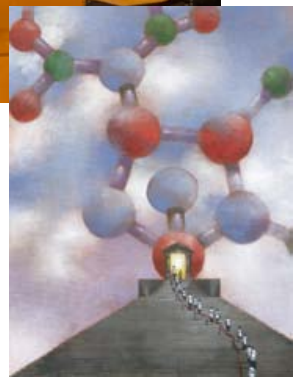
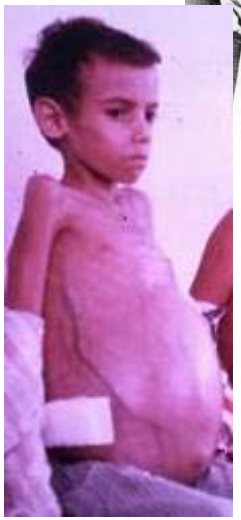


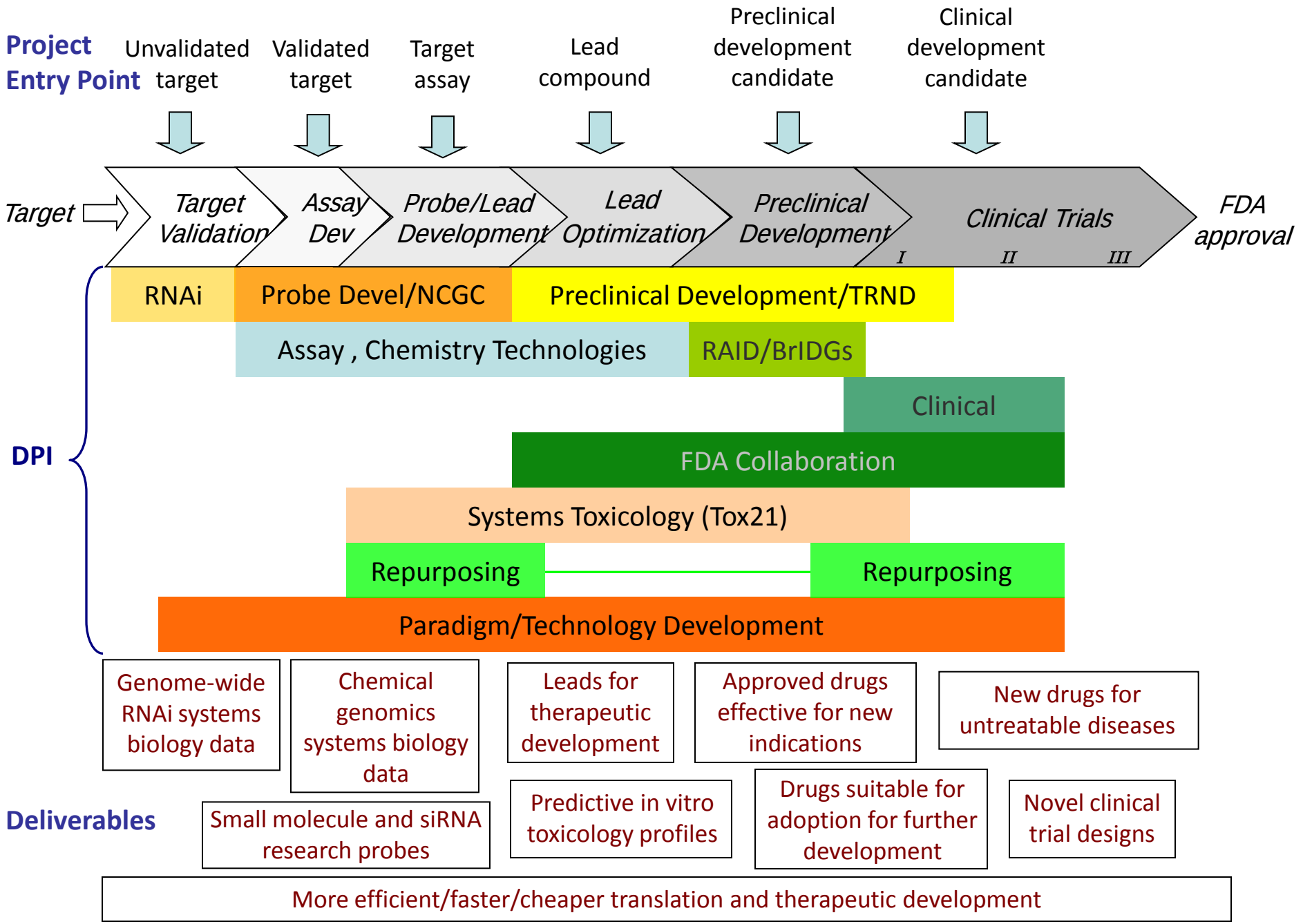
The NCATS Pharmaceutical Collection: Potential Use for Rapid Repurposing Against New/Emerging Threats



Anton Simeonov, Ph.D.
Division of Preclinical Innovation
National Center for Advancing Translational Sciences (NCATS)
National Institutes of Health

BARDA Industry Day, Washington, DC
December 10, 2012

The NCATS Division of Preclinical Innovation: An Integrated Pipeline

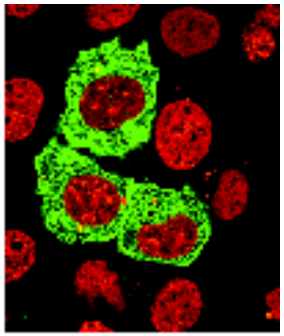


Range of screening assays performed

Extent of reductionism

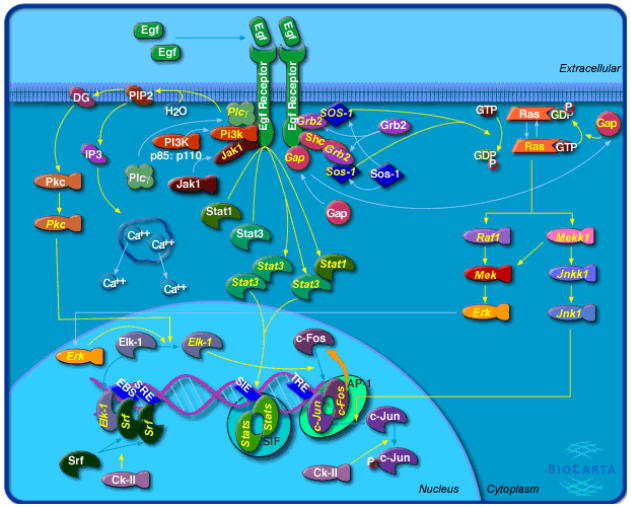
Phenotype

(Image-based HCS, GFP, etc)



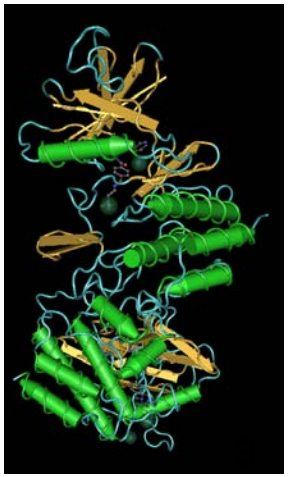
Pathway

(Reporters, e.g., luciferase, β-lactamase)



Protein

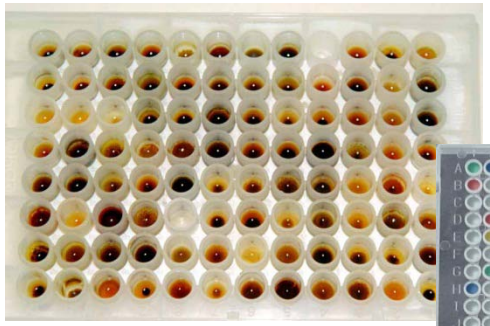
(Enzyme readouts, interactions, etc)



Screening Formats

c

r



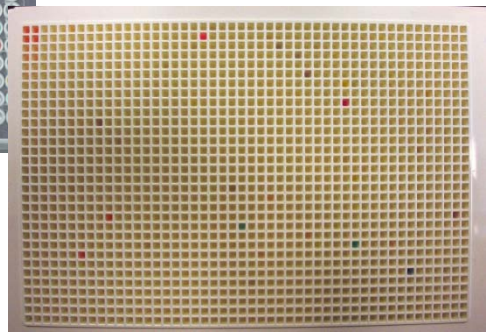
- 8 rows x 12 columns
- 88 test samples

96-well plate

- 16 rows x 32 columns
- 352 test samples

384-well plate

4 x 96-well plates



- 32 rows x 48 columns
- 1,408 test samples

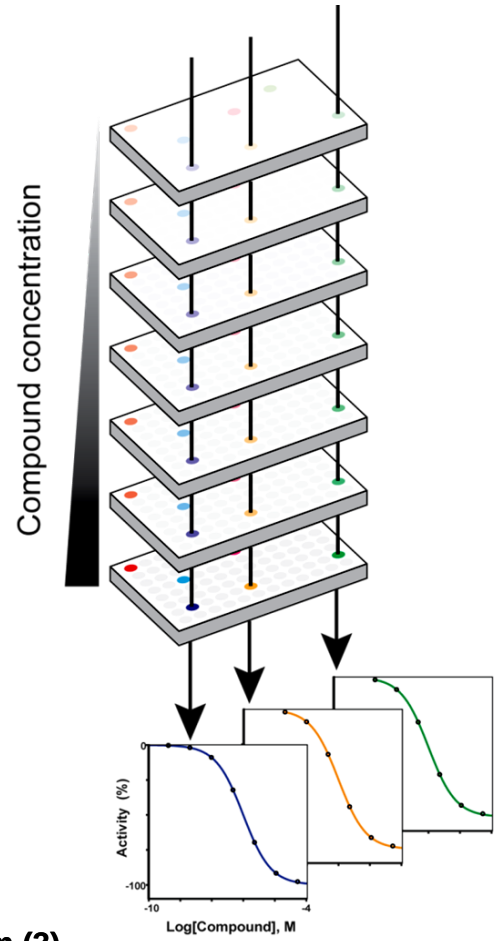
1536-well plate

16 x 96-well plates

If @ 100 microtiter plates per day:

Plate format	samples§/day (wells/day)	Time to screen 1 M samples
96-well	8,800 (9,600)	4 months
384-well	35,200 (38,400)	4 weeks
1536-well	**140,800** (153,600)	**7 days**

Integrated Robotic Screening System



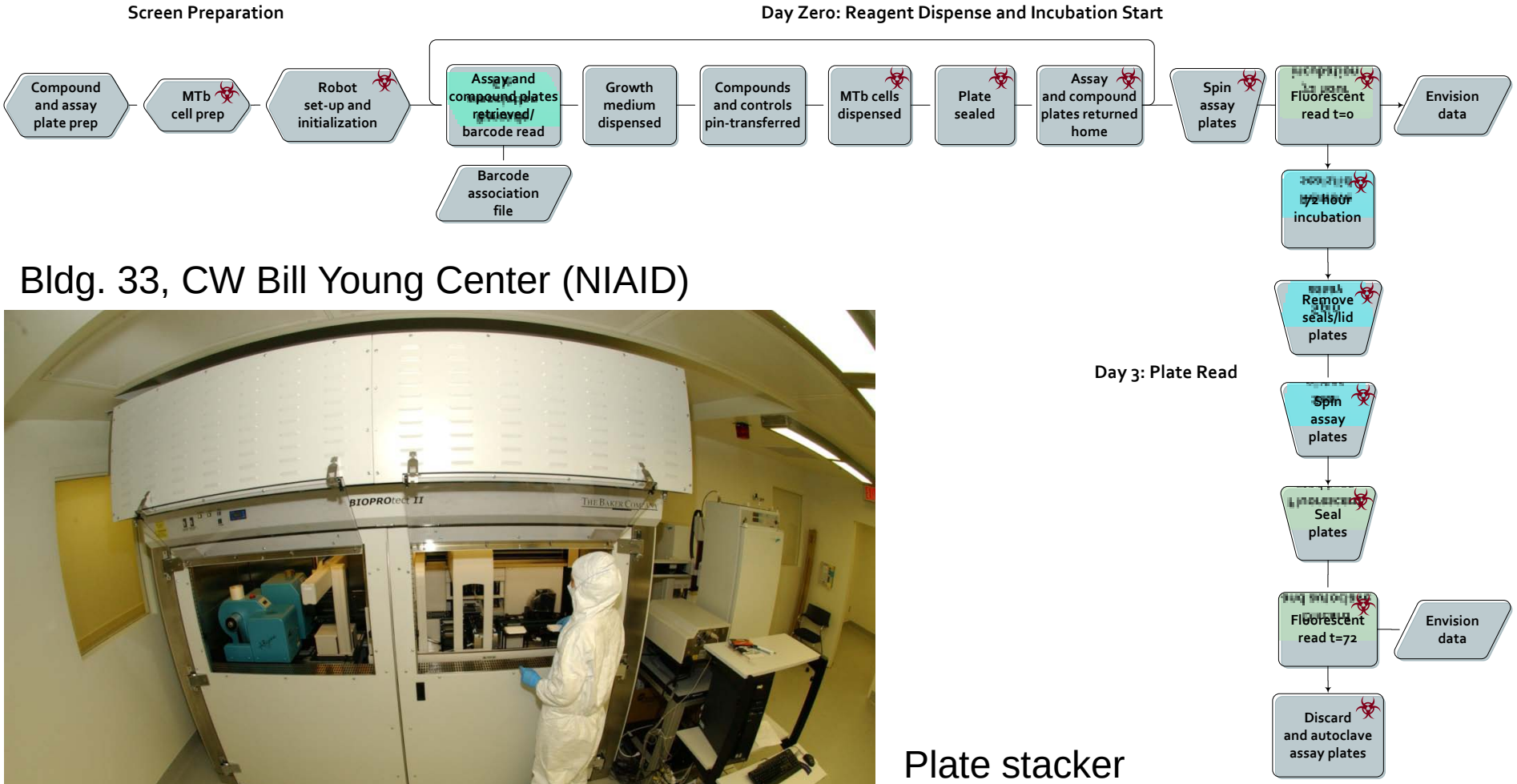
- All screens performed as multipoint titration series
- In total, ~500,000 compounds across multiple sub-libraries
- >250 collaborative projects with investigators worldwide

Automated concentration-response data collection for every sample tested

PNAS, 2006, **103,** 11473-11478

Assay Drug Dev. Technol., 2008, **6,** 637-658

HTS System in BSL-3 Facility, NIH Main Campus

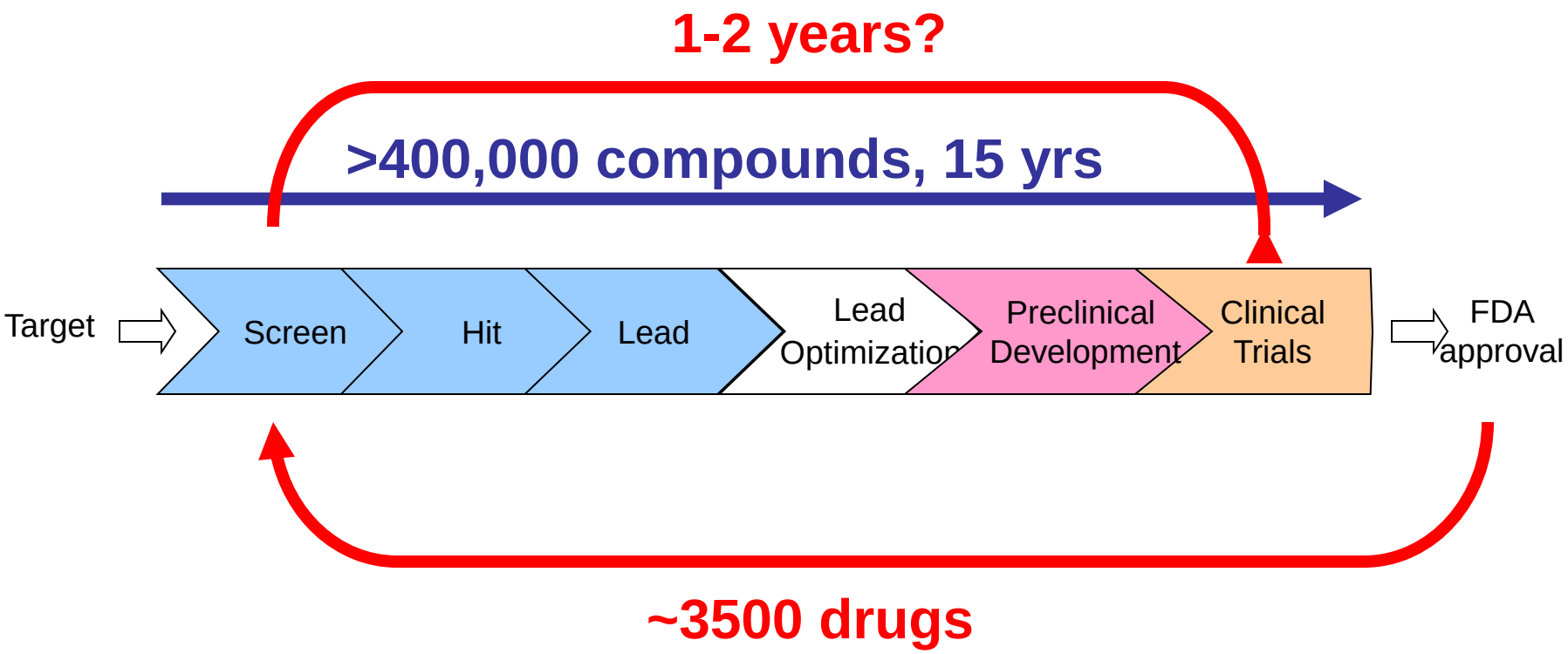


Bldg. 33, CW Bill Young Center (NIAID)

13,000-compound collection screened in dose-response mode against *M. tuberculosis*

Plate stacker
Abgene plate sealer
Biomek NX Single arm: 96MC head
Anaerobic chamber with airlock
EnVision plate reader
Dual bed series nitrogen generator

Two Approaches to Therapeutics



Enabling Comprehensive Drug Repurposing

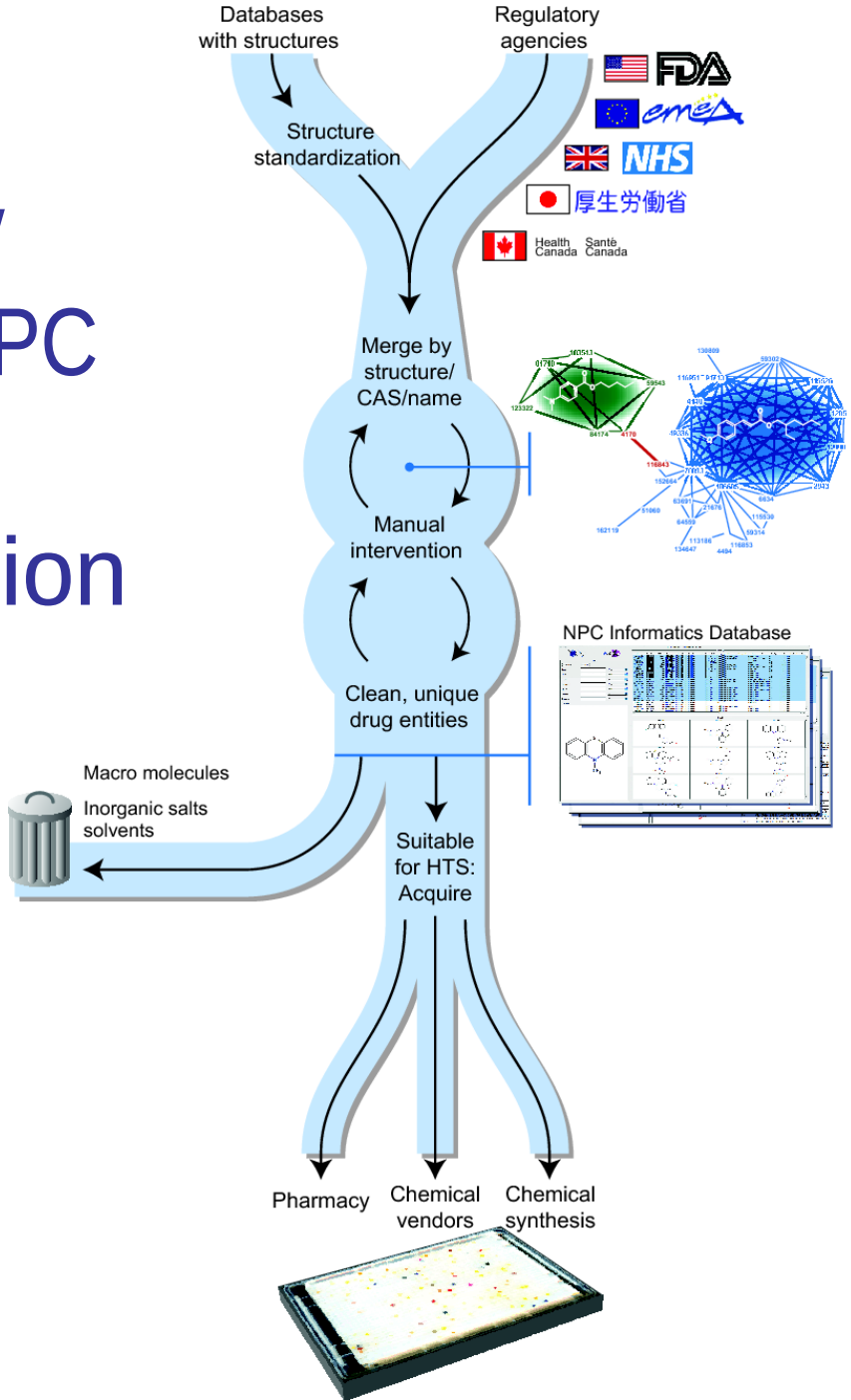
The NCGC Pharmaceutical Collection: A Comprehensive Resource of Clinically Approved Drugs Enabling Repurposing and Chemical Genomics

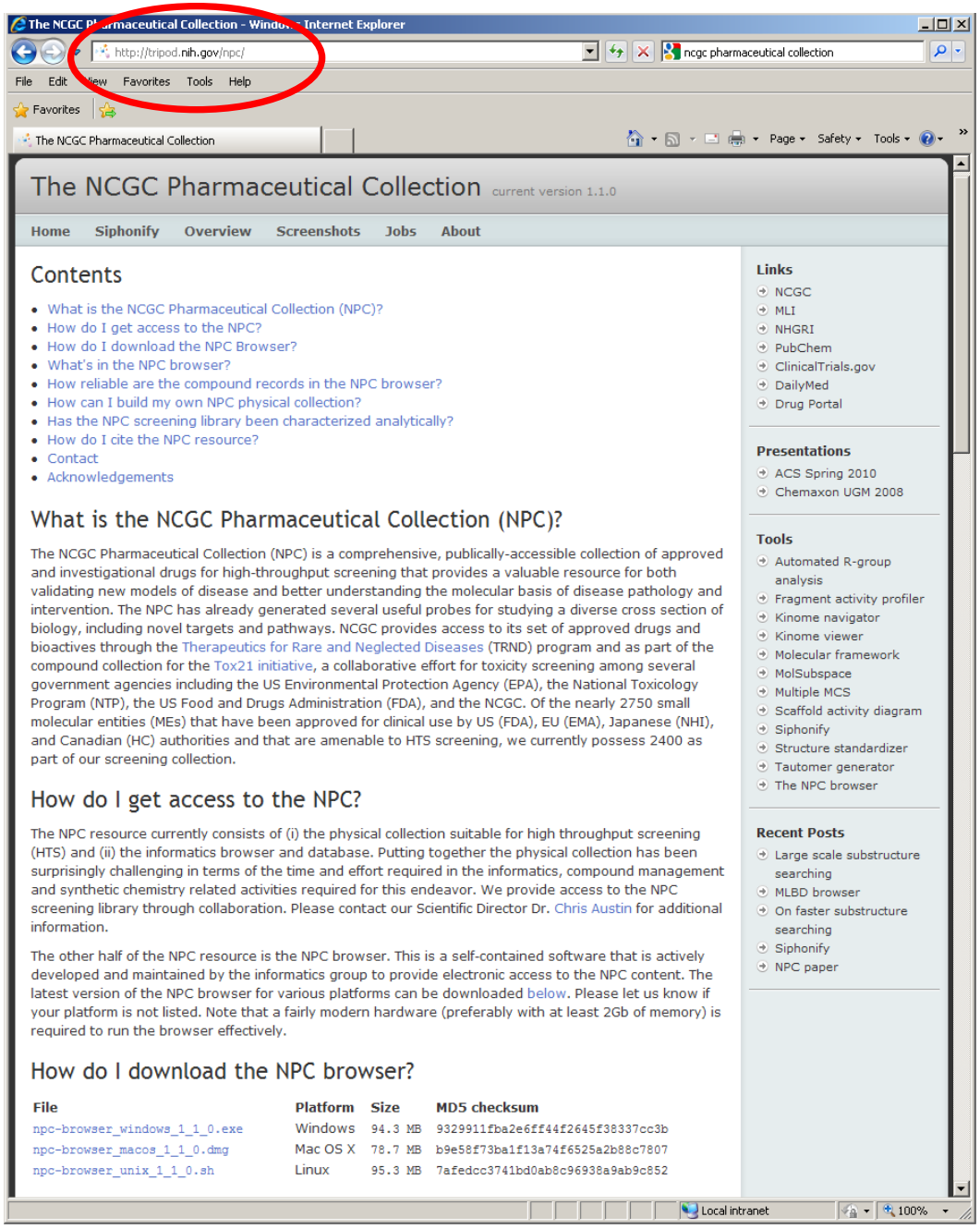
Ruili Huang,* Noel Southall,* Yuhong Wang, Adam Yasgar, Paul Shinn, Ajit Jadhav, Dac-Trung Nguyen, Christopher P. Austin†

Small-molecule compounds approved for use as drugs may be "repurposed" for new indications and studied to determine the mechanisms of their beneficial and adverse effects. A comprehensive collection of all small-molecule drugs approved for human use would be invaluable for systematic repurposing across human diseases, particularly for rare and neglected diseases, for which the cost and time required for development of a new chemical entity are often prohibitive. Previous efforts to build such a comprehensive collection have been limited by the complexities, redundancies, and semantic inconsistencies of drug naming within and among regulatory agencies worldwide; a lack of clear conceptualization of what constitutes a drug; and a lack of access to physical samples. We report here the creation of a definitive, complete, and nonredundant list of all approved molecular entities as a freely available electronic resource and a physical collection of small molecules amenable to high-throughput screening.

www.ScienceTranslationalMedicine.org 27 April 2011 Vol 3 Issue 80 80ps16

Workflow for the NPC library construction process





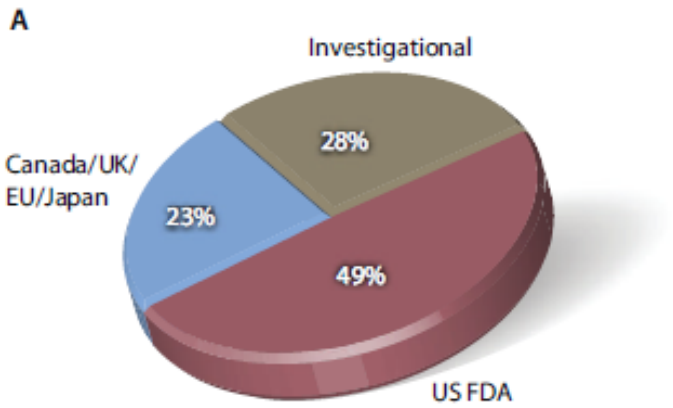
http://tripod.nih.gov/npc/

Procurement Sources

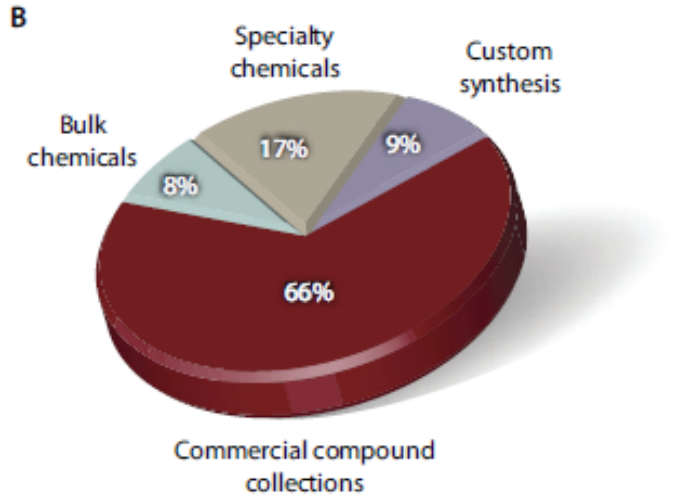
Supplier Name	Supplier Type
Advanced Technology & Industrial Co., Ltd	Specialty Chemicals
AKos Consulting and Solutions GmbH	Specialty Chemicals
Apin	Specialty Chemicals
Apollo Scientific Ltd	Specialty Chemicals
ART-CHEM GmbH	Specialty Chemicals
ASDI Inc.	Specialty Chemicals
Aurora Fine Chemicals	Specialty Chemicals
Beta Pharma Inc	Specialty Chemicals
BioAustralis	Specialty Chemicals
Bionet Research (Owned by Key Organics)	Specialty Chemicals
BIOTREND Chemicals AG	Specialty Chemicals
Bosche Scientific, LLC	Specialty Chemicals
Chemical Block Ltd.	Specialty Chemicals
Chemos	Specialty Chemicals
ChemPacific Corp.	Specialty Chemicals
ChemSampCo	Specialty Chemicals
CHESS	Specialty Chemicals
ClVentiChem	Specialty Chemicals
Epsilon Chimie	Specialty Chemicals
HuskerChem	Specialty Chemicals
INDOFINE Chemical Company, Inc	Specialty Chemicals
Kemprotec Limited	Specialty Chemicals
Labotest	Specialty Chemicals
LKT Laboratories, Inc	Specialty Chemicals
Matrix Scientific	Specialty Chemicals
MDD World Molecules	Specialty Chemicals
Menai Organics	Specialty Chemicals
Molecular Diversity Preservation Intl.	Specialty Chemicals
National Cancer Institute	Specialty Chemicals
Oakwood Producst, Inc.	Specialty Chemicals
Peakdale Molecular Ltd	Specialty Chemicals
Pharmeks LTD.	Specialty Chemicals
PolyPeptide Group (formerly NeoSystem SA)	Specialty Chemicals
Scientific Exchange	Specialty Chemicals
Selleck	Specialty Chemicals
Sequoia Research Product LTD	Specialty Chemicals
SynphaBase AG	Specialty Chemicals
Tripos	Specialty Chemicals
Tyger Scientific, Inc	Specialty Chemicals
Vitas-M Laboratory Ltd.	Specialty Chemicals

Supplier Name	Supplier Type
Alfa Aesar	Bulk Chemicals
Asinex Ltd.	Bulk Chemicals
CalBioChem	Bulk Chemicals
ChemBridge Corporation	Bulk Chemicals
ChemDiv, Inc	Bulk Chemicals
Enamine	Bulk Chemicals
Innovapharm Ltd.	Bulk Chemicals
InterBioScreen Ltd.	Bulk Chemicals
Maybridge	Bulk Chemicals
SigmaAldrich - ALDRICH	Bulk Chemicals
SigmaAldrich - FLUKA	Bulk Chemicals
SigmaAldrich - RIEDEL	Bulk Chemicals
SigmaAldrich - SALOR	Bulk Chemicals
SigmaAldrich - SIGMA	Bulk Chemicals
SigmaAldrich - Sigma DiscoveryCPR	Bulk Chemicals
Specs	Bulk Chemicals
Tocris Bioscience	Bulk Chemicals
American Custom Chemicals Corporation	Custom Synthesis
APAC Pharmaceutical, LLC	Custom Synthesis
Florida Center for Heterocyclic Compounds	Custom Synthesis
GVK Biosciences	Custom Synthesis
NIH Center for Chemical Genomics	Custom Synthesis
Pharmaron	Custom Synthesis
University of Pittsburgh UPCMLD	Custom Synthesis
Henry Schein	Pharmacies
National Instititute on Drug Abuse	Pharmacies
United States Pharmacopeial Convention, Inc.	Pharmacies
Walter Reed	Pharmacies
Ambinter	Screening Libraries
BIOMOL	Screening Libraries
Microsource	Screening Libraries
Prestwick	Screening Libraries
SigmaAldrich - LOPAC	Screening Libraries
Tim Tec, Inc	Screening Libraries
Toronto Research Chemicals	Screening Libraries

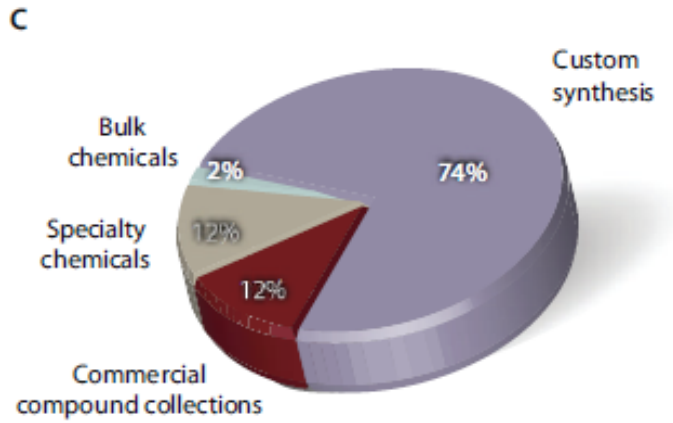
The NPC Screening Resource



Composition



Sources



Cost

NPC Status, 2012

Drug Source	*In house*	*Procurement in process*
US FDA	1635	182
UK/EU/Canada/Japan	756	177
Investigational	928	3953
Total Approved	2391	359
Total	**3319**	**4312**

Repurposing Case Study: Refractory CLL

CLL — Chronic Lymphocytic Leukemia

- 30% of all leukemias
- Standard of care: chemotherapy

Relapse virtually universal; treatments needed for refractory disease

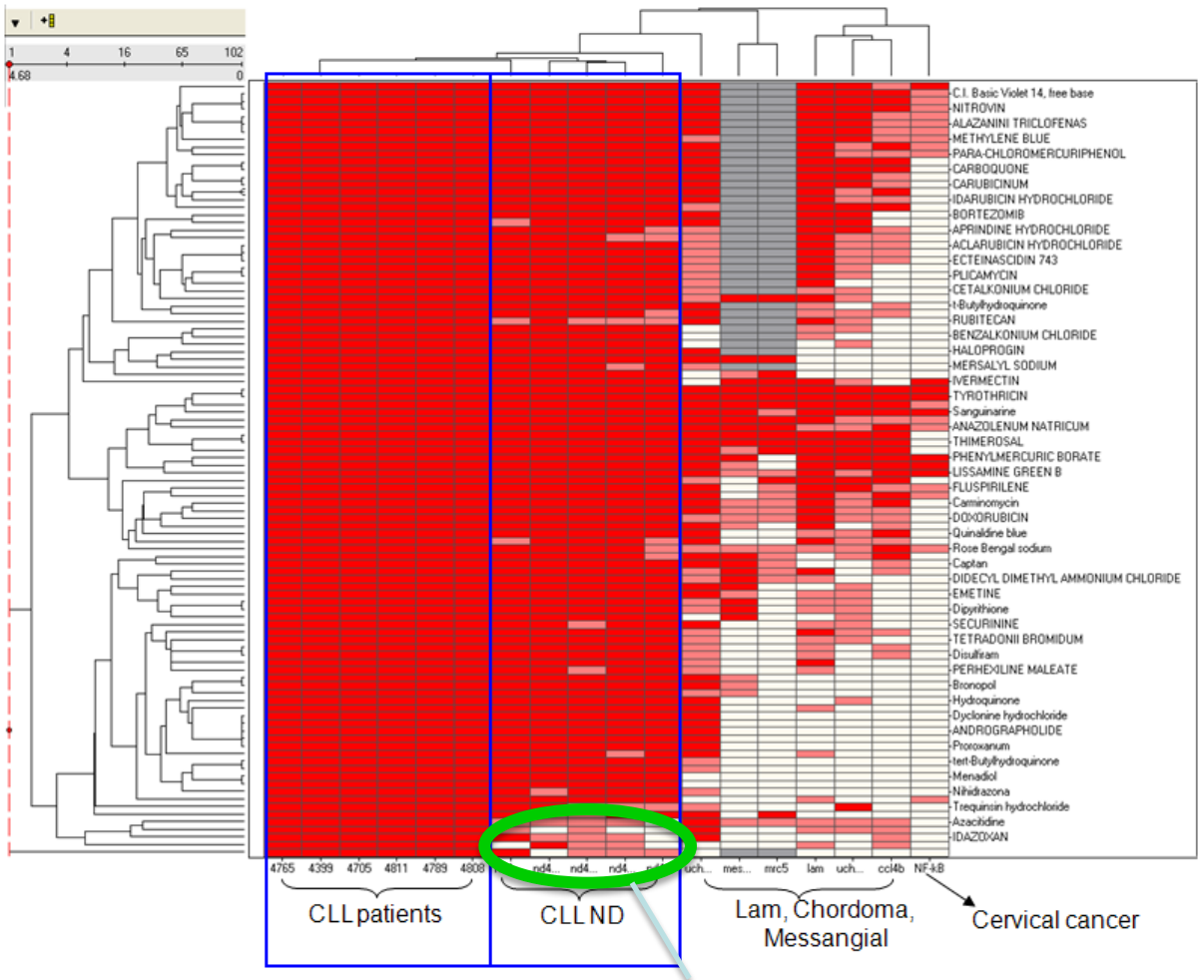
NPC CLL screen

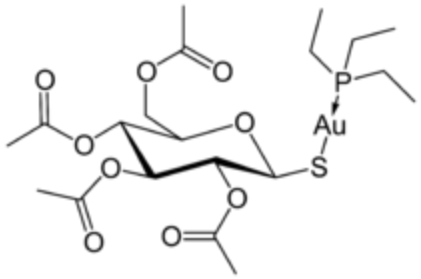
CLL and normal donor B-cells obtained from patients at NIH Clinical Center
- Adrian Wiestner, NHLBI
- Cells from six CLL patients and five normal donors tested

NPC screened at 9 concentrations, 1 nM to 57 µM
- Readout: cell viability (ATP measurement)

Desired compound profile = *Differential* cell killing

Discovery of a CLL-Selective Cytotoxic Agent





Auranofin

Kills CLL but not normal donor B cells

KU's Institute for Advancing Medical Innovation, The Leukemia & Lymphoma Society and NIH begin groundbreaking clinical trial for leukemia patients

November 01, 2011

By KUMC News

Scott Weir, PharmD, PhD, is director of KU's Institute for Advancing Medical Innovation

As part of an aggressive effort to speed delivery of treatments to patients by finding new uses for approved drugs, researchers at the University of Kansas Medical Center have begun a clinical trial targeting the most common form of adult leukemia with a drug first approved to treat arthritis more than 25 years ago.

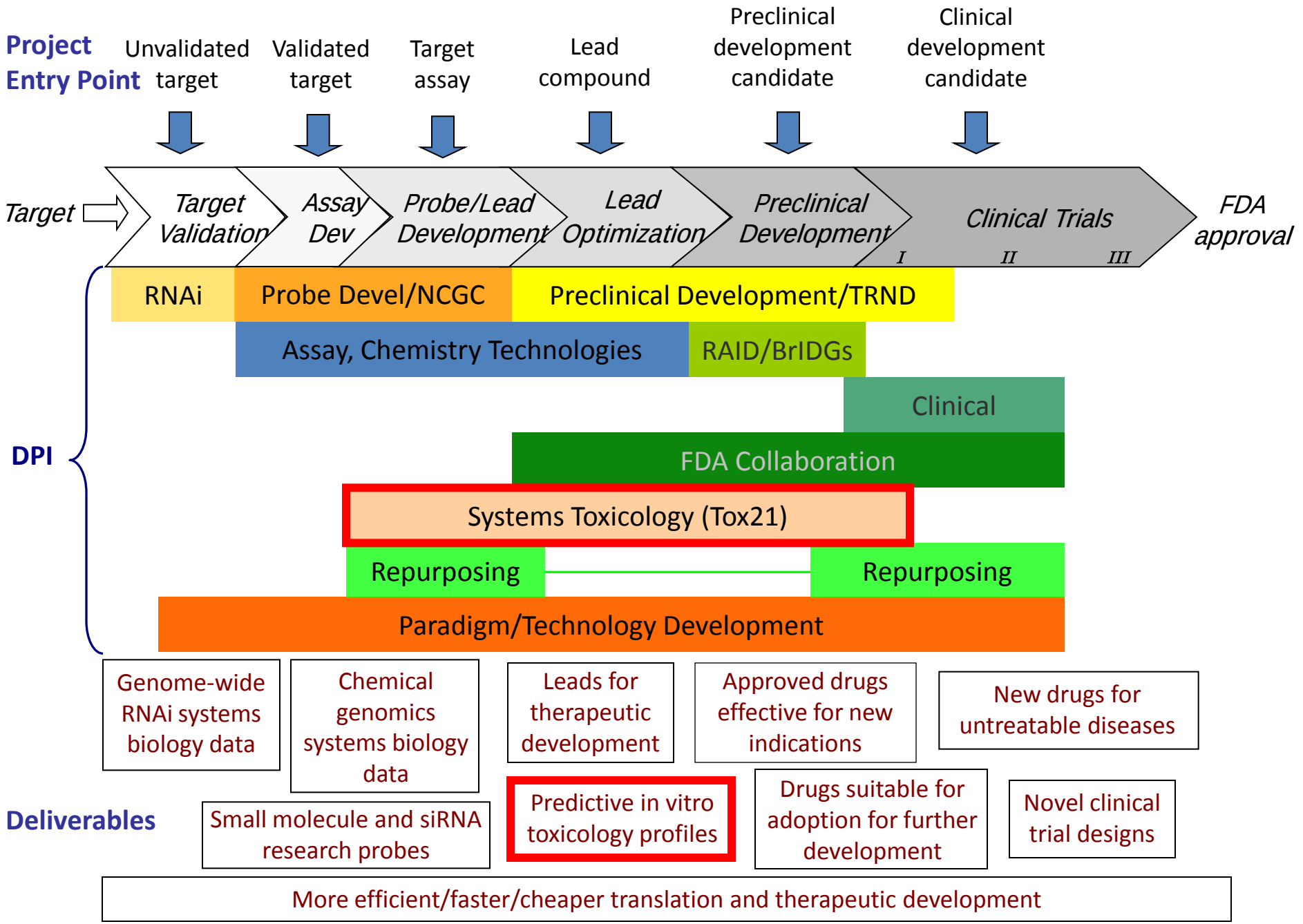
Earlier this month, KU researchers treated the first trial participant, a Kansas City-area patient suffering from chronic lymphocytic leukemia or CLL, with the drug auranofin, which has long been used to treat patients with arthritis.

The trial is one key piece of a larger collaboration between KU, The Leukemia & Lymphoma Society (LLS) and the National Institutes of Health (NIH) to accelerate discovery and development of safe, effective and affordable cancer treatments. Over the last two years, the group discovered that auranofin kills CLL cells in test tubes, and received approval to test the drug in CLL patients.

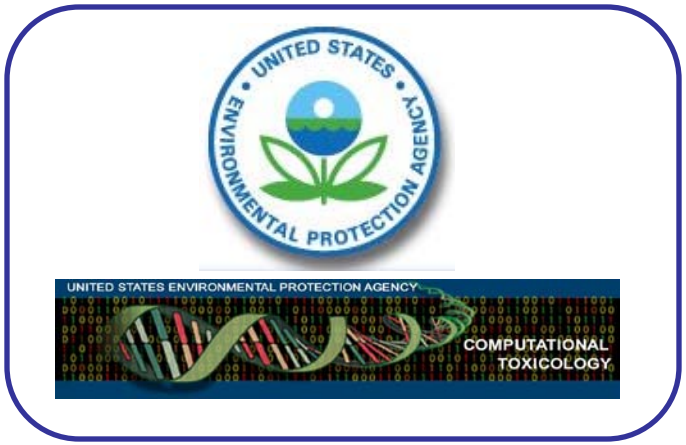
"Today's process of discovering and developing new drugs for patients takes too much time and costs too much money," said Louis J. DeGennaro, Ph.D., executive vice president and chief mission officer, LLS. "The collaboration between KU, LLS and NIH is committed to giving new hope to patients by reducing sharply the time and costs associated with developing new therapies. Auranofin is a great example of what is possible through an effective public-private partnership."

"Spending more than $1 billion and taking more than a decade to deliver new therapies to patients is simply not sustainable," said Scott Weir, PharmD, PhD, director of KU's Institute for Advancing Medical Innovation. "Our group moved this new discovery into a clinical trial in just two years and for about $1 million, representing significant time and cost savings from business as usual."

The NCATS Division of Preclinical Innovation: An Integrated Pipeline



The Tox21 Community

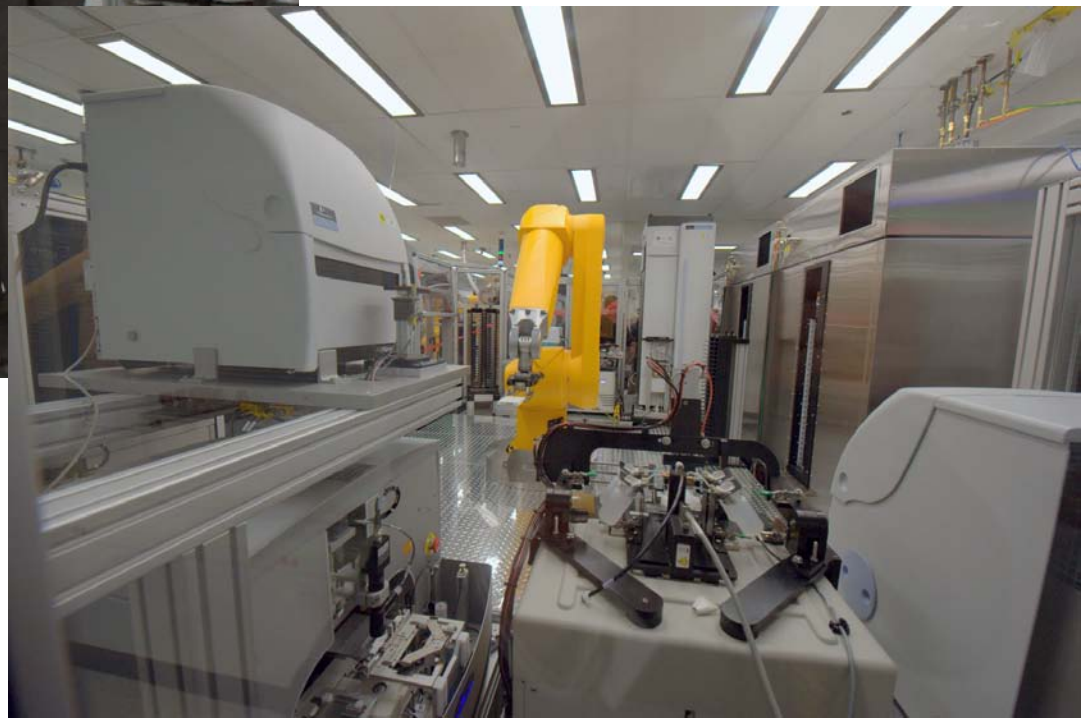


Tox21

- Identify patterns of compound-induced biological response in order to:
  - Characterize toxicity/disease pathways
  - Prioritize compounds for more extensive toxicological evaluation
- Develop predictive models for biological response in humans, while minimizing use of laboratory animals

Tox21 Robot Ribbon-Cutting March 10, 2011

(L to R): Eric Green (Director, NHGRI/NIH), Linda Birnbaum (Director, NIEHS/NIH), Janet Woodcock (Director, CDER/FDA), Lek Kadelli (Asst Administrator, ORD/EPA)



Mobilization of Tox21 Team: BP Oil Spill

Environ. Sci. Technol. **XXXX**, *xxx*, 000–000

Analysis of Eight Oil Spill Dispersants Using Rapid, In Vitro Tests for Endocrine and Other Biological Activity

RICHARD S. JUDSON,*,† MATTHEW T. MARTIN,† DAVID M. REIF,† KEITH A. HOUCK,† THOMAS B. KNUDSEN,† DANIEL M. ROTROFF,† MENGHANG XIA,‡ SRILATHA SAKAMURU,‡ RUILI HUANG,‡ PAUL SHINN,‡ CHRISTOPHER P. AUSTIN,‡ ROBERT J. KAVLOCK,† AND DAVID J. DIX†

National Center for Computational Toxicology, Office of Research and Development, U.S. Environmental Protection Agency, Research Triangle Park, North Carolina 27711, and NIH Chemical Genomics Center, National Institutes of Health, Department of Health and Human Services, Bethesda, Maryland 20892

Received June 25, 2010. Revised manuscript received June 30, 2010. Accepted June 30, 2010.

Energy Services, L.P., Sugar Land, TX). In excess of 1.5 M gallons of dispersant have been released into the Gulf as of June 26, 2010. Oil spill dispersants are complex mixtures of two basic components (*1*). The first component is composed of one or more surfactants that can emulsify oil. The second component is a hydrocarbon-based solvent mixture that helps break up large clumps of high molecular weight, more viscous oil. There is limited information on the potential of dispersants to cause acute or long-term toxicity in aquatic species or humans.

EPA's Office of Research and Development was asked to evaluate the potential toxicity of eight oil spill dispersants, including Corexit 9500. Because of the need for rapid turnaround, it was decided to employ a series of in vitro, cell-based assays. One mode of toxicity that is of concern for dispersants is endocrine disruption (*2*), due to of the fact that nonylphenol ethoxylates (NPEs) are used in some of the dispersants as part of the surfactant component. NPEs can degrade to produce nonylphenol (*3*), which can strongly interact with the estrogen receptor (*4*–*7*). NPEs themselves have been shown to inhibit testicular growth in rainbow trout (*8*). Because of this fact, the focus of our in vitro studies was on measuring potential interaction of the dispersants with the estrogen receptor (ER) and the androgen receptor (AR).

Here we describe the results of a series of rapid in vitro tests to determine the interaction of eight oil spill dispersants with ER, AR, and other receptors and transcription factors.

The Tox21 team was called upon to perform rapid testing of oil dispersants used in the Gulf of Mexico BP oil spill: multiple cell lines revived and associated assays performed during the Memorial Day weekend.

Potential Utilization of the NPC for NETs

- Therapeutics for NETs must be identified rapidly
  - Timeline of NME development (10 yr) incompatible, making repurposing of currently approved drug only rapid route for NET (1-2 yr scale)
  - NCGC is NIH intramural facility so can be activated for national need with very little lead time (as done during the Gulf Oil Spill disaster)
  - NPC is comprehensive informatics and screening collection resource purpose-built for this type of need

- NCGC can screen entire NPC as 15-point dilution series in <1 wk, already >100 assays screened
  - BSL 1-2 at main facility, BSL 3 at NIH Bldg 33

- NPC not screened against NETs to date due to lack of mandate/funding for such activity, all current projects are funded for specific deliverables

Selected Infectious Disease Projects at the Center

- Anthrax internalization
- Botulinum NT (USAMRIID)
- *E. coli:* DNA replication modulators, Beta-lactamase, posttranslational modifications
- Giardia: Fructose-1,6-bisphosphate aldolase, trophozoite viability
- Lassa and Marburg VSV pseudotypes
- HIV nucleocapsid
- Hookworm TGR
- Influenza NS1
- Leishmania pyruvate kinase
- Malaria: Killing/profiling, Plastid replication, delayed-death phenotype
- SARS: Viral protease inhibitors
- Shigella: Toxin modulators
- Schistosomiasis: redox maintenance proteins
- Trypanosomiasis: Cruzain, TbPFK, LmPGAM and TbPGK
- Tuberculosis: Cell killing, profiling (BSL-3)
- Venezuelan equine encephalitis virus (VEEV) (USAMRIID)
- Vaccinia Virus Entry

Practical Issues in Utilizing the NPC for NETs

- Ongoing re-acquisition of collection (100 mg) very expensive so taking some time
  - Total cost >$7M
- To conserve drug, all screening done in-house, we do not send copies of collection except in very exceptional circumstances
  - We utilize only 20 nl -100 nl of compound for each test well
- We cannot do BSL4 screens

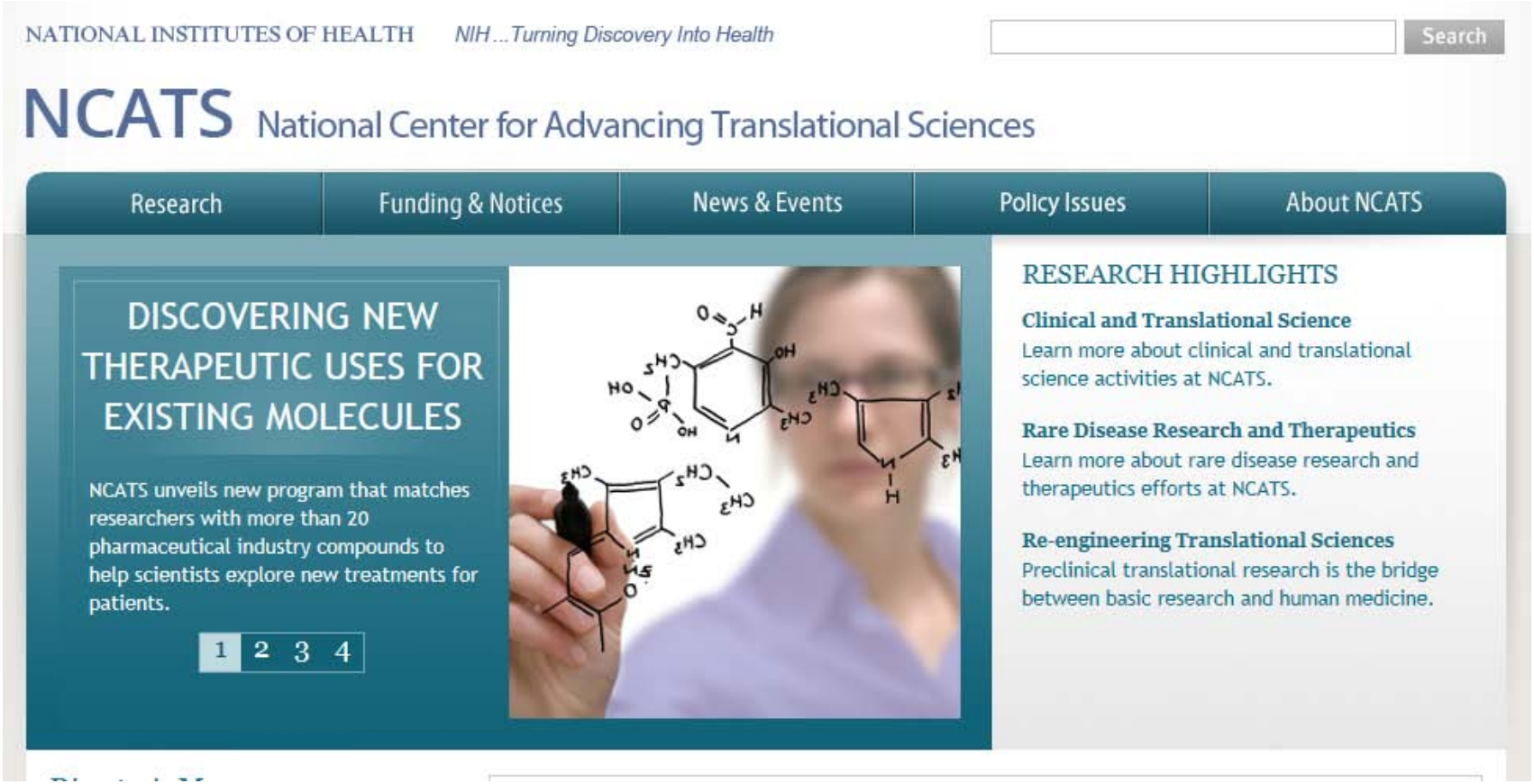
Summary

- Repurposing collection and screening capacity in place, could be used for NETs

- NCATS-DPI would be very interested in working with our Federal partners on this
  - Tox21 is very productive precedent
  - NPC unique to NCGC
  - qHTS unique to NCGC
  - Intramural Federal lab status makes project like this very flexible

- The project would require resources but marginal cost to BARDA and partners would be small as much of investment in collection and assay/screening/informatics capacity already made

Further Information



NCATS.nih.gov asimeono@mail.nih.gov