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Independent Testing of Hand Portable Biodetection Equipment

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Pacific Northwest National Laboratory
Richland, WA

Presentation at:
APHL Annual Meeting
June 2014

U.S. DEPARTMENT OF
ENERGY



Photo Credit: Bryon Marsh, GA CST

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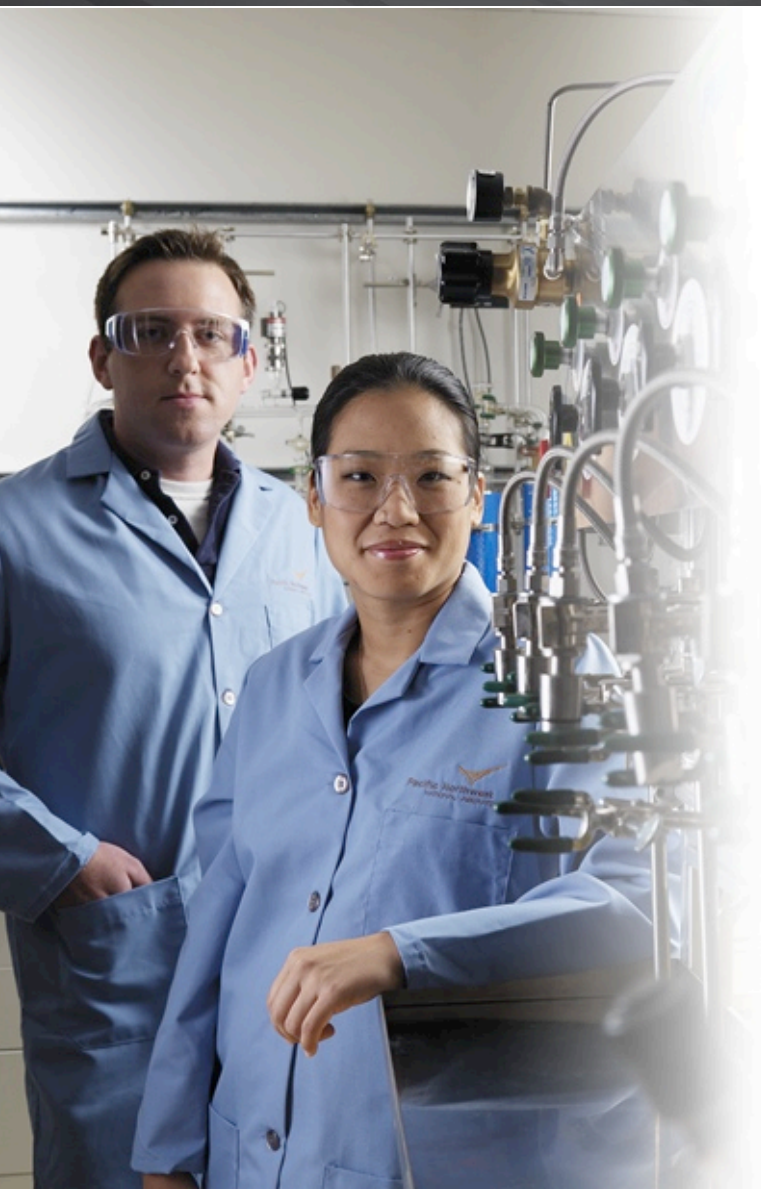
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We
**TRANSFORM
THE WORLD**
through
courageous
DISCOVERY
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INNOVATION.

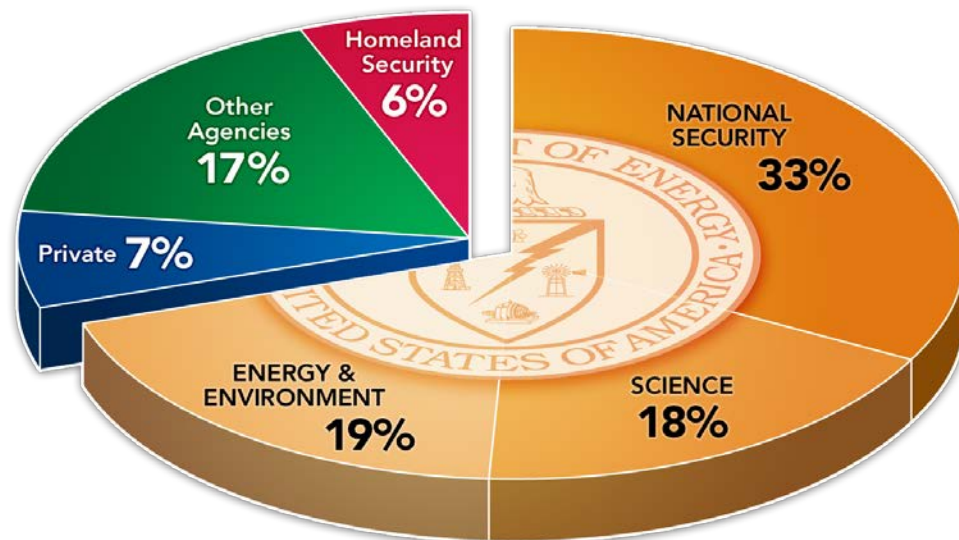
- ▶ Operated by Battelle since 1965
- ▶ Unique S&T capabilities
- ▶ Mission-driven collaborations with government, industry and academia



PNNL at a glance

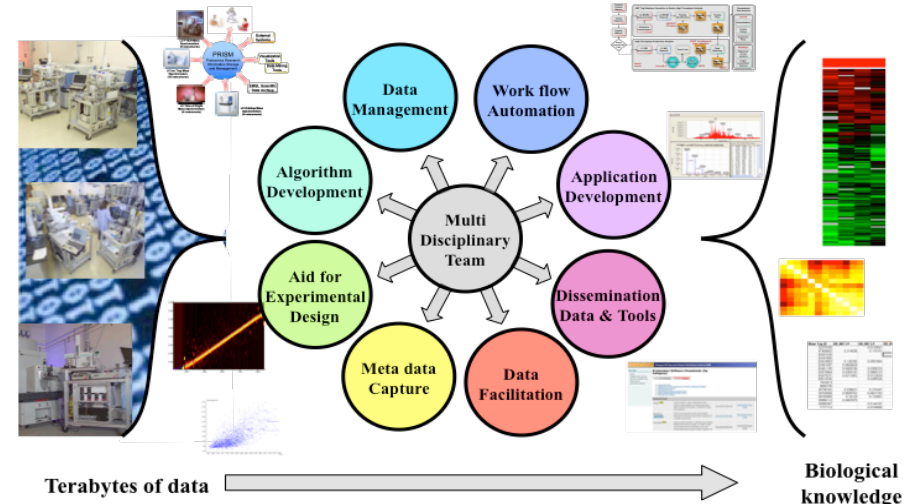


- ▶ \$936M operating budget
- ▶ 4,300 scientists, engineers and non-technical staff
- ▶ Yearly accomplishments include
 - 1,168 peer-reviewed papers
 - 85 U.S. and foreign patents granted; 264 invention disclosures
 - 4 R&D 100 and FLC Awards



Biology Programs at PNNL

- ▶ Total biology programs: ~90M
 - Spans basic science to application
 - Balanced between DOE (BER), NIH, and National Security clients
- ▶ People: ~280 scientists conduct biological research as primary focus
 - Many others involved in biological research
- ▶ World-leading proteomics foundation





Field Biodetection: PNNL Support Role

- ▶ Field biodetection technology is rapidly advancing and is in widespread use, although the quality of results is largely unknown
- ▶ While standards for sampling and CONOPS exist (ASTM E2458-10 & E2770-10), implementation support is limited



▶ Framework for Biothreat Field Response Mission Capability

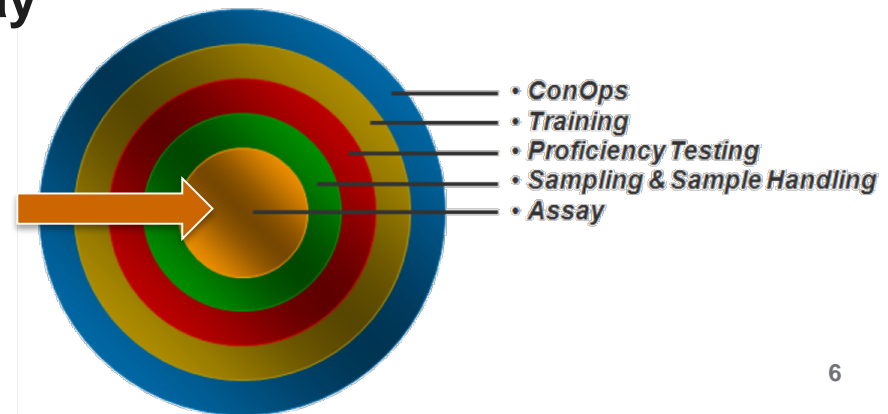
- ConOps
- Training
- Proficiency testing
- Sampling and sample handling
- **Assay**

Framework for a Biothreat Field Response
Mission Capability

April 5, 2011



PNNL's Role



The Suspicious “White Powder” Challenge

- ▶ Thousands of suspicious powder events have been reported since 9-11
 - Events are costly: from local to state and federal level
 - Total estimated yearly cost for suspicious powder events: millions of dollars
 - Does not include impacts due to diverting resources from other needs to bioresponse
 - Can lead to illness and loss of life
- ▶ What instrument(s) can be used to rapidly determine if a powder/unknown sample contains a biological threat?
 - Cost for instrument and analyses?
 - Ease of use and time to result?
 - Confidence of the information/data?



Need for independent testing to
assess instrument performance and use limitations

Ground-Up Approach: Biodetection Technology ID, Assessment, and Transition



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- ▶ **Define** performance requirements with stakeholder and end user input
 - **Conduct** interviews
 - **Bring together** stakeholders, end users, and key agency reps
- ▶ **Perform** technology foraging
- ▶ **Establish** technology test plans
- ▶ **Conduct** 3rd party testing of biodetection technologies

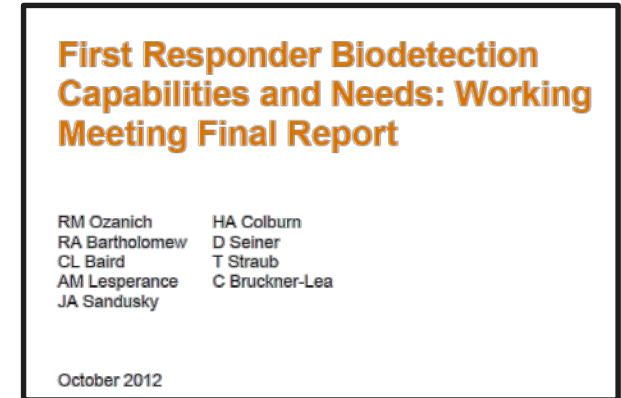


- ▶ **Publish** and disseminate instrument and assay testing results and summaries for instrument use, limitations, cost/benefit
- ▶ **Facilitate** transition of information to first responder community

Meetings with Stakeholders, End Users, and Key Agency Representatives

▶ Working Meeting at Seattle Fire Training Center (May 2012)

- Conducted interviews of first responders in the Pacific NW region
- Brought together end users and others in the hazmat community (FBI, LRN, CST, DHS) to understand and define key biodetection needs and gaps
- Developed recommendations for the near and long term to address gaps and needs



▶ Session at IAFC (May 2014)

- Biodetection 101: Instrument Testing, Use and Limitations, and Needs/Gaps Discussions
 - Provided overview of the biodetection test results
 - Facilitated a panel discussion with hazmat experts from across the Nation, highlighting:
 - ◆ Specific biodetection-related issues
 - ◆ Current limitations with existing equipment
 - ◆ Biodetection and sampling information needs/gaps



Biodetection Technologies Report

- ▶ Biodetection Technology Report available FREE on project website: **biodetectionresource.pnnl.gov**
- ▶ Report on smart phone app available soon
- ▶ Includes sources of information and lists of peer-reviewed references
- ▶ Technologies include:
 - Sampling kits
 - General biological indicator tests:
 - Protein, adenosine tri-phosphate (ATP), and total DNA tests
 - Fourier transform infrared spectroscopy (FTIR)
 - Specific assays:
 - Protein-based: Immunoassays
 - DNA-based: Polymerase chain reaction (PCR)

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Biodetection Technologies for First Responders: 2014 Edition

RM Ozanich HA Colburn
CL Baird TM Straub
RA Bartholomew CJ Bruckner-Lea

March 2014

PROFILE[®] 1 Kit for Detection of Live Cells

General Biological Indicator Test		
Assay	Biological Indicator	LOD*
PROFILE [®] 1	ATP	See comparable value for spores **

* Reported in peer-reviewed reference
** Approximately 2000-10,000 spores/mL

Assay time: ~15 minutes (longer growth times can be used to improve sensitivity).
Basis of Detection: Presence of ATP (requires live, metabolically active biological material); does not distinguish between biothreat and non-threat materials.
Required sample preparation? Moderate.
Automatic results display? User interprets numerical readout.
Unit Weight: <1 lb.
Power: 9V battery (1000+ reads) or AC.
Cost: Assay – \$450 (reagents and materials for 100 tests; \$4.50 ea); instrument – \$5000 (includes carrying kit, micro luminometer, filtravette, and pipettor).
Additional costs: None.
Assay shelf-life: 12 months from date of manufacture (some reagents require refrigeration).

12:34 PM

Biodetection Technologies for First Responders: 2014 Edition

RM Ozanich HA Colburn
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Browse products by categories

- Sample Collection See All >
- Biological Indicator Tests See All >
- Immunoassays See All >
- PCR-Based See All >

Explore other content

Biodetection Tutorial Your Favorites

Prepared for the Department of Homeland Security Science and Technology Directorate
<http://biodetectionresource.pnnl.gov>

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Biodetection Technologies Under Evaluation

- ▶ **Goal:** Understand instrument performance, limitations, and cost/benefit of detection technologies
- ▶ **Test samples:** Common hoax powders, *Bacillus anthracis* spores and DNA (including near neighbor DNA), pure and crude ricin preps (~1% toxin)
- ▶ General Biological Indicator Tests:

■ Protein

- BioCheck Powder Screening (20/20)
- INDIPRO strips (Macherey-Nagel)
- TASKit BioScreener (Field Forensics)

■ Chemical/Spectroscopic

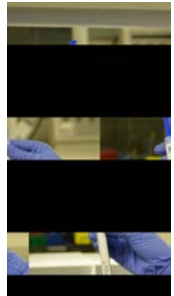
- HazMatID 360 FTIR (Smiths Detection)

■ ATP

- Clean-Trace (3M)
- PROFILE 1 (New Horizons Diagnostics)

■ DNA

- Prime Alert (GenPrime)



Biodetection Technologies Under Evaluation

► Immunoassays and Readers

- 1-agent BADD assays (AdVnt)
- 5-agent ProStrips (AdVnt)
- 5-agent RAID 5 (Alexeter)
- 8-agent RAID 8 (Alexeter)
- 1-agent BioDetect Test Strips and Guardian Reader (Alexeter)
- 3-agent and 4-agent NIDS assays and SAR IV Optical Reader (ANP Technologies)
- 8-agent IMASS assay (BBI Detection)
- 1-agent ENVI assays and Bioassay Optical Reader Module (EnviroNics)
- 3-agent Toxin Screen (GenPrime)
- 1-agent SMART II assays (New Horizons)
- 1-agent Zephyr ID System (Path Sensors)
- 1-agent RAMP Test Strips and Optical Reader (Response Biomedical)
- BioThreat Alert Test Strips and Optical Reader (Tetracore)



Biodetection Technologies Under Evaluation



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► Polymerase Chain Reaction (PCR)

- RAZOR EX (BioFire)
- FilmArray (BioFire)
- T-COR 4 (Tetracore)
- POCKIT (GeneReach USA)
- Bio-Seeq PLUS (Smiths Detection) (no ricin assay)



Criteria for Successful Testing

▶ Criteria

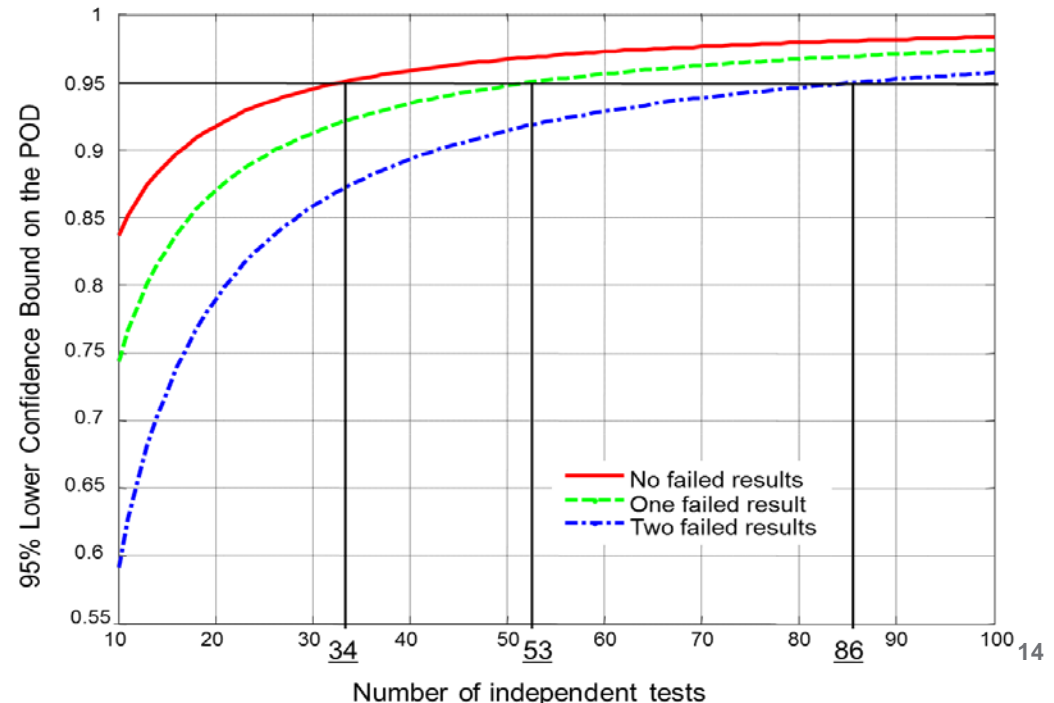
- Achieve a minimum Probability of Detection (POD) of 0.95 with 95% confidence

▶ Assumptions

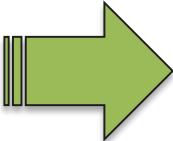
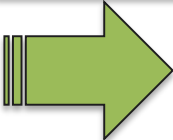
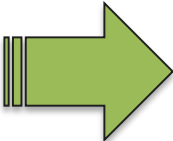
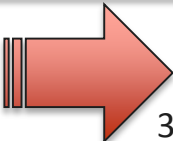
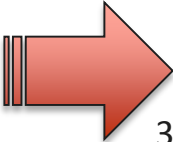
- DNA testing assumes all inclusivity strains equivalent and all exclusivity strains equivalent
- White powder testing of high specificity assays assumes all environmental powders are exclusivity samples and provide equivalent information.

- 34 samples must be tested with no failures
- 53 samples must be tested with ≤ 1 failure
- 86 samples must be tested with ≤ 2 failures

Number of Tests Needed for 95% Lower Confidence Bound on POD



Hand Portable PCR Platform DNA Test Results

Instrument	Type of test	# Analyses	Positive	Negative	Overall # of Failed Tests	
FilmArray	Inclusivity	39	39	0	0	 Pass
	Exclusivity	36	0	36	0	
	Blank	18	0	18	0	
RAZOR EX	Inclusivity	39	39	0	0	 Pass
	Exclusivity	54	1	53	1	
	Blank	24	0	24	0	
T-COR 4	Inclusivity	39	39	0	0	 Pass
	Exclusivity	36	0	36	0	
	Blank	17	0	17	0	
	Positive Controls	6	6	0	0	
Bio-Seq PLUS	Inclusivity	39	36	3	3	 Fail 3 false negative inclusivity samples
	Exclusivity	36	0	36	0	
	Blank	16	0	15	1	
POCKIT	Inclusivity	39	39	0	0	 Fail 3 false positive exclusivity samples
	Exclusivity	60	3	57	3	
	Blank	22	0	22	0	
	Positive Controls	12	12	0	0	

Immunoassay Test Results with Environmental Powders



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Instrument	# Agents per Test	Type of Test	# Tests	Result/Comments	# Positives	PASS/ FAIL
Pro Strips (AdVnt)	5-agent	Ba and Ricin	66	All other signatures negative	0	PASS
BADD (AdVnt)	1-agent	Ba and Ricin	66	0 false positives	0	PASS
RAID 5 (Alexeter)	5-agent	Ba and Ricin	66	All other signatures negative	0	PASS
RAID 8 (Alexeter)	5-agent	Ba and Ricin	66	All other signatures negative	0	PASS
BioDetect (Alexeter)	1-agent	Ba	66	2 false positives each for kaolin with visual and reader	Visual: 2 Reader: 2	FAIL FAIL
BioDetect (Alexeter)	1-agent	Ricin	66	0 false positives	Visual: 0 Reader: 0	PASS PASS
NIDS (ANP Technologies)	3-agent	Ba	66	All other signatures negative	0	PASS
NIDS (ANP Technologies)	4-agent	Ricin	66	All other signatures negative	0	PASS
ENVI (Envionics)	1-agent	Ricin	66	All signatures negative	Visual: 0 Reader: 0	PASS PASS
Toxin Screen (GenPrime)	3-agent	Ricin	66	All other signatures negative	0	PASS
SMART II (New Horizons)	1-agent	Ba and Ricin	66	0 false positives	0	PASS
Zephyr (PathSensors)	1-agent	Ba	66	0 false positives	0	PASS
RAMP (Response Biomedical)	1-agent	Ba	66	0 false positives	0	PASS
RAMP (Response Biomedical)	1-agent	Ricin	66	0 false positives	0	PASS
BioThreat Alert (Tetracore)	1-agent	Ba	66	0 false positives	Visual: 0 Reader: 0	PASS PASS
BioThreat Alert (Tetracore)	1-agent	Ricin	66	Multiple invalid results (no control line)	Visual: 0 Reader: 0	PASS PASS

- Tested 22 environmental powders in triplicate (66 tests).
- All immunoassays pass evaluation, except the BioDetect for *Ba*.

Summary of Environmental Powder Results

- ▶ Tested 22 environmental powders in triplicate (66 tests)
- ▶ **General biological tests** (protein, ATP, DNA, FTIR)
 - **As expected, many gave positive results** with organic and protein containing powders (frequency of detection: protein > ATP > DNA)
 - Some inorganic powders also tested positive with the general test kits
 - Important for users to understand test limitations for sample screening
- ▶ Specific assays test criteria: “Pass” meets minimum probability of no detection of 0.95 with 95% confidence (requires 0 or only 1 false positives for the 66 tests)
- ▶ **Immunoassay results: Overall low false positive rate**
 - Most immunoassays had no false positives and “pass”
 - Only the BioDetect had 2 positives each for the reader and visual assessment and therefore “failed”
 - Need to be careful to follow assay instructions closely
- ▶ **PCR results: Overall low false positive rate**
 - Most systems/assays had no false positives and “pass”
 - Only the pXO2 assay on the POCKIT system “failed”
 - One system (T-COR 4) had many control failures



Agent and Toxin Testing of General Tests

Protein, ATP, DNA, and FTIR Tests



Threat Agent	Origin/ Form	Protein Tests					ATP Tests		DNA Test	FTIR
		20/20 BioCheck		TASKit Bioscreener		INDIPRO Strips	Clean-Trace ATP	PROFILE-1 ATP	GenPrime Prime Alert	HazMatID 360
		Prot.	pH	Prot.	Starch					
Ricin	Native castor control mash	+++	NNN	+++	---	+++	+++	+++	---	+++
	Native castor acetone extract	+++	NNN	+++	---	+++	+++	---	---	+++
	Native castor acetone protein precipitate	+++	BBB	+++	---	+++	+++	---	---	+++
<i>Bacillus anthracis</i> Sterne spores	In-house prep	+++	NNN	+++	---	+++	+++	+++	+++	+++

Green: positive test, as desired (all positive tests desired)

- Protein tests and FTIR: all positive for protein, as expected.
- ATP: Clean trace gives all positive; Profile-1 has some negative results for ricin.
- DNA test: negative results for ricin, and some negative results for *Ba*.

Agent and Toxin Testing of PCR Systems

FilmArray, RAZOR EX, POCKIT, T-COR 4, Bio-Seq PLUS

Threat Agent	Origin/Form	FilmArray	RAZOR EX 10-Threat and BA-3	POCKIT	T-COR 4	Bio-Seq PLUS
Ricin	Native castor control mash	+++ Ricin	+++ Ricin	+++++++ Ricin	+++++ [^] Ricin	No Ricin Assay Available
	Native castor acetone extract	+++ Ricin	++N [^] Ricin	+++++++ Ricin	+++ ^{**^} Ricin	No Ricin Assay Available
	Native castor acetone protein precipitate	+++ Ricin	+- Ricin	+++++++ Ricin	+++++++ ^{**^} Ricin	No Ricin Assay Available
	Pure Ricin Preparation	--- Ricin	--- Ricin	--- Ricin	--- Ricin	No Ricin Assay Available
Ba Sterne spores	In-house prep	+++ <i>Ba</i> Chrom +++ <i>Ba</i> pXO1	+++ <i>Ba</i> pXO1 (assay 1) +++ <i>Ba</i> pXO1 (assay 2) -- [^] <i>Ba</i> pXO2	+++++++ <i>Ba</i> pXO1	+++++++ ^{**^} <i>Ba</i> pXO1	-+ <i>Ba</i> pXO1
Ba Ames spores	Collaborator prep/test	+++++ <i>Ba</i> pXO2	+++++ <i>Ba</i> pXO2	+++++ <i>Ba</i> pXO2	+++++ <i>Ba</i> pXO2	+++++ <i>Ba</i> pXO2

N = No result reported; Inhibition control failure: reaction did not go to completion
**1 or more reactions with low fluorescence

[^]1 or more control failures
[^][^]Did not appear to be a true amplicon

Green = True Positive

- Positive PCR tests as expected for crude ricin and *Ba* samples.
- No positive PCR for pure ricin protein (as expected)
- Control/low signal issues with T-COR 4 and some RAZOR EX samples

Summary of Initial Agent and Toxin Data

- ▶ Initial tests in at least triplicate with:
 - *Bacillus anthracis* spores (*Ba* Sterne or *Ba* Ames)
 - Ricin
 - 3 crude ricin preps for all tests
 - Pure ricin toxin tested with immunoassays and PCR only
- ▶ **General screening tests: Results vary with type of test and sample**
 - Protein, FTIR, Clean Trace ATP gave all positive results
 - DNA test was negative for most samples, and Profile-1 ATP was negative for most ricin preps
- ▶ **Immunoassays: All *Ba* and ricin samples positive as expected**
 - Often also have positives for additional signatures for agents not present in the sample
 - Only Pro Strips and RAID 8 tested so far (others in progress)
- ▶ **PCR: All *Ba* and *crude* ricin preps are positive as expected**
 - Pure ricin protein is not detected, as expected
- ▶ Underway: LOD studies and replicates for evaluating ability to achieve 0.95 POD at 95% confidence

Strategies for Instrument Use for Sample Screening (Potential Examples)



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Biodetection Strategy #1

Protein Test → PCR

Biodetection Strategy #2

FTIR → Protein Test → Immunoassay

Biodetection Strategy #3

Protein Test → Immunoassay → PCR

Biodetection Strategy #4

Protein Test → ATP → Immunoassay

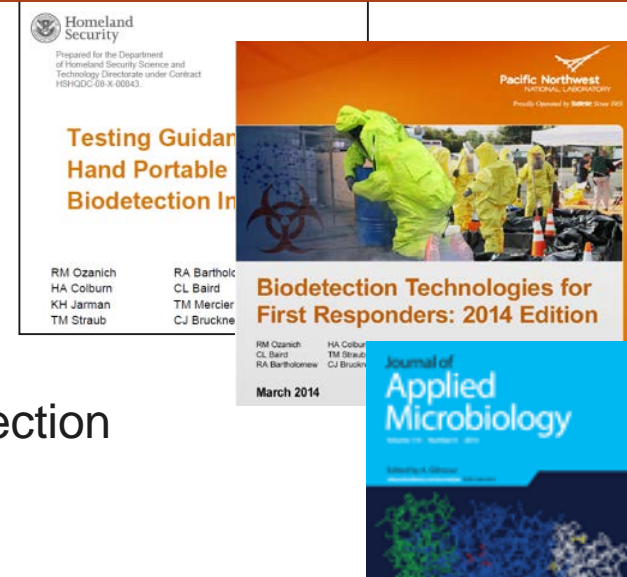
Selected strategy depends on both end user needs and constraints and performance of combined detection platforms

LRN reference lab does confirmatory testing

PNNL Providing Impact

- ▶ PNNL laboratory testing: cost effective assessment of available commercial technologies
 - Establish LOD, dynamic range, potential for false positive/negative results, and impact of other substances such as common hoax powders
 - Identify technology deficiencies/limitations
 - Evaluate strategies for instrument use
 - Provide objective information and guidance for selection and use of biodetection technology
- ▶ Publish and disseminate information
 - Reports, peer-reviewed publications
 - Website, videos, smartphone app, etc.

<http://biodetectionresource.pnnl.gov>



Evaluation of the FilmArray® system for detection of *Bacillus anthracis*, *Francisella tularensis* and *Yersinia pestis*

Volume 114, Issue 4, pages 992–1000, April 2013

Identify & Acquire Technology

Develop Test Plans

Test and Evaluate Technology

Publish Results

Transition to
End Users

It Takes A Village!

▶ **Principle Investigator:** Cindy Bruckner-Lea

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- Terre Mercier
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- Timothy Straub
- Hayley Cardamone
- Pamela Kinsey
- Mary Lancaster



▶ **DHS Program Manager:** Anne Hultgren

- This effort is funded by the Department of Homeland Security Science and Technology Directorate under Contract HSHQDC-08-X-00843.

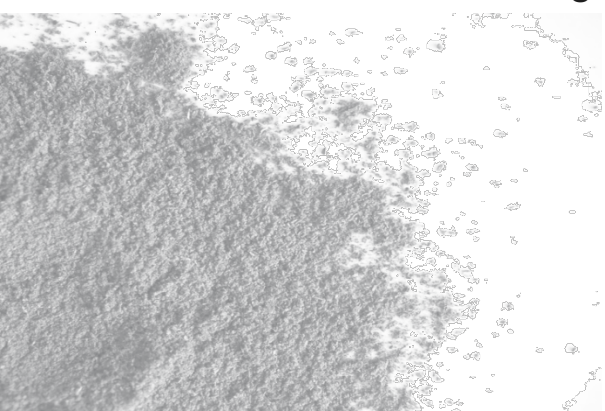
Data Set #2: Screening of Environmental Powders



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- ▶ Goal: understand instrument performance, limitations, and cost/benefit of assays/biodetection platforms
- ▶ Test samples:
 - Environmental material and potentially interfering common powders
 - General indicator tests: 10 mg, except Profile 1 at 1 mg
 - Immunoassays: 1 mg or 0.1 mg/ml
 - PCR: 0.1 mg/ml in assay buffer



Environmental Test Materials	
<i>Bt</i> powder (Dipel)	Cornstarch
Powdered milk	Baking powder
Powdered infant formula	Kaolin
Powdered coffee creamer	Borax
Powdered sugar	Brewer's yeast
Talcum powder	MgSO ₄ (Epsom salt)
White flour	Powdered toothpaste
Baking soda	Popcorn salt
Chalk dust (CaCO ₃)	Acetaminophen
Chalk dust (MgCO ₃)	Instant pectin
Road dust	Miralax

General Biological Indicator Results

Organic Powders



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Class of Powder	Powder Type	Protein Tests				ATP Tests		DNA Test	FTIR	
		20/20 BioCheck		TASKit Bioscreener		INDIPRO Strips	Clean-Trace Surface ATP	PROFILE 1 ATP	Prime Alert	HazMatID 360
		Prot.	pH	Prot.	Starch					
Organic, Biological	Brewers yeast powder	+++	NNN	+++	---	+++	+++	+++	+++	+++
	Dipel dust	---	NNN	---	---	---	---	+++	---	---
Organic, Protein-Containing	Milk powder	+++	AAA	+++	---	+++	+++	+++	---	+++
	Infant formula	+++	AAA	+++	---	+++	---	---	---	---
	White flour	+++	NNN	+++	++I	+++	+++	---	---	+++
Organic, No Protein	Coffee creamer	---	NNN	---	---	+++	---	+-	---	---
	Instant pectin	---	NNN	---	---	++	---	---	---	---
	Acetaminophen	+++	NNN	+++	---	---	---	---	---	---
	Powdered sugar	---	NNN	---	---	---	---	---	---	---
	Corn starch	---	NNN	---	+++	---	---	---	---	---
	MiraLAX (Polyethylene glycol 3300)	---	NNN	---	---	---	---	---	+-	---

- As expected, many biological indicator tests were positive for organic and protein-containing powder (frequency of detection: protein>ATP>DNA)
- Dipel dust (contains ~1% *Bt*) was negative for all screening tests except Profile 1 ATP

General Biological Indicator Results

Inorganic Powders



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Class of Powder	Powder Type	Protein Tests				ATP Tests		DNA Test	FTIR	
		20/20 BioCheck		TASKit Bioscreener		INDIPRO Strips	Clean-Trace ATP	PROFILE 1 ATP	Prime Alert	HazMatID 360
		Prot.	pH	Prot.	Starch					
Inorganic	Toothpaste powder	---	BBB	---	---	+++	---	++	---	---
	Baking powder	---	BBB	---	---	+++	++	---	---	---
	Antacid	---	NBB	---	---	+++	---	---	---	---
	Baking soda	---	BBB	---	---	+++	---	---	---	---
	Epsom salt	++	AAN	---	---	---	---	+	---	---
	Gym chalk	++	BBB	---	---	+++	---	++	---	---
	Borax	---	BBB	---	---	---	---	---	---	---
	Talc	---	NNN	---	---	---	---	+	---	---
	Road dust	---	NNN	---	---	---	---	+	---	---
	Kaolin	---	NNN	---	---	---	---	---	---	---
Salt	---	NNN	---	---	---	---	---	---	---	

- 20/20 and INDIPRO protein tests and the ATP tests gave some positive results
- Bioscreener protein test, GenPrime DNA test, and HazMatID FTIR gave no positives

PCR Test Results with Environmental Powders



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Instrument	Type of Test	# Tests	Result/Comments	# False Positives	PASS/ FAIL
FilmArray (17-agent)	<i>Ba</i> (pXO1, pXO2, chrom) and ricin	66	2 false positives for other signatures	0	PASS
RAZOR EX BA-3 (<i>Ba</i> only, 3 signatures)	<i>Ba</i> (two pXO1, pXO2)	66	3 “no result” due to control failures	0	PASS
RAZOR EX 10-Threat (10- threat agents)	<i>Ba</i> (pXO2) and ricin	66	All other threat signatures negative	0	PASS
T-COR 4	<i>Ba</i> pXO1	66	6 control failures, but all sample results negative	0	PASS
T-COR 4	<i>Ba</i> pXO2	66	6 control failures, but all sample results negative	0	PASS
T-COR 4	Ricin	66	6 control failures, but all sample results negative	0	PASS
BioSeeq PLUS	<i>Ba</i> pXO1	66	3 indeterminate sample results	0	PASS
BioSeeq PLUS	<i>Ba</i> pXO2	66	1 false positive for Miralax (PEG); 2 indeterminate results	1	PASS
POCKIT	<i>Ba</i> pXO1	66	No false positives	0	PASS
POCKIT	<i>Ba</i> pXO2	66	15 false positives; 1 indeterminate result	20	FAIL
POCKIT	Ricin	66	1 false positive for coffee creamer	1	PASS

- **FilmArray, RAZOR EX, and Bio-Seeq PLUS: pass exclusivity powder testing**
- **T-COR 4: passes exclusivity powder testing, but many system control failures**
- **POCKIT: pXO1 and ricin assays pass testing, pXO2 assay fails testing**

Agent and Toxin Testing of Immunoassays

ProStrips and RAID 8

Threat Agent	Origin/Form	Pro Strips (Ricin)	Pro Strips (Ba)	Pro Strips (Other Signatures)	RAID 8 (Ricin)	RAID 8 (Ba)	RAID 8 (Other Signatures)
Ricin	Native castor control mash	+++	+++	+++	+++	---	---
	Native castor acetone extract	+++	+++	+++	+++	---	+++
	Native castor acetone protein precipitate	+++	+++	++-	+++	---	+++
	Pure Ricin Preparation	+++	+++	---	+++	---	---
<i>Bacillus anthracis</i> Sterne spores	In-house prep	---	+++	---	---	+-	+-

Green = True Positive

Red = False Positive

White = True Negative

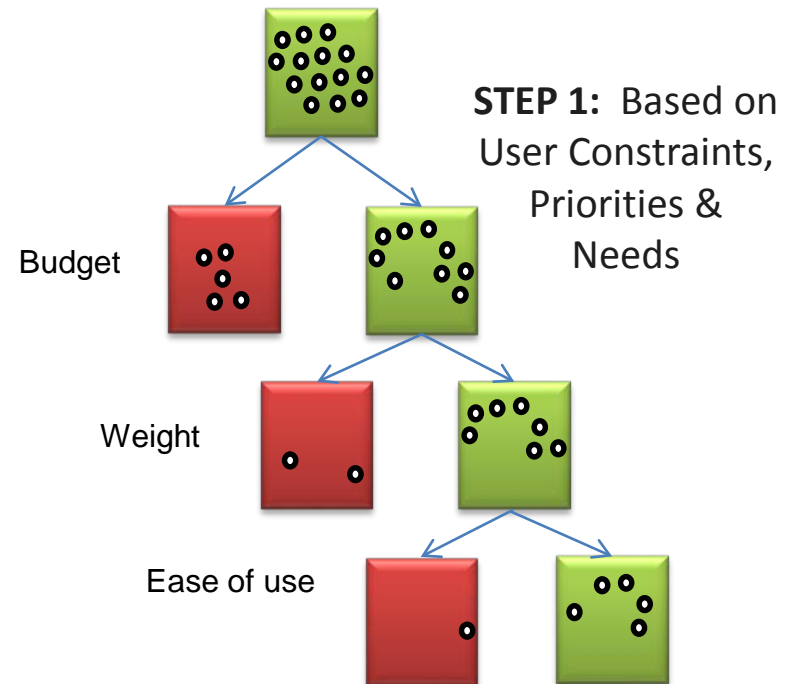
- Pro Strips: *Ba* positive for *Ba* sample, as expected; ricin preps (not pure ricin) show positive for ricin and other signatures in addition to ricin
- RAID 8: Ricin and *Ba* positives as expected; also additional positive for signatures

Strategies for Instrument Selection

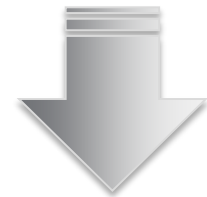
- ▶ Assess limitations, ability to achieve probability of detection goal, cost/benefit
- ▶ Step 1: Instruments are down-selected from pool based on user-specific needs
- ▶ Step 2: Performance comparison/score calculation (per instrument/target) using test results

- Create a score based on probabilities of 3 incorrect decisions:
 - P(FN)=probability of a false negative
 - P(FP)=probability of a false positive
 - P(NP)=probability of a nuisance alarm
- Weight these probabilities by:
 - P(TP)=probability of a true positive
 - P(TN)=probability of a true negative=1-P(TP)

■ Score=
$$P(\text{FN}) * P(\text{TP}) + [P(\text{FP}) + P(\text{NP})] * P(\text{TN})$$



STEP 2: Evaluation of Instrument Performance



Score for performance comparison between platforms