Office of Scientific Coordination

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“The views expressed by the presenter are his alone and do not reflect current or future, position or policy of the U.S. Food & Drug Administration or any other U.S. government organization.”
Office of Scientific Coordination (number of FTEs)
  – OSC (3)

Toxicology Program Support (number of FTEs)
  – Program Support (6)
  – Experimental Liaison Support (4)
  – Document Support (4)
  – Nanotechnology Core Facility (9, +1 to be hired)
  – Inhalation Toxicology Core Facility (2, +1 to be hired)

Veterinary Services (number of FTEs)
  – Veterinary Program (3)
  – Microbiology Surveillance Program (4)
Office Mission

.. support NCTR research programs/protocols by providing support in the following key areas:

– NIEHS/NTP Interagency Agreement (IAG)
– Veterinary Care (animal health surveillance and COR Animal Care Contract)
– MultiGen (Exp. Support Liaisons, Documentation Support Staff);
– Pathology (COR Pathology Contract)
– Nanotechnology (NCTR/ORA Core Facility)
– Inhalation Toxicology (CTP/NCTR Core Facility)
– Laboratory Equipment Repair & Maintenance (COR)
Office Research Themes

1) Developing hazard assessment nominations on behalf of FDA for submission to National Toxicology Program.

2) Nanotoxicology
   Nanotoxicology Core Facility to support:
   • Center- and FDA-wide research
   • Methods development to support research
   • Immuno-nano-toxicology

3) Inhalation Toxicology
   Center for Tobacco Products (CTP)/NCTR Core Facility to support mission of CTP with inhalation toxicology studies.
2013 Accomplishment #1 – NCTR/ORA
Nanotechnology Core Facility

**Equipment**
- Jeol 1400 TEM
- Zeiss Merlin SEM with Gatan 3View2
- Perkin-Elmer NexIon 300 ICP-MS

**Personnel**
- Replacements
  - Taylor Ingle, PhD
  - Alokita Karmakar, PhD
  - Christopher Dugard, MS
  - Trisha Eustaquio, PhD (Comm. Fellow)
- NCTR Director of NanoCore

**Procedures**
- Supporting FDA projects (characterization, detection; Li *et al.*, Chen *et al.*, 2013)
- Ions versus particles (single particle ICP-MS supporting three projects)
- Serial block face sequential SEM imaging and brain tissue mitochondria
- Electron Microscopy Electronic Notebook (EMEN) development
- Immuno-nano-toxicology
2013 Accomplishment #1 – Article in Nanotechnology

Nanotoxicology

ORIGINAL ARTICLE

Cytotoxicity and genotoxicity assessment of silver nanoparticles in mouse

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Abstract

Silver nanoparticles (AgNPs) are among the most commercially used nanomaterials and their toxicity and genotoxicity are controversial. Although many in vitro studies have been conducted to evaluate the genotoxicity of AgNPs, in vivo genotoxicity studies on the nanomaterials are limited. Given the unique physicochemical properties and complex pharmacokinetics behavior of nanoparticles (NPs), in vivo genotoxicity assessment of AgNPs is badly needed. In this study, the clastogenicity and mutagenicity of AgNPs with different sizes and coatings were evaluated using mouse micronucleus (MN) assay, Pig-a assay and Comet assay. Five 7-week-old male B6C3F1 mice per group were treated with 5 nm polyvinylpyrrolidone (PVP)-coated AgNPs at a single dose of 0.5, 1.0, 2.5, 5.0, 10.0 or 20.0 mg/kg body weight (bw) via intravenous injection for both the MN and Pig-a assays; or with 15–100 nm PVP- or 10–80 nm silicon-coated AgNPs at a single or 3-day repeated dose of 25.0 mg/kg bw for the MN assay and Comet assay in mouse liver. Inductively coupled plasma mass spectrometry (ICP-MS)

Keywords

Clastogenicity, in vivo comet assay, in vivo micronucleus assay, in vivo Pig-a gene mutation assay, mutagenicity, silver nanoparticles

History

Received 26 April 2013
Revised 13 August 2013
Accepted 22 September 2013
Published online 22 November 2013
An article titled “Cytotoxicity and genotoxicity assessment of silver nanoparticles in mouse” was published in the journal, *Nanotoxicology*.

The NanoCore provided the characterization and stability of test article in support of the research division’s project.

The key questions were
1. Is nanosilver genotoxic?
2. Did the silver ion or particle get to tissue?

Note: The key scientific questions are not possible to answer without NanoCore equipment and procedures.
Research

Scanning electron micrograph image of rat brain tissue (shown is one section in a sequential series); staff are using 2D and 3D approach to determine mitochondrial morphology and volume (* key scientific question) (T. Eustaquio, A. Paredes).

2013 Accomplishment #1
The Zeiss Merlin scanning electron microscope (SEM) has a Gatan 3View2 serial block face sectioning device. On a small block of tissue, one can image with the SEM, section, reimage, section, etc., for hundreds or thousands of sequential sections. Shown in this slide is one section of rat brain tissue that has been stained with heavy metals for electron microscopy contrast. Using these images, and those from transmission electron microscopy (TEM) of the same tissues, staff are quantifying by 3-dimensional and 2-dimensional analysis, respectively, the effects of treatment on brain mitochondrial morphology and volume.
2013 Accomplishment #1

International Collaboration

• NCTR hosted the 2013 Global Coalition on Regulatory Science Research.

• A component of the Global Coalition was a 1 ½ day Global Summit on Regulatory Science (GSRS13) focusing on “Nanotechnology and Regulatory Science Research”.

• There was an international list of speakers on the role of characterization in product and safety assessment.

• Continued focus at the Global Summit in future years will include regulatory science research on nanotechnology, especially characterization.
Purpose: Critical need within Agency for capability to conduct short-term inhalation toxicity studies on compounds of interest to Agency.

Equipment:
- TSE Systems Inc. nose-only inhalation units (4 +2), installed, validated
- AB SCIEX Q-Trap LC-MS for quantitative analysis (DBT)

Personnel:
Shu-Chieh Hu, PhD (late 2012); Yunan Tang, PhD DABT; Hyun-ki Kang, MS; Estatira Sepehr, PhD (DBT)

Procedures/Protocol:
- Inhalation units validated for GLP protocol
- Started pharmacokinetic studies on CTP primary chemical of interest
- In FY14 repeated-dose toxicity study (OECD 403, 412) and 90-day subchronic toxicity study (OECD 413).
Additional Information About Previous Slide 11

Abbreviations from previous slide were defined as they were presented and included:

- LC-MS = liquid chromatography mass spectrometry
- DBT = NCTR Division of Biochemical Toxicology
- DABT = Diplomat of American Board of Toxicology
- GLP = Good Laboratory Practice
- CTP = FDA Center for Tobacco Products
- OECD = Organization of Economic Cooperation and Development, with the numbers referring to specific internationally accepted protocol for conducting studies.
2013 Accomplishment #3 — Integration of units into OSC following reorganization

Experimental Support Liaisons, Document Support Team
• Support animal study protocol support (MultiGen)
• New database (PRODACS) development
• Documentation for SOPs
• Validation/migration of programs and servers

Veterinary Support
• Animal health and welfare program & microbiology surveillance
• AAALAC accreditation (since 1976)
• Acknowledgement, N.V. Gopee, DVM, PhD, DABT, DACLAM.

Informatics/Bioinformatics (with NCTR DBB)
• FDA-Label
• Adaptation of EMEN for NCTR & FDA use.
Abbreviations from previous slide were defined as they were presented.

- **MultiGen** = NCTR in-house animal database system
- **PRODACCS** = NCTR in-house animal database system being developed
- **SOP** = Standard Operating Procedure
- **AAALAC** = Association for the Assessment and Accreditation of Laboratory Animal Care International
- **DVM** = Doctorate of Veterinary Medicine
- **Ph.D.** = Doctorate of Philosophy
- **DABT** = Diplomate, American Board of Toxicology
- **DACLAM** = Diplomate, American College of Laboratory Animal Medicine
- **EMEN** = Electron Microscopy Electronic Notebook
- **DBB** = NCTR Division of Bioinformatics and Biostatistics
Welcome to the NCTR/ORA NanoCore Database. This EMEN2 database contains nanotoxicity study data submitted by the various NanoCore groups (ORA, EM, PEAS), NCTR divisions and FDA centers etc. Please select a group on the left to find your projects. If your group or project is not listed, please contact the administrator to request access.

Activity and recent records

EM Group-NanoCore/NCTR
- Protocol: (Angel Paredes)
- Protocol: (Patrick Slisco)

Office of Scientific Coordination/NCTR
This slide shows a screenshot of the Dashboard of the NCTR version of the Electron Microscopy Electronic Notebook (EMEN) designated as EMEN2. This notebook will be the repository for data generated by the NCTR/ORA Nanotechnology Core Facility (NanoCore) electron microscopes and other equipment, with secure access for the research project scientists.
**Nanotechnology & Nanotechnology Core Facility**

Continue methods development in anticipation of research needs. Primary research thrusts are:

1. Ion vs. particle *in vitro, in vivo*;  
   *Question:* Is this still the question in 3-5 years?

2. Sequential SEM 3D imaging  
   *Question:* Role 3D-EM in toxicology and regulatory science?

3. Immuno-nano-toxicology (currently focused on complement activation)  
   *Question:* Should immuno aspects of nanomaterials be expanded?

4. Next-generation nanomaterials  
   *Question:* What additional characterization will be required? PK and PBPK of all components?
**Strategy (Future Directions, 2)**

**Inhalation Toxicology Core Facility**
Capacity of nose-only inhalation units (n= 6 units, 46 animals/unit).

*Question 1*: What inhalation issues are we facing in Public Health that can be addressed?

*Question 2*: Is there a strategic need within FDA for more inhalation toxicology?

**Informatics/Bioinformatics**

*Question 1*: How strategic are electronic notebooks (EMEN)?

*Question 2*: Virtual pathology reviews, is this the future?
The End