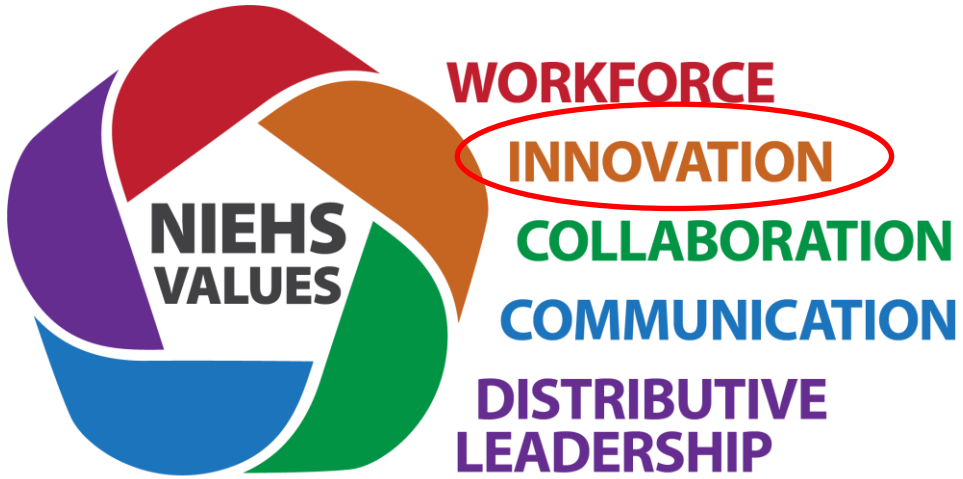
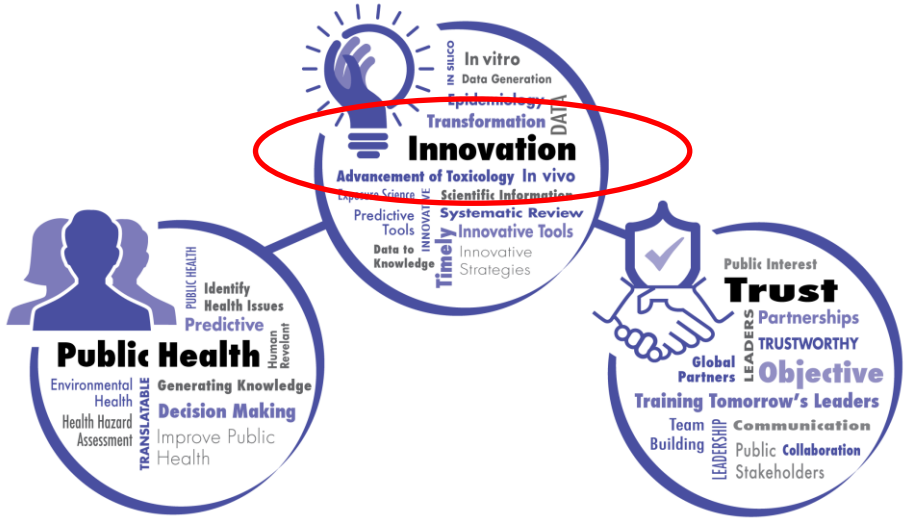
Looking Forward: Innovation in the NIEHS Division of the National Toxicology Program

Brian R. Berridge, DVM, PhD, DACVP
Division of the NTP
National Institute of Environmental Health Sciences

NTP Board of Scientific Counselors Meeting
December 8, 2021

Innovation as a Theme

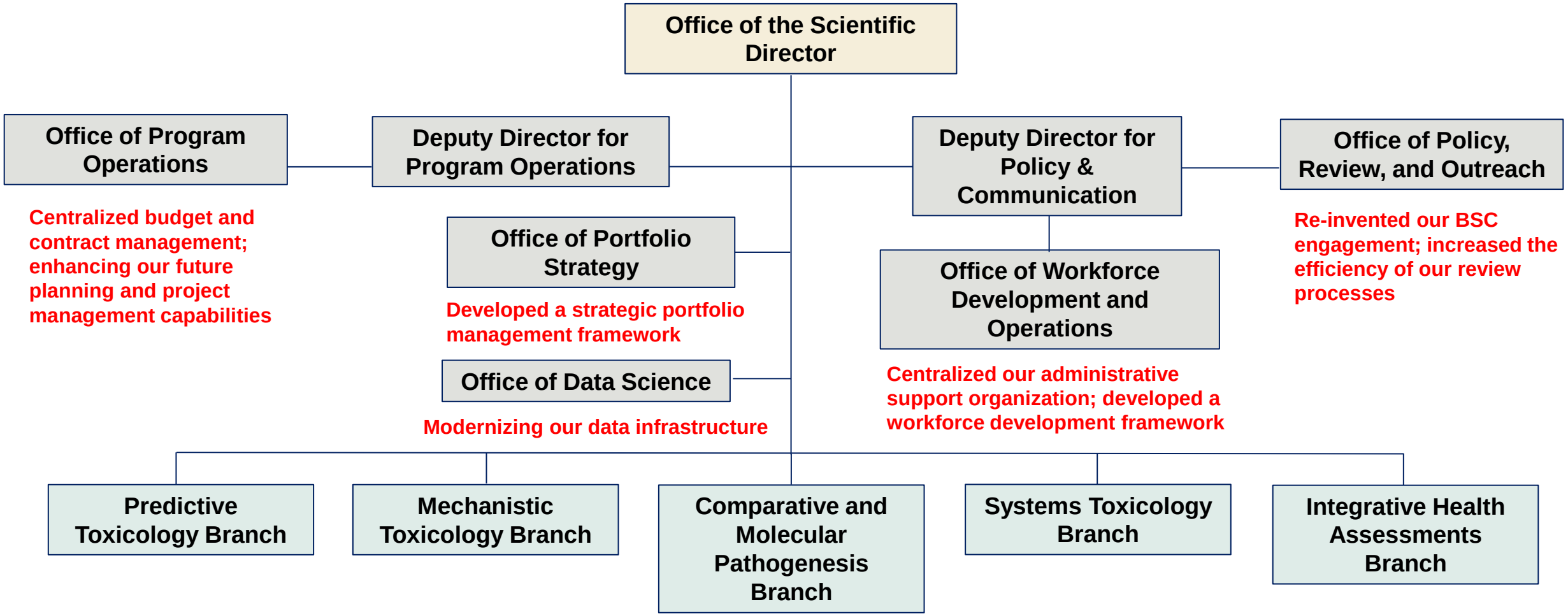




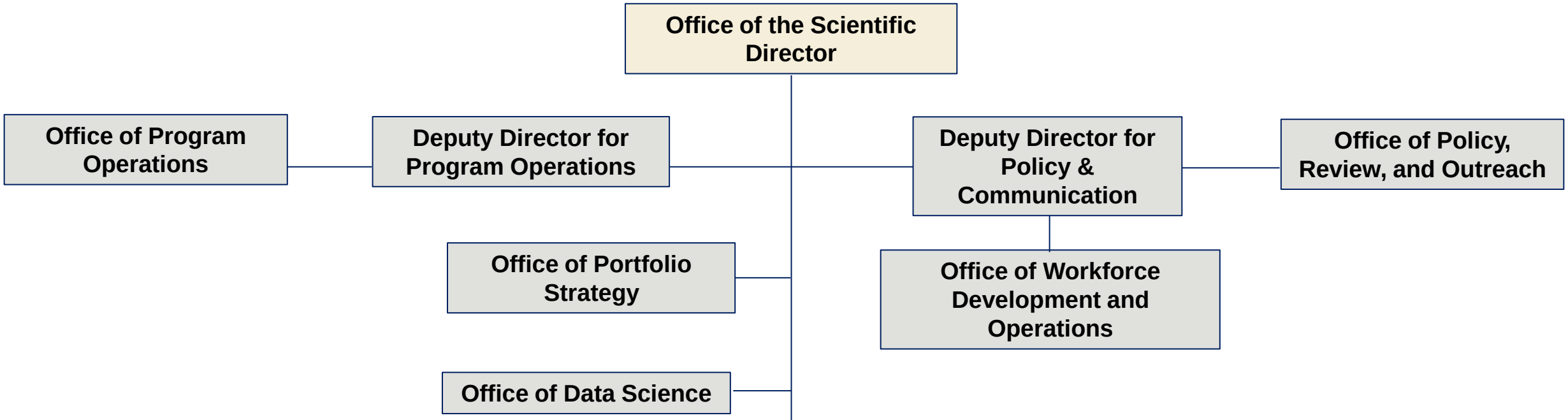
2018 DNTP Brand

"Lead the transformation of toxicology through the development and application of innovative tools and strategies."

History of Innovation



History of Innovation



Predictive Toxicology Branch

Built computational tools that enhance our use of bioactivity data; expanded Tox21; leadership in adoption of NAMs

Mechanistic Toxicology Branch

Added new capabilities and research focus; grew existing capabilities

Comparative and Molecular Pathogenesis Branch

Increased the efficiency of pathology review; expanded training program

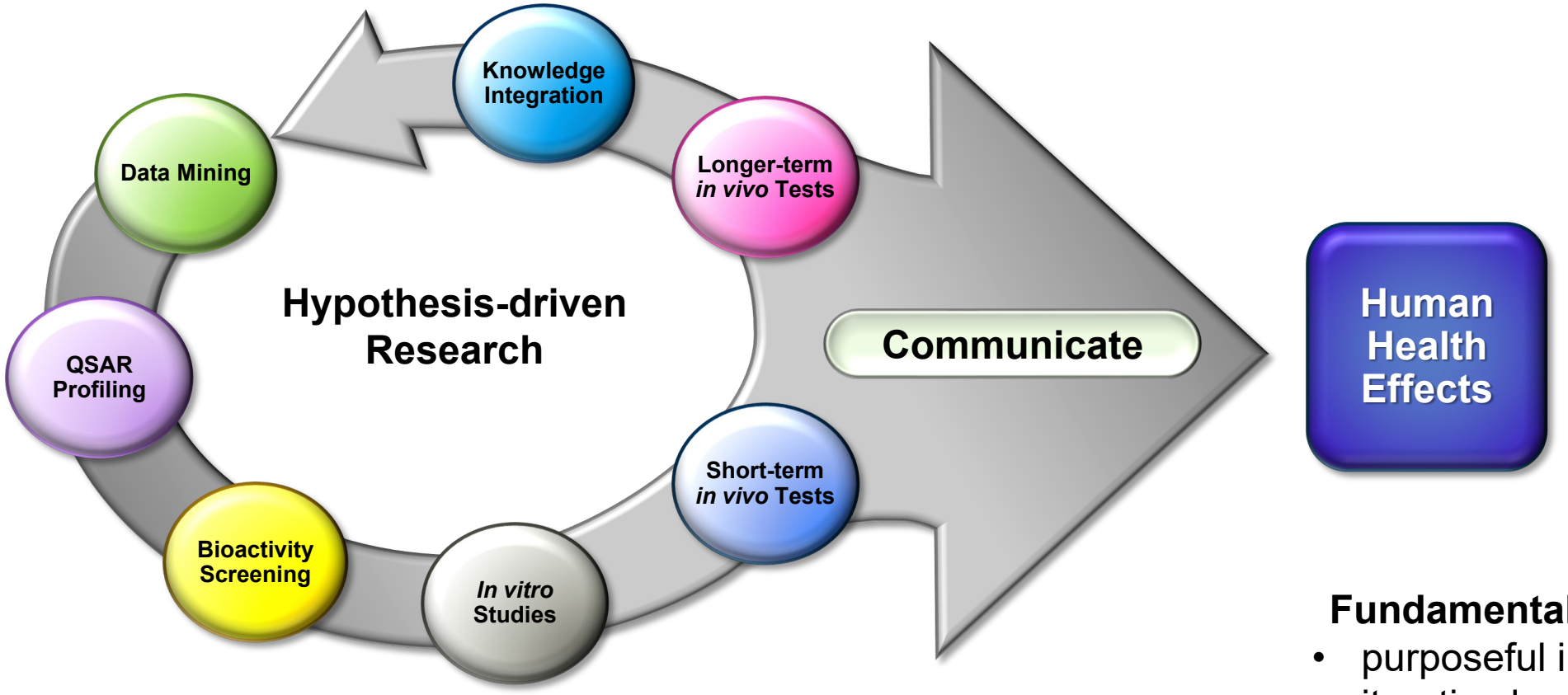
Systems Toxicology Branch

Enhanced assessment of life-stage susceptibility; expanded study type portfolio

Integrative Health Assessments Branch

Expanded literature-based assessment capabilities

Translational Toxicology Pipeline



It's not just about having the tools but also how you use them!

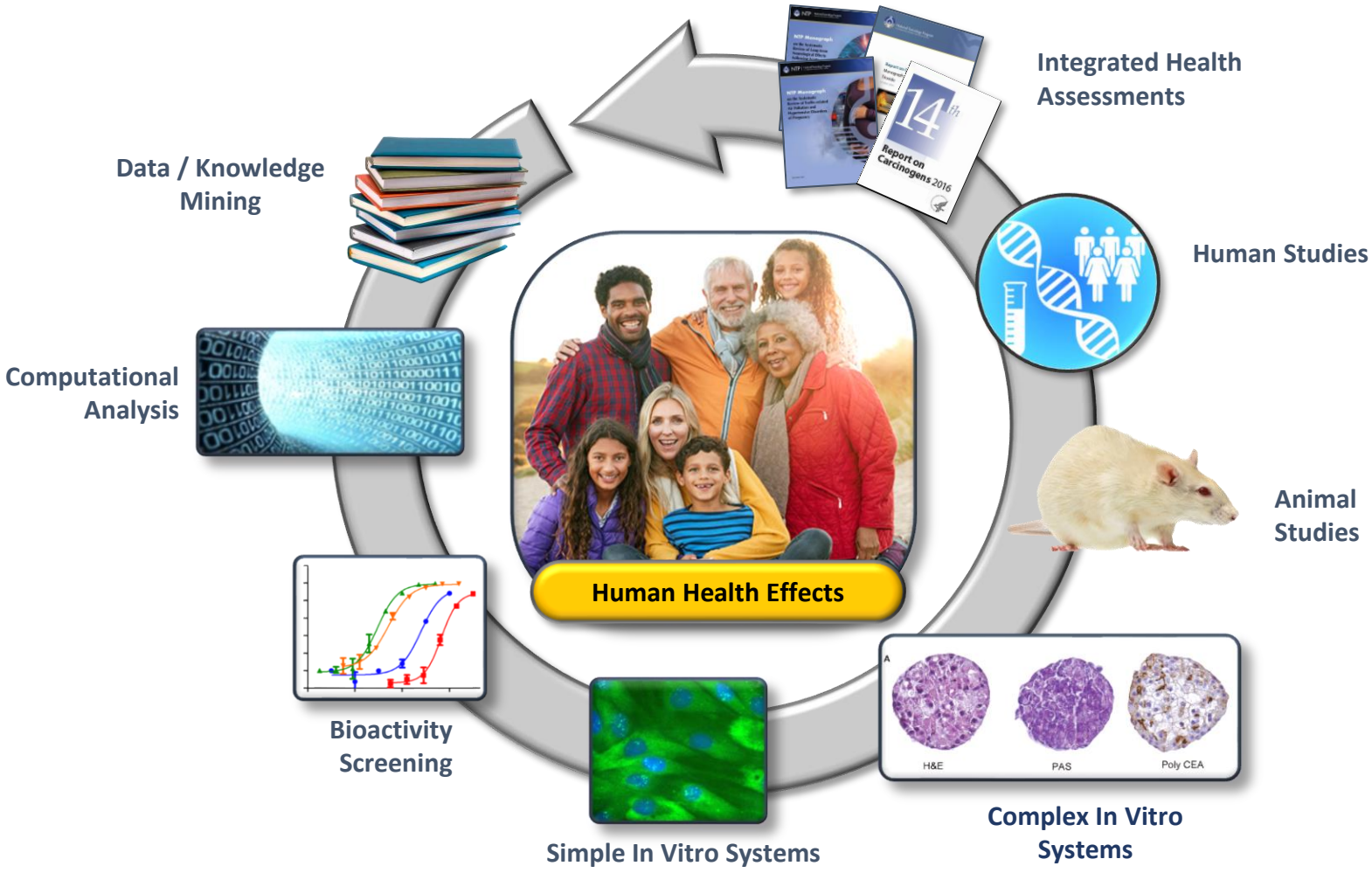
Fundamental changes

- purposeful integration
- iterative learning
- informed progression
- hypothesis-driven

Concept introduced in 2018

National Institute of Environmental Health Sciences
Division of the National Toxicology Program

Translational Toxicology Pipeline 2021



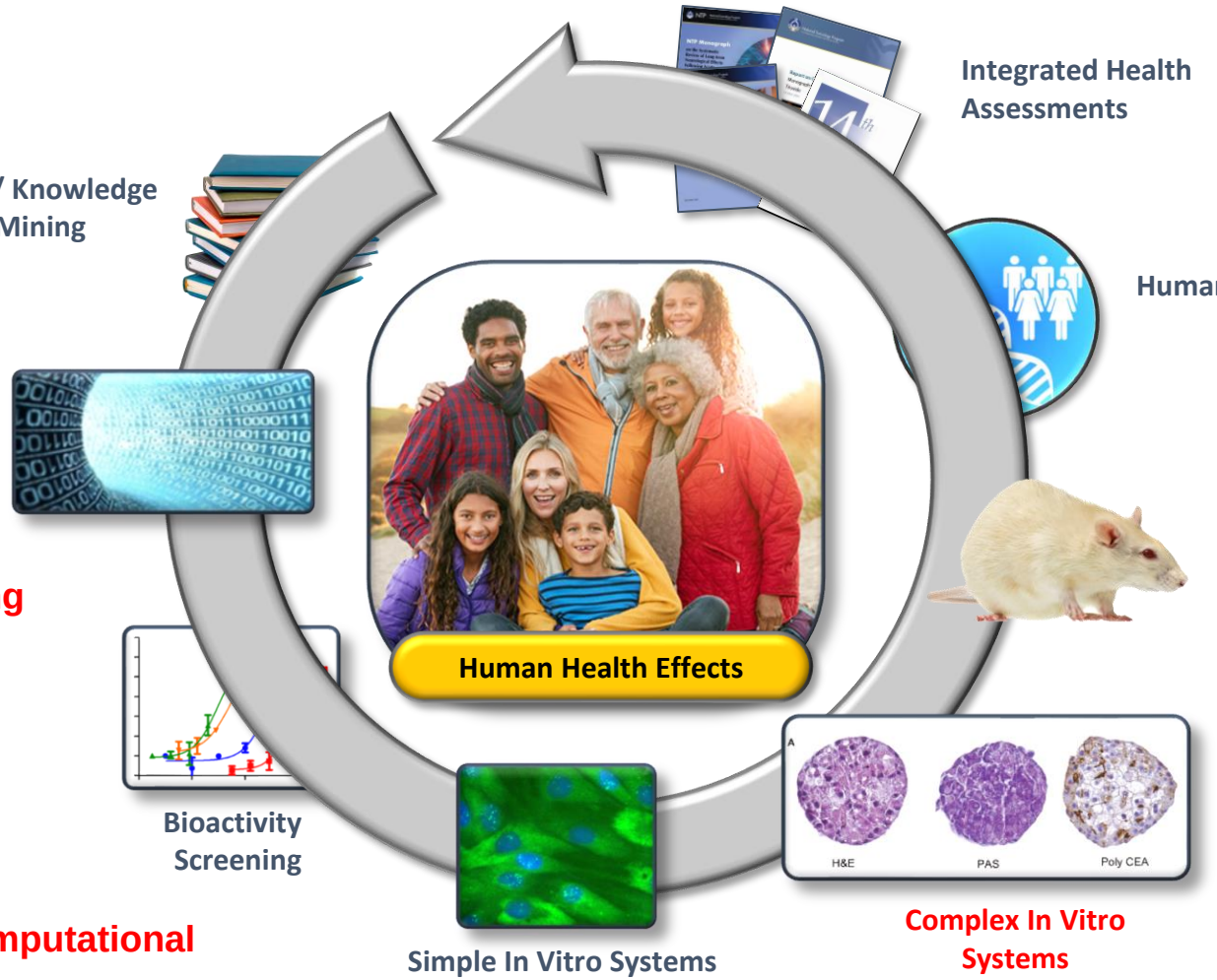
National Institute of Environmental Health Sciences
Division of the National Toxicology Program

Innovating the Translational Toxicology Pipeline

Incremental → Disruptive

Decision frameworks

AI-based automated knowledge mining



Human health assessments

Integrative physiological monitoring

Systems-based HTP screening

Histopathology AI/ML

In vitro models of human disease

Multi-scale computational modeling

Modeling host susceptibility/vulnerability

Advances in Evidence Informatics

- Leveraging informatics to support evidence-based decisions
  - Identify, adapt, and develop a toolbox of informatics approaches to advance our ability to turn data into knowledge for understanding health effects from environmental exposures
  - Improve workflow, reduce manual workload, identify tools for the range of DNTP users
  - Support synthesis through identification and categorization to better link mechanism to experimental and epidemiological data

Systems-based HTP Screening

Chemical Research in Toxicology

pubs.acs.org/crt Article

High-Throughput Screening to Identify Chemical Cardiotoxic Potential

Shagun Krishna, Brian Berridge, and Nicole Kleinstreuer*

Cite This: https://dx.doi.org/10.1021/acs.chemrestox.0c00382 Read Online

Bioactivity data sources

EPA's ToxCast- 1800 chemicals, 700+ assays

Tox21- 10K chemicals, 70+ assays

CV-relevant Bioactivity Targets

Defined biological framework

Literature mining

Slice	Target name	Effect	Reference	Slice Color
ADORA	Adenosine Receptor	Vasodialation, alterations in BP	Bowes et al., 2012	
ADR	Adrenergic Receptor	Arrhythmia, Alterations in BP	Bowes et al., 2012	
CHRM	Muscarinic Acetylcholine Receptor	Alterations in BP and HR, tachycardia	Bowes et al., 2012	
DRD	Dopamine Receptor	Alterations in BP and HR, Vascular relaxation	Bowes et al., 2012	
EDNR	Endothelin Receptor	Alterations in BP, Can exert adverse effects during	Bowes et al., 2012	
HTR	Serotonine Receptor	Alterations in BP, Potential cardiac valvulopathy	Bowes et al., 2012	
AVPR	Vasopressin Receptor	Alterations in BP and HR, Cardiac hypertrophy	Bowes et al., 2012	
HRH	Histamine Receptor	Positive inotropy	Bowes et al., 2012	
OPR	Opioid Receptor	Alterations in BP and Cardiac contritility	Bowes et al., 2013	
CHRNA	Cholinergic receptor	Alterations in BP and HR	Bowes et al., 2012	
SCN1A	Voltage-gated Sodium Channel	Slowed cardiac conduction; prolonged QRS interval	Bowes et al., 2012	
CACNA	Voltage-Gated Calcium Channel	Alterations in BP, QT prolongation, Arrhythmia	Bowes et al., 2012	
KCNH2	Potassium Voltage Gated Channel	QT prolongation	Bowes et al., 2012	
VEGF	Vascular Endothelial Growth Factor	Alterations in BP , Cardiac Ischemia	Touyz & Herrmann, 2018	
VascularTissue	Vascular Tissue	Myocardial ischemia, cardiac Arrhythmias		
OxidativeStress	Oxidative Stress	Cellular Hypertrophy; Cardiac Cell Death	Takimoto & Kass, 2007	
MtDysfunction	Mitochondrial Dysfunction	Cardiac dysfunction; Cardiomyopathy	Marin-Garcia, 2003	
TissueFactor	Tissue Factor	Alterations in BP and ventricular hypertrophy	Bode & Mackman, 2015	
PDE	Phosphodiesterase	Alterations in cardiac contractility, HR and BP	Bowes et al., 2012	
MAO	Monoamine Oxidase	Alterations in BP	Bowes et al., 2012	
JNK	c-Jun N-terminal kinase	Vascular injury, cardiac hypertrophy	Muslin, 2008	
TyrKinase	Tyrosine Kinase	Alterations in BP, LV dysfunction, conduction abnormalities, QT prolongation	Lamore, Kohnken, Peters, & Kolaja, 2020	
AroPro	Aromatase Protein	Ischemic heart disease	Khosrow-Khavar et al., 2017	
ACHE	Acetylcholinesterase	Alterations in BP and HR	Bowes et al., 2012	
COX	Cyclooxygenase	Myocardial infarction; Alteration in BP; Ischaemic stroke; Atherothrombosis	Bowes et al., 2012	
ERAlpha	Estrogen receptor Alpha	Abnormal cardiac contractility, cardiac hypertrophy	Pugach, Blenck, Dragavon, Langer, & Leinwand, 2016	
NR3C1	Glucocorticoid receptor	Alterations in BP; Arrhythmia	Bowes et al., 2012	
PPARG	Peroxisome Proliferator Activated Receptor Gamma	Cardiac hypertrophy , Atherosclerosis	Das & Chakrabarti, 2006	
AHR	Aryl hydrocarbon receptor	Endothelial dysfunction, Atherosclerosis	Wu et al.,2011	
AP1	Activator protein1	Atherosclerosis	Meijer et al., 2012	
HIF1A	Hypoxia Inducible Factor1Alpha	Ischaemia disease	Semenza, 2014	
NFKB	NF Kappa B	Atherosclerosis	Fiordelisi et al, 2019	
TP53	Tumor Protein p53	Alteration in cardiac function	Mercer & Bennett, 2006	
ICAM-1	Intercellular adhesion molecule-1	Markers of endothelial dysfunction	Boyd et al., 2008	
IL6	Interleukin 6	Markers of inflammation and oxidative stress	Chu et al., 2020	
t-PA	Tissue Type plasminogen activator	Markers of endothelial dysfunction	Mason, 2017	
PAI -1	Plasminogen activator inhibitor	Markers of endothelial dysfunction	Mason, 2017	
NPA	Natriuretic peptide A	Release in response to elevation in LV filling pressure and wall stress		
SAA1	Serum amyloid A1	Direct promotion of vascular dysfunction throu vascular tissues		
SLC6A	Serotonin transporter	Pulmonary Hypertension,Cardiac Arrhythr Valve Abnormalities	Bowes et al., 2012	

Krishna, S., et al.. (2020). *Chemical Research in Toxicology*.

e.g., Tributyltin Chloride Bioactivity

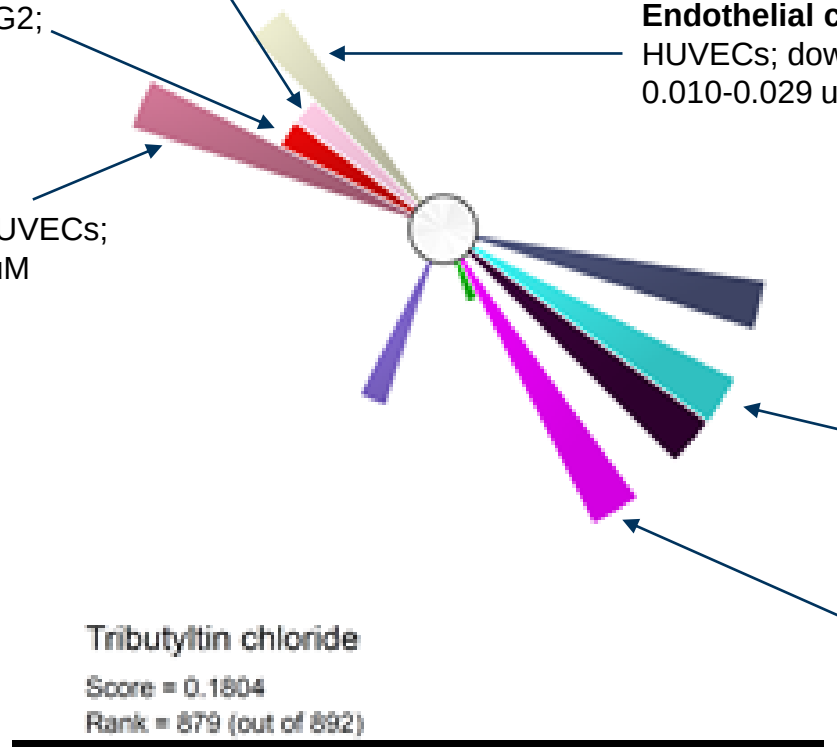
Uses: marine anti-fouling paints, antifungal, plastics stabilizer

Oxidative stress; HepG2; AC50 = 0.097 uM

Mito Dysfunction; MMP in HepG2; AC50 = 0.086 uM

Endothelial cell activation; activated HUVECs; down-regulation; AC50 = 0.010-0.029 uM

Tissue Factor; LPS-activated HUVECs; down-regulation; AC50 = 0.012 uM



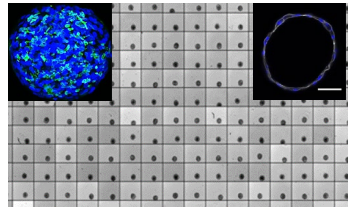
PAI-1; bronchial epi cells; down-regulation; AC50 = 0.023 uM

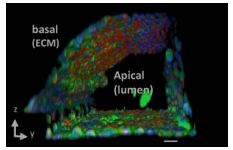
ICAM-1; cytokine-activated HUVECs; down-regulation; AC50 = 0.014 uM

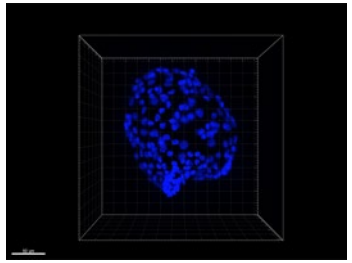
National Institute of Environmental Health Sciences
Division of the National Toxicology Program

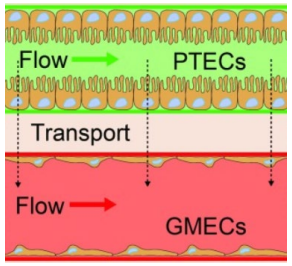
Complex In Vitro Systems

Human Microphysiological System Exploration & Qualification → **Assays & Analytics** → **Application & Human Translation**

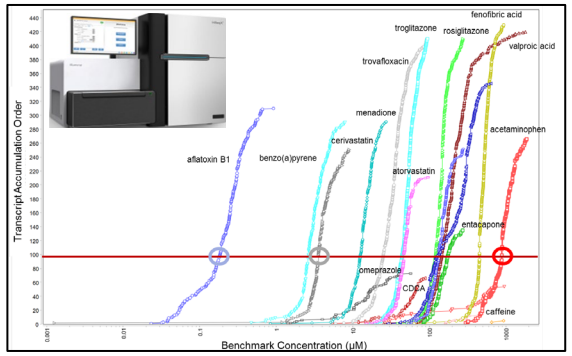


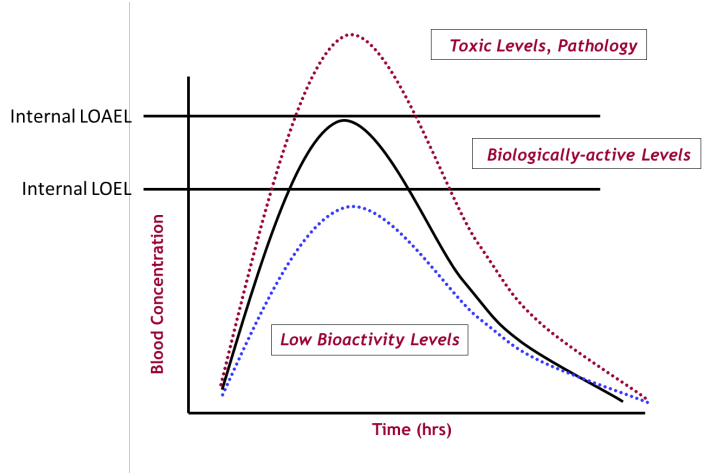


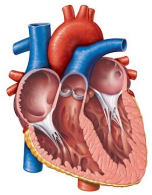




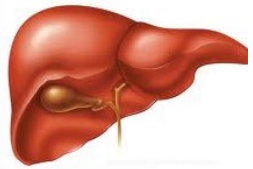
Lin et al. 2019



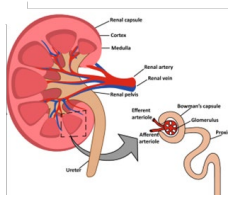


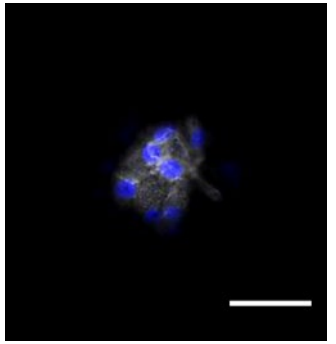


- Cardiovascular injury
- Cellular & molecular mechanisms of pathophysiological response
- Human translation



- Metabolic activation of toxicity
- Hepatic filtration & metabolism
- Cellular & molecular mechanisms of pathophysiological response
- Human translation

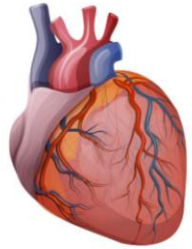




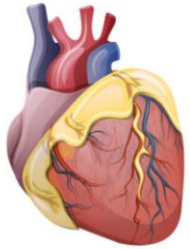
- Renal toxicity of the proximal tubule
- Proximal tubule transport & metabolism
- Renal clearance & bioaccumulation
- Human translation

National Institute of Environmental Health Sciences
Division of the National Toxicology Program

In Vitro Models of Human Disease

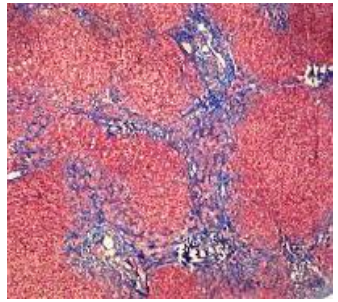


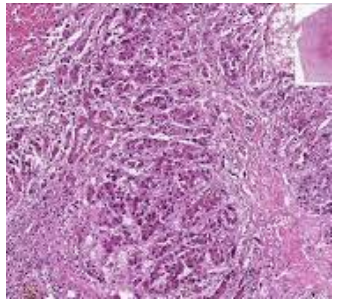
healthy heart

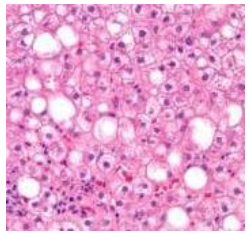


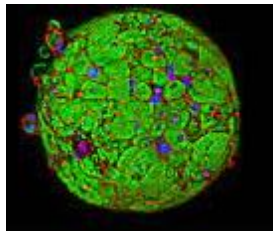
obesity heart

- Disease state susceptibility (Hypertrophic Cardiomyopathy, Long QT syndrome, dilated cardiomyopathy etc.)
- Inflammation
- Fibrosis
- Hypoxia, I/R injury

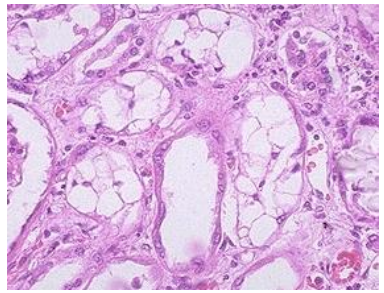


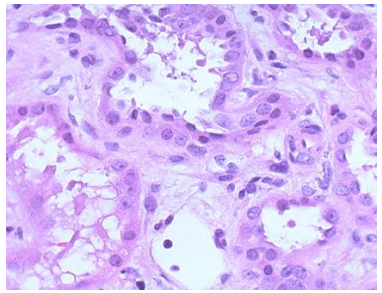


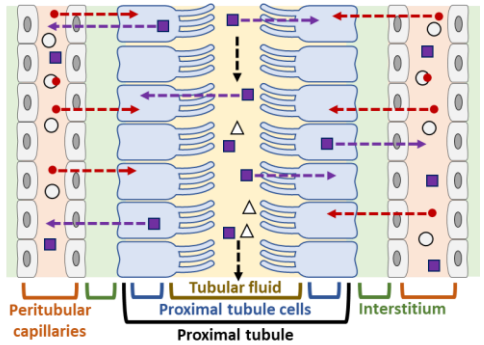




- Hepatocellular carcinoma (HCC)
- Susceptibility from non-alcoholic fatty liver disease, hyperglycemia
- Fibrosis
- Genetic disorders & metabolism
- Interindividual susceptibility

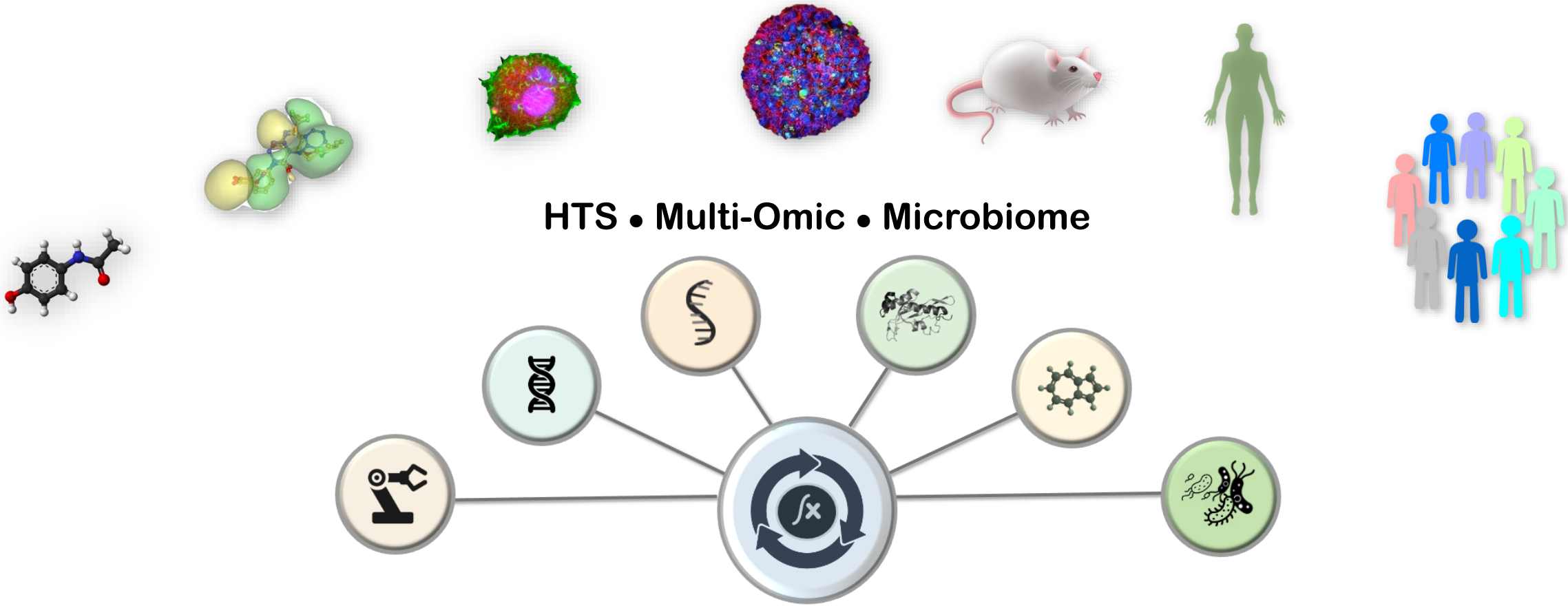






- Renal proximal tubule
  - Chronic hyperglycemia
  - Hypoxia
  - Dehydration & CKDu
  - Fibrosis
- Renal cell carcinoma (RCC)
- Genetic disorders

Multiscale Modeling



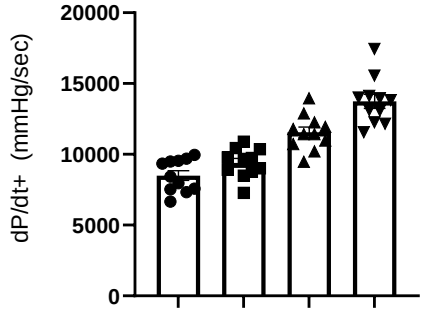
- Translation
- Prediction
- Genome-Scale
- Kinetic
- Sex as a biological variable
- Population variability and susceptibility

Functional Reserve as a Toxicologic Endpoint

CV Hazards of HIV Therapeutics

SD Rats dosed by gavage beginning at GD6

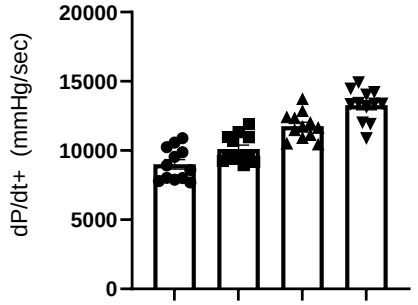
Vehicle Group



- no DOB
- 1 mg/Kg/min DOB
- 3.3 mg/Kg/min DOB
- 10 mg/Kg/min DOB

Dams @ weaning

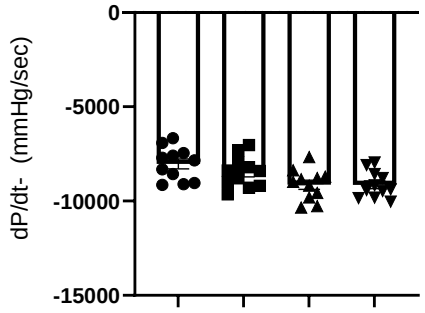
High Dose Group



- no DOB
- 1 mg/Kg/min DOB
- 3.3 mg/Kg/min DOB
- 10 mg/Kg/min DOB

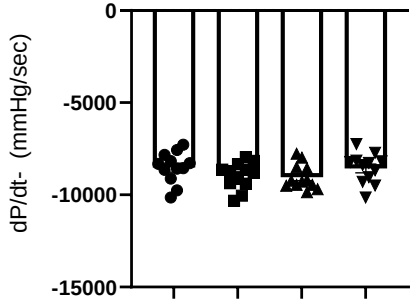
Test agent = HIV triple drug combination
Dobutamine = beta-adrenergic agonist; positive inotrope
dP/dt+ = measure of contractile performance
dP/dt- = measure of myocardial relaxation

Vehicle Group



- no DOB
- 1 mg/Kg/min DOB
- 3.3 mg/Kg/min DOB
- 10 mg/Kg/min DOB

High Dose Group



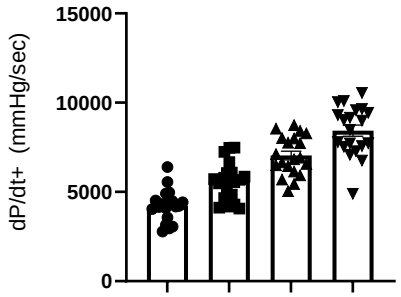
- no DOB
- 1 mg/Kg/min DOB
- 3.3 mg/Kg/min DOB
- 10 mg/Kg/min DOB

Functional Reserve as a Toxicologic Endpoint

CV Hazards of HIV Therapeutics

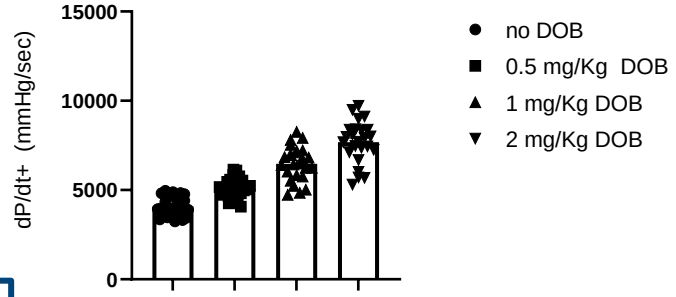
SD Rats dosed by gavage beginning at GD6

Vehicle Group



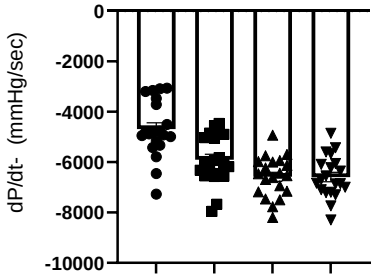
Pups @ weaning

High Dose Group

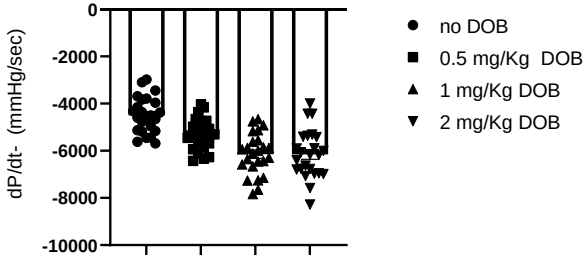


Test agent = HIV triple drug combination
Dobutamine = beta-adrenergic agonist; positive inotrope
dP/dt+ = measure of contractile performance
dP/dt- = measure of myocardial relaxation

Vehicle Group

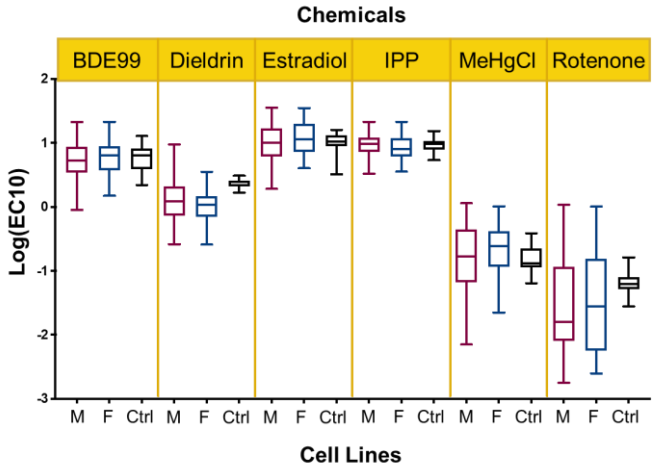


High Dose Group



Genetic Susceptibility

- Tox21 partner project testing an *in vitro* DO panel of unique neural progenitor cell lines for determining empirically based toxicodynamic variability factors.
  - Variability in cytotoxicity data, was comparable to that seen with human cells lines
  - Current work using cell painting data in over 100 DO cell lines to investigate sub-cytotoxic effects that can be quantified using high content imaging.
- Utility of DO to assess population variability *in vivo* for select agents for hazard ID, characterization and dose-response
  - JAX collaboration for short term toxicity studies in DO mice



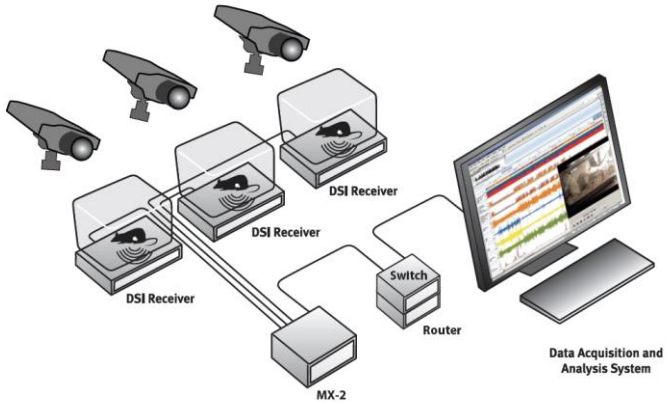
Chemical	TDVF05 (90% CI)	
	DO Mouse NPCs	Human LCLs[1]
IPP	1.71 (1.60, 1.86)	-
Estradiol	1.82 (1.66, 2.05)	-
BDE 99	2.39 (2.00, 2.96)	-
Dieldrin	2.80 (2.42, 3.33)	3.76
Default factor = 3.16		
Rotenone	11.2 (7.51, 19.1)	-
MeHgCl	26.9 (10.3, 109)	16.03

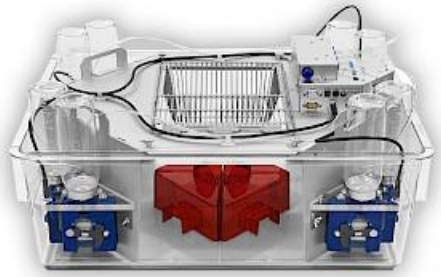
Unpublished data provided by Alison Harrill (NICHD)

Integrative Physiological Monitoring

Animal study outcomes can be significantly improved by contextualizing mechanistic outcomes in a dynamic and continuous assessment of physiology and behavior

- Utilizing a holistic approach that is
  - Integrative
    - Expanded focus beyond a single organ system
  - Translational
    - Focused on translational clinically relevant endpoints
  - Continuous and automated
    - Leveraging emerging technologies to collect more data in an automated way
- Collaboration with DIR investigators and other ICs
  - Establishment of a dedicated animal research core at NIEHS

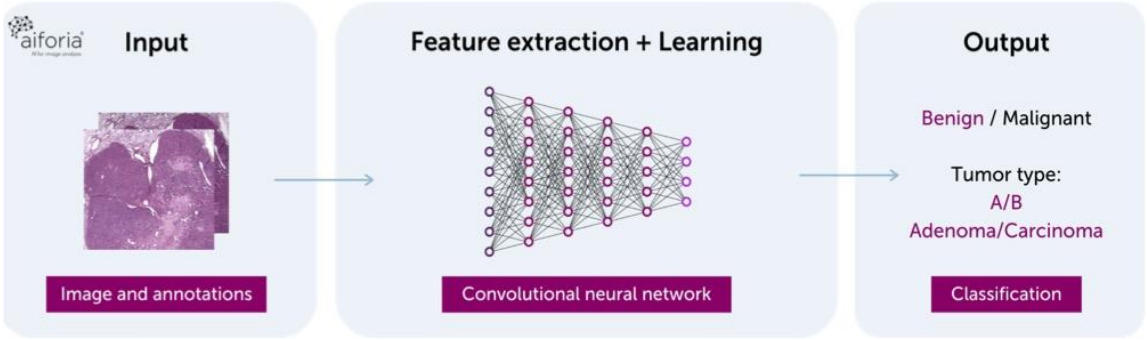


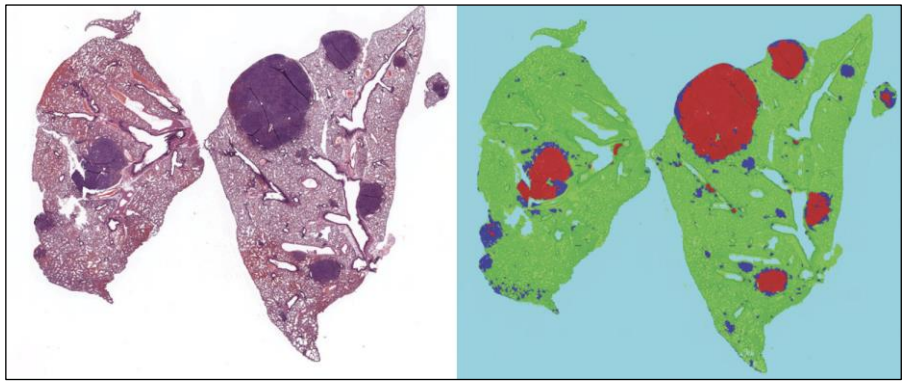


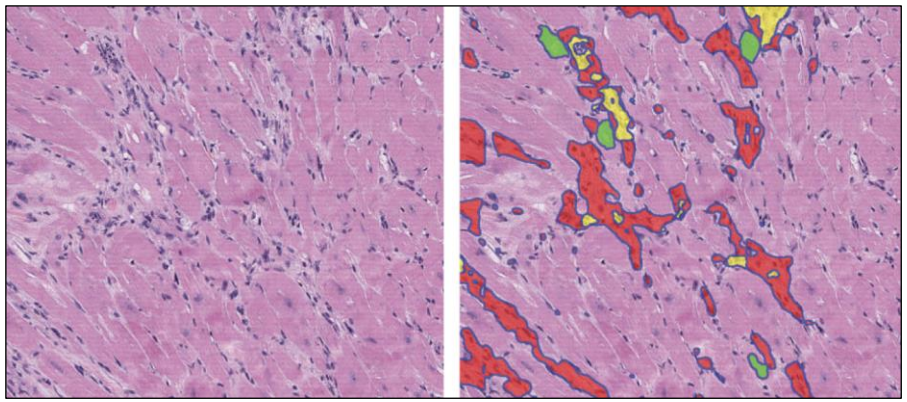
National Institute of Environmental Health Sciences
Division of the National Toxicology Program

Artificial Intelligence in Toxicologic Pathology

- Artificial Intelligence (AI) now routinely used in diagnostic toxicologic pathology
- Automated, faster, reduce costs, improve diagnostic accuracy, consistency, and workflow
- Being used to screen, detect, and diagnose histopathological lesions
- Establishing an AI Core in CMPB to use this innovative technology for DNTP/NIEHS
  - PhD contractor scientist hired to provide expertise
  - Initiated a continuing education seminar series in AI in summer 2021
- Completed a study developing and training an AI algorithm to diagnose mouse lung tumors

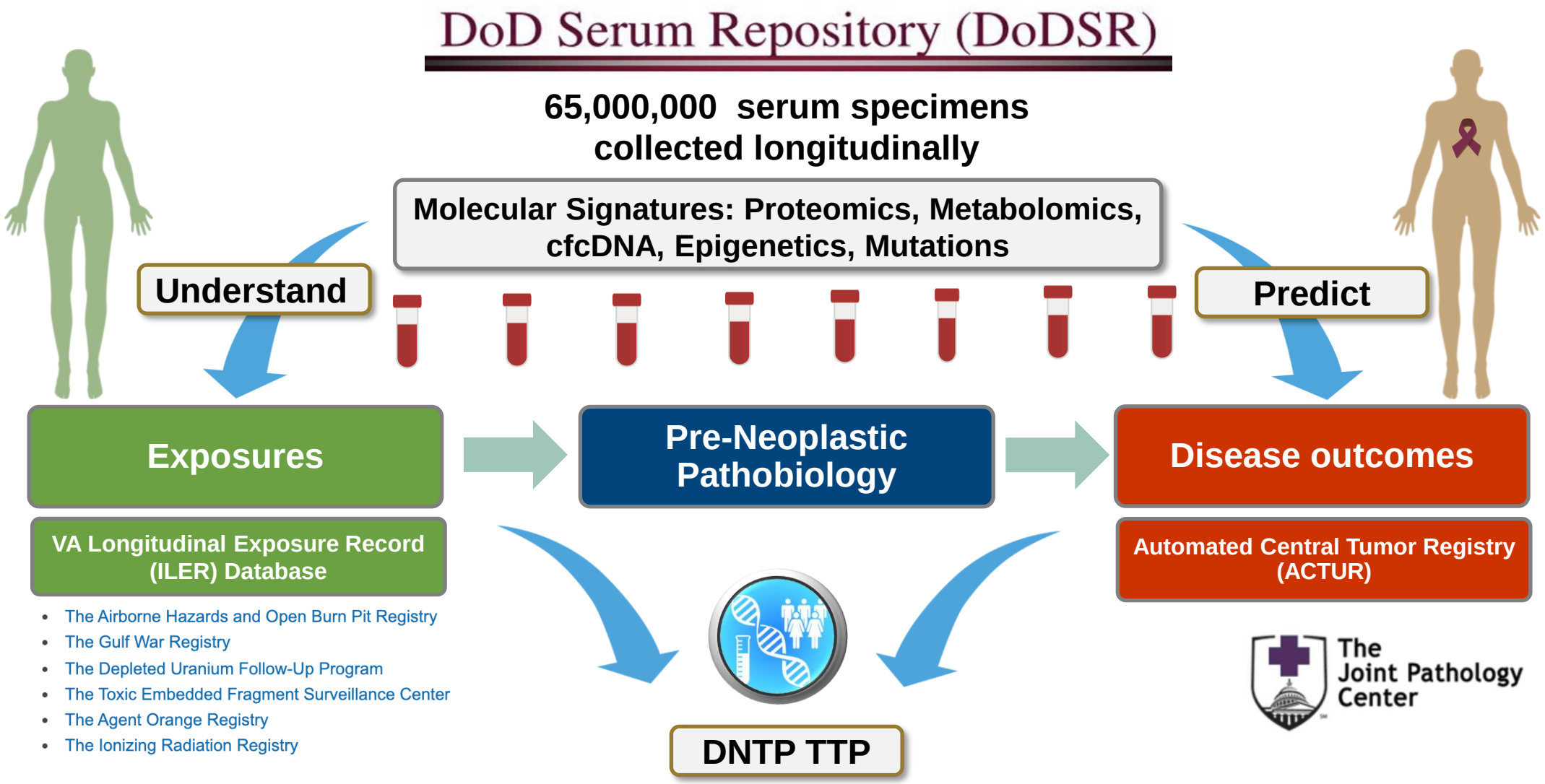






Translational Human Research

Linking Exposures to Disease



Summary

- There is a long history of innovation at DNTP where we've actively and continuously refined our organizational processes and structure as well as been leaders in developing novel approaches to environmental hazard assessment.
- Despite the innumerable distractions of the last few years, we've significantly increased our efforts to innovate the way toxicology is applied in hazard identification and characterization.
  - We've embraced our intent to be more predictive and translational.
- All of our efforts to innovate the way we operate and execute our science are aligned to contemporary problems we're trying to solve.
- We look forward to sharing the outcomes of these efforts as we share the progress and outcomes of our strategic and prioritized portfolio.

Thank you!

Questions?