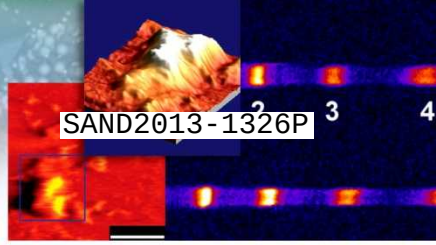
SANDIA BIOSCIENCE



Sandia's Bioscience Program

Jim Carney
Manager
Bioenergy and Defense Technologies

U.S. DEPARTMENT OF ENERGY

Sandia National Laboratories is a multi-program laboratory managed and operated by Sandia Corporation, a wholly owned subsidiary of Lockheed Martin Corporation, for the U.S. Department of Energy's National Nuclear Security Administration under contract DE-AC04-94AL85000.

NNSA

Sandia's bioscience program exists to address the Nation's biodefense and biofuels needs

"Developing the next generation of biofuels is key to our effort to end our dependence on foreign oil and address the climate crisis – while creating millions of new jobs that can't be outsourced."

"...terrorists are more likely to be able to obtain and use a biological weapon than a nuclear weapon…The US government needs to move aggressively to limit the proliferation of biological weapons and reduce the prospect of a bioterror attack."

Our goal is to provide actionable biological information and solutions required to meet National Security challenges in biodefense and biofuels production.

SANDIA BIOSCIENCE

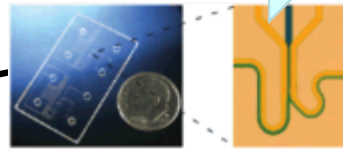
We have extensive experience with developing μbioanalytical systems

1996 → 2012

- **Infectious disease diagnostics**
- **Environmental monitoring**
- **Early threat detection**
- **Toxin diagnostics**

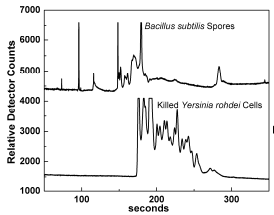
DHS CBNP
- Sample Prep
- Viruses

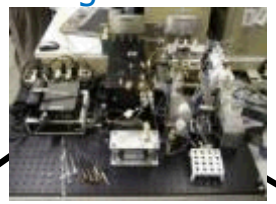
NIAID RapiDx Toxin Diagnostics



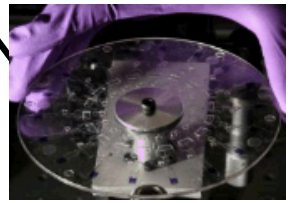
NIDCR Human Oral Microbiome

DoD TTP
- Sample prep
- Bacteria/spores





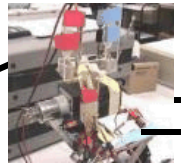
DHS BioBriefcase



SpinDx

SNL LDRD
Combined Gas-phase Chemical agent and Liquid-phase Explosives Detector

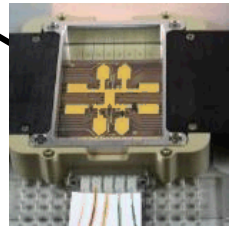
DOE CBNP
- Separated Gas/Liquid
- Liquid-phase Biotoxins



Waters CRADA
- EK-HPLC

Tenix CRADA UWS sensor

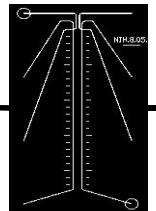
DoD DTO.50
- Autonomous operation
- Bio-aerosols



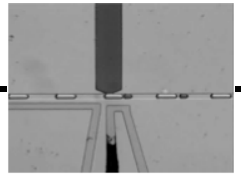
Automated Molecular Biology

- **Oligosaccharide analysis**
- **Enzyme screening**
- **Synthesis and screening of large combinatorial DNA libraries**

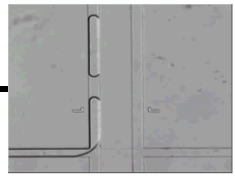
Sugar CE



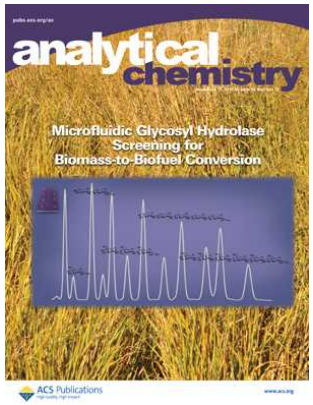
Enzyme Chip

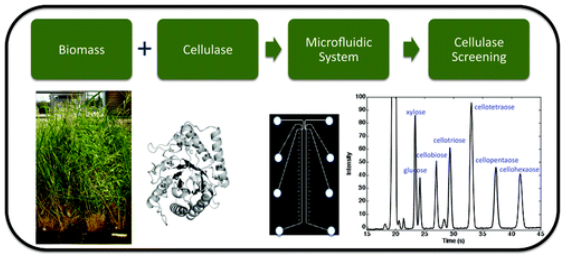


SynBio Chip



Our biologists and engineers work together to design new methods and systems to acquire biological information



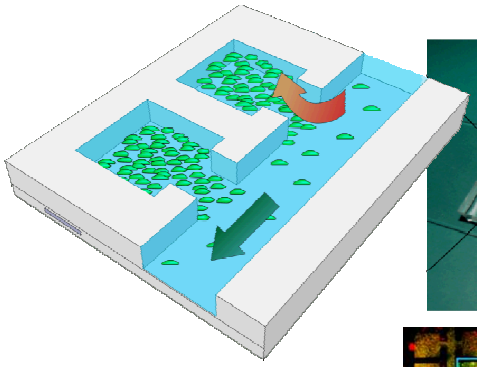


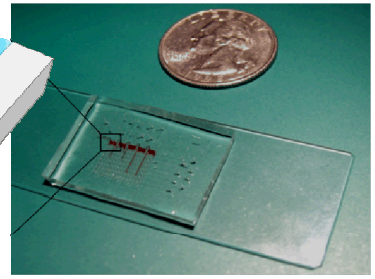
Microfluidic Glycosyl Hydrolase Screening for Biomass-to-Biofuel Conversion

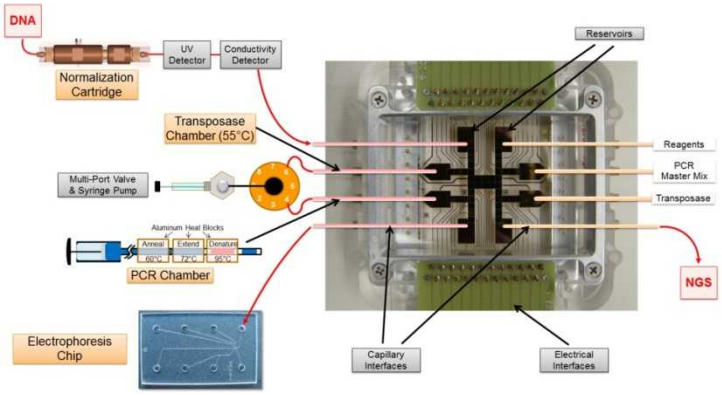
Bharadwaj et al. Anal Chem 2010.

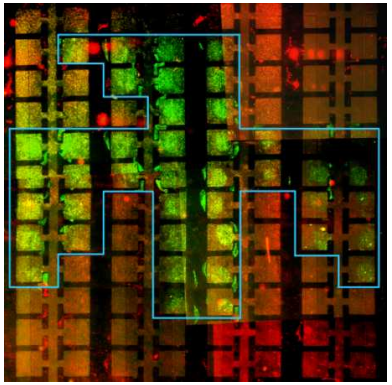
Genome-wide RNAi screening platform for RVFV.

Harmon et al. J Virol 2012.







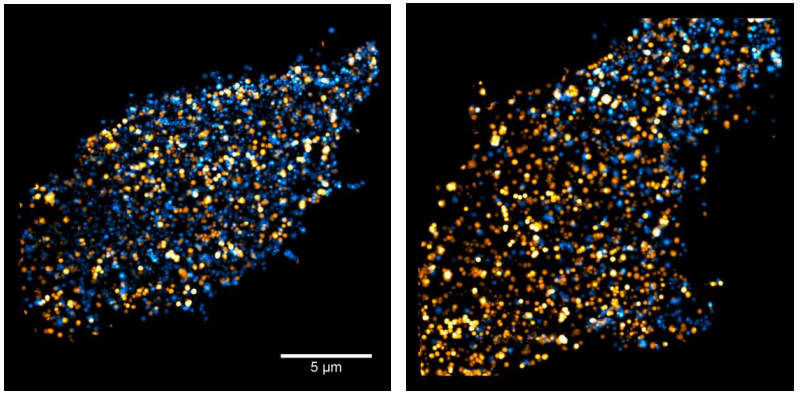


DMF platform for SGS library prep won SLAS 2011 Innovation Award and 2012 R&D 100 award. *Kim et al. JALA 2011.*

We focus our investigations at the appropriate biological scale and at the elements of interest in a sample

Physiological effects of free fatty acid production in genetically engineered *Synechococcus elongatus* PCC 7942.

Ruffing et al. Biotechnol. Bioeng. 2012

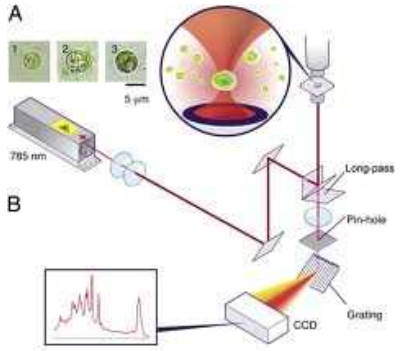


Optical super-resolution reveals pathogen specific TLR4 reorganization.

Aaron et al. Small 2012

In vivo lipidomics using single-cell Raman spectroscopy.

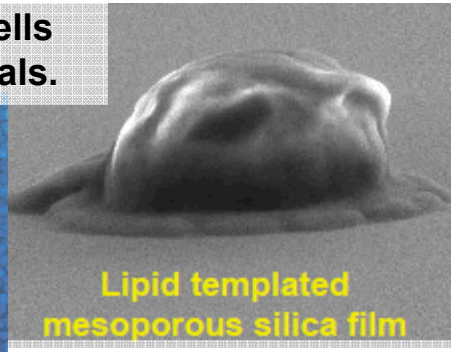
Singh et al. PNAS 2011



Membrane bending by protein–protein crowding.

Stachiowiak et al. Nature Cell Biol 2012

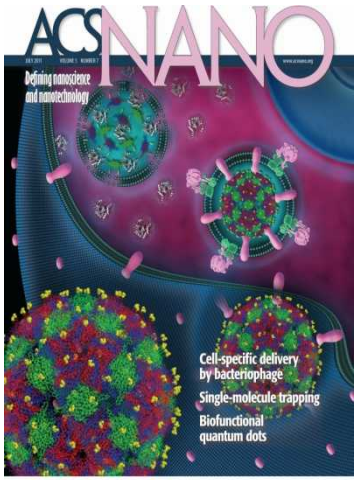
Integrating living cells into 3D nanomaterials.



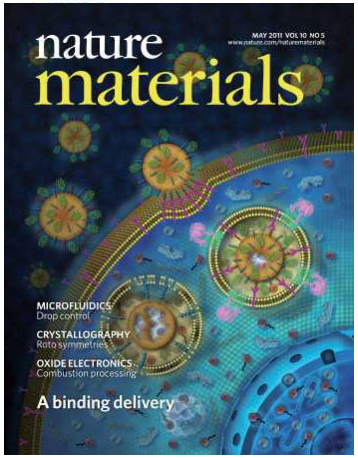
Carnes et al. Nature Chem Biol 2010
Harper et al. ACS Nano 2010
Harper et al. Small 2012

We modify organisms and cells to create biology-inspired solutions

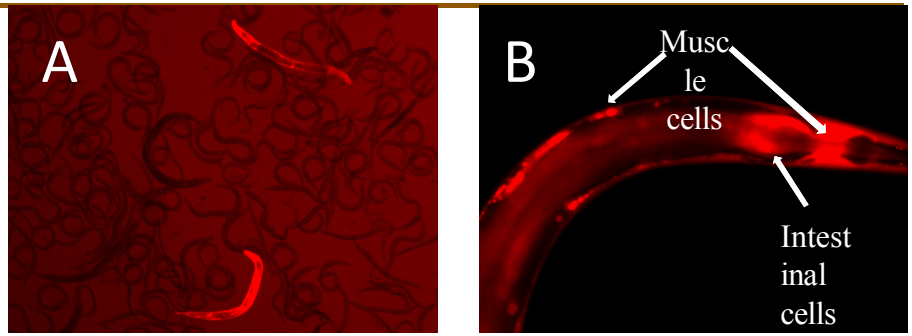
Cell-Specific Delivery of Diverse Cargos by Bio-inspired Engineered Nanoparticles



MS2 virus-like particles and protocells deliver nanoparticles, chemotherapeutic drugs, siRNA to target cells.

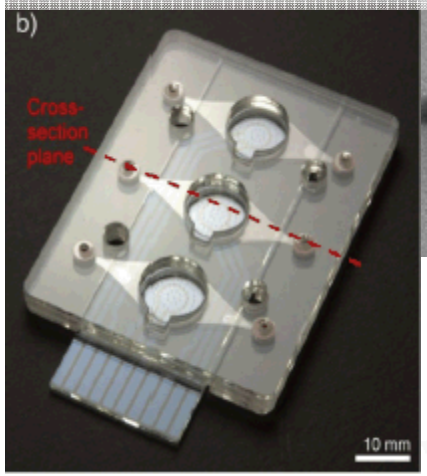


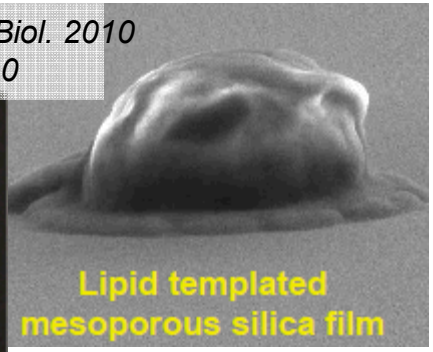
Integrating Living Cells into 3D Nanomaterials. Once sealed within a device, living cells can *remain viable and responsive when stored under ambient conditions for over 2 months*



Clearance testing with *C. elegans.* *C. elegans* infected with a recombinant Vesicular Stomatitis Virus expressing mCherry.

Carnes *et al. Nature Chem. Biol. 2010*
Harper *et al. ACS Nano 2010*





Microfluidic cartridge fabricated from inexpensive plastics via laser ablation allowing rapid prototyping

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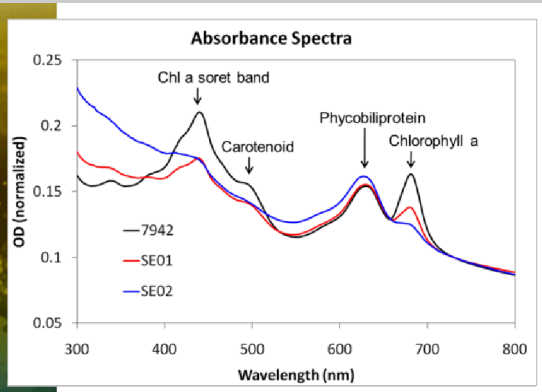
Genetic Engineering of Cyanobacteria for Biodiesel Feedstock

Anne M. Ruffing – Bioenergy & Defense Technologies – aruffin@sandia.gov

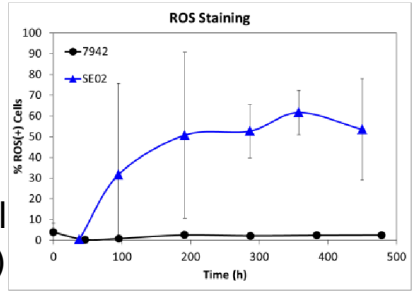
Physiological Effects of FFA Production[1]



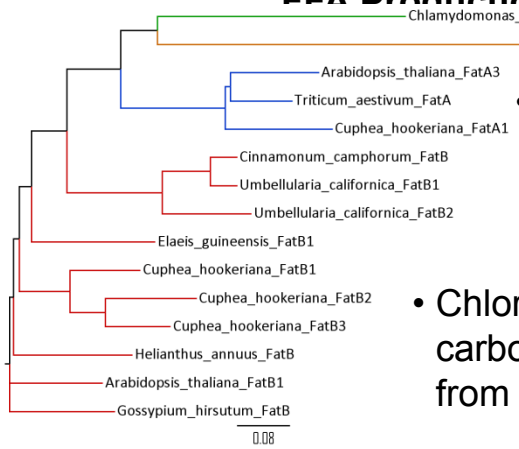
- Reduced cell growth
- Decreased photosynthetic yields
- Chl-*a* degradation
- Aggregation of phycobiliproteins at the cell poles

RNA-seq Analysis to Determine Genetic Responses to FFA Production

- Strains:
  - Wild type (7942)
  - *tesAΔaas* (SE02)
- Time-points:
  - 100h (low FFA, exponential)
  - 240h (high FFA, stationary)

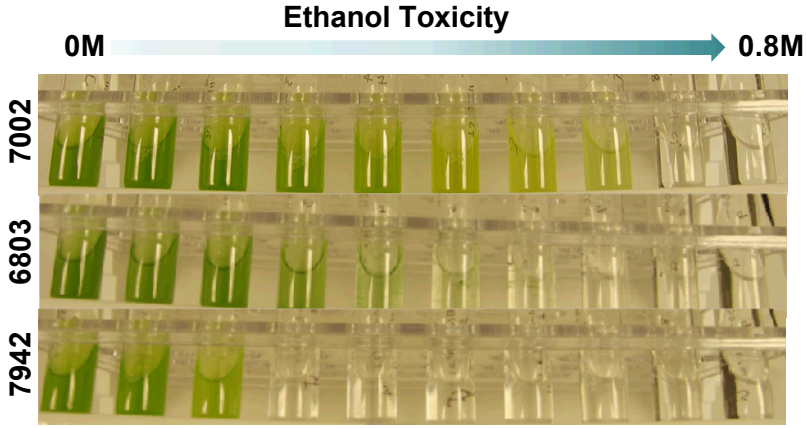


Borrowing Genes from Green Algae to Improve FFA Production[2]



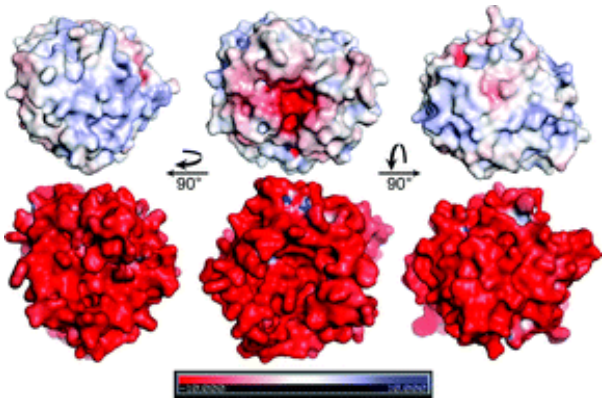
- Acyl-ACP thioesterase (Fat1) from *Chlamydomonas reinhardtii*
- Chloroplastic acetyl-CoA carboxylase (*accBCDA*) from *C. reinhardtii*

Biofuel Toxicity for 3 Model Cyanobacteria



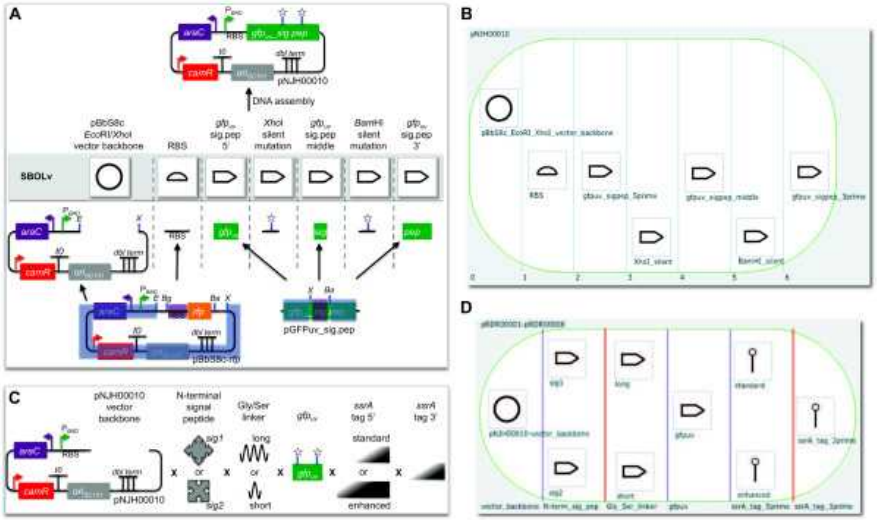
1 Ruffing AM, Jones HDT. Physiological effects of free fatty acid production in genetically engineered *Synechococcus elongatus* PCC7942. *Biotech. & Bioeng.* 2012 100(9):2190-9.

We have developed bioinformatics and computational pipelines to analyze complex data



Identification of a haloalkaliphilic and thermostable cellulase with improved ionic liquid tolerance.
Zhang et. al. Green Chemistry 2011.

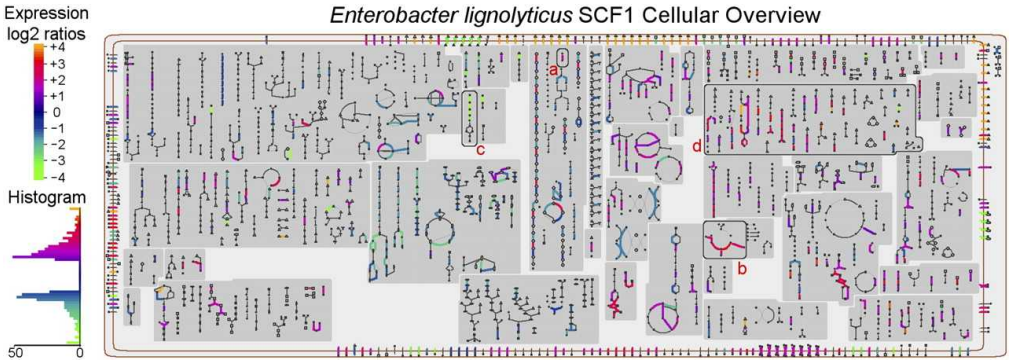
DeviceEditor visual biological CAD canvas
Chen et. al. J. Biol. Eng. 2012



RNAcentral: A vision for an international database of RNA sequences.
Bateman et al. RNA 2011.

Selection of a phylogenetically informative region of the norovirus genome for outbreak linkage.
Verhoef et al. Virus Genes 2012.

Global transcriptome response to ionic liquid by *Enterobacter lignolyticus.*
Khudyakov et. al. PNAS 2012.



Research investments at Sandia come from a variety of sources

NNSA

Industry Partners

NIH

DOE/Office of Science

DOE/Energy

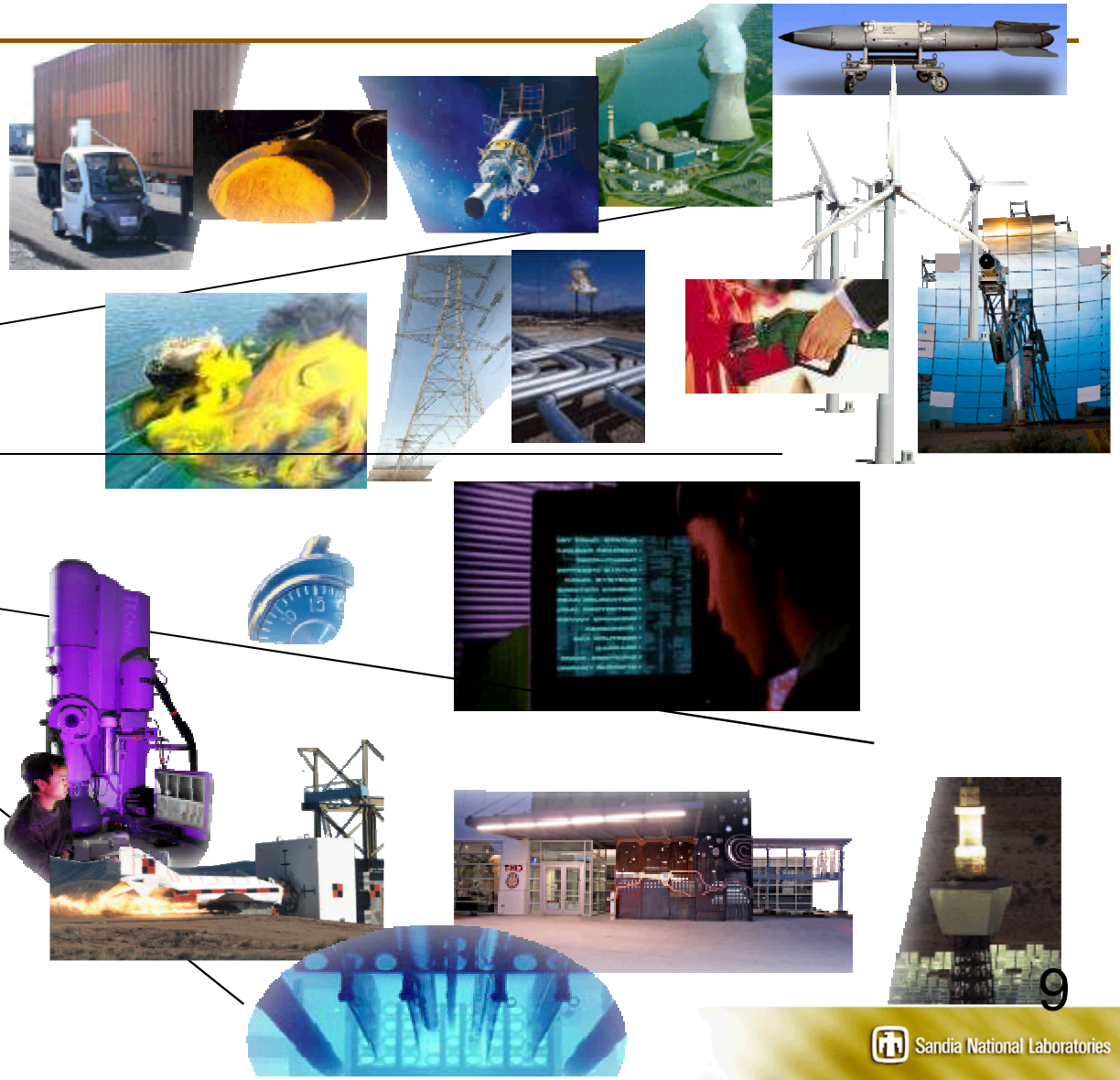
DoD/ Research

DHS

DARPA

Other Government Agencies

LDRD



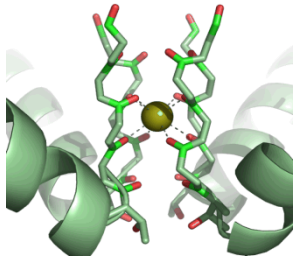
Sandia's NIH projects are at the interface of physical and computational science, engineering, and biology

Computational biology and chemistry

R01 NCRR C-MS3D (Pancerella)
R21 NIGMS Fusogenic Properties of Lipid Bilayers (Stevens)
K25 NHLBI Basis of MTB Latency/ Reactivation (May)
Nanomedicine Development Center (Rempe)

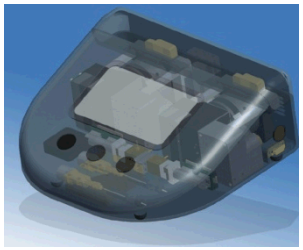
μBioanalytical Platforms

NIDCR U01 Oral Diagnostics (Singh)
NIAID U01 Biotoxin Diagnostics (Singh)
NIDCR R01 Human Oral Microbiome (Singh)



Modeling biological pores (Rempe)

NIGMS P50 Spatiotemporal Modeling Center
(UNM PI: Oliver, SNL PI: Singh)
http://stmc.health.unm.edu



Biotoxin diagnostics (Singh)

Materials science

R21 NIBIB Fluorescent Ceramic Nanoprobes (Boyle)
UNM-led NCI R01 Signaling of IgE Subunits (PI: Oliver, Boyle)
Cancer Nanotechnology Platform Partnership (PI: Brinker)

Imaging

NIH New Innovator Award (Timlin)
NIH RO1 HIV Nef protein structure (led by NE U, Kent)

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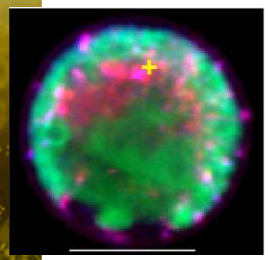
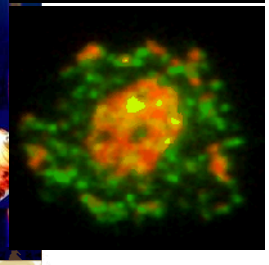
Timlin Lab Research Focus

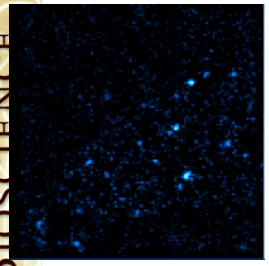
http://bio.sandia.gov/people/timlin.html

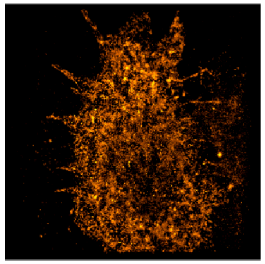


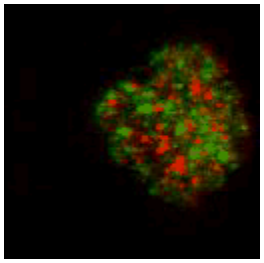


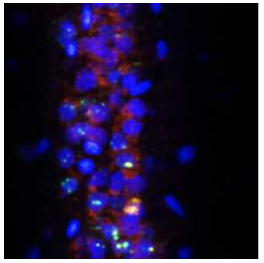
Unraveling Spatial-Temporal Relationships in Complex Multicomponent Biological Systems at Multiple Scales

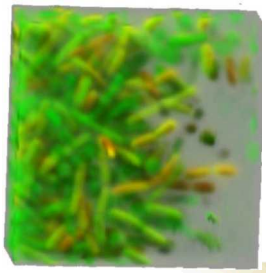
- Advanced spectroscopy
- Innovative imaging technologies
- Chemometric data analysis tools
  - Multidisciplinary
  - Cell biology, immunology, and microbiology
  - Biodefense and Bioenergy











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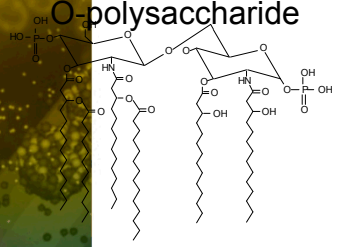
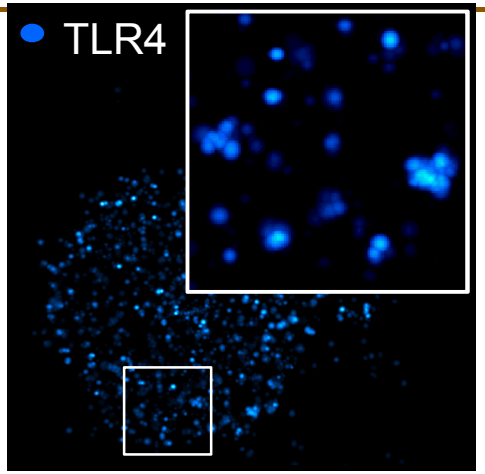
Receptor Cluster Formation in Immune Response

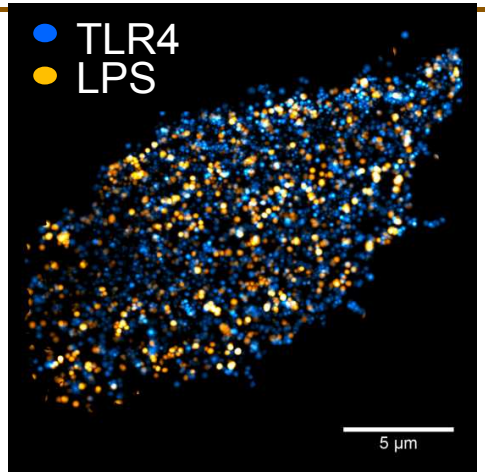
E. coli LPS

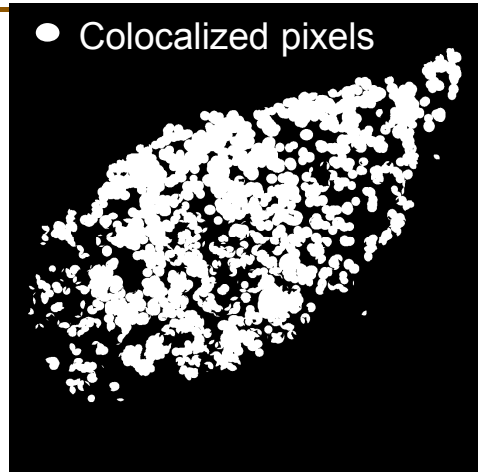
Smooth O-polysaccharide



Bind surface, stimulatory

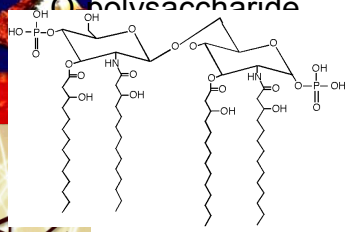




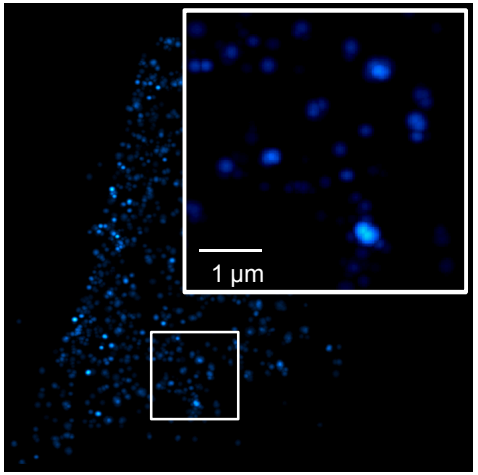


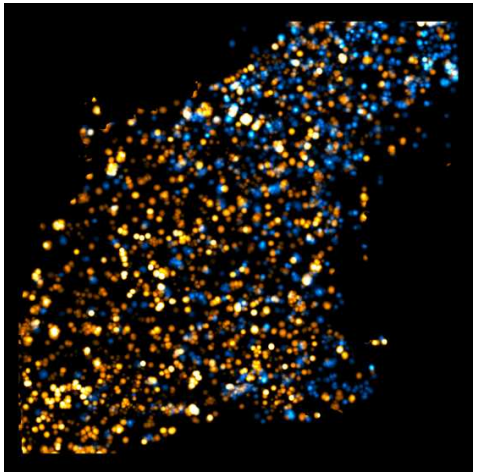
Y. pestis LPS (37 º)

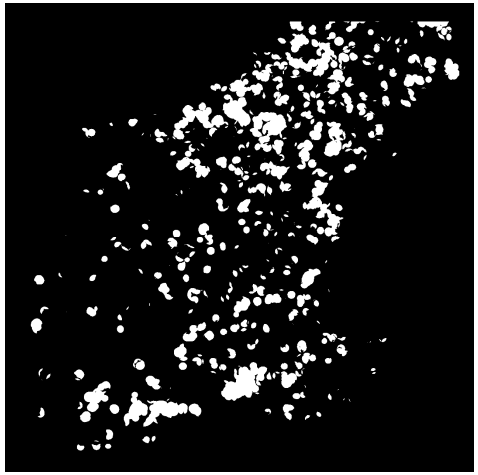
Rough O-polysaccharide



Bind surface, non-stimulatory







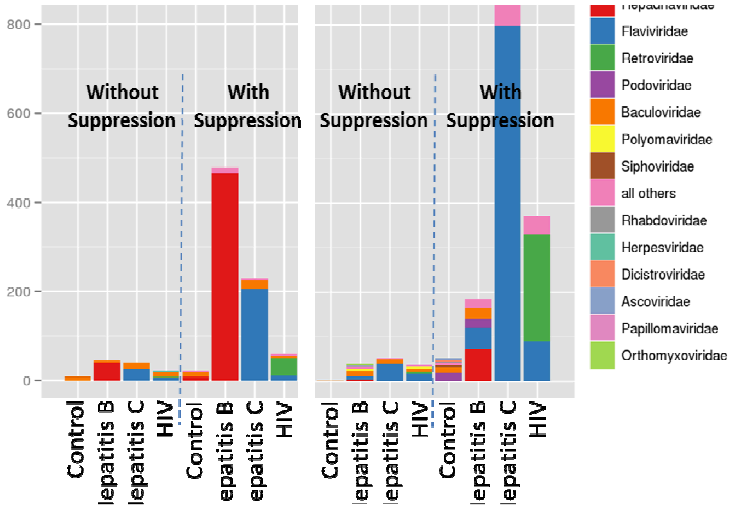
Aaron JS, Carson BD, Timlin JA, Characterization of Differential Toll-Like Receptor Responses below the Optical Diffraction Limit. *Small* 2012; 8(19): 3041-3049.

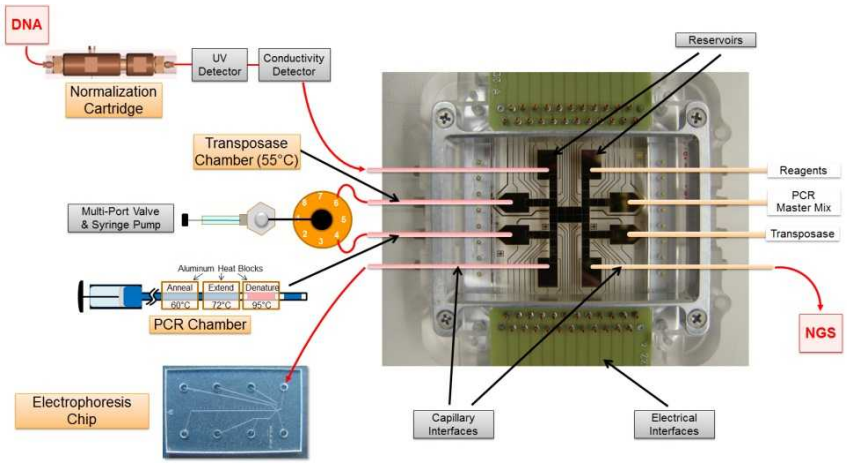
NIH DIRECTOR'S NEW INNOVATOR AWARD

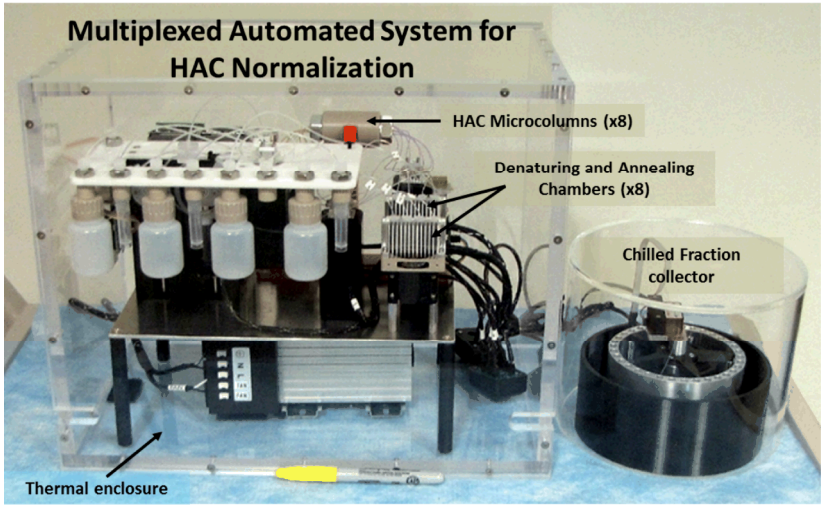
Sandia National Laboratories

Rapid Threat Organism Recognition LDRD Grand Challenge

RapTOR rapidly and automatically applies suppressive molecular biology methods, next generation sequencing and bioinformatics to enable the identification and characterization of unknown pathogens in clinical samples, thereby enabling a faster and more effective public and military health response.







SANDIA BIOSCIENCE

We are transitioning RapTOR technologies and methods to industry, DOE, CDC, DOD

RapTOR

Bioforensics

USACIL-funded Battlefield Automated DNA Analysis and Sampling System ($610K/18 mos)

1. USACIL Phase 2 project ($650K/12 mos)
2. DTRA CX bioforensics project ($1.6M/24 mos)
3. Collaboration with Advanced Liquid Logic (User Agreement signed)
4. IARPA BIC program (approved) ($12.5M/60 mos)

Public Health

DTRA-funded CDC BRRAT Lab transition ($350K/24 mos)

State Dept-sponsored project with Central Public Health Laboratory, Cairo, Egypt (approved) ($300K/12 mos)

SGS

Eureka Genomics CRADA

DeRisi Laboratory (UCSF)

Bioenergy

DOE OBP-funded Algal Pond Crash Forensics (Lane, $800K/24 mos)

ATP3 proposal to DOE ASAP FOA (awarded) ($1.6M to Sandia/60 mos)

~$18M in follow-on funding

SANDIA BIOSCIENCE

SpinDx Overview

- **Rapid:** < 20 min sample-to-answer
- **Inexpensive:** < $500 per instrument, < $2.00 per consumable disk
- **Multiplexed:** Up to 64 parallel assays
- **Ultra-Sensitive:** 1 – 2 orders of magnitude more sensitive than ELISA
- **No Sample Preparation:** Direct analysis of clinical (blood, serum, etc.) or non-clinical (foods, powders, etc.) samples
- **Minimally-Invasive:** 2-µL sample per assay
- **Broad assay menu:** Proteins, nucleic acids, cells

Designed for Biodefense

- **Radiation Biodosimetry**
- **Toxin & Pathogen Diagnostics**

SANDIA BIOSCIENCE

Sandia has extensive experience with developing and fielding CB defense solutions

Detection Systems

BioWatch Gen 2
BioWatch Gen 3
Tenix Water Monitor
RDCDS
μGasAnalyzer
PROTECT
RapTOR

ConOps Development & Support

BioNet
BioWatch Incident Characterization (BWIC)
BioWatch Indoor Reachback Center (BIRC)
PROACT

Decontamination & Restoration

Sandia Decon Foam
DF-200
IBRD
WARRP
Validated Sampling Procedures
BROOM
AWARE

Medical Diagnostics

Toxin Diagnostics
Oral Diagnostics
Radiation Biodosimetry
Rickettsial Diagnostics
NASA astronaut diagnostics

SANDIA BIOSCIENCE

Joint Bioenergy Institute Overview:

JBEI is focused on the efficient conversion of lignocellulosic biomass into fuels

Unified Research & Operations

-$134M, five-year program

-Highly focused research agenda

-Single operation and facility

Six Partners

- Three DOE National Laboratories
- Two Universities
- One Foundation

Berkeley Lab | Sandia National Laboratories | Lawrence Livermore National Laboratory | UC Davis University of California | Carnegie Institution for Science

Four Divisions:

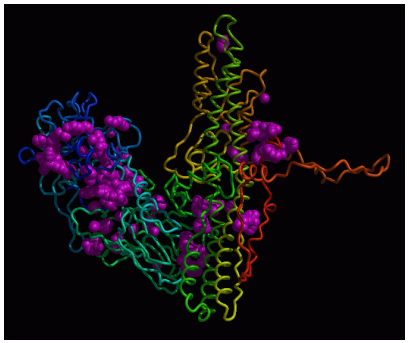
- Feedstocks
- Deconstruction
- Fuels Synthesis
- Cross-cutting Technologies

Industry Partnership Program

- Underpin growth of biofuels industry
- Ensure technology transfer to the developing biofuels industry

Sandia Bioscience

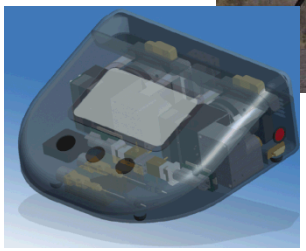
Our objective is the development of biology-enabled solutions …

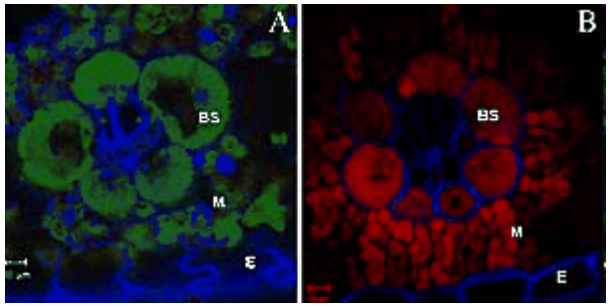


Assays

Biomass deconstruction

Biodetection





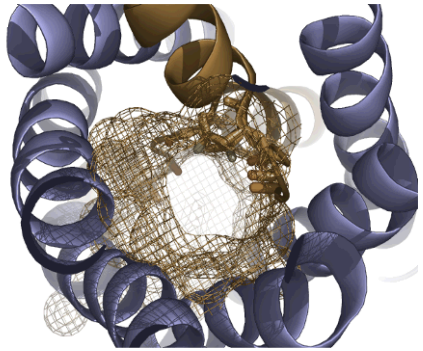
Biomass analysis

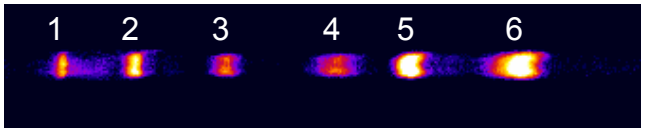
Medical diagnostics

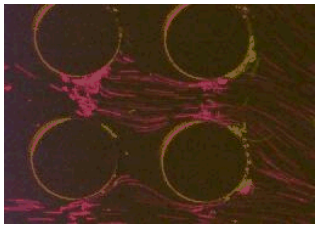
SANDIA BIOSCIENCE

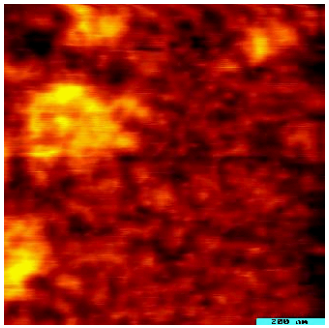
Sandia National Laboratories

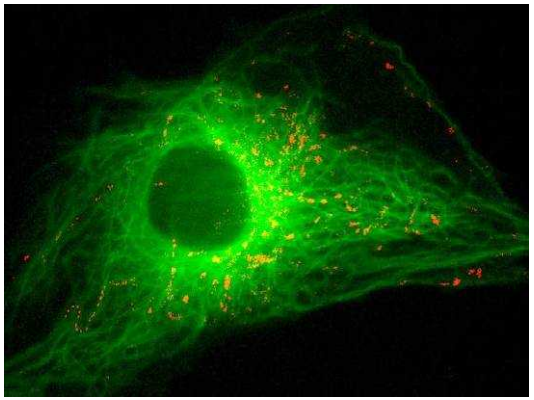
…enabled and differentiated by the underlying science

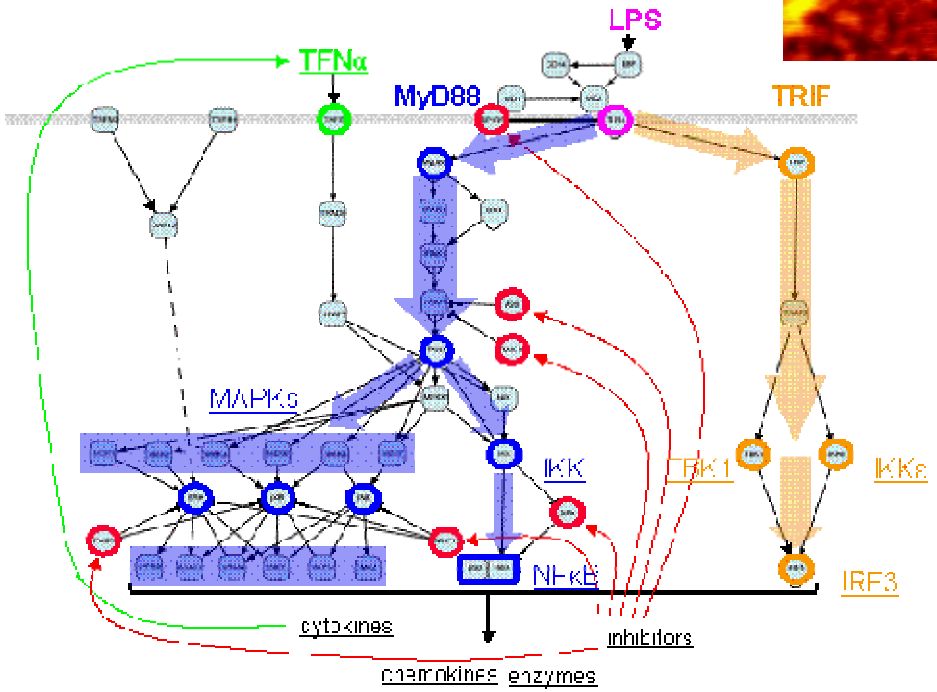


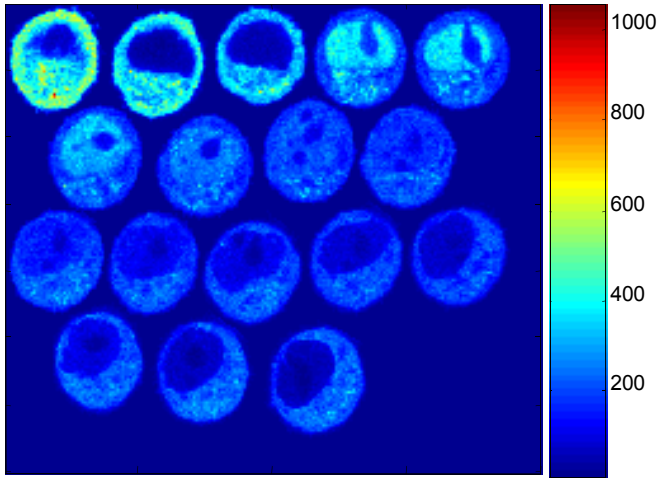












SANDIA BIOSCIENCE

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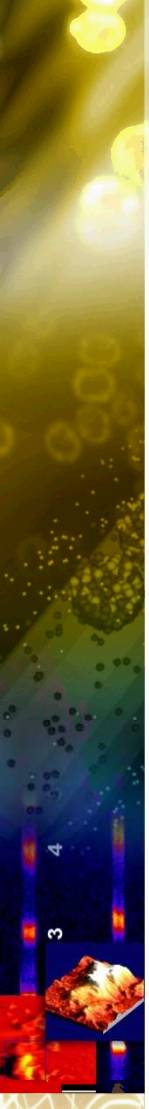
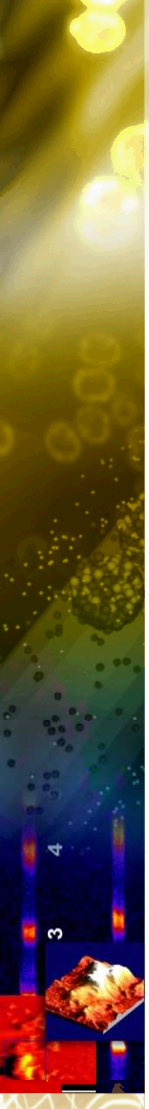


SANDIA BIOSCIENCE

Sandia National Laboratories

We partner with other institutions to achieve our goals



UTMB The University of Texas Medical Branch

UCDAVIS

Massachusetts Institute of Technology

UCSF University of California San Francisco

Lovelace

Statoil

Sapphire Energy

Columbia University

USDA

Berkeley Lab

Carnegie Institution for Science

UNM

BIO-RAD

Cal

Monsanto

New Mexico Department of Health

CDC Centers for Disease Control and Prevention

ASU Arizona State University

Total

Biodefense ⟷ Biofuels

Sandia Bioscience