Puerto Rico Testsite for Exploring Contamination Threats (PROTECT)

NIEHS SRP P42 Research Program
Northeastern University; University of Puerto Rico; University of Michigan
West Virginia University, Silent Spring Institute, EarthSoft

Directors: Akram N. Alshawabkeh & José F. Cordero

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Outline

• PROTECT Center Overview
• PROTECT Approach
• PROTECT Projects and Cores
• Acknowledgements
PROTECT Center

• Started in April 2010
• Involves many institutions and partners:
  – Northeastern University, University of Puerto Rico, University of Michigan, West Virginia University, Silent Spring Institute, EarthSoft Inc.
• Holistic source-to-outcome approach
• Diverse expertise
  – engineers, biochemists, electrochemists, toxicologists, epidemiologist, biostatisticians, pediatricians, agronomist, hydrogeologists, and social scientists.

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Key Aspects

• Preterm Births
• Superfund Sites in Northern Puerto Rico
• Karst Hydrogeology
• Contamination focus
  – Chlorinated Solvents
  – Phthalates
Babies born before 37 completed weeks of gestation are considered preterm. **Puerto Rico** has the highest rate (17.7%) of any U.S. jurisdiction. Below only Malawi (18.1%) globally.
Preterm Birth

• Preterm birth (PTB) is the leading cause of neonatal mortality in the US, contributing to over one-third of infant deaths.

• Results in high incidence of health complications that can lead to lifelong disabilities.

• Preterm birth is a major, costly health problem in the US.

• Known risk factors for prematurity do not explain the marked increase in preterm births in the US and Puerto Rico.
Contamination in Puerto Rico

- 200+ Hazardous Waste Sites
- 16 Active Sites listed on the National Priority List (NPL); 22 Historical Sites
- Many sites include unlined landfills above aquifer in karst geologic formations
- Aquifer is primarily limestone with highly permeable karst aquifers from which most of the wells draw water
Karst

2012 Field Trip, PR

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About 40% of the groundwater used for drinking comes from karst aquifers. Other parts of the world with large areas of karst include China and Europe.
Research Questions

- What is the contribution of environmental contamination to preterm birth in PR?
- How significant is karst water as a route of exposure?
- Can we develop better strategies for detection and green remediation to minimize or prevent exposure to environmental contamination?
PROTECT Components

• 5 Projects
  – 3 Biomedical (Projects 1, 2 and 3)
  – 2 Environmental (Projects 4 and 5)

• 2 Research Support Cores
  – Human Subjects and Sampling Core
  – Data Management and Modeling Core

• 4 Enrichment Cores
  – Administrative Core
  – Research Translation Core
  – Training Core
  – Community Engagement Core
PROTECT Projects

- Project 1: Molecular epidemiology study
- Project 2: Mechanistic pathways study
- Project 3: Non-targeted analysis study
- Project 4: Fate and transport study
- Project 5: Remediation study
PROTECT Approach

Scientific Knowledge – Technology Transfer – Information for Public – Trained Workforce

A Administrative
   - 5 Green Remediation
   - 4 Fate and Transport

B Research Translation
   - D Data Management and Modeling

C Human Subjects and Sampling
   - Environmental
   - Biomedical

F Community Engagement
   - 3 Nontargeted Chemical Analysis
   - 2 Mechanistic Toxicology

E Training
   - 1 Targeted Epidemiology

Contaminant exposure analyzed from source to outcome

Key:
- Records
- Data
- Questionnaires
- Blood
- Urine
- Water
- Placenta

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PROTECT Team

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Civil Engineering

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Civil Engineering

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Biochemical Sciences

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Toxicology

P. Brown
Sociology & Health Sciences

D. Kaeli
Computer Engineering

I. Padilla
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MD, Pediatrics

J. Meeker
Epidemiology

C. Velez Vega
Social Work

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Human Subjects Core and Biomedical Projects (1, 2 and 3)

Presented by José Cordero
Human Subjects and Sampling Core

Leader: José Cordero

• Maintain the infrastructure for recruitment and follow-up.
• Conduct sequential interviews, abstract medical records, and collect biological and environmental samples
• Process, archive, and distribute collected samples to project investigators;
• In collaboration with the Data Core, maintain a repository of samples with an integrated database.
• Relatively large area (~1000 mi$^2$) with significant socioeconomical diversity

  => Requires strong community engagement component
Participant Follow-up – Human Subjects Core -

Screening & Recruitment

Study Subject

First Visit (Clinic) 16-20 weeks
- V1 Interview
- Medical Record Abstraction
- Biological Samples
- Product Use

Second Visit (Clinic) 20-24 weeks
- V2 Interview
- Home Geographical Coordinates
- Environmental and Biological Samples
- Product Use

Second Visit (Clinic) 24-28 weeks
- V3 Interview & Food Frequency
- Medical Record Abstraction
- Biological Samples
- Product Use

Third Visit (Clinic) 24-28 weeks

Delivery & Postpartum (Clinic)
- Interview
- Medical Record Abstraction
- Cord Blood Samples, Placental tissue

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PROTECT Recruitment
As of July 2015

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Withdrawal Rate

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Project 1: Molecular Epidemiology Study of Phthalate Exposure and Preterm Birth in Puerto Rico
Leader: John Meeker; University of Michigan

- Investigate associations between exposure to phthalates during pregnancy and preterm birth.
- Identify connections between environmental chemicals and markers of inflammation, oxidative stress, and endocrine disruption.
- Determine factors associated with increased phthalate exposure to inform effective exposure and risk reduction efforts.
Project 1 Selected Results

✓ Urinary phthalate biomarkers can be detected in all women in the PROTECT cohort.

✓ Levels for certain phthalates are elevated in the PROTECT cohort compared to women of reproductive age in the United States (NHANES).

✓ Specific behaviors (use of perfume, makeup, and other personal care products) and conditions (drinking water source) may lead to elevated phthalate exposure levels and may represent points of intervention.

✓ Project 1 recently found strong and significant positive relationships between multiple phthalates in urine and markers of oxidative stress.
Project 1 Selected Results

- Drinking or cooking with water from private wells associated with higher DEHP metabolites, but not statistically significant (small N thus far).
- Increased MEP associated with: Use of perfume; Use of colored cosmetics and Use of nail polish.
- Increased MCNP or MCOP associated with:
  - Plastic cistern for water storage
  - Microwaving food/drinks in plastic containers
  - Consumption of ice cream or chicken
Project 2: Toxicant Activation of Pathways of Preterm Birth in Gestational Tissues
Leader: Rita Loch-Caruso, University of Michigan

- Delineate the role of reactive oxygen species (ROS) in adverse pregnancy outcomes and tissue responses in rodents exposed to toxicants
- Develop and use in vitro models of human placenta and extraplacental membranes to identify mechanistic links between toxicant exposures and preterm birth
- Determine how immune cells contribute to toxicant-induced responses relevant to preterm birth
- Identify toxicant-induced modification of host defense against microbial infection of gestational tissues as a potential contributing factor to preterm birth
Diverse toxicants may contribute to preterm birth risk through an oxidative stress mechanism.
Project 2 Selected Findings

• A phthalate metabolite (MEHP) stimulates ROS generation and prostaglandin expression in human placental cells (trophoblasts & macrophages) in vitro

• The trichloroethylene metabolite DCVC inhibits bacteria-stimulated host defense responses important for tissue resistance to microbial infection in human extraplacental membranes in vitro

• Pregnant rats exposed to TCE had litters with decreased fetal weight, placental oxidative DNA damage, and maternal inflammation
The TCE metabolite DCVC inhibits TNF-α production which is important for tissue defense against infection.

The phthalate metabolite MEHP stimulates freshly isolated human placental macrophages to increase production of prostaglandins, important activators of labor.

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**Reproductive Toxicology**

The trichloroethylene metabolite S-(1,2-dichlorovinyl)-L-cysteine but not trichloroacetate inhibits pathogen-stimulated TNF-α in human extraplacental membranes in vitro

*Erica Boldenow*, Iman Hessen, Mark C. Chames, Chuanyu Xi, Rita Loch-Caruso

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**Reproductive Biology and Endocrinology**

Mono-ethylhexyl phthalate stimulates prostaglandin secretion in human placental macrophages and THP-1 cells

Lauren M Tetz, David M Aronoff, and Rita Loch-Caruso


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**Graphs**

- **A** PGE₂
- **B** PGF₂α

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Project 3: Discovery of Xenobiotics Associated with Preterm Birth
Leader: Dr. Roger Giese, Northeastern Univ.

- Discover xenobiotics such as toxicant metabolites that contribute to preterm birth
- Explore xenobiotic profiles in the urine, placental tissues and water
- Compare patterns of DNA adducts in human placenta and laboratory-stressed placental cell cultures
Project 3 Selected Results

✓ Introduced PROTECT-developed xenobiotic detection technology called the Porous Extraction Paddle (PEP) for convenient extractions at remote sites – Patent application filed

✓ Developed CAX-B, a novel mass tag for ultrasensitive detection – Provisional patent application filed

✓ Increased detection of the urinary sulfateome by 75-fold (up to 1129 nonpolar sulfates)
Project 3 Selected Results

Two LC-UV chromatograms from 2 PEP extracts of urine 6 weeks apart: high reproducibility is seen.

Detection of 160 amol of thymidine by CAX-Mass Spectrometry

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Environmental Projects (3 and 4) and Data Management Core

Presented by Ingrid Padilla
Project 4: Dynamic Transport and Exposure Pathways of Contaminants in Karst Groundwater Systems
Leader: Ingrid Y. Padilla, Univ. of Puerto Rico, Mayagüez

- Characterize fate and transport of contaminants in karst groundwater (conduit and diffusion dominated flow)
  - Fundamental Processes at Lab Scale
  - Applied Technologies at the Field Scale
- Assess spatial and temporal (historical and current) variability in water quality in groundwater and tap water
- Study contaminant distribution resulting from changes in contaminant sources, hydrologic conditions, remedial activities, and site management
- Develop new predictive tools to reduce exposure
Project 4 Selected Results

✓ Spatiotemporal analysis of groundwater data reflects extensive contamination

✓ Refined spatiotemporal analysis show significant variability in the distribution of CVOCs

Padilla et al., 2015

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Yu et al., 2015
Project 4 Selected Results

- Higher detection frequencies and concentrations of phthalates are associated with regions of highest aquifer permeabilities and sinkhole density.

- Marked differences in detection frequencies and concentrations between source water and tap water.
Project 4 Selected Results

✓ Laboratory-Scale
  ✓ Develop statistical characterization of preferential flow paths and quantified transport parameters that are to be used for predictive purposes

✓ Spring Watershed characterization
  ✓ Translates what we learn from lab-scale experiments into what is happening at the field scale

(Anaya et al. 2014)
Project 5: Green Remediation by Solar Energy Conversion into Electrolysis in Groundwater
Leader: Akram Alshawabkeh, Northeastern Univ.

- Evaluate electrolysis for manipulating redox conditions in groundwater
- Evaluate transformation of TCE and other contaminants in pore fluid by electrolysis
- Assess toxicity evolution
- Engineer system for field implementation
Project 5 - Electrochemical Transformation Mechanisms

- Use electrolysis to promote oxidation in groundwater
- Use electrolysis to promote reduction in groundwater

Figure 1. Proof-of-concept of invention
Project 5 Selected Results

✓ Transformation of all dissolved TCE from groundwater

✓ Delineation of transformation mechanisms

✓ Demonstrated simultaneous transformation of contaminant mixtures

✓ Patent application filed for novel two electrode remediation system

✓ Working on pilot-testing
Data Management and Modeling Core
Leader: David Kaeli, Northeastern Univ.

Data Sources
- Questionnaires
- Information Abstracted by Core, Raw Data Imported
- Records
- Placenta
- Blood
- Samples Analyzed by Projects, Raw Data Imported
- Urine
- Water

PROTECT Data Repository

End Users
- PROTECT Researchers and Trainees
- Approved External Stakeholders (via RTC)
- Public (via CEC)
Data Management and Modeling Core - By the Numbers

• Human Subject Data
  – 3,193 total fields/participant; Presently 15 different forms
  – Close to ~1.5M records!

• Environmental Data
  – 1048 wells (14 of them include water contaminant data)
  – 35 springs (3 of them include water contaminant data)
  – Field data; 9 wells and 2 springs are sampled twice a year
  – Tap water data: 13 contaminants

• Targeted Exposure Data
  – 51 targeted chemicals * ~8 fields * # of participant
    • 19 Phthalates and Phenols
    • 18 Trace Metal
    • 14 Pesticides

• Non-targeted Biological Data
  – 5 fields, >1B data points in 6 urine samples
    • Mass-to-charge values
    • Data peaks
Community Engagement Core
Leaders: Carmen Velez Vega, UPR
Phil Brown, Northeastern Univ.

- PROTECT Wins the 2015 People’s Choice Award at the EPA Community Involvement Training Conference
- PROTECT Researchers Partner with March of Dimes in San Juan
- The CEC has brought a number of community partners together to form a Community Advisory Board that include Ciudadanos en Defensa del Ambiente (CEDDA; Citizens for Environmental Defense), Ciudadanos del Karso (Citizens of the Karst), and COTICAM (Steering Committee for Environmental Quality).
Partners and Collaborators

- Collaboration with local stakeholders
- Health care professional groups
- Local Community Health Centers
- Environmental advocacy groups
Acknowledgments

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For More Information

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– Email: protect-info@coe.neu.edu
– or contact Rachel Grashow
  Phone: (617) 373-4153
  r.grashow@neu.edu

– Previous CLU-IN presentation:
  Integrating Data from Multidisciplinary Research,
  Session I: Introducing the Big Picture
  Sponsor: NIEHS SRP
  https://clu-in.org/conf/tio/IntegratingData1/
Upcoming Conferences

• NIEHS SRP Annual Conference; Nov. 18 – 20, 2015; San Juan, PR
  – http://www.northeastern.edu/srp2015/

• Karst, Groundwater Contamination & Public Health; Jan 27 – 30, 2016; San Juan, PR
  – http://karstwaters.org/conferences/kgcph/