The exposome and exposomics in inhalation toxicology

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Rollins School of Public Health

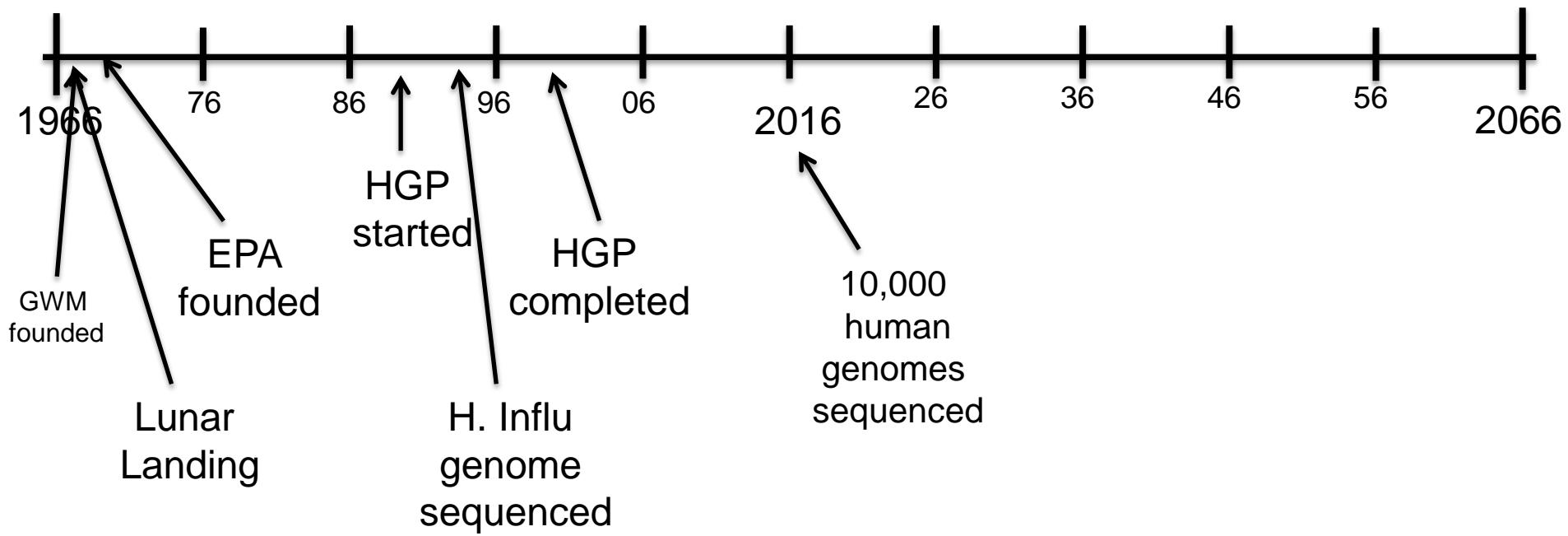Emory University

Atlanta, GA

Outline

- The exposome concept
- Current approaches
- Applications to inhalation toxicology

G x E = P

- Genes x environment = phenotype

- John Locke's (1690) view that we are born *tabula rasa*- a blank slate

- Without input from our environment we are feeble. Humanity requires the environment.

> 1 million

1966   76   86   96   06   2016   26   36   46   56   2066

GWM
founded

Lunar
Landing

EPA
founded

HGP
started

H. Influ
genome
sequenced

HGP
completed

10,000
human
genomes
sequenced

Nature vs. Nurture

- ## Truly a false dichotomy
(see "Nature vs. Nurture: Death of a Dogma" in Neuron, 2010 by Traynor and Singleton)

- ## Whether you call it nature vs. nurture or genes vs. environment, life occurs at the interface.

- ## For the sake of human health, we need a richer understanding of nurture

The Exposome: a Wild idea

Complementing the Genome with an "Exposome": The Outstanding Challenge of Environmental Exposure Measurement in Molecular Epidemiology

Christopher Paul Wild

Molecular Epidemiology Unit, Centre for Epidemiology and Biostatistics, Leeds Institute of Genetics, Health and Therapeutics, Faculty of Medicine and Health, University of Leeds, Leeds, United Kingdom

Defined the "*Exposome*" as all exposures from conception onwards, including those from lifestyle, diet and the environment.
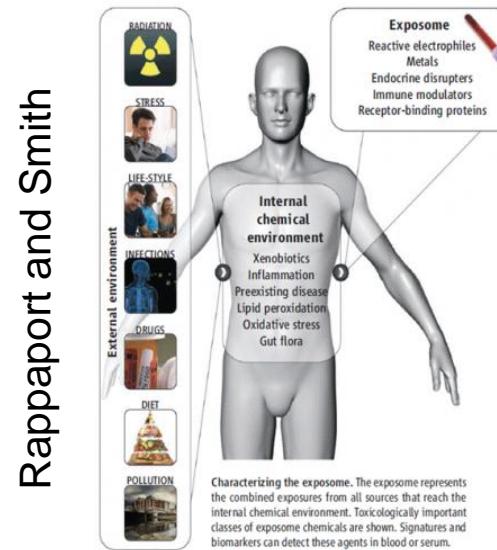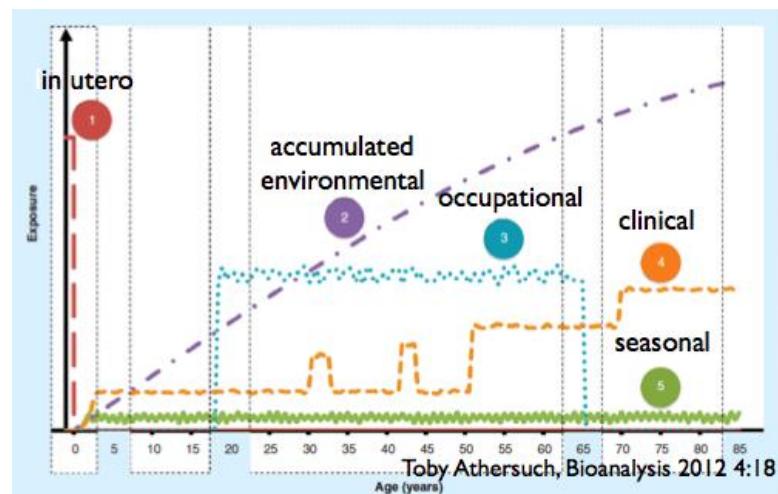
Rappaport and Smith

Science, 2010

Table 3.1 Omics, Omics, Omics. The Major Omic Technologies and their Usage

-ome (Year Coined)	No. of Citations	Root Term	No. of Citations	Fold-Difference
Genome (1920)	857,443	Gene	1,787,215	21
Proteome (1994)	27,875	Protein	5,278,564	189
Transcriptome (1997)	14,826	Transcript	58,146	3.9
Epigenomics (1950s)	1,214	Epigenetics	6,723	5.5
Toxicogenomics[a] (1999)	1,117	Toxin	273,954	245.3
Exposome (2005)	54	Exposure[b]	583,775	10,811

A search of PubMed for the -ome term and its corresponding root word was conducted. The ratio between the -ome term and the root term gives a sense of the acceptance and use of the -ome term. Genome and proteome are well represented, while toxicogenomics and exposome are not. There is a strong denominator effect here in that there were only 54 citations of the work exposome as of July, 2013.
[a]Toxicogenomics, like pharmacogenomics, never used the -ome suffix, only the -omic suffix.
[b]Substituting environment (1,101,485) for exposure yields a 20,398-fold differential.

Toby Athersuch, Bioanalysis 2012 4:18

The Nature of Nurture: Refining the Definition of the Exposome

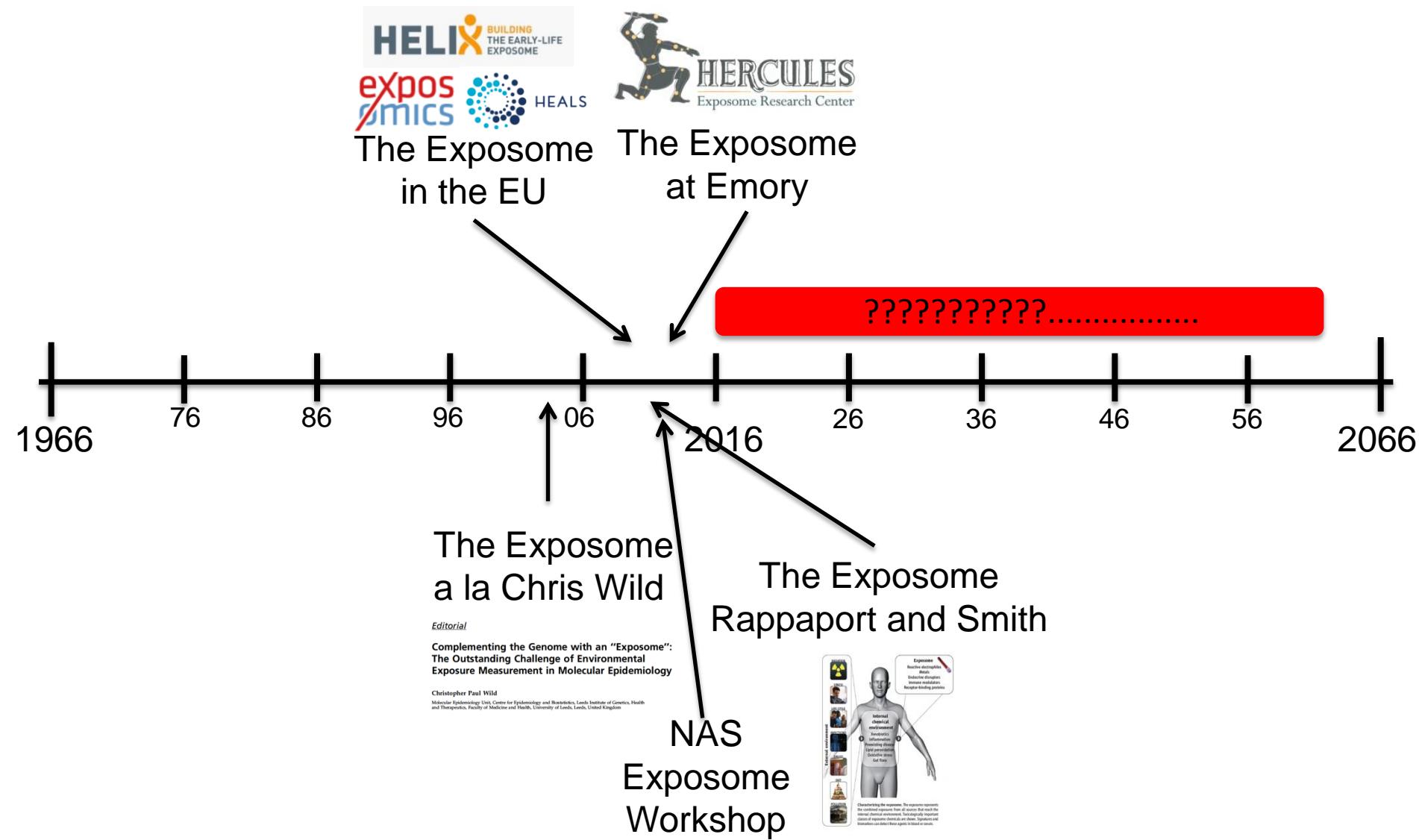Gary W. Miller[1] and Dean P. Jones

Rollins School of Public Health, School of Medicine, Emory University, Atlanta, Georgia 30322
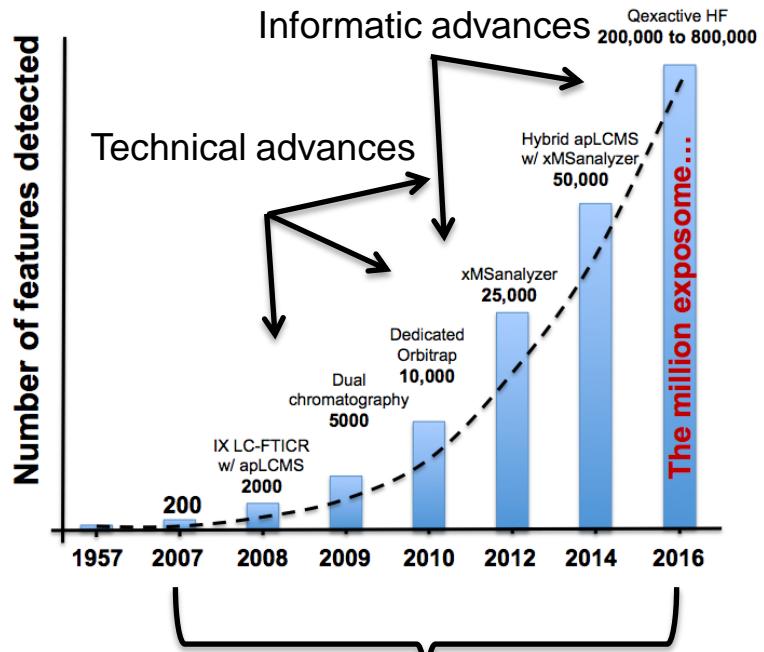
Exposome: The cumulative measure of environmental influences and associated biological responses throughout the lifespan, including exposures from the environment, diet, behavior, and endogenous processes

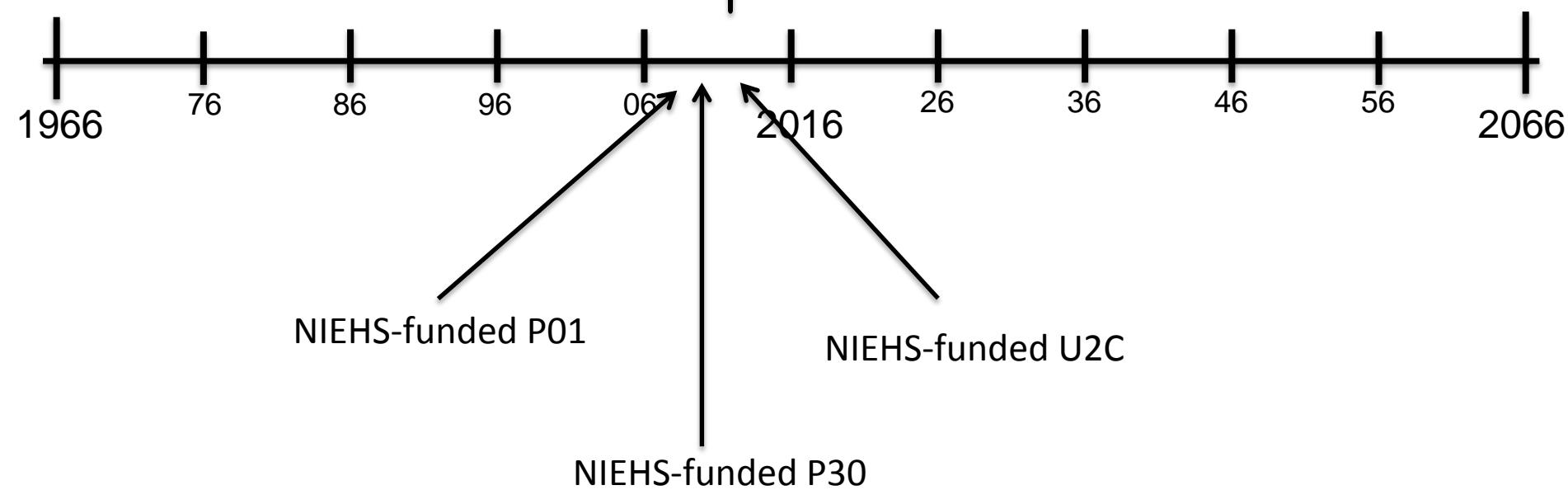The exposome as a foil to the genome

Oxford English Dictionary: Foil (noun):

A person or thing that contrasts with and so emphasizes and enhances the qualities of another.
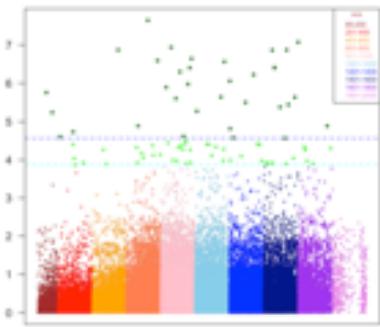
The Exposome
in the EU

The Exposome
at Emory

???????????..................

76   86   96   06   26   36   46   56

1966   2016   2066

The Exposome
a la Chris Wild

Editorial

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The Exposome
Rappaport and Smith

NAS
Exposome
Workshop

Number of features detected

Informatic advances
Qexactive HF
200,000 to 800,000

Technical advances

Hybrid apLCMS
w/ xMSanalyzer
50,000

xMSanalyzer
25,000

Dedicated
Orbitrap
10,000

Dual
chromatography
5000

IX LC-FTICR
w/ apLCMS
2000

200

The million exposome...

1957 2007 2008 2009 2010 2012 2014 2016

HRM Metabolomics

2500 encoded
1000+ pharmaceutical
80,000 synthetic
200,000 plant-based
100,000+ microbial

1966   76   86   96   06   2016   26   36   46   56   2066

NIEHS-funded P01

NIEHS-funded U2C

NIEHS-funded P30

Affordable analysis of thousands of analytes: exogenous, endogenous, biological pathways

plasticizers or pesticides
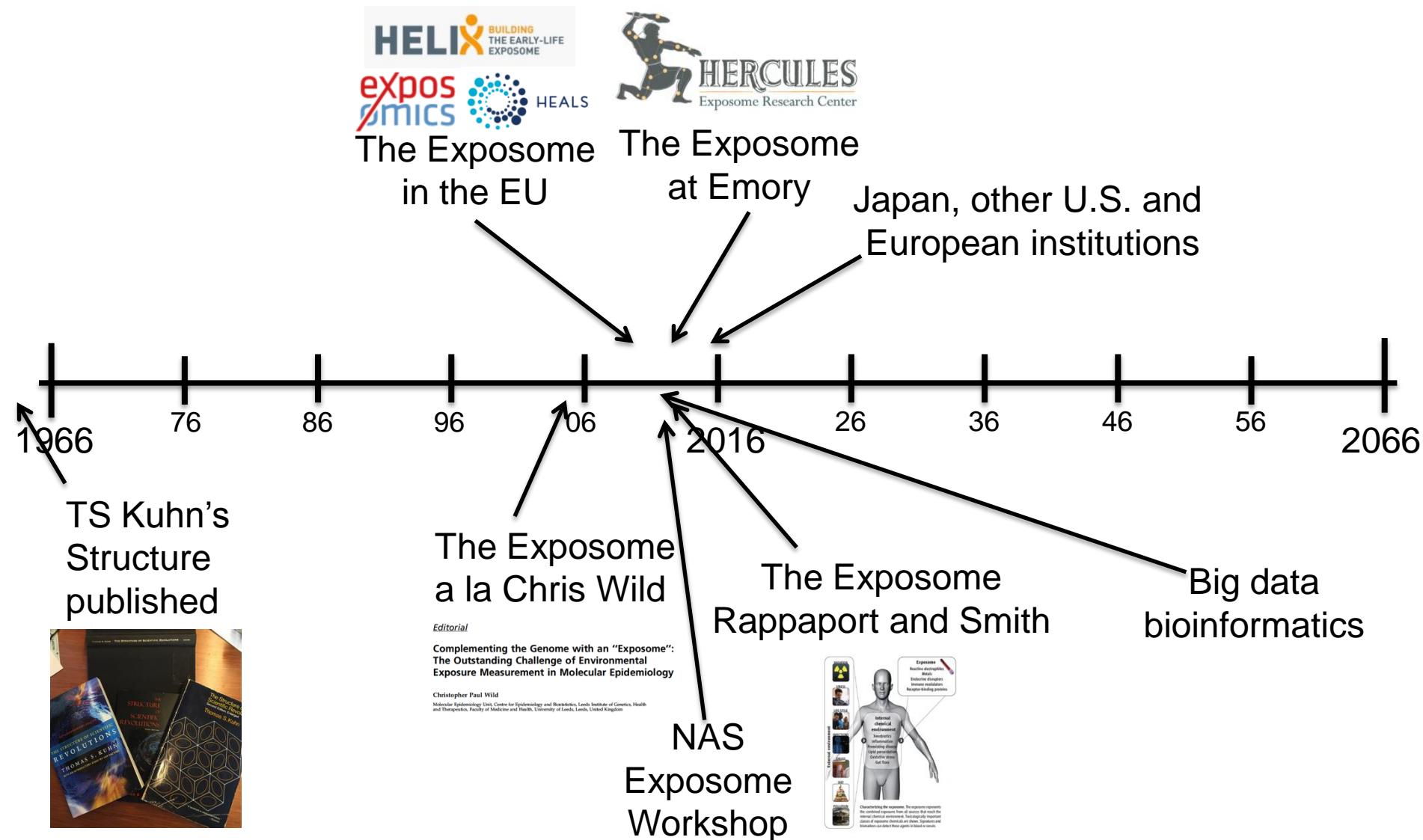microbial metabolites of medicines
tyrosine or tryptophan pathways

~$250/sample for over
20,000 well-characterized and
reliable features

HELIX BUILDING THE EARLY-LIFE EXPOSOME

exposomics HEALS

HERCULES
Exposome Research Center

The Exposome
in the EU

The Exposome
at Emory

Japan, other U.S. and
European institutions

1966   76   86   96   06   2016   26   36   46   56   2066

The Exposome
a la Chris Wild

Editorial

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and Therapeutics, Faculty of Medicine and Health, University of Leeds, Leeds, United Kingdom

NAS
Exposome
Workshop

The Exposome
Rappaport and Smith

Big data
bioinformatics

Are we entering the early phase of a paradigm shift?

The Exposome in the EU

The Exposome at Emory

Japan, other U.S. and European institutions

TS Kuhn's Structure published

The Exposome a la Chris Wild

The Exposome Rappaport and Smith

Big data bioinformatics

NAS Exposome Workshop

Timeline: 1966 · 76 · 86 · 96 · 06 · 2016 · 26 · 36 · 46 · 56 · 2066

Editorial

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Molecular Epidemiology Unit, Centre for Epidemiology and Biostatistics, Leeds Institute of Genetics, Health and Therapeutics, Faculty of Medicine and Health, University of Leeds, Leeds, United Kingdom

Postscript to 2nd edition-1969
Kuhn on the term "paradigm"

"…it stands for the entire constellation of beliefs, values, techniques, and so on shared by the members of a community."

"…it denotes one sort of element in that constellation, the concrete puzzle-solutions which, employed as models or examples, can replace explicit rules as a *basis for the solution of the remaining puzzles* in normal science."

How do we assess our surroundings?

- Global
- National
- Regional
- State
- County
- City
- Neighborhood
- Home
- Personal space

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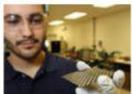
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5 days ago

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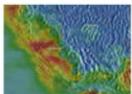
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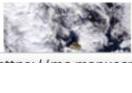
NASA's Terra Satellite Sees Alaskan Volcanic
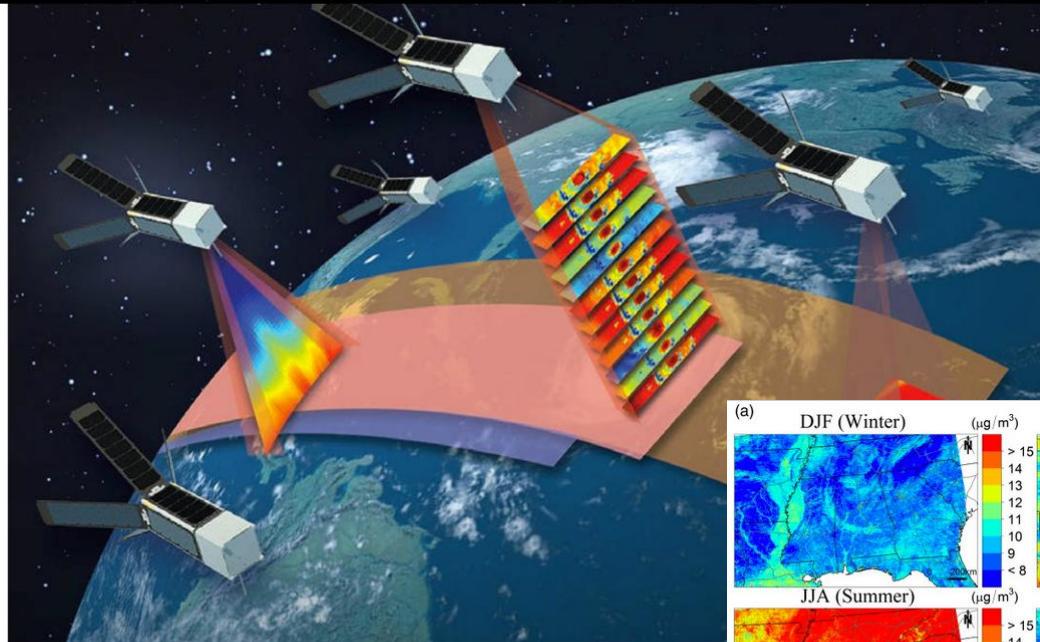
The Time-Resolved Observations of Precipitation structure and storm Intensity with a Constellation of Smallsats (TR... will study the development of tropical cyclones by taking measurements of temperature, precipitation and cloud pro...
Credits: MIT Lincoln Laboratory

NASA has selected two proposals for new Earth science investigations that will put new ins... particulate air pollutants and study the development of tropical cyclones.
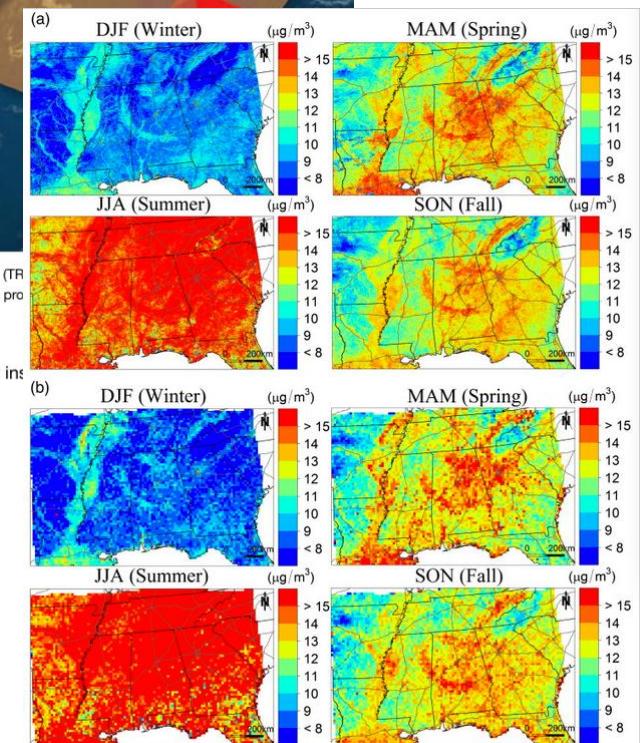
Fig. 8.
Seasonal mean PM$_{2.5}$ estimated using MAIAC (a) and MODIS (b).

Images from NASA and Dr. Yang Liu laboratory

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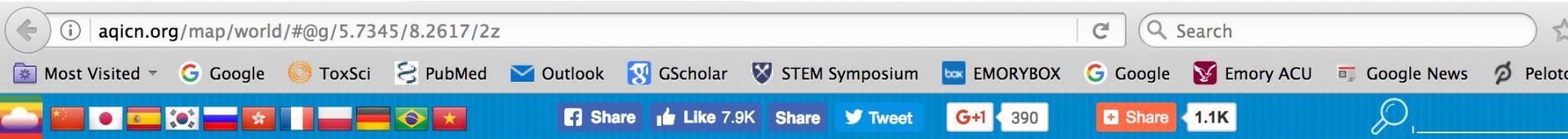
Air Pollution in World: Real-time Air Quality Index Visual

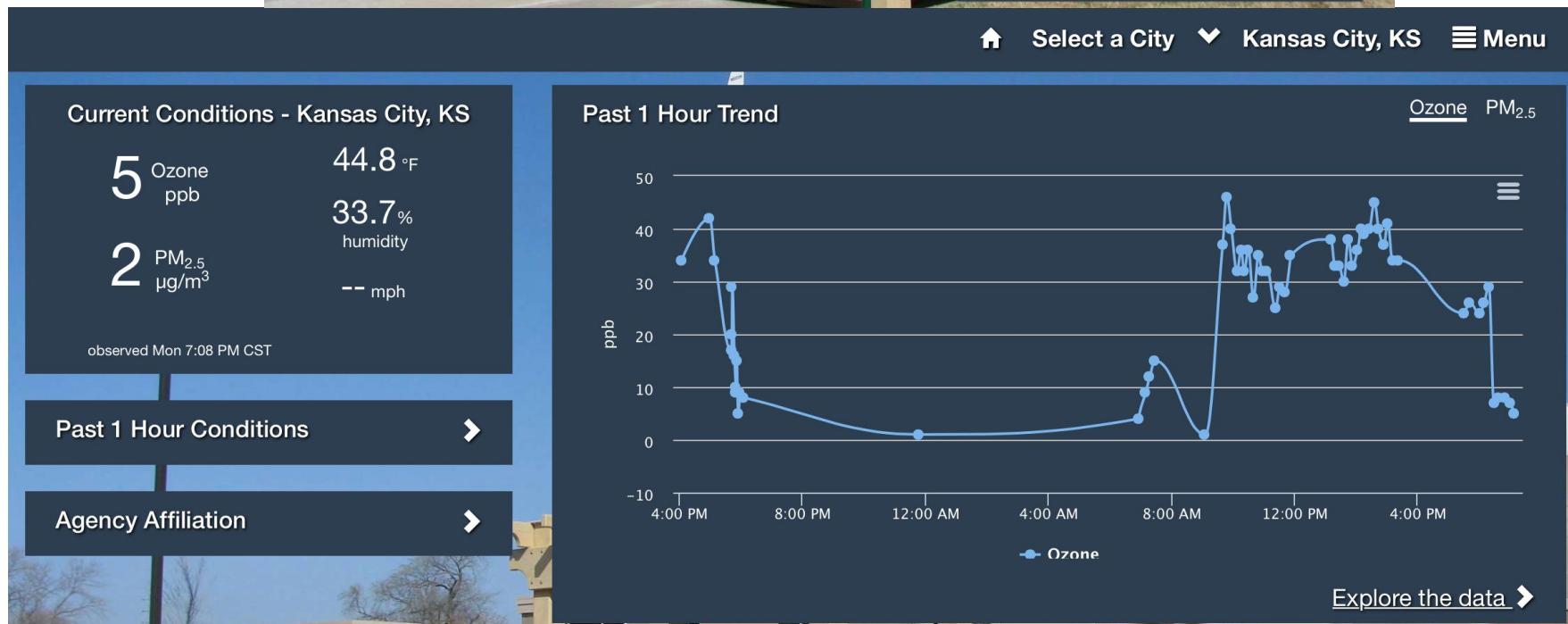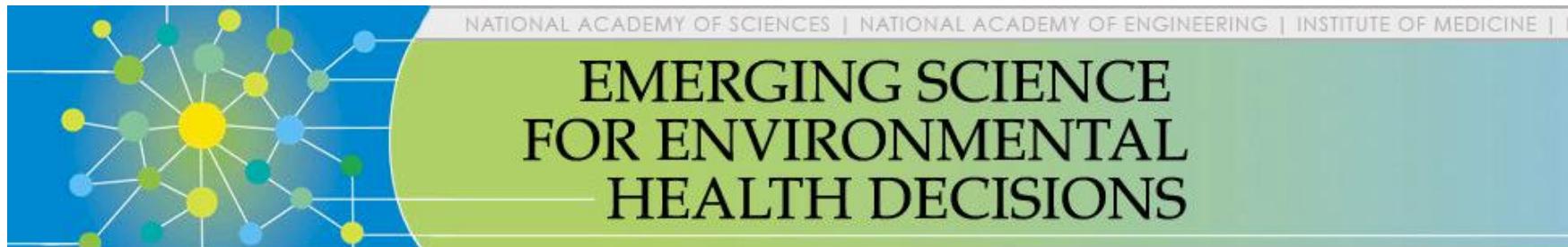| World | Asia | Europe | North America | South America | Africa | Australia | Middle East | India | China |

Similar transformative technologies, such as remote sensing and wearables, are expanding our definition of exposure science.

Personal Environmental Exposure Measurements:
Making Sense and Making Use of Emerging Capabilities

November 16-17, 2016

National Academies of Sciences, Engineering, and Medicine
Keck Center, Room 100
500 Fifth Street, NW
Washington, DC 20001

http://nas-sites.org/emergingscience/meetings/personal-exposure-measurement/workshop-agenda/

Meet Speck.
The indoor air quality monitor empowering you to breathe easier.

▶ Watch the Video

17 w
Good

Air Quality Matters

How Can Speck Help You?

Protecting you and your family from harmful air

SPARROW™ wearable air monitor with a powerful app

0 PPM SAFE
24% RH   84°F   45%
TODAY'S FORECAST
85°F   33 AQI

SPARROW is a **wearable environmental health and safety monitor** that measures Carbon Monoxide (CO), along with temperature, pressure and relative humidity. It can be attached to your smartphone case, clothes, bag, purse, stroller, bike, and even placed in your car.

GET SPARROW

SPARROW also works in conjunction with the OtterBox uniVERSE Case System, the innovative smartphone protection that allows for interchangeable accessories to slide directly onto the back of the phone case, making it easier to protect yourself from toxic air quality wherever you go.
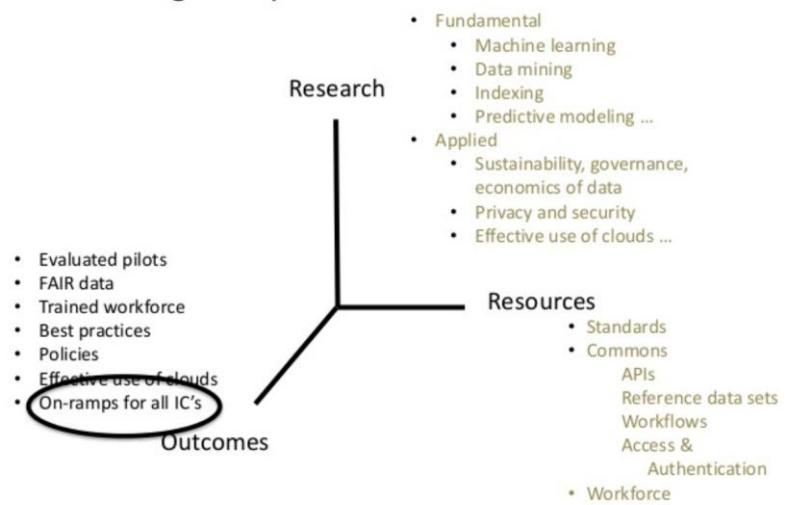
BD2K @ NIH – A Vision Through 2020

Philip E. Bourne, PhD, FACMI

Associate Director for Data Science

philip.bourne@nih.gov

A Strategic Response

Research

- Fundamental
  - Machine learning
  - Data mining
  - Indexing
  - Predictive modeling ...
- Applied
  - Sustainability, governance, economics of data
  - Privacy and security
  - Effective use of clouds ...

Outcomes
- Evaluated pilots
- FAIR data
- Trained workforce
- Best practices
- Policies
- Effective use of clouds
- On-ramps for all IC's

Resources
- Standards
- Commons
  APIs
  Reference data sets
  Workflows
  Access & Authentication
- Workforce

There is intense competition for data scientists. Toxicology/Environmental Health must invest in data science infrastructure and aggressively recruit, train, and retain those individuals who will be solving our problems in the next 50 years.

Exposome-outside in

- The exposome includes our complex surroundings. The source, timing, duration, and physiochemical properties of these exposures are critical.

- At Emory, we have been focusing most of our efforts on the components of the exposome that can be measured inside the body.

HERCULES
Exposome Research Center

Funded in 2013 (ES P30-019776, renewal pending)
Core facilities for:
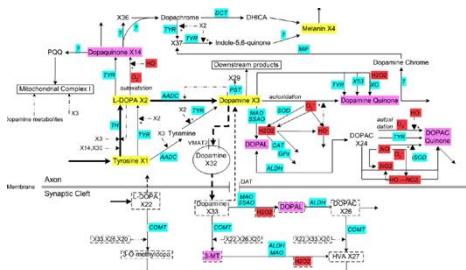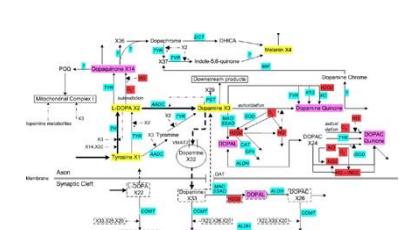data sciences (Waller, Voit, and Kemp)
targeted exposures (Barr)
untargeted exposures (Jones)
patient studies (Ziegler)
pilot awards (Morgan)
career development (Tolbert)
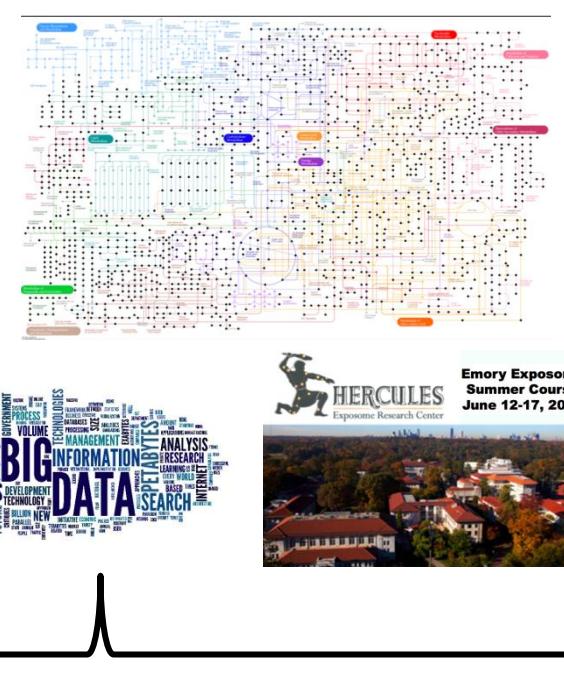community engagement (Kegler/Pearson)

HERCULES
Exposome Research Center

Emory Exposome Summer Course June 12-17, 2016

High-resolution metabolomics workflow

Databases (design, maintenance)

Bioinformatics
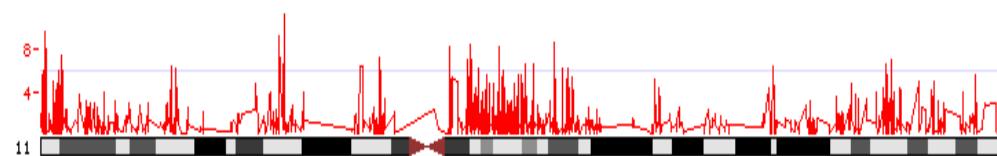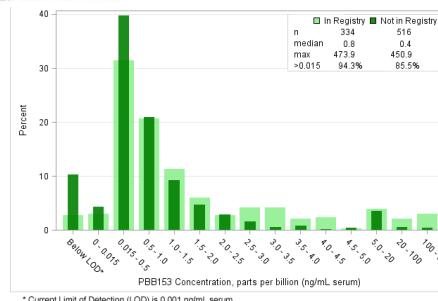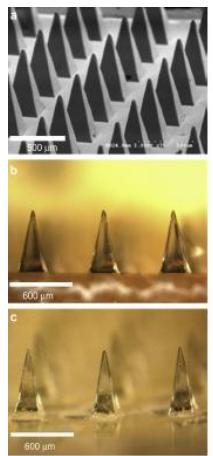
Computational toxicology

Biostats

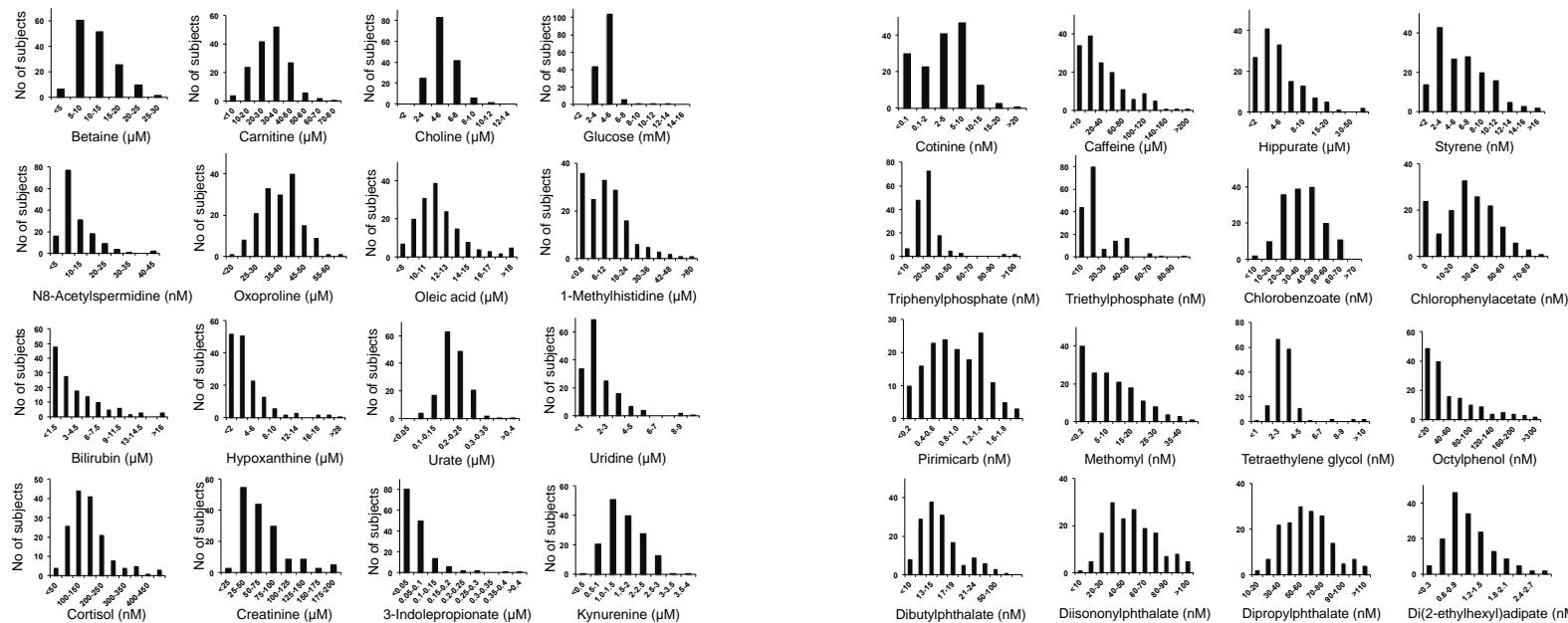High performance computing

Machine learning

Systems biology
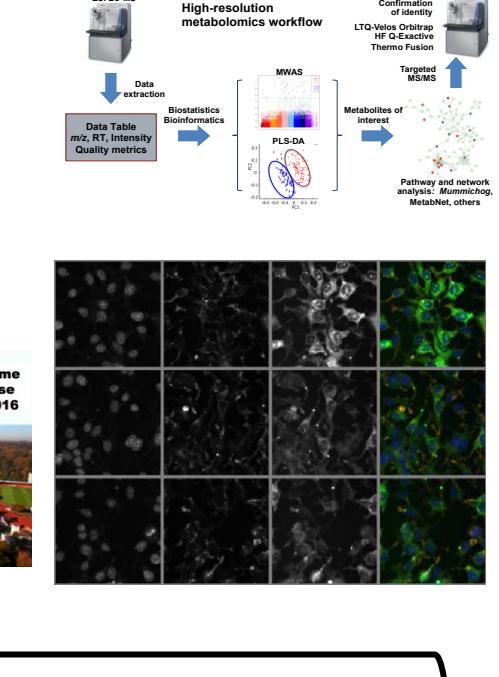
Environmental chemicals are present at 3-4 orders of magnitude lower concentration than endogenous metabolites *Rappaport S. et al., Environ Health Perspect, 2014*
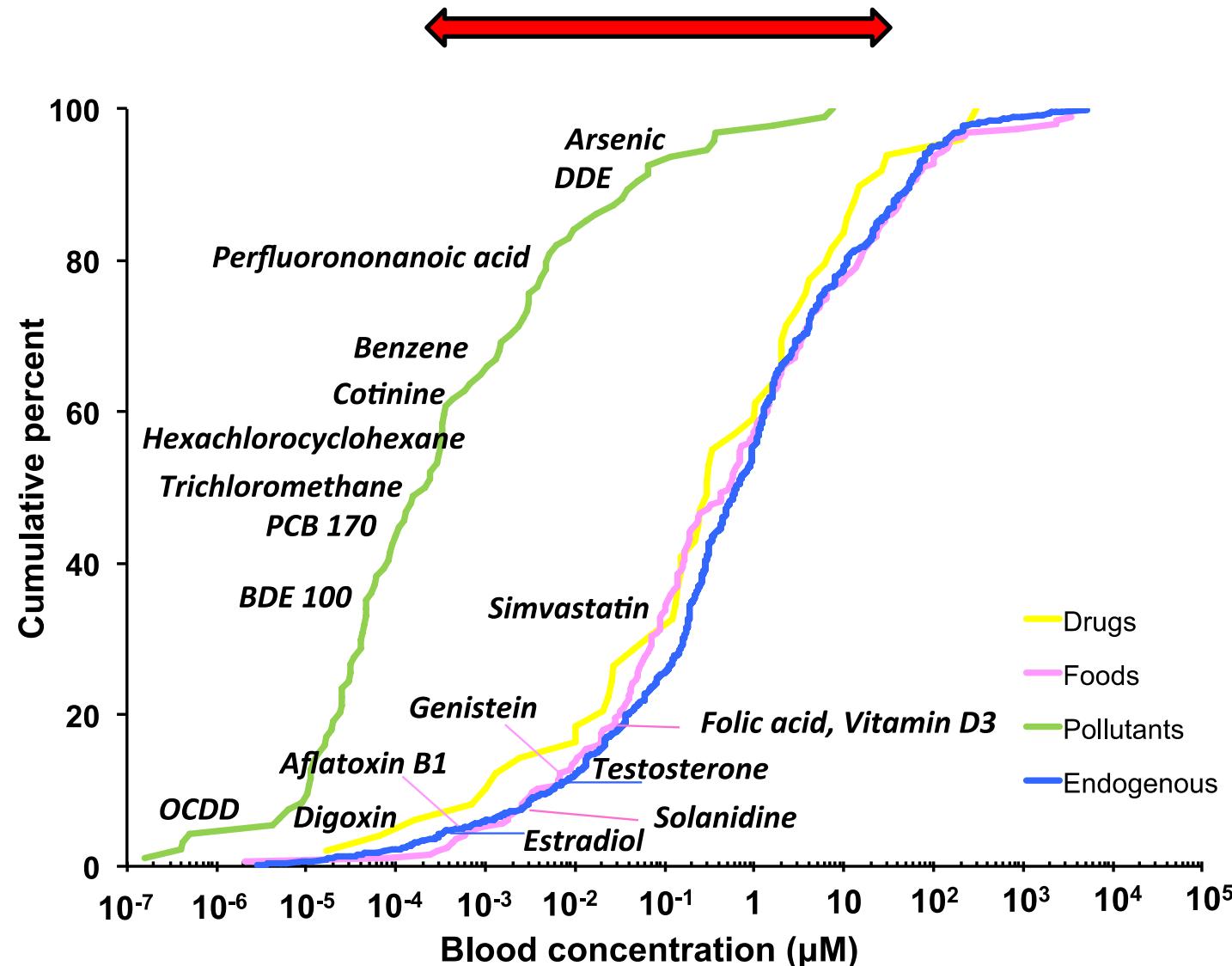
High resolution metabolomic methods provide reliable metabolite measurements in human plasma in millimolar and micromolar range…

and reliable environmental and dietary chemical measurements in *nanomolar* and *subnanomolar* range

Go et al 2015 Toxicol Sci

Environmental chemicals are present at 3-4 orders of magnitude lower concentration than endogenous metabolites *Rappaport S. et al., Environ Health Perspect, 2014*

HRM measures metabolites over >7 orders of magnitude *Go et al Tox Sci 2015*
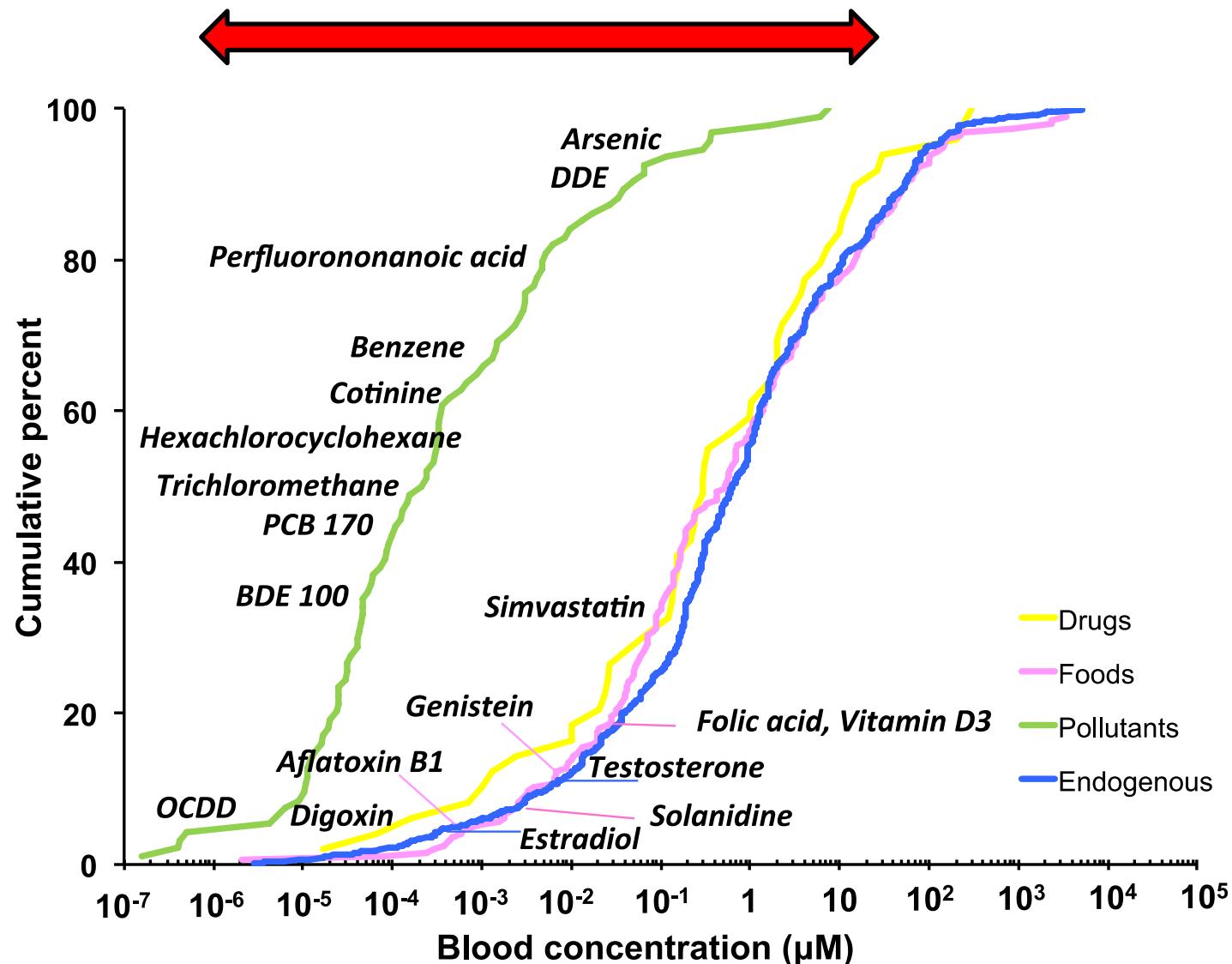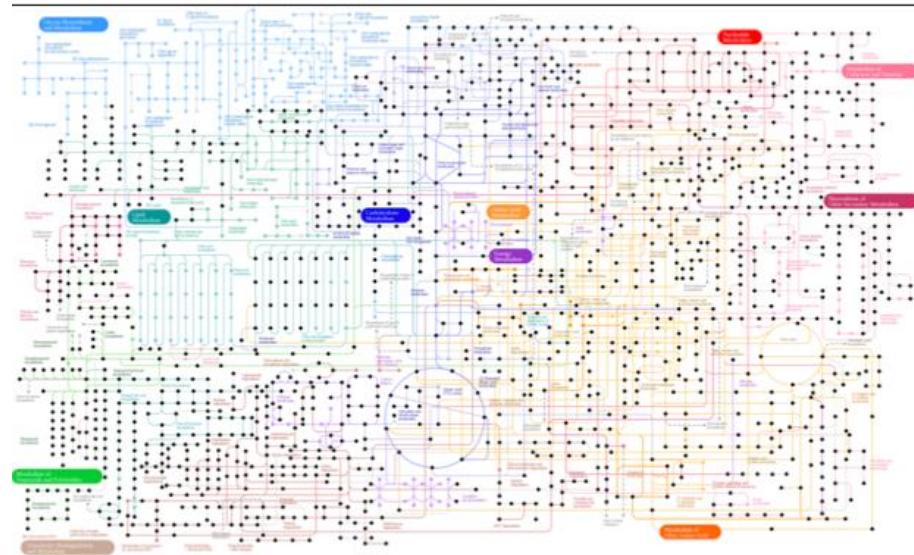
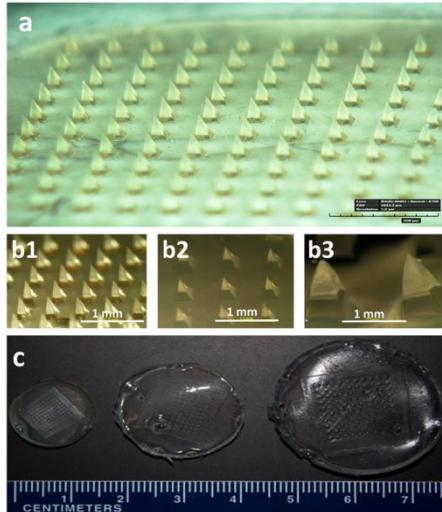# Analyte collection using microneedle patch

Detected 7,044 total peaks

6,615 in plasma; 6,012 in ISF (peak present in ≥ 1 sample in matrix)

Plasma

1,032 (15%)

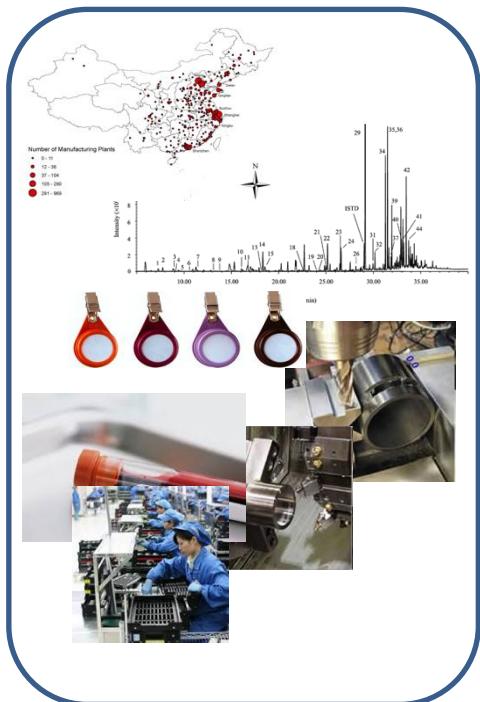**Common**

5,583 (79%)

ISF

429 (6%)
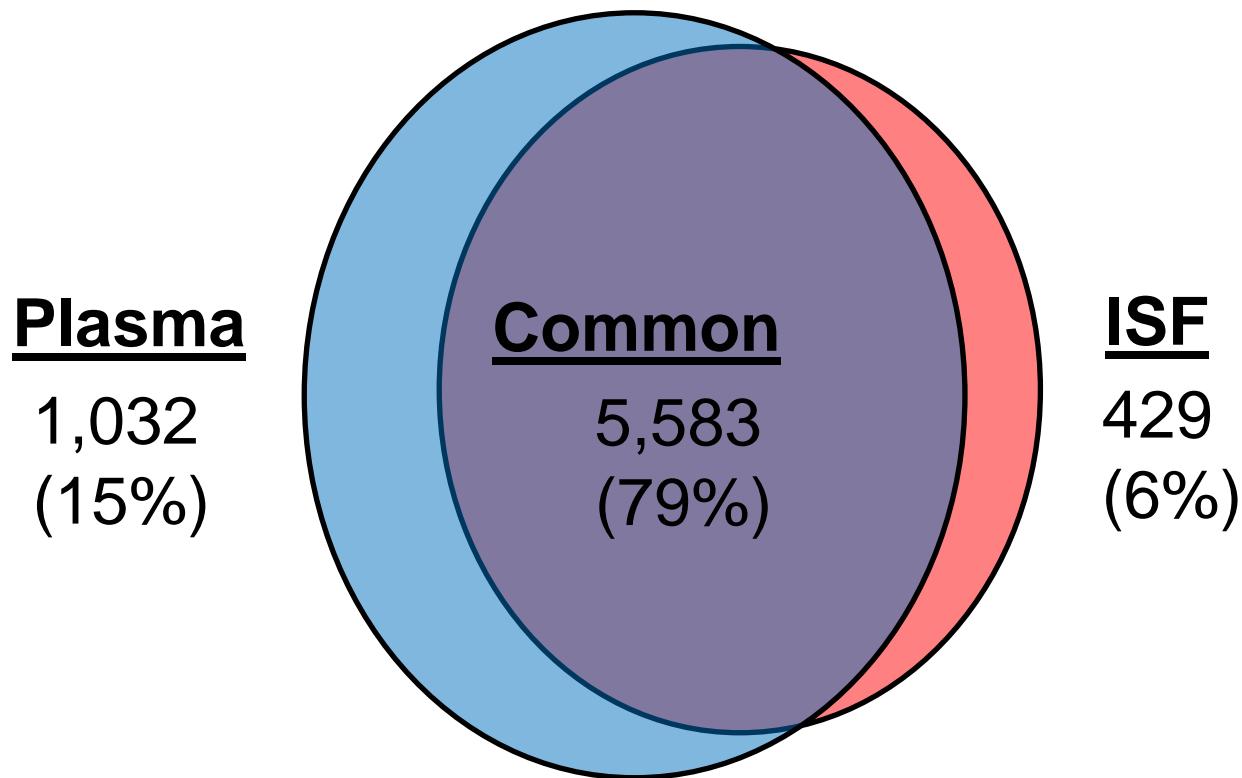
Strongly-correlated features between plasma and ISF (peak present in ≥ 4 samples in both plasma and ISF)

m/z	Retention time	Spearman r	Compound	Adduct	Source	No. positive in plasma	No. positive in ISF
136.0425	87.441	0.98	Homocysteine	M+H	xMSannotator	10	10
141.0715	118.369	0.96	Dipropyl sulfide	M+Na	HMDB	10	10
269.1255	88.818	0.94	Lenticin	M+Na	HMDB	9	7
248.1474	86.775	0.92	Hydroxybutyrylcarnitine	M+H	xMSannotator	5	5
129.0659	87.709	0.92	Glutamine	M+H-H2O	known	10	10
104.0710	88.573	0.90	Dimethylglycine	M+H	HMDB	10	10
119.0162	88.079	0.90	Dihydro-4-mercapto-3(2H)-furanone	M+H	HMDB	10	10
195.0874	68.647	0.89	Caffeine	M+H	HMDB	6	4
522.2577	559.475	0.89	LysoPE(0:0/20:5(5Z,8Z,11Z,14Z,17Z))	M+Na	HMDB	10	8
182.0811	84.235	0.88	L-Tyrosine	M+H	HMDB	10	10
478.2920	560.465	0.88	LysoPE(0:0/18:2(9Z,12Z))	M+H	HMDB	10	10
138.0549	120.073	0.87	m-Aminobenzoic acid	M+H	HMDB	7	9
116.0709	112.113	0.85	L-Proline	M+H	HMDB	10	10
157.0834	65.384	0.85	Diethylene glycol monoethyl ether	M+Na	HMDB	10	10
110.0273	128.640	0.85	Hypotaurine	M+H	HMDB	4	10
204.1229	65.499	0.84	L-Acetylcarnitine	M+H	HMDB	10	10
310.2010	72.742	0.84	L-Octanoylcarnitine	M+Na	HMDB	10	10
136.0756	84.022	0.83	5-Acetyl-2-methylpyridine	M+H	HMDB	10	10
144.1018	125.060	0.83	Proline betaine	M+H	xMSannotator	10	10
342.2635	86.528	0.83	trans-2-Dodecenoylcarnitine	M+H	xMSannotator	10	6
205.1263	66.469	0.82	L-Acetylcarnitine	M+H(C13)	xMSannotator	9	10
317.2510	85.138	0.81	Decanoylcarnitine	M+H(C13)	xMSannotator	10	4
120.0656	121.825	0.81	L-Threonine	M+H	xMSannotator	10	10

High resolution metabolomics provides a **<u>central reference platform</u>** to link exposure data, internal burden and biological response to exposure

Exposure assessment

Metabolomics

Core Biological Metabolome

Microbiome-related Chemicals

Endogenous metabolome

Non-nutritive Chemicals in Diet

Supplements and Pharmaceuticals

Environmental metabolome

Commercial Products

Environmental Chemicals

Molecular response

6-Thioinosine 5'-monophosphate

Adenine

Xanthine

IMP

Uracil

UMP

Guanosine

Deoxyadenosine

Deoxyguanosine

Deoxycytidine

Adenosine

Uridine

0.0   0.02   0.04
Fold Change

Courtesy of Dean Jones

A genetic basis for the variable effect of smoking/nicotine on Parkinson's disease

EM Hill-Burns[1], N Singh[1], P Ganguly[1], TH Hamza[1], J Montimurro[1], DM Kay[1], D Yearout[2], P Sheehan[1], K Frodey[1], JA Mclear[1], MB Feany[3], SD Hanes[1,4,5,6], WJ Wolfgang[4,5], CP Zabetian[2], SA Factor[7] and H Payami[1,5]

SV2C Genotype	Frequency	Relative PD risk in smokers
Homozygous, major alleles	23%	(0.4) ⬇⬇⬇
Heterozygous	75%	(.8-1.9) ⬆ or ⬇
Homozygous, minor alleles	2%	(3.3) ⬆⬆⬆

GWAS for protective effect of nicotine in human PD and fly PD model both identified SV2C/ortholog

The Synaptic Vesicle Protein SV2 Is a Novel Type of Transmembrane Transporter

Mel B. Feany,* Sandra Lee,† Robert H. Edwards,† and Kathleen M. Buckley*

Identification, characterization, and molecular cloning of a novel transporter-like protein localized to the central nervous system

Jay A. Gingrich[a], Peter H. Andersen[a,*], Mario Tiberi[a], Salah El Mestikawy[a], Per N. Jorgensen[b], Robert T. Fremeau Jr.[a] and Marc G. Caron[a]

SV2C IS A SYNAPTIC VESICLE PROTEIN WITH AN UNUSUALLY RESTRICTED LOCALIZATION: ANATOMY OF A SYNAPTIC VESICLE PROTEIN FAMILY

R. JANZ* and T. C. SÜDHOF†

## SV2, a Brain Synaptic Vesicle Protein Homologous to Bacterial Transporters

Sandra M. Bajjalieh, Karen Peterson, Rajesh Shinghal, Richard H. Scheller*

Synaptic vesicle glycoprotein 2C (SV2C) modulates dopamine release and is disrupted in Parkinson disease

Amy R. Dunn[a], Kristen A. Stout[a], Minagi Ozawa[a], Kelly M. Lohr[a], Carlie A. Hoffman[a], Alison I. Bernstein[a], Yingjie Li[a,b], Minzheng Wang[a], Carmelo Sgobio[c], Namratha Sastry[c], Huaibin Cai[c], W. Michael Caudle[a], and Gary W. Miller[a,b,d,e,1]

[a]Department of Environmental Health, Rollins School of Public Health, Emory University, Atlanta, GA 30322; [b]Center for Neurodegenerative Diseases, Emory University School of Medicine, Atlanta, GA 30322; [c]Transgenics Section, National Institute on Aging, National Institutes of Health, Bethesda, MD 20892; [d]Department of Pharmacology, Emory University School of Medicine, Atlanta, GA 30322; and [e]Department of Neurology, Emory University School of Medicine, Atlanta, GA 30322
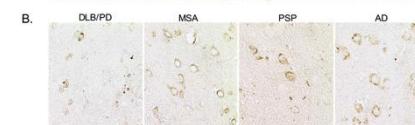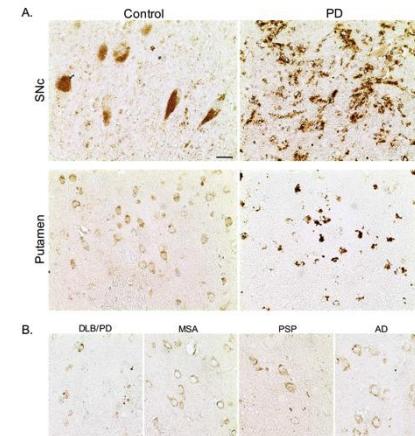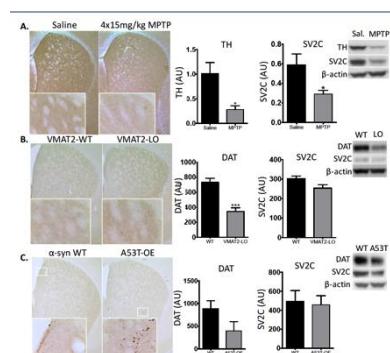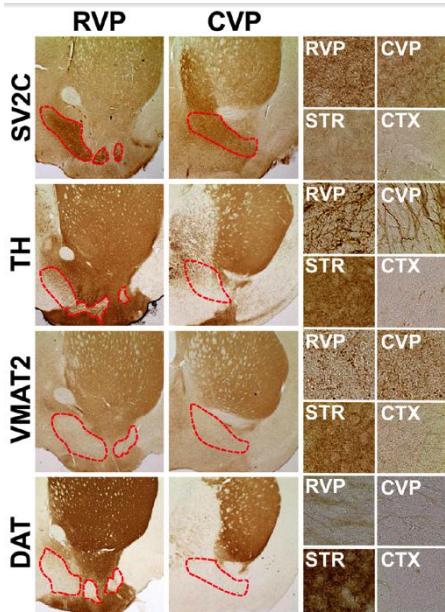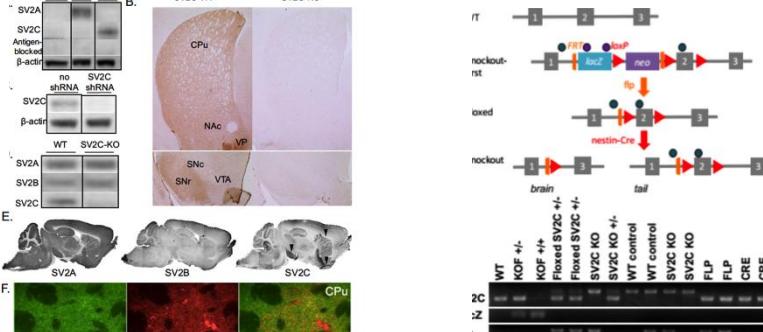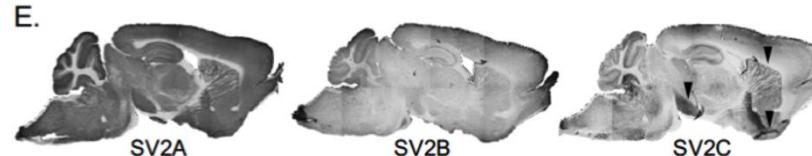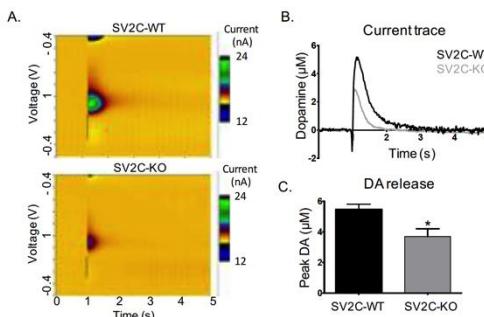
A new avenue of research because a single geneticist decided to include a couple of "environmental" factors into the analysis

When it comes to the exposome we do not need to measure everything. We need to provide a valid set of parameters for "E" that can be used in conjunction with existing studies that employ genomics, epigenomics, proteomics, etc. to study diseases and health outcomes.

Examples of exposomics for inhalation toxicology
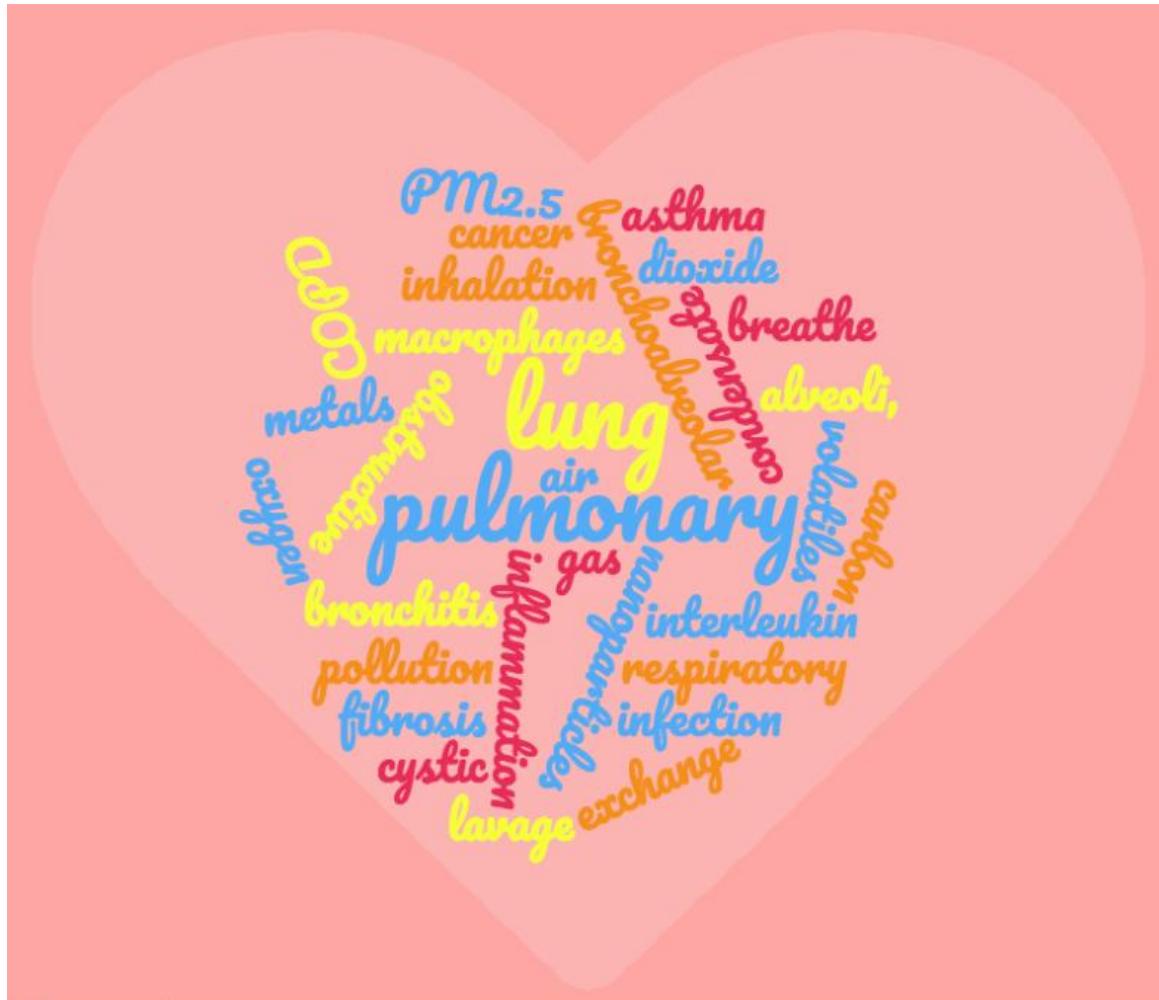
Lung inflammation after H1NI infection

Chandler et al. AJPRICP, 2016
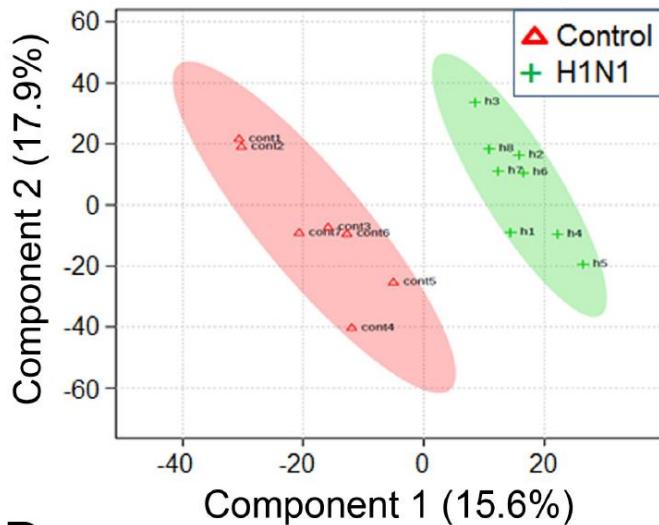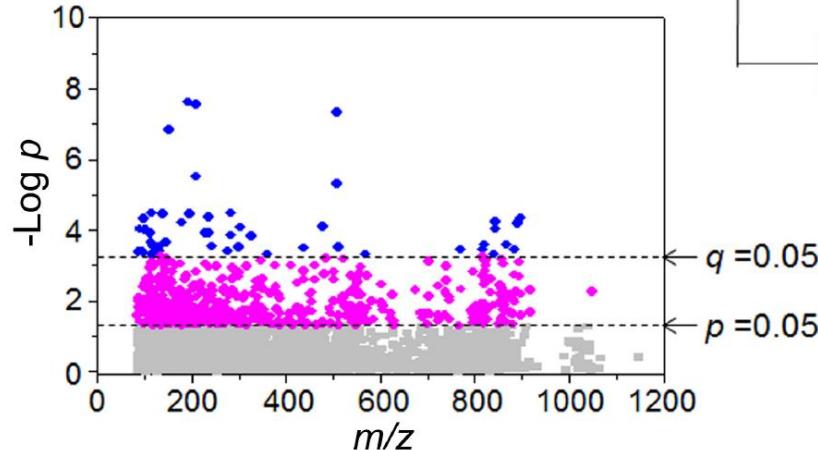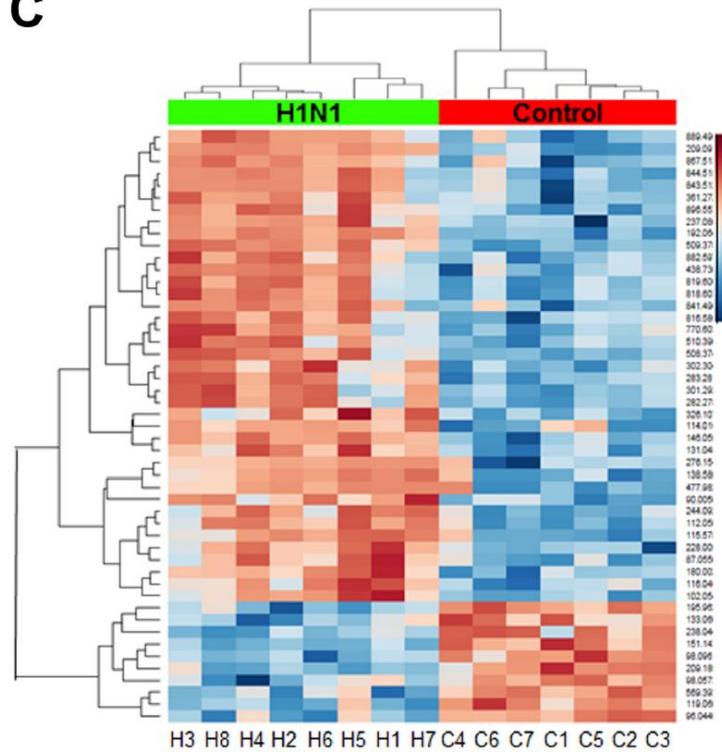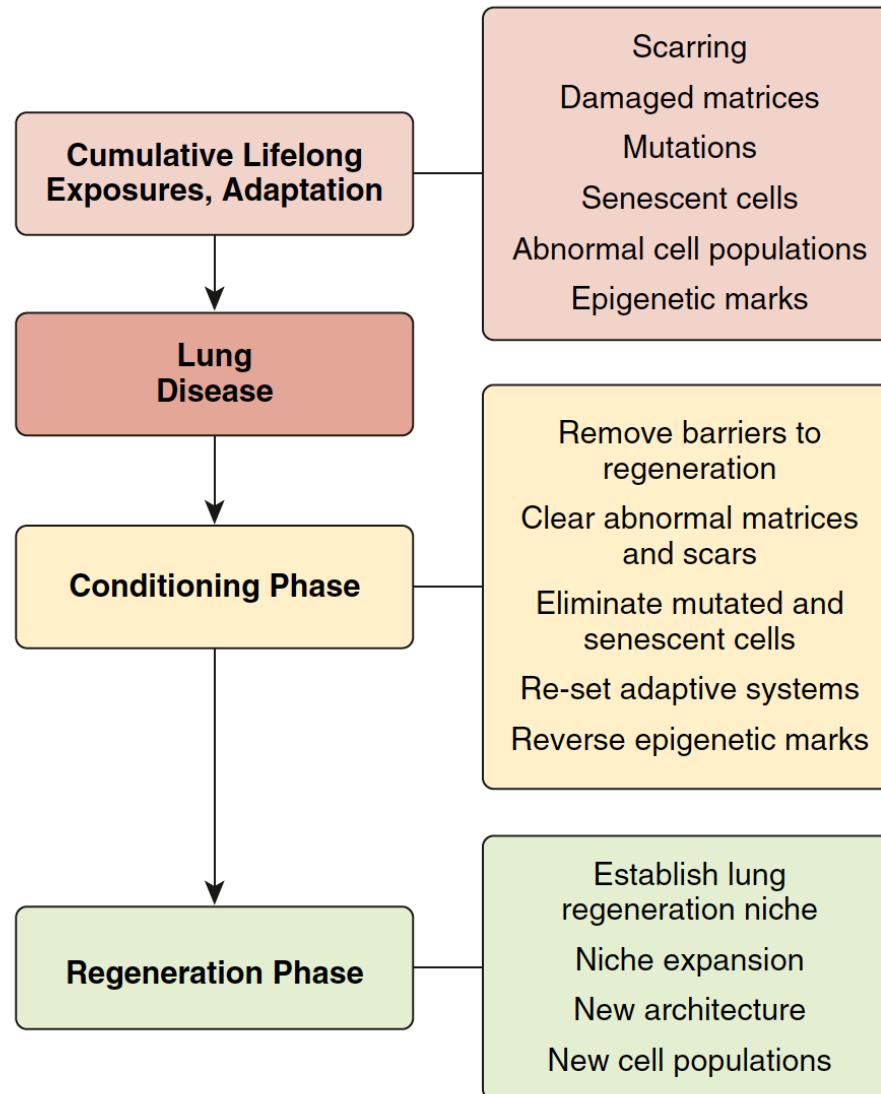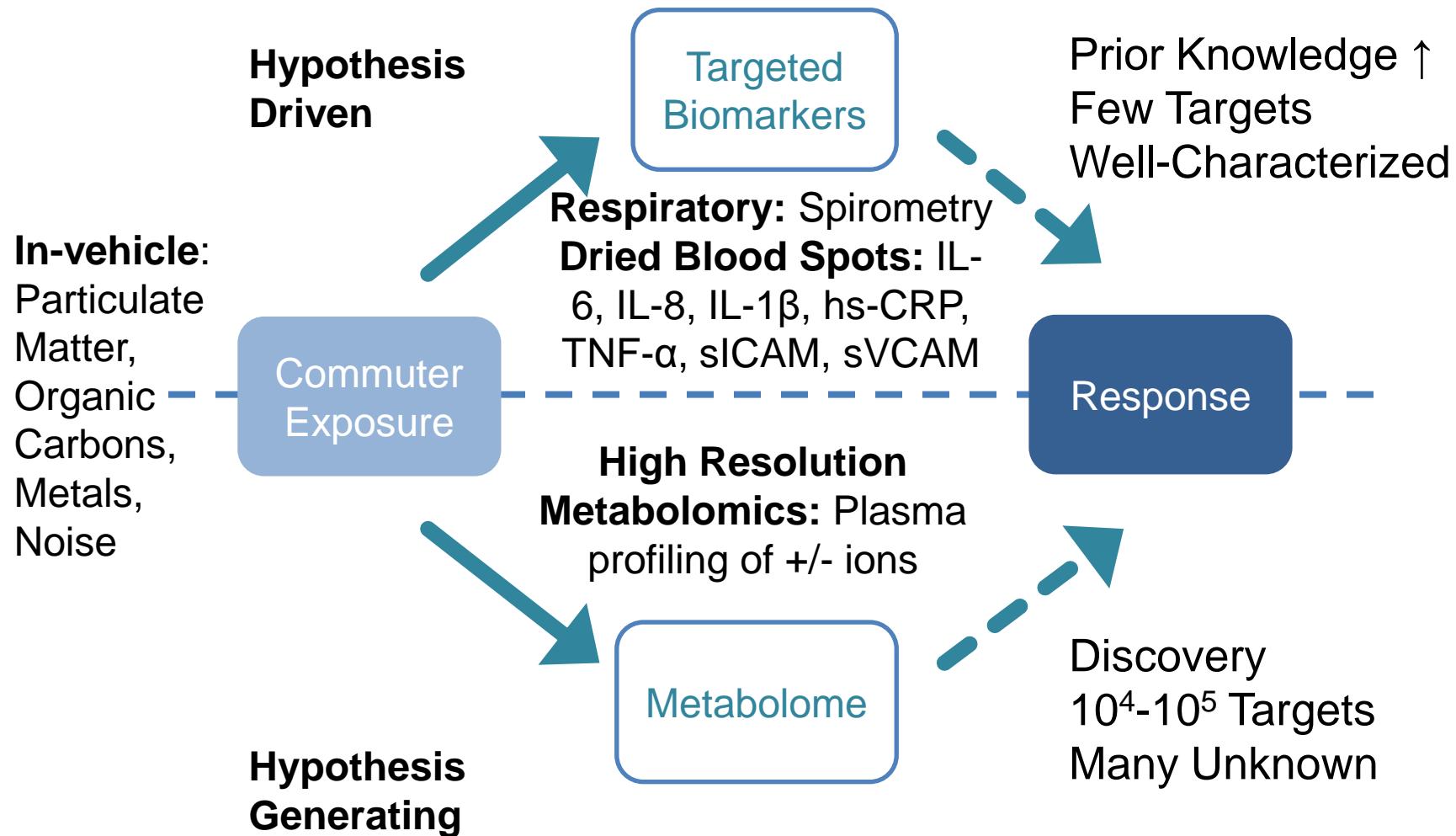
Figure 1. Conceptual diagram for lung regeneration. After accumulation of lifelong exposures and adaptive responses in disease development, a conditioning phase is needed to enable efficient regeneration of lung function.

Go and Jones, 2016

Short-Term Exposures and Acute Biological Responses

Goal: Identify pathways of human metabolism that change with respect to traffic air pollution exposure in commuting settings

Hypothesis Driven

Targeted Biomarkers

Prior Knowledge ↑
Few Targets
Well-Characterized

In-vehicle: Particulate Matter, Organic Carbons, Metals, Noise

Commuter Exposure

Respiratory: Spirometry
Dried Blood Spots: IL-6, IL-8, IL-1β, hs-CRP, TNF-α, sICAM, sVCAM

Response

High Resolution Metabolomics: Plasma profiling of +/- ions

Metabolome

Hypothesis Generating

Discovery
10^4-10^5 Targets
Many Unknown

Courtesy of Dr. Chandresh Ladva and Dr. Jeremy Sarnat

Short-Term Exposures and Acute Biological Responses

In-vehicle: Particulate Matter, Organic Carbons, Metals, Noise

Commuter Exposure

1 → Targeted Biomarkers

Respiratory: Spirometry
Dried Blood Spots: IL-6, IL-8, IL-1β, hs-CRP, TNF-α, sICAM, sVCAM

2 → Metabolome

High Resolution Metabolomics: Plasma profiling of +/- ions

Response

Short-Term Exposures and Acute Biological Responses

Biological Response:
Metabolic Pathways,
Oxidative Stress,
Systemic Inflammation

Particulate Metals Perturb Metabolome

Lead (Pb)

Aluminum (Al)
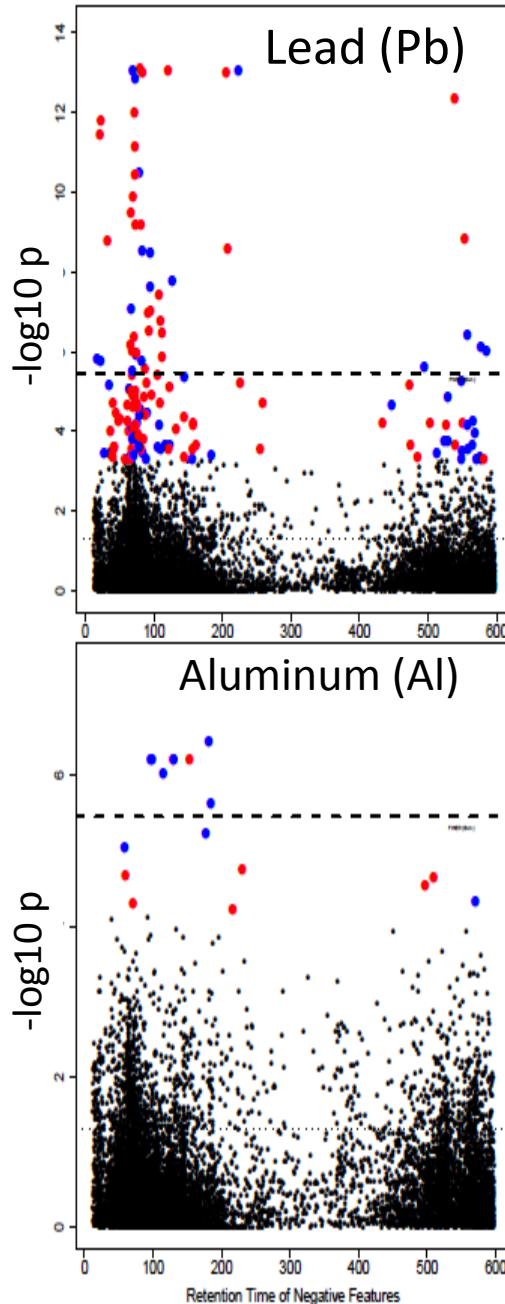
- Null associations for many pollutant models

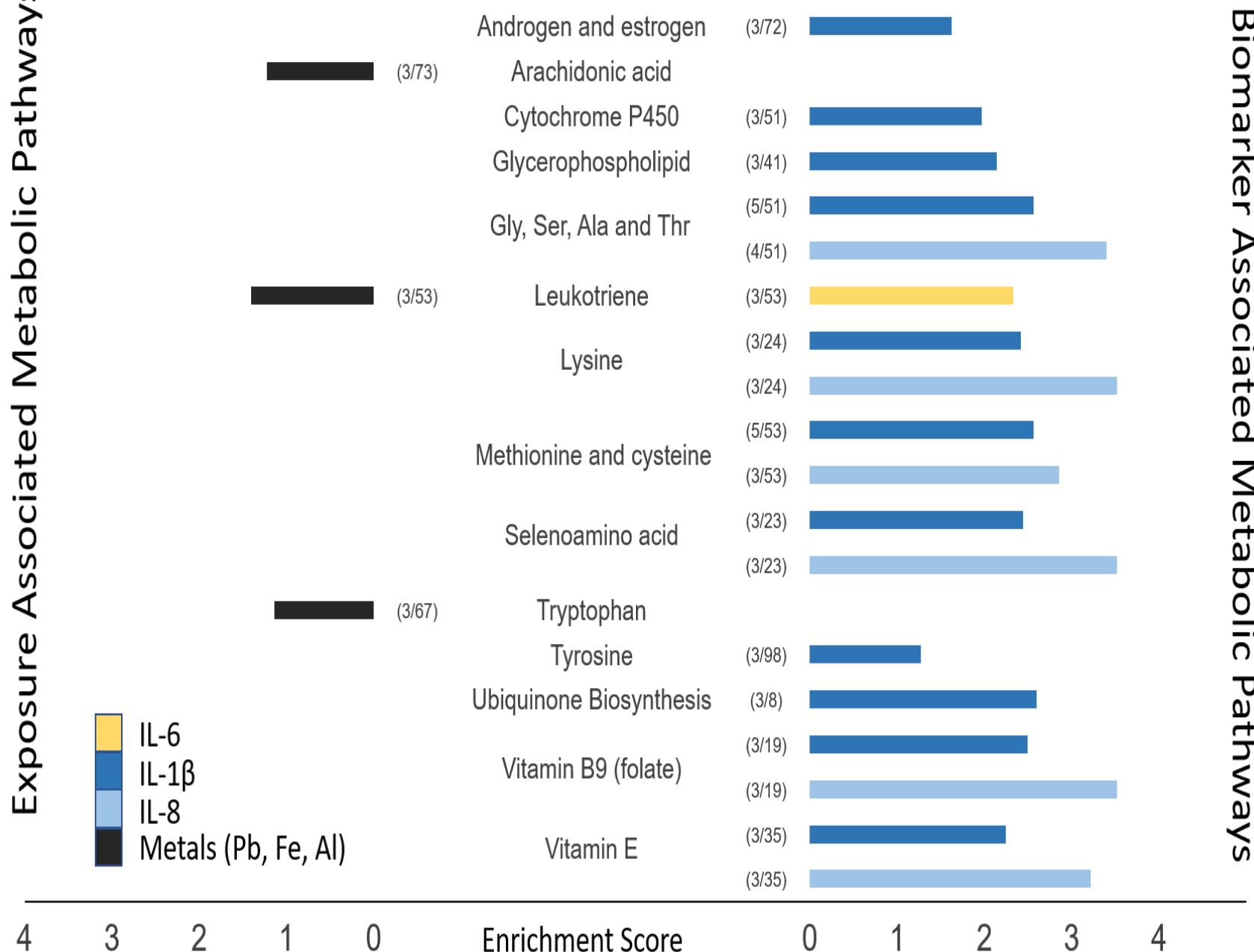- Particulate **Pb, Al, Fe** associated with metabolic changes
  - Pb: 150 features differed
  - Components indicative of road dust

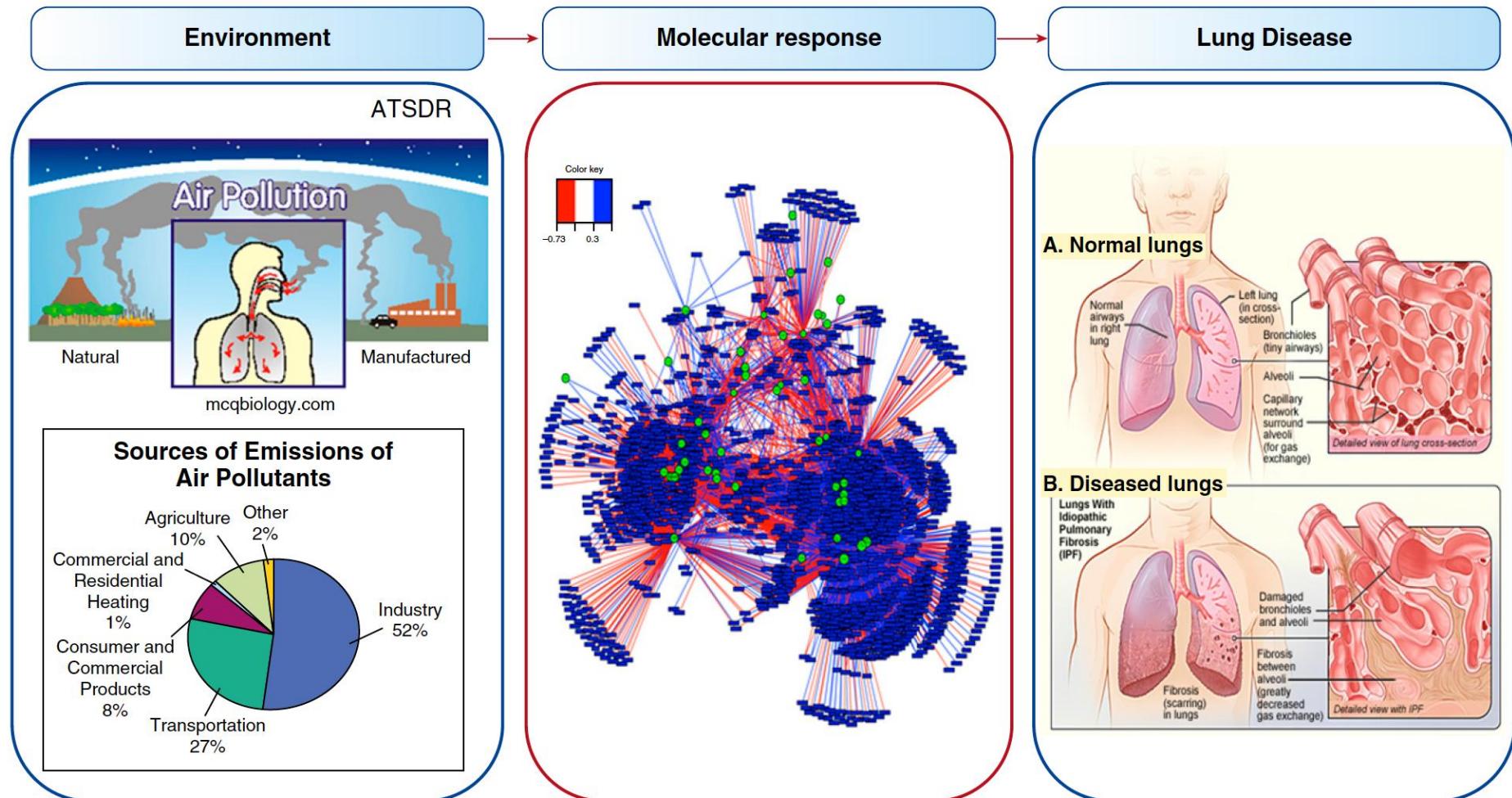- First evidence of pollutant-mediated metabolic response in ACE panel

- Y-axis: Significance of pre-post metabolite intensities
  - $(-\log_{10}(p))$
- X-axis: Retention time of individual ions (N = 14,000)

- Thick Dashed line: Bonferroni ($\alpha < 0.05$)
- Colored Points: FDR < 0.05
  - Direction of average pre-post changes in metabolite intensities indicated by red (decrease) and blue (increase)

Human Metabolic Pathway Enrichment

Exposure Associated Metabolic Pathways

Biomarker Associated Metabolic Pathways

Pathway	Exposure	Biomarker
Androgen and estrogen		(3/72)
Arachidonic acid	(3/73)	
Cytochrome P450		(3/51)
Glycerophospholipid		(3/41)
Gly, Ser, Ala and Thr		(5/51) (4/51)
Leukotriene	(3/53)	(3/53)
Lysine		(3/24) (3/24)
Methionine and cysteine		(5/53) (3/53)
Selenoamino acid		(3/23) (3/23)
Tryptophan	(3/67)	
Tyrosine		(3/98)
Ubiquinone Biosynthesis		(3/8)
Vitamin B9 (folate)		(3/19) (3/19)
Vitamin E		(3/35) (3/35)

Legend:
- IL-6
- IL-1β
- IL-8
- Metals (Pb, Fe, Al)

Enrichment Score

4 3 2 1 0   0 1 2 3 4

Figure 4. The cumulative lifelong perturbations in metabolic and cellular functions contribute to lung disease. The lungs are exposed to airborne particulates, environmental chemicals, and infectious agents. Within the lungs, the body burden of these environmental agents causes many types of molecular responses. *Central panel* reprinted by permission from Reference 85; *right panel* is from wikipdedia.org for idiopathic pulmonary fibrosis (https://en.wikipedia.org/wiki/Idiopathic_pulmonary_fibrosis). ATSDR = Agency for Toxic Substances and Disease Registry.

Go and Jones, 2016

Are we entering the early phase of a paradigm shift? YES
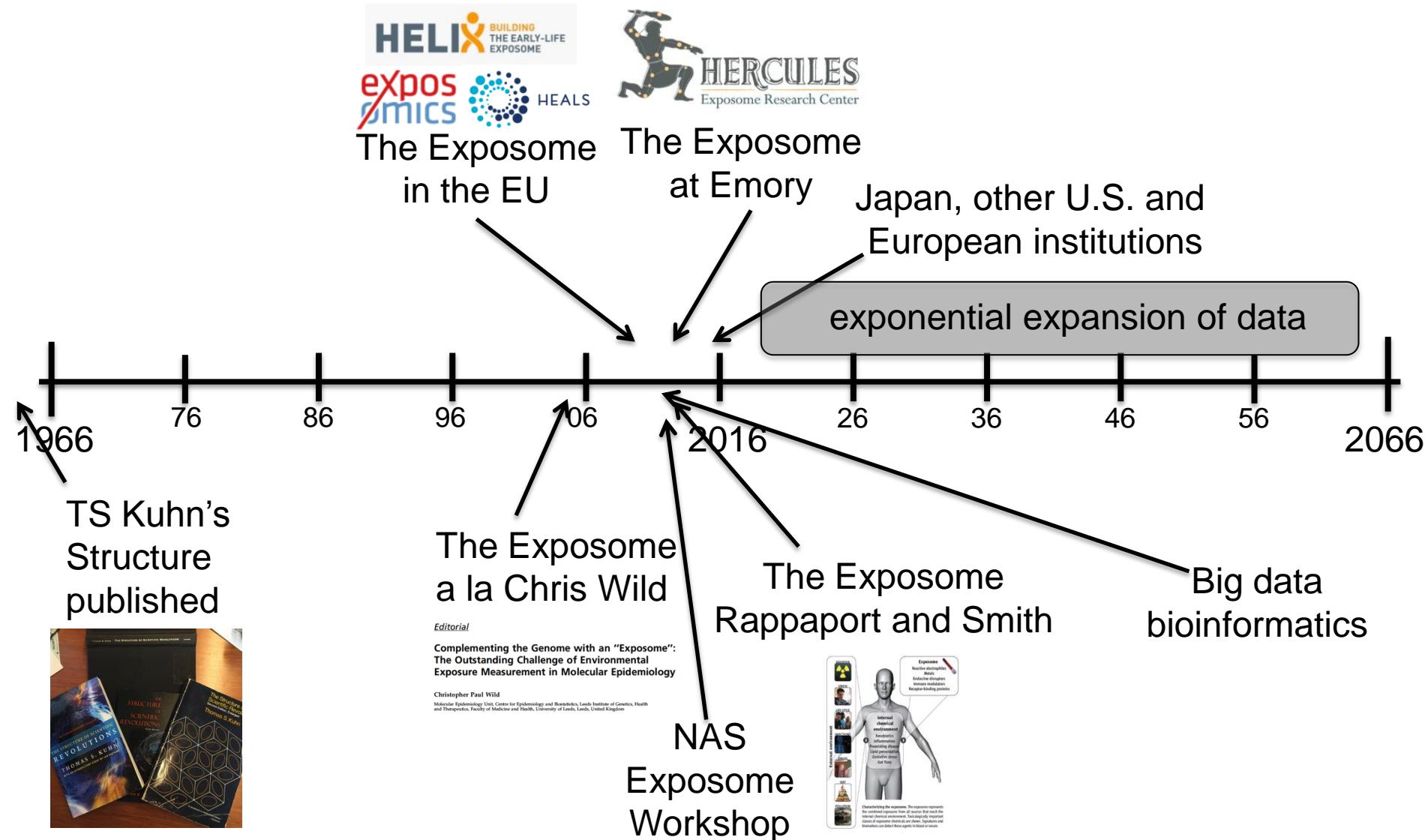
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EDITORIAL
Data Sharing in Toxicology: Beyond Show and Tell
Gary W. Miller[1]

EDITORIAL
More than Manuscripts: Reproducibility, Rigor, and Research Productivity in the Big Data Era
Lance A. Waller[1] and Gary W. Miller[2]

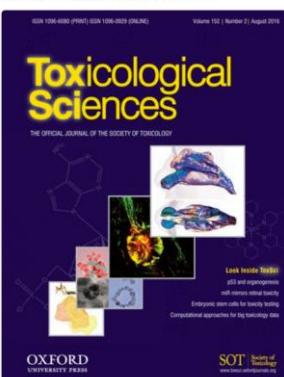EDITORIAL
Making Data Accessible: The Dryad Experience
Gary W. Miller[1]

EDITORIAL
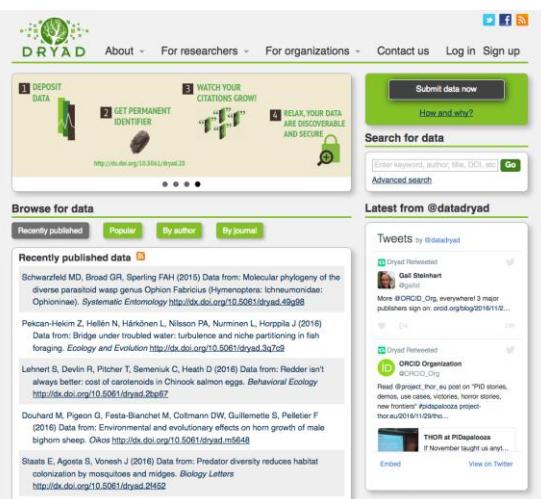## Young Investigators in Toxicology: Is There a Crisis?
Gary W. Miller

Superfund @ Duke @DukeSuperfund · Aug 1
Congrats Tony Luz & Joel Meyer (co-I on #Duke SRC's Proj. 3)! Their work is featured on cover of #ToxSci this month.

EDITORIAL
Preprints in Toxicology
Gary W. Miller[1,*,†]

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