Nanotechnology – Human Health and Environmental Impacts

Jim Rice, Robert Hurt, Agnes Kane

Brown University, Providence, Rhode Island

NEWMOA Webinar, January 24th, 2012

The Superfund Research Program at Brown University

• Focus on Mixed Exposures
  – Environmental health and engineering research
  – Technology development

• 4 Biomedical and 3 Engineering Research Projects
  – Tackle reuse of contaminated land in RI and other post-industrial states
  – Seek to understand the health effects of complex exposures
  – Focus on Nanotechnology & Metals, Molecular Epidemiology & Reproduction, and Semi-Volatile Organics & Vapor Intrusion

• Research Translation and Community Engagement Cores
  – Provide knowledge and services to professional and community stakeholders
  – Address basic and translational research issues, management decisions, and communication complexities inherent to reuse of hazardous waste sites

Rhode Island is Our Laboratory

Mission - To provide Rhode Island (and other states) with a responsive center of technical excellence that takes a research-oriented approach to resolving the complex scientific, engineering, and societal issues that arise when considering the reuse of hazardous waste sites
1/30/2012

2001-2
Study Group

Spring 2005
funding of
Superfund
grant- 4 yrs

Spring 2008
competing
renewal
application for
Superfund

Spring 2004
submission of
Superfund
application

Spring 2009
funding of
Superfund
grant- 5yrs

2002 2003 2004 2005 2006 2007 2008 2009
Partnerships at Regional NPL Sites

• Fisherville Mill NPL Site, Grafton, MA
  – In partnership with our Analytical core, the Fisherville Redevelopment Company, and Clark University, Jim Rice leads an effort aimed at helping to develop an efficient and reliable analytical testing method to determine whether bioremediation efforts are improving water quality in the Blackstone River.

• Centredale Manor NPL Site, N. Providence, RI
  – Attended October 2011 Dialogue Group Meeting with EPA, RI DEM, RI DOH, community activists, lawyers, etc.
  – Exploring opportunities for research; interested in partnering with Narragansett EPA labs to determine how remediation activity influences bioavailability of sediment contaminants
  – Partnering with WRWC through Community Engagement Core efforts – Questions about the Oxbow area; community education and risk communication; and what is happening downriver?

How Can You Interact With Us?

• We provide seek two-way professional to professional communication of relevance
  – Official State partners (Rhode Island Departments of Health and Environmental Management)
  – Other non-governmental partners (RI Bar Association, NEWMOA, Metcalf, etc.)

• We host translational seminars, conferences, webinars, symposiums. For example:
  – Addressing the Complex Site: Mixed Pollutants Across Environmental Media Symposium at the American Chemical Society National Meeting (Fall 2012)
  – Social, Economic, and Psychological Costs of Contamination Workshop (May 2012)
  – Fate and Transport and Toxicology of Nanomaterials Webinar co-sponsored by NEWMOA (Jan. 2012)
  – Epigenetics and Fetal Origins of Health and Disease Symposium (Oct. 2011) - 79 attendees (DOH, academia, healthcare, non-profits); 23 RIs awarded continuing education credits
  – Environmental Health and Chemical Exposures: Law and Science Seminar co-sponsored by The RI Bar Association (2010)

• We aim to involve our investigators & students in translational research/communication activities.
  – Monthly seminar series
  – Planning informal lunches with State partners, i.e., RIDEM and RIDOH
  – We would like to get students more engaged with actual hazardous waste sites

• We search for creative and practical research activities and funding opportunities/partnerships.

• Contact Us:
  – Website: http://www.brown.edu/Research/SRP/
  – James Rice, PhD, Engineering State Agencies Liaison, James_Rice@brown.edu
Carbon nanotube coating improves neuronal recordings
Keeser et al., 2008

The Nanotechnology Movement

Definition
the systematic manipulation of matter on the length scale 1-100 nm to produce useful new engineered structures, materials, or devices.

History
- R. Feynman, 1959, “There’s plenty of room at the bottom”
- Kroto, Smalley, Curl, Fullerene C60 synthesis, 1985
- Sumio Iijima, 1991, nanotube synthesis

Nanotechnology today
- over 60,000 scientific papers / yr
- over 1,300 industrial products (as of 2010)

The U.S. National Nanotechnology Initiative (NNI)
- first passed in 2001, reauthorized in 2008
- now ~ 2.1 billion $ / yr (2012)
- four phases envisioned:
  1. passive nanostructures
  2. active nanostructures
  3. three dimensional nanosystems
  4. molecular nanosystems
Example: History of Carbon Nanomaterials

- **Fullerene discovery** (1985)
- **Soot, carbon black**
- **C60**
- **Single-wall carbon nanotube discovery** (1991)
- **Start of US NNI** (2001)
- **Graphene synthesis** (2004)
- **Fluorescent carbon dots** (2006)

Nanofabrication approaches: “Top Down”

- **Mask** (passes light only in non-patterned areas)
- **Photoresist** (a polymer, either polymerized or depolymerized upon exposure to UV light)
- **Spin coated film**
- **Substrate** (silicon)
- **Chemical etching**
- **Circuit pattern**
Bottom-up, or self-assembly, approaches use chemical or physical forces operating at the nanoscale to assemble basic units into larger structures.

Inspiration for bottom-up approaches comes from the biological world, which self-assemble into complex systems that even replicate and evolve.

Example

organometallic reagents, e.g. Me$_2$Cd, (TMS)$_2$Se

preheated coordinating solvent e.g. tri-octadecylphosphine (TOP)

Nanomaterials Today

Geometry x Chemistry = almost limitless diversity, complexity

- Equi-axed forms (nanoparticles)
- One-dimensional forms (fibers and tubes)
- Two-dimensional forms (plates and disks)
- Nanostructured surfaces
- Nanostructured solids

Periodic Table of Elements
### Some specific examples of nanomaterials

<table>
<thead>
<tr>
<th>Equi-axed forms (particles)</th>
<th>1D materials (fibers, tubes)</th>
<th>2D materials (plates, disks)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>metals</strong></td>
<td>Silver, gold nanoparticles</td>
<td>nanowires</td>
</tr>
<tr>
<td>Iron, cobalt, nickel magnetic nanoparticles</td>
<td></td>
<td></td>
</tr>
<tr>
<td>copper nanoparticle conducting inks</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>semiconductors</strong></td>
<td>CdSe quantum dots</td>
<td>Si, ZnO semiconducting nanowires, nanorods</td>
</tr>
<tr>
<td><strong>ceramics</strong></td>
<td>zinc oxide, titanium dioxide pigments and sun screens, cerium oxide catalysts</td>
<td>electrosyn ceramic nanofibers for composite fillers</td>
</tr>
<tr>
<td><strong>carbons</strong></td>
<td>fullerences, carbon black, nanohorns</td>
<td>carbon nanotubes nanofibers</td>
</tr>
<tr>
<td><strong>polymers</strong></td>
<td>biodegradable polymer nanobeads for medical applications, dendrimers</td>
<td>electrosyn polymer nanofibers</td>
</tr>
</tbody>
</table>

### Nanotechnology in Industry

Now (1,300 products)

Components for next-generation batteries, fuel cells, computer chips, displays, structural materials, cosmetics, implants, drug delivery vehicles, paints, self-cleaning windows, and much, much more……..

Human Exposures

Four primary exposure routes are:
- inhalation
- dermal (skin) exposure
- ingestion
- injection / implantation

Some nanomaterials become easily airborne
- dry powders, mists from sonication, spraying

Particles < 5-10 um are respirable

Nanomaterials may permeate the skin, or migrate up the olfactory nerve to the brain, but possibly in low doses

Nanoparticles can be ingested in food, beverage, or by swallowing after clearance from the lung

Exposure Example: Graphene

Graphene – the single-atom-thick sheet of sp2-hybridized carbon first isolated from graphite in 2004

Above: monolayer epitaxial graphene silicon substrate for nanoelectronics → likely not a significant exposure risk

Typically produced as bulk dry powder

Typically produced as aqueous suspension, which can be used in biomedicine or aerosolized

Related forms, however......
Many “Few-Layer-Graphene” Samples are Potentially Respirable

...and some are larger than macrophages and may not be cleared from the lung

Graphene Oxide Solutions May Be Aerosolized to form Respirable “Crumpled Graphene Nanoparticles”
**Environmental Impacts and Environmental Health**

**Exposure Scenarios**

Occupational / consumer (inhalation, ingestion, skin contact)
- of most immediate concern

Medical (injection for imaging, drug delivery…)
- toxicity characterization already a major part of drug / therapy development

Environmental
- emerging issue: are nanomaterials the pollutants of the future?

**Some environmental issues**

- What is the toxicity of high-volume nanoparticles to aquatic / terrestrial organisms, microbial communities?
- Are nanoparticles bioavailable to organismal tissues? Do they bioaccumulate, biomagnify through trophic transfer?
- How/where are nanoparticles transported in the environment?
- Do nanoparticles persist in the environment? Do they transform?

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**Example of Environmental Transformation and Fate: NanoSilver**

**Synthesis**

Sodium citrate
\( \text{NaBH}_4 \)
\( \text{AgClO}_4 \)

Washed by deionized water twice

40 mg/L nAg colloid

**Two questions:**

1. Will these noble metal nanoparticles be stable (persistent) in the environment?

2. In suspension, Ag NPs usually co-exist with Ag\(^+\) ions, which determine much of their biological / environmental activity. What determines this ion concentration?

Citric-acid stabilized AgNPs
Average size: 5~8 nm

Jingyu Liu, Ph.D. student, Brown University
A general measurement method for ion-nanoparticle partitioning

Environmental or biological simulant fluid or buffer

incubation

ultrafiltration

nAg

cellulose membrane
3K Da, (1-2 nm pore size)

separation

quantification

Total ionic silver by AAs (graphite furnace atomic absorption spectroscopy) or ICP

Results: ion-particle partitioning in nanosilver

nAg particles are not persistent in the presence of O₂ !

nAg particles are not persistent in the presence of O₂ !

nAg

O₂, H⁺

Ag⁺ (slow)

H₂O₂, O₂⁻

Ag⁺ (fast)

This reaction produces active peroxide intermediates

Is inhibited by natural organic matter

Leads to complete particle dissolution in aerobic environments

Results: ion-particle partitioning in nanosilver

nAg

15% of total silver

Ag⁺ (slow)

H₂O₂, O₂⁻

Ag⁺ (fast)

This reaction produces active peroxide intermediates

Is inhibited by natural organic matter

Leads to complete particle dissolution in aerobic environments
If nano-silver oxidatively dissolves, why doesn't bulk silver?

Answer: it can! (but slowly – about 1 nm/day)

Leaching, or ion-particle partitioning, is a major theme in nanomaterial safety

Quantum dots

Antibacterial nanosilver

Carbon nanotubes

ZnS, CdSe, Cd\(^{2+}\), Se\(^{2-}\), Zn\(^{2+}\)

Fe\(^{2+}\), Ni\(^{2+}\), Y\(^{3+}\), Co\(^{3+}\), Zn\(^{2+}\), Ni\(^{2+}\), NiO
Nanosilver “corrodes” by dissolution – does it also “tarnish”?  

In the lab, nanosilver reacts with sulfide in the presence of O₂.

Silver sulfide nanoparticles detected in waste water treatment sludge [Kim et al., ES&T, 2010]

Competing Pathways of Environmental Transformation

Indirect Route

Oxidative dissolution

Ag²⁺

H₂S/HS⁻ (O₂)

Precipitation

Direct Route

Solid-fluid heterogeneous reaction

Nano-Enabled Consumer Products

Example: Quantum Dot Composites for Energy-Efficient Lighting

CdSe quantum dots absorb short-wavelength light and emit longer wavelength light -- thus they red-shift the LED spectrum without the losses that simple filters would cause

“A quantum dot optic”

“Emotionally satisfying” LED lighting

Seth Coe-Sullivan, Sc.B. eng. ’09
Chief Technical Officer, QDVision

1/30/2012
**Nano-enabled Products**

**Example: End-of-Life Behavior of the Quantum Dot Optic**

- Do Cd, Se leach from the optic after disposal?
- Are QD nanoparticles released to the environment through degradation processes?

The QD optic releases small amounts of Cd and Se through water infiltration and quantum dot dissolution.

There is no evidence of nanoparticle release, or unique “nanospecific” risk.

**Results**

- **Total Cd release**
- **Soluble Cd only**

**Example environmental Application:** Nanomaterials for Hg vapor capture

Some types of nano-selenium have ultrahigh activity for Hg vapor capture.

![Graph showing absorption capacity and sorption rates of different materials](image-url)
Nano-selenium formulations offer self-sensing and mercury detection

Nano-sorbent has the advantage of optical reporting:

*Self-sensing function* - is the sorbent spent?

*Mercury finder function* - where was the spill / break?

Smart boxes, bags for safe recycling

Technologies for permanent Hg sequestration

Power plant flue gas capture

Retail drop-off boxes

Cartridge-based air-cleaning for NASA spaceflights

Consumer spill and disposal kits
Financial support from the US EPA Science to Achieve Results Program, the NSF NIRT program, and the NIEHS Superfund Research Program grant at Brown is gratefully acknowledged.

The Laboratory for Environmental and Health Nanoscience

ENVIROMENTAL AND HUMAN HEALTH IMPACTS OF NANOTECHNOLOGY

1. Environmental bioaccumulation and toxicity to aquatic organisms
2. Human exposure and disease

Biswas & Wu, J Air & Waste Management Association, 55, 708, 2005
1. ENVIRONMENTAL BIOACCUMULATION AND TOXICITY TO AQUATIC ORGANISMS

October 28, 2011: The Environmental Protection Agency issued a proposed $101 million cleanup plan for the Centredale Manor superfund site in North Providence.

- Carbon black nanoparticles and carbon nanotubes have sp²-hybridized graphenic carbon surfaces
- High surface area for adsorption of PAHs by π-π bonding


NANOPARTICLE-BASED DISPERSANTS FOR ENVIRONMENTAL REMEDIATION OF POLYCYCLIC AROMATIC HYDROCARBONS

Hypothesis: Nanoparticle-based dispersants will stabilize oil-water emulsions, adsorb aromatic hydrocarbons, and decrease their toxicity and bioavailability.
2. INHALATION AND SYSTEMIC TRANSLOCATION OF NANOPARTICLES

Exposure — Dose — Response

Respiratory Tract
- Nose
- Conducting Airways
- Airspaces or Alveoli

GI Tract
- Brain
- Elimination in feces

Blood and Lymph
- Liver
- Elimination in bile
- Excretion

Skin
- Soft Tissues and Bone

Bone Marrow
- Spleen
- Heart
- Kidneys

Exposure, systemic translocation, and elimination

POTENTIAL CHRONIC TOXICITY OF NANOMATERIALS

NEL et al. SCIENCE 311: 622-627, 2006

<table>
<thead>
<tr>
<th>TISSUE TARGET</th>
<th>DISEASE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Macrophages and inflammatory cells</td>
<td>granulomas</td>
</tr>
<tr>
<td></td>
<td>chronic inflammation</td>
</tr>
<tr>
<td></td>
<td>fibrosis or scarring</td>
</tr>
<tr>
<td>Lungs</td>
<td>cancer, mesothelioma</td>
</tr>
<tr>
<td>Blood vessels</td>
<td>stroke, heart attack</td>
</tr>
<tr>
<td>Immune system</td>
<td>autoimmune disease</td>
</tr>
<tr>
<td></td>
<td>leukemia, lymphoma</td>
</tr>
<tr>
<td>Nervous system, brain</td>
<td>heart arrhythmia</td>
</tr>
<tr>
<td></td>
<td>brain injury</td>
</tr>
</tbody>
</table>
Occupational Carcinogens

<table>
<thead>
<tr>
<th>Agent</th>
<th>Industry</th>
<th>Target Site</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arsenic</td>
<td>Glass, metal, pesticide</td>
<td>Lung, Skin</td>
</tr>
<tr>
<td>Asbestos</td>
<td>Construction</td>
<td>Lung, Pleura, Larynx</td>
</tr>
<tr>
<td>Benzene</td>
<td>Chemical</td>
<td>Leukemia</td>
</tr>
<tr>
<td>Beryllium</td>
<td>Aerospace</td>
<td>Lung</td>
</tr>
<tr>
<td>Cadmium</td>
<td>Dyes, batteries</td>
<td>Lung, Prostate, Kidney</td>
</tr>
<tr>
<td>Chromium (VI)</td>
<td>Metal plating, welding</td>
<td>Lung, Nasal Sinus</td>
</tr>
<tr>
<td>Nickel</td>
<td>Metallurgy, alloys</td>
<td>Lung, Nasal Sinus</td>
</tr>
<tr>
<td>Crystalline silica</td>
<td>Mining, glass, pottery</td>
<td>Lung</td>
</tr>
<tr>
<td>Sulphuric acid mists</td>
<td>Metallurgy</td>
<td>Larynx</td>
</tr>
<tr>
<td>Benzo[a] pyrene</td>
<td>Coal gasification, paving and roofing</td>
<td>Lung</td>
</tr>
</tbody>
</table>


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**THE ASBESTOS-CARBON NANOTUBE ANALOGY**

**History of Asbestos-Related Diseases**


<table>
<thead>
<tr>
<th>Disease</th>
<th>Established Casual Association</th>
</tr>
</thead>
<tbody>
<tr>
<td>Asbestosis</td>
<td>1930</td>
</tr>
<tr>
<td>Lung Cancer</td>
<td>1955</td>
</tr>
<tr>
<td>Mesothelioma</td>
<td>1965</td>
</tr>
<tr>
<td>Cancer of Larynx</td>
<td>2006</td>
</tr>
<tr>
<td>Nanodiseases</td>
<td>????</td>
</tr>
</tbody>
</table>

- The Potential Environmental Impact of Engineered Nanomaterials.  
  Vicki L. Colvin, Nature Biotechnology, October, 2003

- Ethical and Scientific Issues of Nanotechnology in the Workplace  
  Paul A. Schulte and Fabio Salamanca – Buentello, Environ Health Perspect, 2007

- Researchers Find Nanotubes May Pose Health Risks Similar to Asbestos  
## The Asbestos-Carbon Nanotube Analogy

<table>
<thead>
<tr>
<th>Asbestos Fibers</th>
<th>Carbon Nanotubes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chrysotile</td>
<td></td>
</tr>
<tr>
<td>crocidolite</td>
<td></td>
</tr>
<tr>
<td>anthophyllite</td>
<td></td>
</tr>
<tr>
<td>Winchite-richlite</td>
<td></td>
</tr>
<tr>
<td>tremolite</td>
<td></td>
</tr>
<tr>
<td>amosite</td>
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</tbody>
</table>

**U.S. Geological Survey**

Properties of Fibers Relevant for Biological Activity
1. Surface reactivity
2. Durability and biopersistence
3. Fibrous shape and dimensions

## Properties of Fibers Relevant for Biological Activity

1. **Surface Reactivity of Asbestos Fibers and Carbon Nanotubes**

   ![Fenton chemistry](image)

   **Surface e- donor:**

   ![Surface e- donor](image)

   Reilly et al. (2007) JNM, 48: 1039-1042

## Surface Defects and Impurities in Carbon Nanotubes

**Iron Catalyst Residues**

![Topological defects](image)

- 5 nm
- 10 nm
Properties of Fibers Relevant for Biological Activity

2. Biopersistence

<table>
<thead>
<tr>
<th>Fiber</th>
<th>$T_{1/2}$ in vivo (days)</th>
<th>Carcinogenicity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crocidolite asbestos NaFe$_2$O$_3$</td>
<td>&gt; 600</td>
<td>+</td>
</tr>
<tr>
<td>Wollastonite CaSiO$_3$</td>
<td>600</td>
<td></td>
</tr>
<tr>
<td>Fiberglass</td>
<td>21</td>
<td>-</td>
</tr>
<tr>
<td>Biopersistent mineral fibers are toxic and carcinogenic in rodents</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Bernstein et al., Inhalation Toxicol, 2005

Phagocytic Vacuole:
- Low pH (4.5)
- Endogenous ROS, Fe

Carboxylated Carbon Nanotubes are Oxidatively Degraded at Low pH
Liu et al. Carbon, 2010

Enzymatic Degradation by Horseradish Peroxidase

Properties of Fibers Relevant for Biological Activity

3. Fibrous Shape and Dimensions

Long Fibers Induce Frustrated Phagocytosis and Impair Clearance

Incomplete or Frustrated Phagocytosis by Macrophages

Penetration into Alveolar Walls

Intratracheal Instillation of CNTs Causes Impaired Lung Clearance and Translocation to the Pleural Space

Lung Clearance Mechanisms
Daniele, Immune defenses of the lungs, 1988

Frustrated phagocytosis of a carbon nanotube by an alveolar macrophage after 7 days
Penetration Into the Pleural Space

Long carbon nanotubes on the mucociliary escalator after 7 days

Robert Mercer, NIOSH SEM

PROPERTIES OF FIBROUS NANOMATERIALS AND CARCINOGENICITY
Asbestos Fibers and Carbon Nanotubes Induce Micronuclei And Mitotic Abnormalities in Human Lung Epithelial Cells

Carbon nanotube localized in midbody.
Confocal microscopy- fluorescence and DIC.

Carbon nanotubes induce multinucleated daughter cells.

Carbon nanotubes interfere with chromosomal segregation.
Confocal microscopy- fluorescence and DIC.

Asbestos fibers induce multipolar mitoses.

Lagging chromosomes produce micronuclei in daughter cells 48 hours after exposure to CNTs.


Microtubules-green fluorescence; nuclei and chromosomes-blue fluorescence
**Physicochemical Properties of Fibers Associated with Carcinogenicity**

Asbestos fibers/ + macrophages

Carbon nanotubes

Fiber geometry

Frustrated phagocytosis

Redox activity

Biopersistence

Macrophage recruitment and activation

Translocation to lung epithelial cells and pleura

growth factors cytoalbum oxidants

Cell proliferation, persistent inflammation, DNA damage

Lung cancer

Malignant mesothelioma

**Unresolved questions:**

- Do carbon nanotubes translocate to the pleura following inhalation?
- Is their surface activity modified in vivo?
- Does acute toxicity predict chronic disease endpoints?

**Summary**

1. Nanotechnology has significant promise for energy generation, electronics, medicine, aerospace, defense, and environmental remediation.

2. Environmental toxicity and adverse human health effects are potential implications of nanotechnology.

3. The physical and chemical properties of nanomaterials responsible for cell toxicity are related to geometry, length, surface redox reactivity and biopersistence.

4. Commercial carbon nanotubes induce DNA damage and mitotic abnormalities in lung epithelial cells and mesothelial cells similar to asbestos fibers.

5. Engineers will be able to design less toxic, biocompatible and biodegradable nanoparticles for commercial applications that will minimize adverse environmental and health impacts.
Collaboration Between Kane and Hurt Labs:

Annette von dem Busshe, Ph.D.  Indrek Kulaots, Ph.D.
Charlie Vaslet, Ph.D.  Megan Creighton
Norma Messier  Xinyuan Liu
Paulette Ferland  Yang Qui
Ashish Jachak, Ph.D.  Jingyu Liu
April Rodd  Fei Guo

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