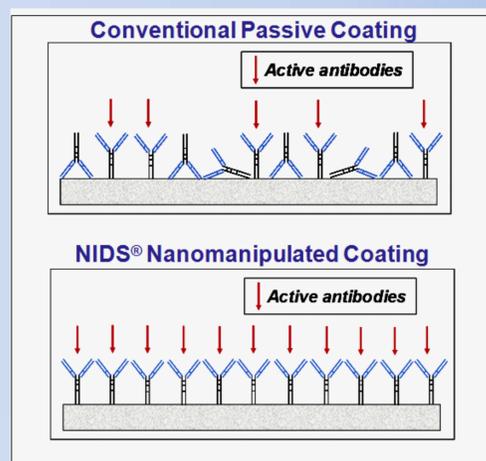


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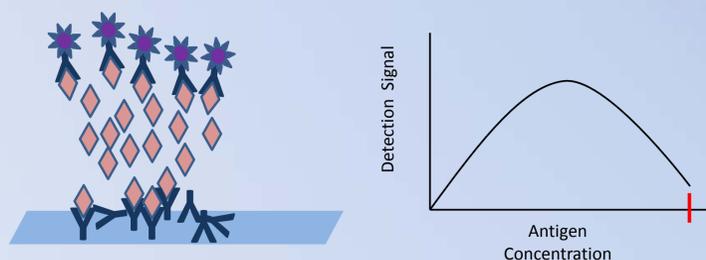
Introduction: ANP Tech has developed multiplexed lateral flow assays for the detection of biothreat agents in waters and powders for the U.S. Department of Defense. The same technology has now been used to produce assays for use by civilian first responders, offering the following advantages over traditional lateral flow assays:

- High Sensitivity
- Low False Positive Rate
- No Hook Effect
- Long Term Room Temperature Stability
- Multiplex Capability
- Ease of Use

The Advantage of Nanotechnology: The NIDS[®] system uses innovative chemistry to orient bioactive molecules such as antibodies, antigens, nucleic acid oligomers, and other binders on a wide variety of surfaces. This orientation ensures the optimal configuration for binding interactions, producing more active antibodies for binding interactions.



The Hook Effect: The Hook Effect is a false negative result in the presence of high concentration of biothreat antigen. The excess antigen binds on both the capture and detector antibodies, preventing a sandwich from being formed, and producing a false negative result.



Stand Alone Reader: ANP Tech has developed a hand-held reader that automatically records and interprets assay results for a user. The reader has eliminated human to human variations when reading assay results in the field, particularly under low light conditions.

The Reader features include:

- Runs on standard alkaline AA batteries
- No tools required to access the following:
 - Battery compartment
 - Removable base for easy cleaning and decontamination
 - Sealed micro-SD card and mini-USB port
- Removable 2 GB microSD flash memory
- Large, color LCD display with variable brightness control (5 levels of intensity)
- Intuitive device User Interface (UI) with context-sensitive button functions

Interferent Compound Testing: The NIDS[®] assays were challenged with the following interferent compounds and white powders, both neat and with antigen at 2X the Detection Limit. No false positives from the neat compounds and no false negatives from suppression of the antigen signal resulted.

Water	Malathion	Clay Soil	Sandy Soil
Loamy Soil	Vero Cell Supernatant	G Media w/ trace minerals	PBS
Tween 80	Burning Vegetation	Burning Diesel	Burning Fog Oil
Burning Rubber	HC Smoke Gunshot	Yellow Signal Smoke	Red Signal Smoke
Green Signal Smoke	Violet Signal Smoke	Nutrient Broth	BHI Broth
Brucella Broth	TSB Broth	Burning Rags	<i>Aspergillus</i>
Dipel	Cleaner	Powdered Sugar	Dry Milk
Baking Soda	Bentonite	Aerosil R812S	Flour
Salt	Kaolin	Foot Powder	Creamer
Yeast	Chalk	Talcum Powder	Spackling Powder
----	Sage Pollen	Bovine Serum Albumin	-----

Biothreat Agents: Current targets include bacteria and toxins, with additional reagents under development to expand the number of targets detected. The multiplexed assays can detect:

- Bacteria 3-Plex:** *B. anthracis*, *Y. pestis*, and *F. tularensis*
- Toxin 4-Plex:** Botulinum toxin A, Botulinum toxin B, Ricin, and Staphylococcal Enterotoxin B

Overcoming the Hook Effect: The nano-orientation technology used in the NIDS[®] assays has eliminated the need for serial dilutions of samples to avoid the Hook Effect. The table below shows concentration levels of biothreat targets tested, with positive results for all targets and no cross reactivity with any other agent on the assay.

Target and Concentration	Bacteria		Toxin	
	Reader	Visual	Reader	Visual
<i>B. anthracis</i> 7.3E8 cfu/mL	Pos	Pos	--	--
<i>Y. pestis</i> 3.6E8 cfu/mL	Pos	Pos	--	--
<i>F. tularensis</i> 9E8 cfu/mL	Pos	Pos	--	--
Bot toxin A 0.1 mg/mL	--	--	Pos	Pos
Bot toxin B 0.1 mg/mL	--	--	Pos	Pos
SEB 0.1 mg/mL	--	--	Pos	Pos
Ricin 0.1 mg/mL	--	--	Pos	Pos

Summary: ANP Technologies continues to develop and produce the next generation of lateral flow assays for both military and civilian first responders. The military assays have been extensively validated and are currently fielded as part of the JCBRAWM Water Test Kit, in addition to experiencing limited fielding through special forces. The civilian assays use this same proven technology that eliminates the Hook Effect, allows multiplex capability, and offer a two year room temperature shelf-life. Smiths Detection is the world-wide distributor of this technology, and has performed internal validation studies of the assays' capabilities.