CVM Perspective

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Center for Veterinary Medicine
Office of Research
CVM Organizational Chart

Office of the Center Director

**Acting Director**
Tracey Forfa, J.D.

**Deputy Director**
Tracey Forfa, J.D.

**Deputy Director, Science Policy**
William Flynn, D.V.M., M.S.

Office of Management
**Director**
Roxanne Schweitzer

**Deputy Director**
Lynnette Riggio

Office of New Animal Drug Evaluation
**Director**
Steven D. Vaughn, D.V.M.

**Deputy Director**
Elizabeth A. Luddy, D.V.M.

Office of Surveillance and Compliance
**Director**
Daniel G. McChesney, Ph.D.

**Deputy Director**
Martine Hartogensis, D.V.M.

Office of Research
**Director**
John S. Graham, Ph.D., MBA, DABT

**Deputy Director**
Mary E. Allen, Ph.D.

Office of Minor Use Minor Species Animal Drug Development
**Director**
Margaret Oeller, D.V.M.

Mission

“Protecting Human and Animal Health”
Office of Research

• Division of Residue Chemistry  
  • Dr. Phil Kijak, Director

• Division of Applied Veterinary Research  
  • Dr. Mike Myers, Acting Director

• Division of Animal and Food Microbiology  
  • Dr. Maureen Davidson, Director

• NARMS  
  • Dr. Patrick McDermott, Director  
  • Dr. Heather Green Tate, Coordinator

• Vet-LIRN  
  • Dr. Renate Reimschuessel, Director
Division of Residue Chemistry

- NADA Method Trials
- Milk projects
- Detection of mycotoxins in animal feed
- Antibiotics in distiller’s grains
- Hormones in animal muscle
Division of Applied Veterinary Research

- Milk and meat safety
- Biomarker research
- Antimicrobial resistance
- Aquaculture research
- GE Animals
- Stem cell research

Aquaculture research
Division of Animal and Food Microbiology

- AMR mechanisms and evolution
- Microbiology
  - NARMS routine testing
  - AMR, feed, veterinary pathogens
- Plasmid sequencing
- Contamination and AMR in Feeds
- Whole genome sequencing
- Metagenomics
- Bioinformatics
NARMS Objectives

1. **Monitor trends** in antimicrobial resistance among foodborne bacteria from humans, retail meats, and animals

2. **Disseminate timely information** on antimicrobial resistance to promote interventions that reduce resistance among foodborne bacteria

3. **Conduct research** to better understand the emergence, persistence, and spread of antimicrobial resistance

4. **Assist the FDA** in making decisions related to the approval of safe and effective antimicrobial drugs for animals
Structure of NARMS--2016

Human Population
- Physician Visit
  - Local Lab
    - State Lab

Retail Meats
- Random stratified sampling in 14 States
- ORA Imported Foods

Animal Population
- Random cecal sampling of national production at slaughter

CDC

FDA

USDA FSIS

Data Integration

(E. coli O157:H7, S. Typhi, Shigella, Vibrio)

Campylobacter
Salmonella
Enterococcus
E. coli

Integrated Report

National Antimicrobial Resistance Monitoring System
Veterinary Laboratory Investigation and Response Network (Vet-LIRN)

To promote human and animal health by collaborating with veterinary diagnostic laboratories to provide scientific information, build lab capacity and investigate issues with CVM regulated products.

- 2010 – Concept
  2011 – Stakeholder meeting
  2014 – Network of 38 laboratories
- Develop mechanisms for conducting investigations.
  - Confidentiality agreements
  - Grants/Contracts
  - Collaborate with other networks
- Activities:
  - Proficiency Testing
  - Emergency Response Exercises
  - Investigate Consumer Complaint Cases, including
    - Jerky Pet Treat Cases
    - Product testing
    - Fanconi testing
    - Necropsy examinations
Roxarsone Studies

- Organic-based arsenical compounds have been used in chickens since March 21, 1944, when the drug 3-Nitro® was approved.
- The active ingredient in 3-Nitro® is a chemical called roxarsone.
- Roxarsone and other organic arsenicals (nitarsone, arsanilic acid, and carbarsone) were approved for use in chickens for growth promotion, feed efficiency and improved pigmentation.
- The organic arsenicals, especially roxarsone, were approved in combination with other drugs such as narasin or salinomycin to prevent coccidiosis, a parasitic disease infecting the intestinal tracts in chickens which can lead to death.
- When 3-Nitro® (roxarsone) and the other organic arsenicals were approved it was assumed that only organic arsenic and not inorganic arsenic would be excreted from the chickens.
  - Organic arsenic compounds are much less toxic than inorganic arsenic, which is a known human carcinogen.
- Inorganic arsenic exists in two forms, arsenic(III) and arsenic(V)
Roxarsone Studies

- Humans (and other animals) can convert arsenic(V) to arsenic(III), which increases arsenic’s toxicity and retention by the body.
- **2009-2011**: Can an approved organic arsenical (3-Nitro®) when incorporated into chicken feed and fed to chickens according to approved label directions, result in the presence of inorganic arsenic in edible tissues?
- FDA scientists were able to develop and validate a new analytical method that had the necessary sensitivity and specificity to detect and quantify the low levels of inorganic arsenic that were expected to be in edible tissues.
Roxarsone Studies

• Results: livers of chickens given feed containing 3-Nitro® had concentrations of inorganic arsenic that were higher than the inorganic arsenic concentrations in the livers of chickens given control (non-medicated) feed

• Results identified four questions that warranted additional follow-up investigation
Questions Addressed by These Studies

• What is the homogeneity and stability of roxarsone in medicated feed?
  – Does roxarsone settle over time?
  – Is there homogeneity in the mixing?

• Could the inorganic arsenic found in the livers of birds in the 2009 study have come from a source other than the roxarsone in the medicated feed used in that study?
  – Drinking water, control feed, or contaminants in the Type A medicated article

• Does the solution used to extract roxarsone and other arsenic species from tissue affect the stability of some arsenic species?
  – Speculation that the alkaline tetramethylammonium hydroxide (TMAH) based solvent degraded some As species into inorganic As

• What is the stability of roxarsone and other arsenic species when stored for prolonged periods of time at -80°C?
  – No testing over storage time in 2009 study
Stability of Roxarsone in Medicated Feed

Analyses of Water, Control Feed, and Medicated Feed
Feed Stability & Homogeneity

• Medicated feed prepared in three 50-lb batches
  – Samples from Top, Middle & Bottom of each batch collected & probe samples from entire lot

• Stability over time assessed at days 0, 3, 10, 17, 24, 31, 39, 45, 52, 66
Conclusions: Feed

• Roxarsone Type A medicated article (specifically, 3-Nitro® 20) can be homogeneously incorporated into feed and is stable in feed matrix for over two months.

• Water is not the source of the inorganic arsenic observed in the livers of poultry given roxarsone-medicated feed in the 2009 study.
Generation of Roxarsone-Incurred Poultry Liver

Study Design and Sample Collection
Poultry Liver Incursion

• Poultry fed either control (non-medicated) feed or roxarsone medicated feed.
• Water given to birds analyzed prior to use
• Roxarsone added at highest approved concentration
  – As species assessed in feed at start & end of study
• Birds on study for 42 days
  – Total of 18 birds, 6 Control; 12 Roxarsone
• Animals euthanized on day 42. Only livers collected for analysis
Impact of Extraction Solution on Roxarsone and As Species Stability

Study Design and Sample Collection
Distilled Water and Tetramethylammonium Hydroxide (TMAH)

- 2009 study used TMAH in the extraction buffer
- Speculation that inorganic arsenic in livers from poultry feed roxarsone came from degradation caused by TMAH
- A water extraction method was developed and validated to eliminate this possibility
Arsenic Speciation Analyses

• Water extraction works well with feed and fortified tissue samples but not with incurred tissues
  – Poor extraction of total arsenic samples

• All discussion of incurred tissue results from TMAH data only
  – TMAH extraction conducted in parallel with water extraction post day 0.
Stability of Roxarsone & As species stored for prolonged periods of time at -80°C

Analyses of Liver Samples Stored over Time
Arsenic Species Detected by LC-ICP-MS

• Detected the following Arsenic species:
  – Roxarsone, As(III), As(V), dimethylarsinic acid (DMA), monomethylarsonate (MMA), 4-arsanilic acid (4-arsan), 4-hydroxy-3-aminobenzeneearsonic acid (3-amino), acetarsone (Acet)
• Only species in red had sufficient precision, accuracy and an established LOQ
Results: Incurred Liver

- Dimethylarsenic Acid (DMA) detectable in all tissue samples. Concentration doesn’t change with storage at -80°C
- As(III) and As(V) detectable in most, but not all individual liver samples
Results: Incurred Liver (con’t)

• Storage does not change concentration of these iAs species over time
• 3-amino Roxarsone not stable in TMAH extracts
  – Break-down to iAs is slight
• Brief freeze (-80°C)/thaw (RT) does not appreciably impact the concentration of the various As species
• As species not stable when stored under refrigeration
Conclusions

• No appreciable iAs in our drinking water  
(Supports 2009 study findings)
• Roxarsone-medicated feed was stable for duration of the study  
(Stability of test article in feed)
• Detectable levels of total As in livers of birds given medicated feed  
(Extraction of As species from incurred samples)
• Long-term storage (-80°C) of incurred livers does not impact As species concentration  
  – Refrigeration negatively impacts stability  
   (Sample storage)
• Roxarsone was homogeneous in medicated feed, with no evidence of settling of roxarsone  
(Feed homogeneity/stability)
Overall Conclusion

Results from current set of studies supports and extends the findings of the 2009 study.

Results also address some key issues raised in the 2009 study.

http://www.fda.gov/AnimalVeterinary/SafetyHealth/ProductSafetyInformation/ucm257540.htm
Regulatory Impact

• On February 27, 2014, Zoetis, Inc. voluntarily withdrew the new animal drug application for 3-Nitro®, as well as arsanilic acid and carbarsone (two other arsenical new animal drugs) for use in animal feed (including all combinations with other approved new animal drugs).

• On April 1, 2015, Zoetis announced that it would discontinue marketing Histostat (nitarsone), the only remaining arsenic-based animal drug on the market, by Fall 2015.
  – approved for the prevention of histomoniasis (blackhead disease) in turkeys and chickens.
CVM Key Strategic Initiatives

• Food Safety Modernization Act (FSMA) Implementation
• Antimicrobial Resistance Strategy
• Unapproved & Compounded Animal Drugs
• Animal Drugs
• Emerging Technologies