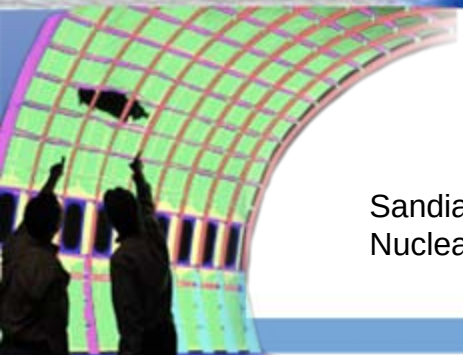
Microfluidic platforms for real-time analysis of clinical and environmental samples

Anup K. Singh, Ph.D

Manager, Biotechnology and Bioengineering Department

Sandia National Laboratories, CA



Sandia is a multiprogram laboratory operated by Sandia Corporation, aNational Nuclear Security Administration under contract DE-AC04-94AL85000.

NCS: Examples of Exposure Areas of Interest

- Exposure to industrial chemicals and byproducts in the air, water, soil and commercial products
- Exposure to natural products in the air, water, soil and commercial products
- Exposure to pharmaceuticals used for therapy and in the environment
- Radiation exposure
- Effects of proximity to manufacturing, transportation and processing facilities

Adapted from Dr. Hirshfeld's talk

THE NATIONAL CHILDREN'S STUDY
HEALTH GROWTH ENVIRONMENT

NCS: Examples of Outcome Areas of Interest

- Interpersonal relationships and bonding
- Inflammatory processes including allergies, asthma and infections
- Genetic and epigenetic status
- Epilepsy and other neurologic disorders
- Cardiovascular screening and function
- Childhood cancer
- Multidisciplinary multidimensional aspects of sensory input, learning and behavior
- Precursors and early signs of chronic diseases such as obesity, asthma, hypertension and diabetes

Adapted from Dr. Hirshfeld's talk

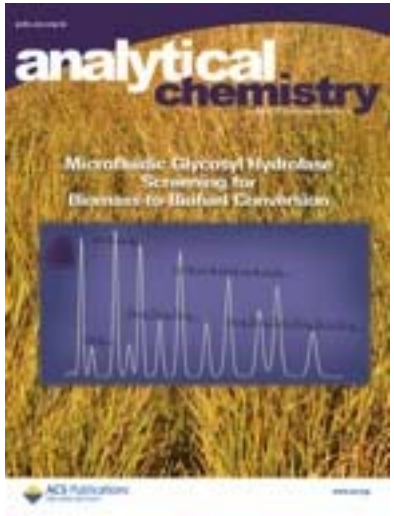
THE NATIONAL CHILDREN'S STUDY
HEALTH GROWTH ENVIRONMENT

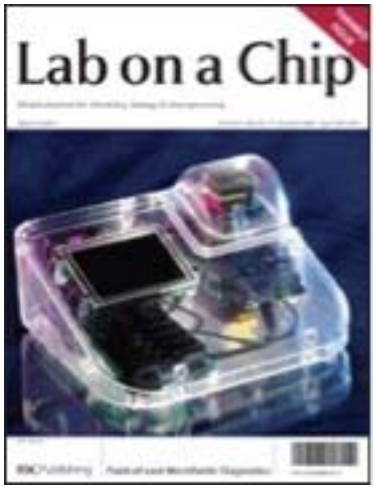
Overview

- NCS is a data-driven study and generation of robust, quantitative data requires sensitive analytical platforms. I will discuss Sandia technologies for two applications:
  - Field-deployable human biomonitoring
  - Field-deployable environmental sensing

Sandia has a long history of developing microscale analytical systems

A number of methods have been adapted to microchips at Sandia:

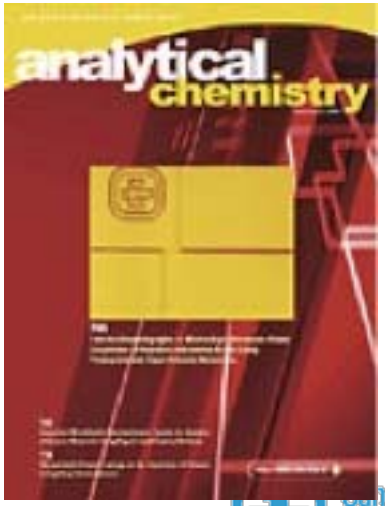




Separation Methods:
Gas chromatography
Liquid Chromatography
Electrophoresis

Others:
Immunoassays
PCR

For applications including:
Environmental biodetection
Chemical Agent detection
Medical diagnostics
Water monitoring



Sandia National Laboratories

SpinDx: A Microfluidic Device for Medical Diagnostics

Team: Anup Singh, Greg Sommer, Ulrich Schaff, Chung-Yan Koh, Matt Piccini, Ron Renzi

FUNDING:
NIDCR
NIAID
Sandia LDRD Program

Sandia National Laboratories

State-of-the-art for environmental sensing and biomonitoring

- **Broad-spectrum detection of chemicals by GC-MS**
  - Sensitive but expensive, limited to specialized labs
- **Monitoring of individual chemicals by EPA methods**
  - Separate test required for each chemical
  - Tests are expensive ($ 20- >100 per assay) and not suitable for field applications
- **Monitoring of a very limited set of biomarkers of exposure can be done by central lab-based tests (e.g., blood panel)**
  - Limited multiplexing
  - Expensive, slow and not field deployable

Next generation clinical diagnostics require new functionality

Centralized laboratory

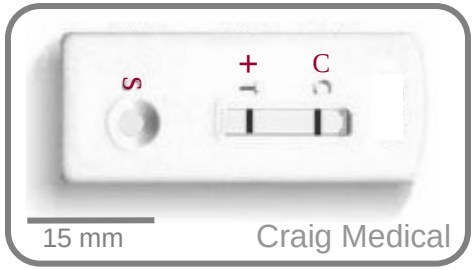
Large instruments requiring certified operators

60 – 120 minute assay times

Quantitation of biomarker content

Gold-standard methods

Point-of-care: home use



Simple to use

3 – 20 minute assay times

Qualitative binary output

Limited relevance to complex diseases

Next generation clinical diagnostics require new functionality

Centralized laboratory

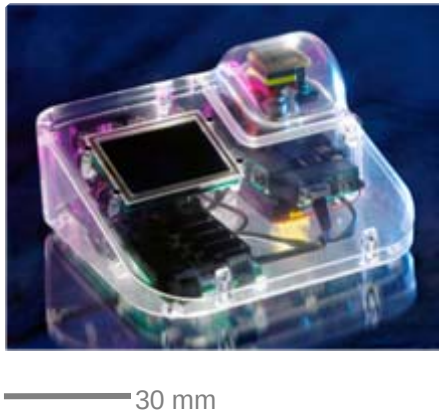
Large instruments requiring certified operators

60 – 120 minute assay times

Quantitation of biomarker content

Gold-standard methods

Clinician's office



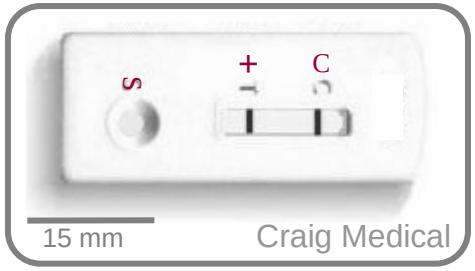
Automated (integrated sample preparation)

5 – 30 minute assay times

Quantitation of biomarker content

Multi-analyte panels relevant to complex diseases

Point-of-care: home use



Simple to use

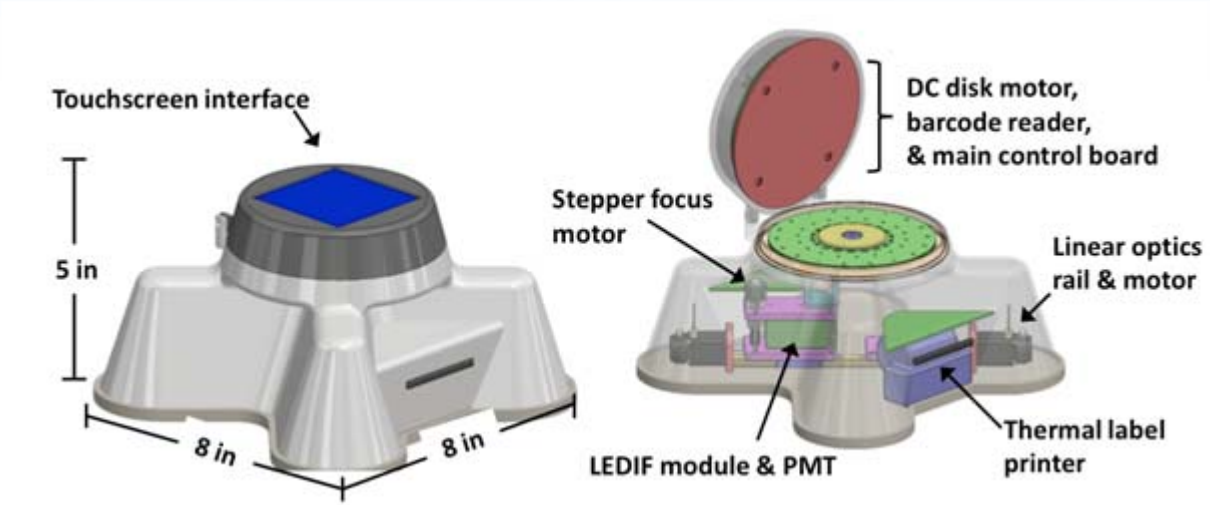
3 – 20 minute assay times

Qualitative binary output

Limited relevance to complex diseases

Sandia National Laboratories

SpinDx



- Bead-based immunoassays in a centrifugal disc
- Up to 36 immunoassays in one disc
- ~15 min assay time per disc
- LOD ~10fM, better than most immunoassay platforms
- No off-device sample prep needed for whole blood, serum, water, food (peanut butter, milk, …)
- Less than cents per assay

Sample-to-Answer Immunoassay in a Disc

1. Metered whole blood input

2. Transfer to mixing chamber

3. Mix with Ab'-coated beads and labeled Ab'' (magnetic stir bar)

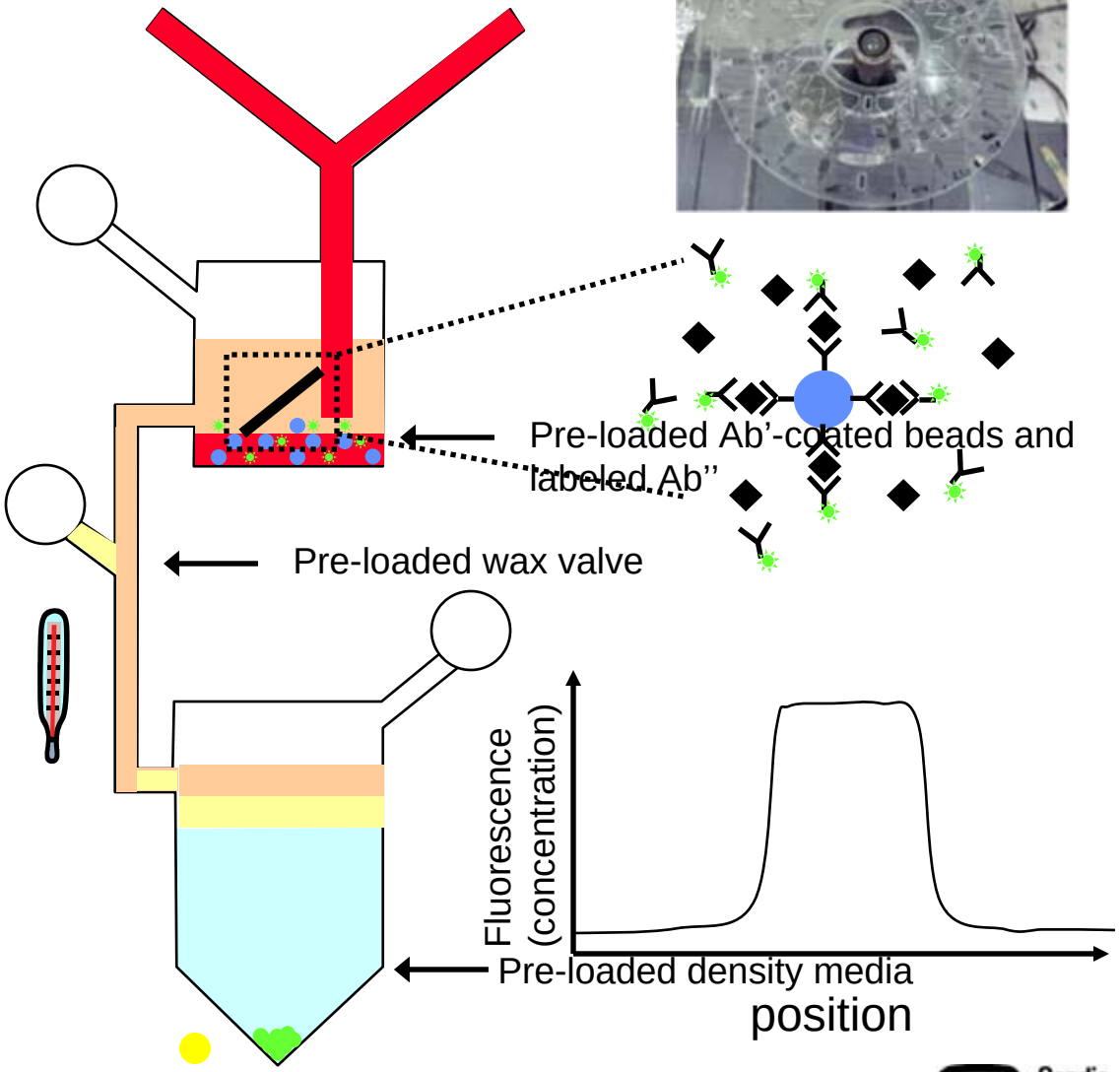
4. Pellet RBC's from plasma

5. Heat, melt wax valve

6. Transfer plasma and beads to separation column

7. Wash and pellet beads from plasma

8. Scan with laser



Sandia National Laboratories

SpinDx– Field testing & FDA approval plan

- Device is at Concept Demo stage
- Prototypes built at Sandia; discs are produced by a company
- Testing with animal model samples already in progress
- Plan to deploy devices at multiple external sites to field-test
- Working with a commercial partner to develop plan for 510(k) filing

Sandia National Laboratories

SpinDx for Human Biomonitoring

- Develop assays for multiple targets (up to 32) in one disc– e.g., PCBs, dioxin, PAH, pesticides, antibiotic residues
- Assays will be validated against EPA methods (e.g, 4000 series)
- Build 10-30 prototype SpinDx devices to be tested at NCS sites

A GCxGC for Fieldable Gas Analyzer

R.J. Simonson
P.C. Galambos, J.W. Lantz, Douglas Read, M.W. Moorman, A.W. Staton
Sandia National Laboratories

J.J. Whiting

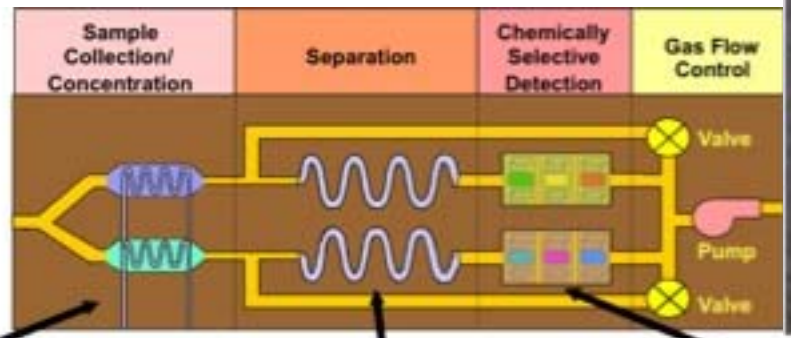
3 Degrees of Separation
Advanced Analytical Technologies

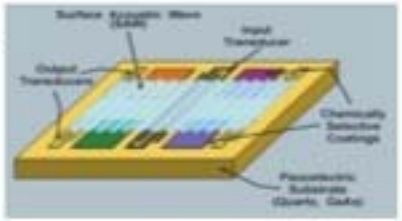
- Collaborators:
  - Prof. Michael Roukes and Dr. Edward Myers (Caltech)
  - Patrick Lewis and Dr. Doug Adkins: Defiant Technologies
  - Darin Graf: Gentech Concepts
  - Philippe Andreucci, Pierre Puget, Eric Colinet: CEA-LETI

- Funding: DARPA, DTRA

Sandia National Laboratories

Sandia's Handheld Gas Analyzer

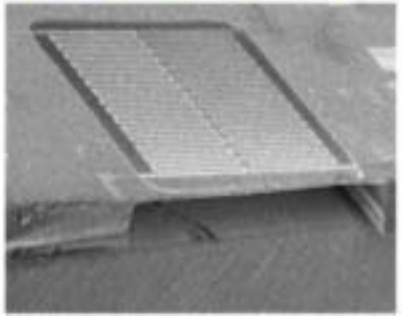


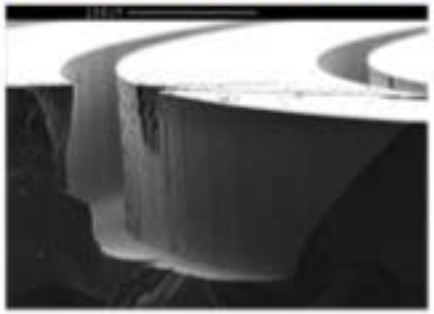


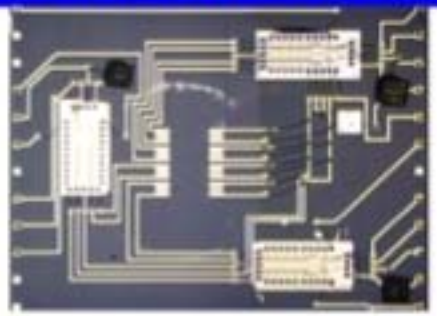
Preconcentrator accumulates species of interest

Gas Chromatograph separates species in time

Acoustic Sensors provide sensitive detection







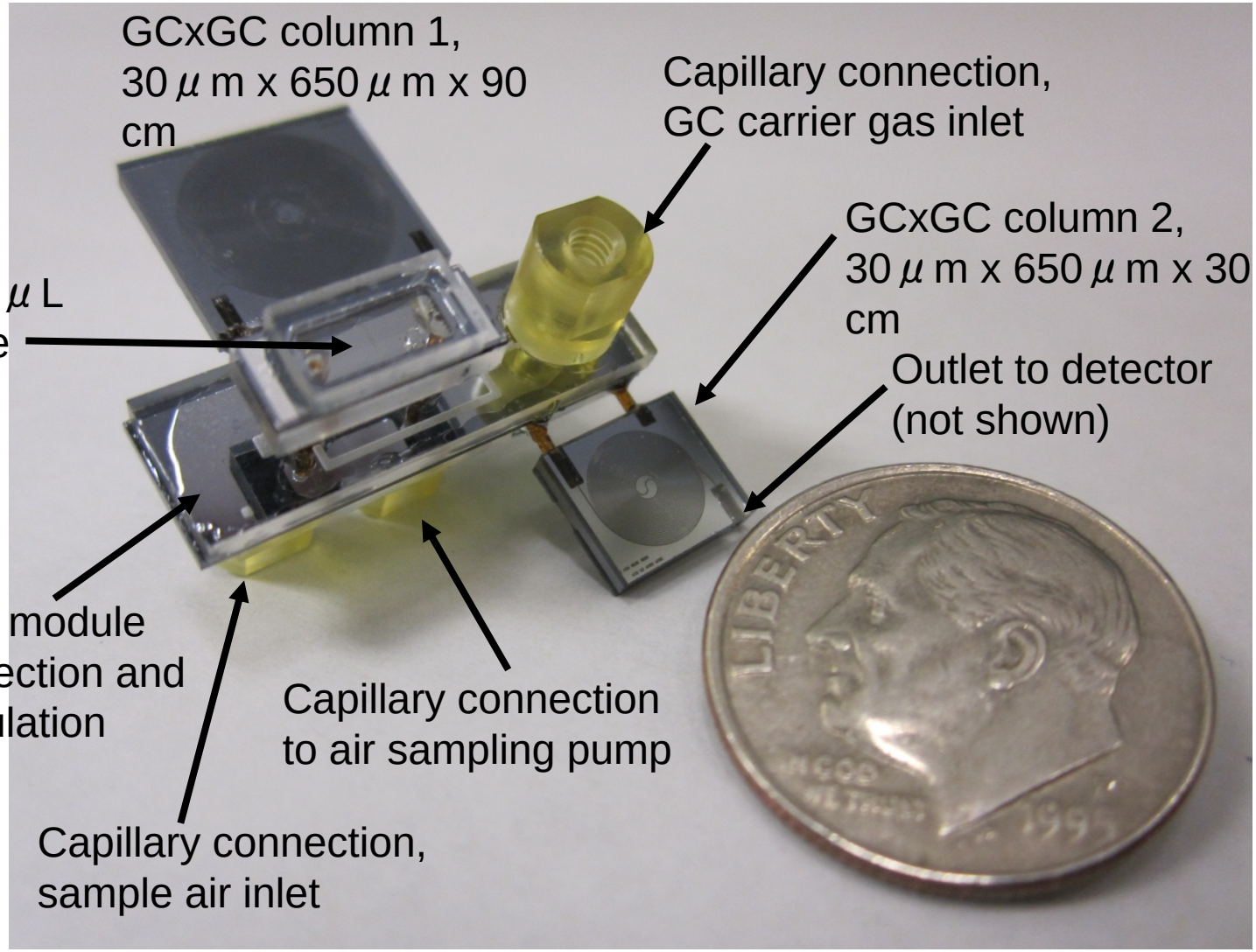
1. Trap mixed vapors

2. Separate by GC

3. Quantify eluents

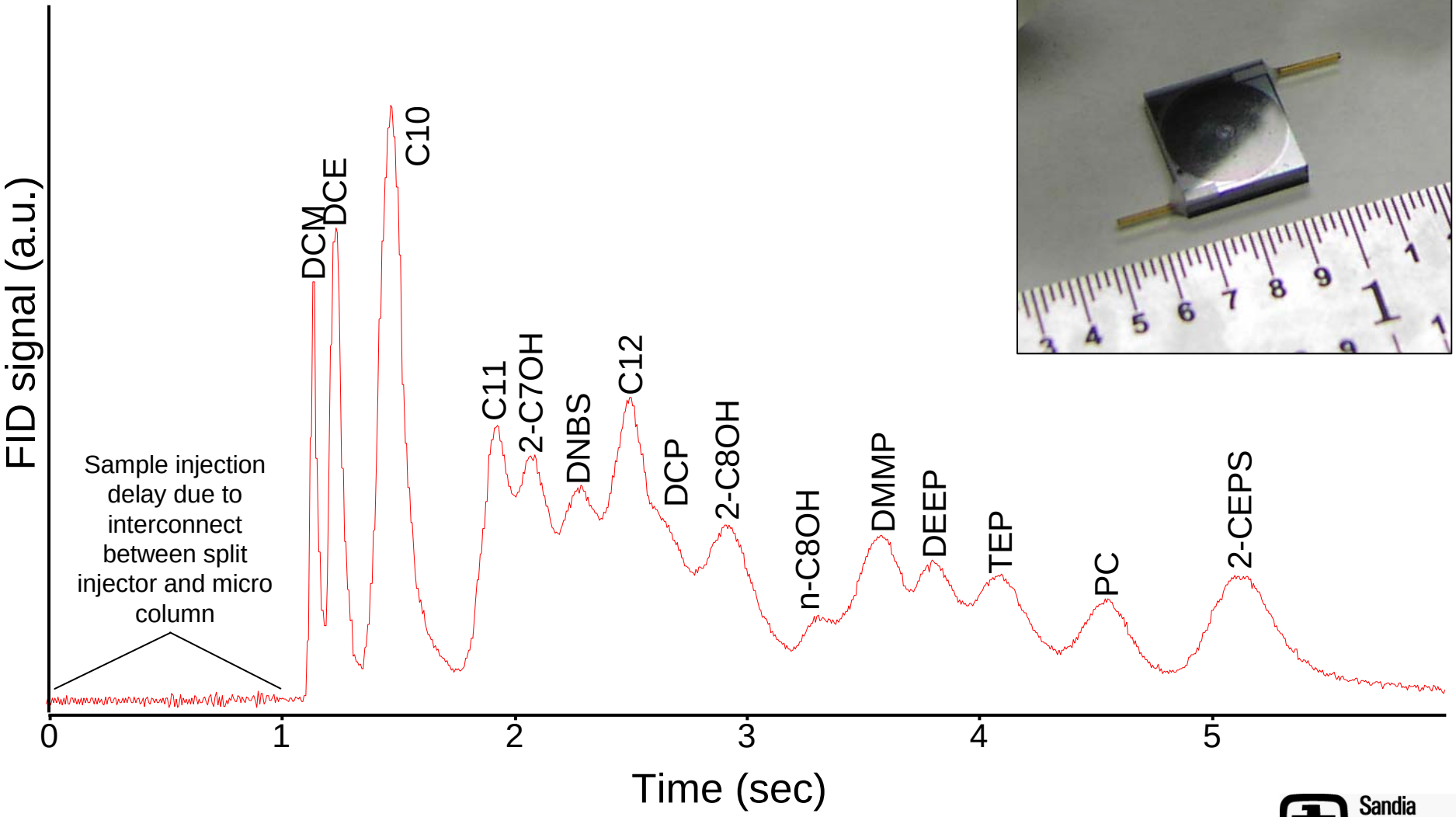
Sandia National Laboratories

Microfabricated GCxGC: What We're Building



Sandia National Laboratories

Fast 1D GC Separation

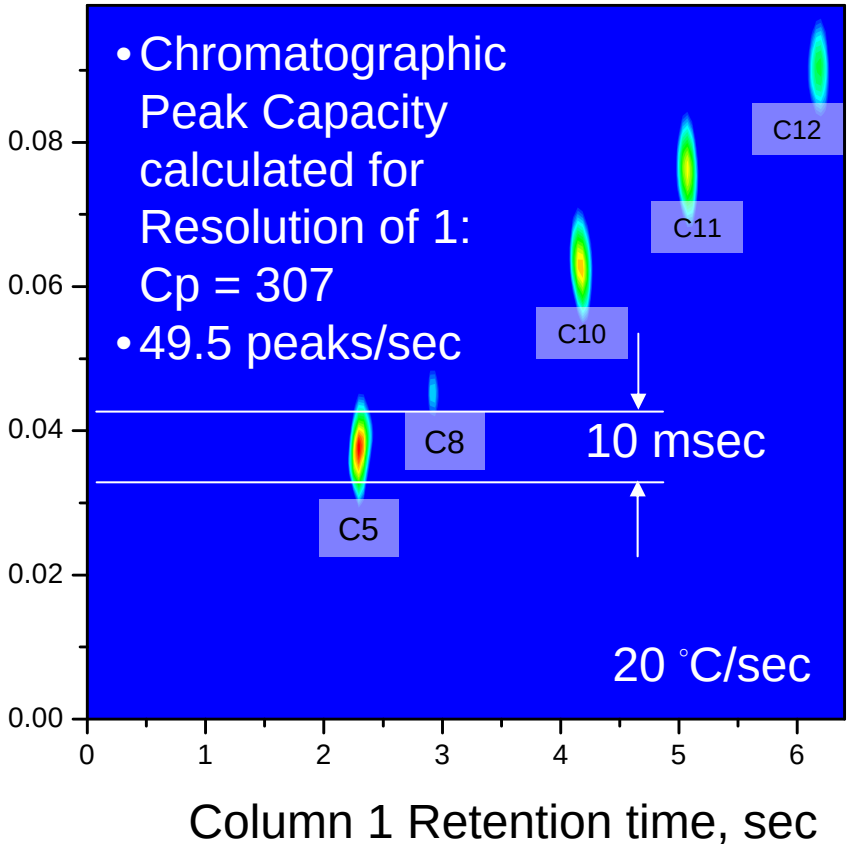


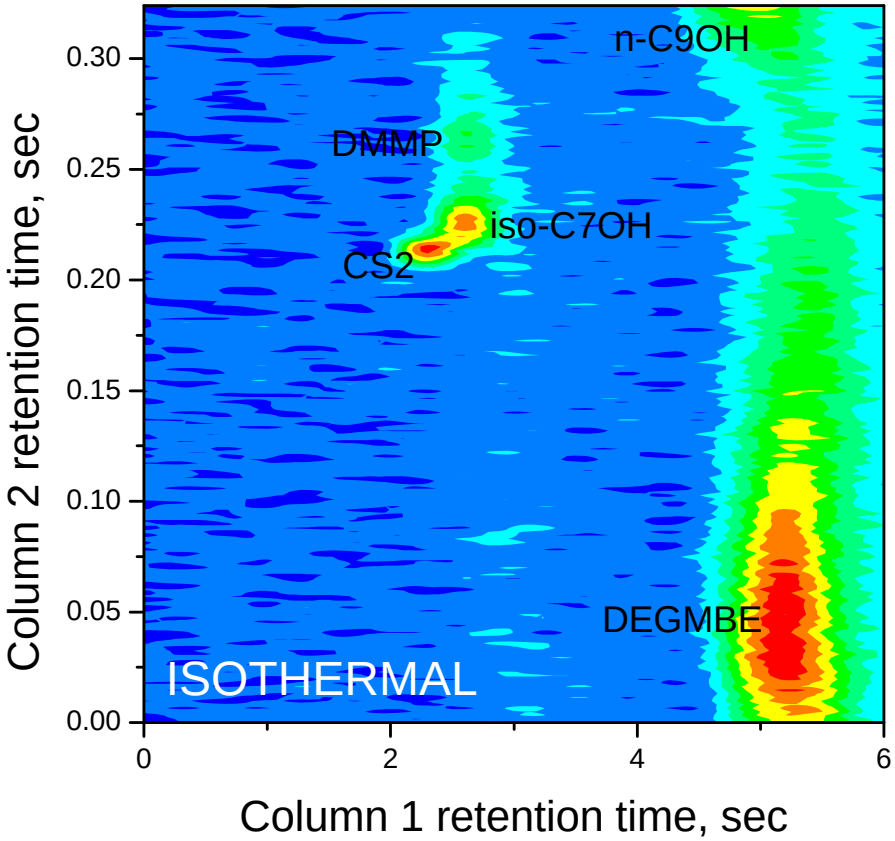
Sandia National Laboratories

GCxGC Separation

Peak capacity estimated using alkane series and FID

CW simulant separated from polar interferents.





Sandia National Laboratories

Development of Fieldable Micro Gas Analyzer Systems: Summary

- Sandia MicroGC and GCxGC have been developed for detection of chemical agents but can be adapted, with further development, for other small molecules such as PAH, dioxins etc.

- GCxGC can be used to provide positive identification of a chemical if corresonding calibrators are available or it can be used to provide a “signature”

- Size and weight of “peripheral” components dominate
  - Batteries, pumps, carrier gas source, data analysis and displays

Sandia National Laboratories

Conclusions

- We can add value to NCS by providing field-deployable analytical platforms for monitoring health of children and for detecting chemicals/drugs

- Platforms available to analyze both environmental and clinical samples

- These platforms can be integrated with mobile phone systems being developed for operation of the device and data acquisition, analysis and transmission

Next Steps

1. Define the analytes and biomarkers to be measured real-time or near real-time together with types of samples (both environmental and clinical)
2. Define performance specifications e.g., sensitivity desired, speed of assays, cost requirements for the device and assay etc.
3. Evaluate Sandia platforms to meet the requirements outlined in 1 and 2; compare against other platforms available to NCS
4. Develop concept-of-operation for technologies deemed suitable– e.g., NCS sites where they will be deployed, duration of deployment, number of devices, number of assays/time etc.
5. Develop and demonstrate assays at Sandia and NCS sites