

Exposure to zinc oxide and titanium dioxide nanoparticles affects markers of reproductive aging in female mice

Jessica Alonso, Ramsés Santacruz-Márquez, Adira M. Safar, and Jodi A. Flaws

Department of Comparative Biosciences, University of Illinois Urbana-Champaign, Urbana, IL

Zinc oxide nanoparticles (ZnO NPs) are widely used in paints, cosmetics, and medications, and titanium dioxide nanoparticles (TiO₂ NPs) can be found in food additives and cosmetics. ZnO and TiO₂ NPs are released into the environment, posing a risk to animal health. As females age, their reproductive system also ages and naturally starts to slow down, as evidenced by lower levels of sex steroid hormones, ion imbalance, increased fibrosis, oxidative stress, and inflammation. Interestingly, ZnO and TiO₂ NPs induce oxidative stress and inflammation in several tissues and reach the ovaries. However, the effect of ZnO and TiO₂ NPs on reproductive aging markers is unclear. Thus, this study tested the hypothesis that ZnO and TiO₂ NPs affect markers of reproductive aging in female mice. Adult CD-1 female mice (32-42 days old) were exposed daily to ZnO (5 and 50 mg/kg) and TiO₂ (5 and 50 mg/kg) NPs orally for 10 days. After 10 days, the ovaries and sera were collected from the mice. Then, ovaries were subjected to analysis of inflammatory, antioxidant, collagen, and ion transporter gene expression. Sera were subjected to analysis of systemic inflammatory profile. Both ZnO (50 mg/kg) and TiO₂ (50 mg/kg) NPs increased the inflammatory markers *Icam1* and *Il-1β* compared to control. TiO₂ (5 mg/kg) increased the antioxidant gene *Sod1* compared to the control. ZnO (50 mg/kg) increased the collagen marker, *Colla1*, compared to the control. ZnO and TiO₂ NPs modified 35 serum immune factors compared to control. Collectively, these data suggest that ZnO and TiO₂ NP exposure increases inflammation, collagen, and oxidative stress, potentially contributing to the early onset of reproductive aging in mice.

Research grant: NIH, R01 ES034112

Student support: Office of the Director, NIH, T35 OD011145

Behavioral analysis of CD-1 mice prenatally exposed to Imidacloprid

Anna Batey¹, Selin. Aktuna², Megan. Mahoney^{1,2}.

¹Department of Comparative Biosciences, University of Illinois Urbana Champaign,

²Neuroscience Program, University of Illinois Urbana Champaign

Imidacloprid (IMI) is a neonicotinoid insecticide commonly used in the United States due to the low risk of toxicity in mammals while remaining potent in insects. Despite being known to have effects on the brain, IMI can be found in products such as topical parasite prevention for pets along with outdoor pest control. Currently, it is still not known how IMI affects fear responses in mammalian species with chronic, low-level exposure. Three groups of CD-1 mice were exposed to either 0.5 or 5.7 mg/kg of IMI or an oil control to investigate how no-observed-adverse-effect level (NOAEL) doses impact the central nervous system. Animals were exposed from gestation day one until birth. Beginning at 6 weeks old mice began behavioral testing. In the fear conditioning test females exposed to high levels of IMI had an increased fear response in the tone environment compared to control animals. Additionally, we are testing motor function, and anxiety-like behavior. Finally, our plan is to use fiber photometry to analyze calcium release in an open-field test where we predict we will see heightened activity in the amygdala. As IMI is so commonly used, it is important to understand how neonicotinoids impact the central nervous system. New information can be beneficial in determining accurate and updated safe exposure levels and in determining if IMI is harmful to mammals.

Research Grant: NIH ES035189

Student Support: Office of the Director, NIH, T35 OD011145

Effects of temperature and salinity on fry survival in *Lucania parva*, *Lucania goodei* and their hybrids

Alyssa Blackmon, Timothy Legare, and Becky Fuller

School of Veterinary Medicine (Blackmon) and Department of Evolution, Ecology, and Behavior (Legare, Fuller), University of Illinois Urbana-Champaign, Champaign IL

Bluefin killifish (*Lucania goodei*) and rainwater killifish (*L. parva*) are two closely related species that occur sympatrically in Florida. *L. goodei* is found in freshwater, while *L. parva* is euryhaline. Both occur in slightly brackish environments. Previous work shows that hybrids are rarely found in nature, suggesting strong reproductive isolating barriers. However, the reason why hybrids are uncommon is unclear. The purpose of this study was to determine the effect of both temperature and salinity on offspring survival in *L. parva*, *L. goodei*, and their hybrids. Offspring were raised until hatching under various temperature and salinity conditions. During the egg stage, offspring were raised at either room temperature (~73°F) or high temperature (~83°F) in salinities of 0ppt, 4ppt, or 8ppt. Upon hatching, half of the room temperature larvae were transferred to a cold treatment (58°F) and all other larvae remained in the same temperature and salinity treatments. Survival and mortality rates were monitored throughout development. The identity of the dam had a significant effect on egg diameter and hatch length; offspring with *L. parva* dams were smaller than those with *L. goodei* dams. The hot treatment had a slightly significant effect on hatch rates and a large effect on hatch time. Results comparing length after 14 days and total growth are still being analyzed. The findings of this study will continue to shape our understanding of the relationship between abiotic factors and the reproductive barriers that maintain speciation in this genus and provide insight into whether reproductive barriers will change as salinities and temperatures fluctuate with climate change.

Research Grant: Lab IRC Funds

Student Support: Office of the Director, NIH, T35, OD011145

EPIDEMIOLOGY OF EMYDID HERPESVIRUS-1 IN CHELONIANS IN NORTHERN ILLINOIS

Jenna Camargo, BS, Laura A. Adamovicz, DVM, PhD, Kaitlin Moorhead, DVM, PhD, Gary A. Glowacki, BS, MS, William Graser, Chris Anchor, Kathryn McCabe, Ryland Darling, BS, and Matthew C. Allender, DVM, MS, PhD, Dipl ACZM

University of Illinois Wildlife Epidemiology Laboratory, Urbana, IL (Camargo, Adamovicz, Moorhead, Darling, Allender); Lake County Forest Preserves, Libertyville, IL (Glowacki).

Herpesviruses are commonly detected pathogens in chelonians and variably associated with morbidity and mortality. It is proposed that an increase in the shedding of these viruses occurs during periods of physiological stress, meaning their detection can be used as indicators of overall population health relevant for both epidemiologic and ecological monitoring. Emydid herpesvirus-1 (EmyHV1) has been detected in both captive and free-ranging turtle species, with clinical signs ranging from none to fatal necrotizing pneumonia and hepatocellular and splenic necrosis. While small-scale surveillance efforts in free-ranging painted turtles have been performed, the virus has shown potential for environmental cross-species transmission, indicating that a broader effort may be necessary. This study used quantitative PCR to test eight species of semiaquatic turtles in six counties in northern Illinois for EmyHV1 across a six-year period (2019-2024). Painted turtles showed the highest overall incidence of EmyHV1 detection (8.55%), followed by common musk turtles (4.2%), red-eared sliders (0.53%), and Blanding's turtles (0.12%). Detection peaked in 2024 in Cook County, and almost all positive results occurred in the spring, coinciding with breeding onset. Turtles with clinical signs of upper respiratory disease were 3.85 times more likely to test positive for EmyHV1. Detection was also positively associated with heterophil to lymphocyte ratio, packed cell volume, and monocyte count. Detection was not significantly associated with sex, age class, or other physical examination abnormalities. This study supports that EmyHV1 is primarily associated with painted turtles and may lead to clinical disease. While inter-species transmission appears low, other sympatric species may be susceptible.

Research grant: Illinois Department of Natural Resources State Wildlife Grant, A Walder Biota grant

Student Support: Office of the Director, NIH T35, OD011145

Metabolic glycan labeling allows targeted surface modulation of canine dendritic cells for various applications

Harry Cheong, Johanna Mettler, and Matthew Berry

University of Illinois Urbana-Champaign, Department of Veterinary Clinical Medicine, College of Veterinary Medicine

Cells can metabolize unnatural monosaccharides bearing chemical “tags” (e.g., azide) and express them in the form of glycoproteins/lipids. The cell surface azido sugars then enable targeted conjugation of cargos of interest via click chemistry. Notably, this technology permits targeted conjugation *in vitro* and *in vivo*, and the conjugated cargo may serve a tracking purpose or exert a biological function. This study aims to demonstrate effective azido sugar labeling of canine dendritic cells (DCs) for various downstream applications. Of particular interest, murine research has demonstrated azide labeled and cytokine-conjugated DCs can enhance the antitumor activity of DC vaccines. However, the application of this approach in dogs remains unexplored. Here, we hypothesize that canine DCs will be effectively labeled with azido sugar at non-toxic concentrations and that azido sugar labeling will alter their activation status. One azido sugar, tetraacetyl-N-azidoacetylmannosamine (AAM), was used to label two different canine immune cell types *in vitro*: DH82 cells, a canine histiocyte/macrophage cell line, and primary monocyte-derived dendritic cells (mo-DCs). Effective AAM labeling was confirmed by conjugating dibenzocyclooctyne (DBCO)-functionalized Cy5 and assessing fluorescence via flow cytometry and confocal microscopy. Cytotoxicity of AAM was assessed using SRB and CCK-8 assays. AAM successfully labeled DH82 and primary canine mo-DCs at nontoxic concentrations within 72 hours *in vitro*. Experiments are ongoing to evaluate the activation status (CD1a, CD40, CD80, CD86, CCR7, and MHC class II) of canine mo-DCs +/- AAM labeling. *In vitro* AAM labeling kinetics of canine DCs closely resembles prior research using murine DCs. Our data provides the basis for exploring metabolic glycan labeling applications in dogs. Specifically, we are positioned to develop strategies to enhance DC-based cancer immunotherapy strategies in canine patients.

Research Grant: University of Illinois

Student Support: Boehringer Ingelheim Veterinary Scholars Program, Illinois CVM

Wild to Friendly! Understanding the genetic mechanisms of domestication in the red fox (*Vulpes vulpes*)

Saisrinivas Ennam^[1], Jennifer L. Johnson^[2], Victoria L. Wills^[3], Emma Schulz^[3], Joseph Del Rio^[2], Halie M. Rando^[4], Lyudmila N. Trut^[5], Anna V. Kukekova^[2]

Tuskegee University College of Veterinary Medicine, Tuskegee, AL, 36088^[1]; Department of Animal Sciences, UIUC, Urbana, IL 61801^[2]; UIUC CVM, Urbana, IL 61802^[3]; Department of Computer Science, Smith College, Northampton, MA, 01060^[4]; Institute of Cytology and Genetics of the Russian Academy of Sciences, Novosibirsk 630090 (Russia)^[5]

The red fox (*Vulpes vulpes*) is an ideal species for domestication studies because its domestication process began recently, with the first successful breeding in captivity dating back to the late 18th century; this step took place in Eastern Canada, where foxes were bred for their fur. Initially, the foxes had difficulty adapting to captivity; however, as more generations were produced, their offspring became better adapted to human interaction. Thus, this first stage of fox domestication is both an act of artificial selection and natural selection for survival in captivity. These foxes were then shipped to various parts of the world, some as founder populations for producing foxes with more domestic behavior in the Farm-fox experiment in Novosibirsk, Russia. Previous work aimed to identify genomic regions involved in fox domestication with shallow sequencing for 30 farm and 20 wild foxes and discovered differing haplotype frequencies between farm and wild foxes. Later, 105 farm and 63 wild foxes were used to identify haplotypes enriched in farm foxes in two regions of chromosome 4 (regions 25 and 37). Upon deep sequencing 15 foxes (9 farm, 5 wild), we discovered high-confidence single-nucleotide polymorphisms (SNPs) in these regions. With this data, we searched for polymorphisms that were likely selection targets. With SNPeff and PROVEAN, we identified the effect of each SNP and determined any possible direct effect. We also examined regulatory effects by analyzing the conservation between species. With our results, we can identify the genes responsible for domestic behavior in red foxes and further develop an understanding of the genetic mechanisms behind domestication and apply them to a broader range of species.

Research Grant: NIH grant GM144276, PI: Kukekova

Student Support: Boehringer Ingelheim Veterinary Scholars Program, University of Illinois College of Veterinary Medicine, Tuskegee University College of Veterinary Medicine

A retrospective study of inflammatory arthritis in dogs with blastomycosis (2006-2019): 12 cases

Amanda Finn, Amy Schnelle, Audrey Billhymer, Jennifer M. Reinhart

Department of Veterinary Clinical Medicine, College of Veterinary Medicine, University of Illinois Urbana-Champaign, Urbana, IL

Blastomycosis, a systemic mycosis in dogs, disseminates broadly from the lungs, the initial site of infection, to multiple organ systems. Although uncommon, articular involvement exists as a possible extrapulmonary site of infection. Cases of blastomycosis involving the joints are poorly described in the literature. Therefore, we conducted a comprehensive evaluation of dogs diagnosed with blastomycosis with inflammatory arthritis. A keyword search of the University of Illinois Veterinary Teaching Hospital's electronic medical records system for 'blastomycosis' or '*Blastomyces*' and a charge search for arthrocentesis was ultimately refined to include dogs with evidence of inflammatory arthritis in at least one joint. Twelve dogs with 25 sampled joints were identified. The cytologic diagnosis was inflammatory arthritis in 21 joints (12 dogs), degenerative joint disease in 1 joint (1 dog), unremarkable in 3 joints (2 dogs). The inflammation type was categorized as either suppurative in 11 dogs (17 joints) or pyogranulomatous in 1 dog (4 joints). Intraarticular *Blastomyces dermatitidis* organisms were observed in 6 dogs (10 joints). Although pyogranulomatous inflammation is classically associated with mycosis, 11 out of 12 dogs in this study had suppurative inflammation; suppurative inflammation should not rule out blastomycosis as a differential diagnosis. Of the 6 dogs with multiple joints sampled, 3 dogs had only 1 joint with intraarticular organisms. This, combined with the fact that 4/25 joints were not inflammatory, suggests that multiple joints should be sampled to increase diagnostic yield in the evaluation of arthritis associated with canine blastomycosis.

Research Grant: None

Student Support: Boehringer Ingelheim Veterinary Scholars Program, University of Illinois College of Veterinary Medicine

Comparative analysis of the infectivity of *Cryptosporidium parvum* strains

Dinah A. Foley, Zoë Reynolds, Maria G. Nava, Sumiti Vinayak Alam

Department of Pathobiology, College of Veterinary Medicine, University of Illinois at Urbana-Champaign, Urbana, Illinois, USA

Cryptosporidium parvum is a protozoan parasite and a common leading cause of diarrheal disease and mortality in young ruminant animals (calves, goat kids) and infants. Despite its global health risk, there are currently no effective drugs to treat or prevent cryptosporidiosis in agricultural animals, children, and immunocompromised individuals. There is a significant gap in our knowledge regarding the function of parasite genes in host infectivity and disease pathogenesis. Understanding the role of parasite factors that underlie infectivity would pave the way for future development of effective drugs and vaccines. In this study, we investigated the importance of a novel *Cryptosporidium parvum* secreted protein (SEP) on parasite infectivity using the immunocompromised mouse infection model. Four *Cryptosporidium* strains (namely SEP-tag, SEP-KO, SEP-IId, and SEP-hom) that all expressed a nanoluciferase reporter were available in the laboratory. Interferon- γ -knockout (IFN- γ KO mice) were infected with an equal number of oocysts from all strains, and luminescence measurements of collected feces were taken to measure parasite infectivity. Our results showed that SEP-tag had the highest parasite infectivity, as indicated by the highest relative luminescence, while SEP-KO appeared to infect less, as indicated by its lowest relative luminescence. This data suggests that SEP plays a role in parasite infectivity and possibly host-parasite interaction. We will use quantitative PCR to measure oocyst shedding in fecal samples and conduct histological analysis of infected intestinal tissue to evaluate any changes in villi architecture upon infection with these strains.

Research Grant: United States Department of Agriculture (USDA) Hatch Grant (to SVA)
Student Support: Office of the Director, NIH, T35 OD01114

Chronic exposure to di(2-ethylhexyl) phthalate (DEHP) and diisononyl phthalate (DiNP) on the colon of mice

Amanda LaBarge¹, Angel Martinez¹, Lyda Yuliana Parra Forero¹, Sarah Ibrahim¹, Adriana Andrus¹, Shih-Hsuan Hsiao², Mary Laws³, Jodi Flaws³, Romana Nowak¹

¹Department of Animal Sciences, College of Animal Consumer and Environmental Sciences, ²Veterinary Diagnostic Laboratory, College of Veterinary Medicine, ³Department of Comparative Biosciences, College of Veterinary Medicine at the University of Illinois Urbana-Champaign (UIUC), Urbana, USA

Di(2-ethylhexyl) phthalate (DEHP) and diisononyl phthalate (DiNP) are commonly used plasticizers. They are found in most everyday items including medical equipment, food packaging, and toys. While previous research has been done on subchronic effects of these plasticizers, there is a lack of knowledge regarding potential consequences of chronic exposure. This study looked at the impact of chronic DEHP and DiNP exposure on the morphology and function of the colon in female CD1 mice. Mice (n=88) were fed chow containing DEHP or DiNP (0.15, 1.5, or 1500 ppm) (n=12 per group) for 9 months; control groups (n=16) received corn oil. Urine samples were collected monthly for metabolic analysis to confirm the presence of phthalates. Colonic tissues were collected, embedded in paraffin, and analyzed via immunohistochemistry for altered mucus production (MUC2 antibody), T-cell proliferation (CD3 antibody), and epithelial cell presence (VIL1 antibody). Tissue slides were imaged using NDP. scan 3.2.15 software and Hamamatsu NanoZoomer 2.0 HT. Statistical analyses were performed using GraphPad Prism 9.4.0, and significance levels were set at p<0.05. Colon histopathology was analyzed by the Veterinary Diagnostic Laboratory. This analysis indicated non-significant changes consisting of minimal lymphoplasmacytic colitis, mild goblet cell loss, and mild crypt hyperplasia. MUC2 analysis demonstrated that DEHP did not significantly alter mucus production. Other analyses are still undergoing assessment. Once completed, these results will give new insight into how DEHP and DiNP chronically influence the colon. This can later be expanded to other parts of the gastrointestinal system in mice, with potential extrapolation to other species.

Research Grant: National Institutes of Health (NIH) ROI ES034112 Student

Support: National Institutes of Health (NIH) 5T35OD011145-20 Field of

The role ESR2 expression plays in regulating ovulation in granulosa cells

Natalie Lopez, Ji-Eun Oh, Chanjin Park, Po-Ching Lin, Mary Bunnell, and CheMyong J. Ko

Department of Comparative Biosciences, College of Veterinary Medicine, University of Illinois, Urbana-Champaign, IL 61802, USA. Epivara Inc., Champaign, IL.

Ovulation is the process by which a mature follicle releases an oocyte, a critical event for female fertility. Granulosa cells (GCs) within the follicle are essential for this process, serving both as structural support and as the primary site of estrogen synthesis. Estrogen mediates its effects primarily through nuclear estrogen receptors (ESRs), which function as transcription factors to regulate target gene expression. Prior studies showing global ablation of ESR2 results in ovulatory failure leading us to hypothesize that ESR2 plays a key role in regulating the expression of Pgr and Cox2 - two genes critically required for ovulation. To test this, we generated a GC-specific ESR2 knockout mouse (Esr1-iCre;Esr2-flox/flox, referred to as Esr1-Esr2KO) using the Cre-LoxP system. We compared the expression levels of PGR and COX2 proteins via immunohistochemistry (IHC) and quantified Pgr and Cox2 mRNA levels using reverse transcription PCR (RT-PCR) in Esr1-Esr2KO and wild-type (WT) mice, at 6 hours after hCG treatment under superovulation induction. Our results showed a marked decrease of both PGR and COX2 expression in the Esr1-Esr2KO ovaries, in contrast to robust expression in WT controls. These findings provide strong evidence that ESR2 is essential for the transcriptional regulation of Pgr and Cox2 in granulosa cells, revealing a coordinated ESR2-PGR-COX2 signaling axis critical for ovulation. Understanding this regulatory pathway offers valuable insight into the molecular mechanisms underlying ovulatory dysfunction and may inform future strategies for the diagnosis and treatment of female infertility.

Research grant: NIH, R21 HD094296

Student support: Office of the Director, NIH, T35 OD011145

Mapping dairy herd contact patterns for simulation of infectious disease spread

Amelia Lucas, Ben Blair

University of Illinois Urbana-Champaign College of Veterinary Medicine, Urbana, Illinois

Highly Pathogenic Avian Influenza (HPAI), traditionally seen in wild birds and poultry, crossed into dairy cattle in May 2024. As of May 2025, over 1,000 herds across the United States have been impacted. This emerging disease poses a critical threat to livestock health and production systems. However, modeling of intra-herd contact networks is limited, leading to significant gaps in our understanding of their role in disease transmission. In this study, we developed a comprehensive contact network modeling system to characterize intra-herd interactions within a commercial dairy farm. Using observational data, digital farm maps, and time logs, we systematically tracked direct and indirect interactions among cattle, personnel, and equipment. These data hope to inform the construction of static and temporal network representation of the farm's interconnectivity, illuminating various potential transmission pathways. This network model attempts to build a foundational framework for computational simulations of infectious disease spread, particularly HPAI, within commercial dairy production systems. Preliminary analysis demonstrates dense interconnectivity between cattle, humans, and equipment, highlighting potential vulnerabilities in biosecurity practices. Ongoing analysis will further quantify this risk and evaluate the impact of potential intervention strategies. Collectively, this work establishes a framework from which future disease modeling, biosecurity enhancement, and outbreak response planning at the individual farm level may begin.

Student Support: Boehringer Ingelheim Veterinary Scholars Program, University of Illinois CVM

Vaccine hesitancy, antimicrobial use, and trust in parents and pet-owners: a cross sectional survey

G. Lunaburg, I. Noor, M. Pardo, D. Eldreth, J. Dariotis, R. Smith

Department of Pathobiology, College of Veterinary Medicine (Lunaburg, Pardo, Smith); Department of Health and Kinesiology, College of Applied Health Sciences (Noor); Department of Human Development and Family Studies, College of Agricultural, Consumer, and Environmental Sciences (Eldreth, Dariotis); University of Illinois Urbana-Champaign, Urbana, IL

How humans behave, and their reasons for that behavior, have the potential to impact the spread of disease. Whether or not a person trusts their health provider could affect access to care and preventative strategies are used across human and animal health for both vaccines and antimicrobial products. Understanding behavior and perceptions allows public health professionals to create tailored communication and outreach to counteract disease spread. For this reason, researchers have created standardized methods to study vaccine hesitancy, prescribed and over-the-counter (OTC) use of antimicrobial medication, and trust in health professionals using pre-validated scales. These methods are not yet available for veterinary medicine, however. This project aims to adapt a set of metrics previously validated for people and their children for use in their pets; these include vaccine hesitancy (Vaccine Hesitancy scale), antimicrobial use (Parental Perceptions on Antibiotics (PAPA) scale and PSM-OTC scale), and trust in health professionals (Trust in Doctors in General (T-DiG) Scale, Trust in the Health Care Team (T-HCT) scale). Additional questions were created to collect data on the frequency of use across common OTC antimicrobials as well as preferences for claims on cleaning product labels. We will also compare the results in a population of US parents to determine if the relationship between vaccine attitudes, antimicrobial use, and medical trust is the same among children and pets. A cross sectional analysis of these behaviors allows a one-health approach to vaccine hesitancy, antimicrobial resistance, and medical mistrust, and can inform public health outreach on these topics.

Research grant: None

Student Support: Office of the Director, NIH, T35 OD011145

Tracking and quantifying changes in fitness for transport in cull sows across the marketing channel

Lila Minnick, Sean Dullard, Amelia Lucas, Benjamin Blair University of Illinois
Urbana-Champaign

The pork industry replaces approximately 50% of breeding females in production annually. These females, referred to as cull sows, composed of approximately 3.2 million animals generate 6% of the pork products produced each year within the US. Despite this scale, the cull sow market remains one of the industry's most understudied and poorly understood segments, creating a critical need to investigate the welfare and traceability of these animals from farm to buying station to slaughter. In this study, sow welfare was assessed longitudinally at 2 sow farms supplying a single buying station. Assessment was performed utilizing standardized scoring systems: 1-5 points body condition score (BCS), a 0-3 point lameness scale, and a 0-2 point skin lesion scale. Utilizing these observations, a fitness for transport metric was generated as a combination of all 3 scales including general observed health. We hypothesize that fitness throughout the marketing channel is strongly associated with fitness at transport from the initial farm location. The data generated will support the development of clear, evidence-based guidelines for industry stakeholders to assess welfare and transport fitness of cull sows. By establishing standardized and specific criteria along with scoring scales of significant welfare factors, transport fitness can be positively influenced early in the process to prevent deterioration along their journey. Given the lack of existing research and data on cull sow transport fitness and welfare, this study intends to highlight the critical state of cull sow welfare within farm in an attempt to drive further research to positively impact managerial practices within this segment of the industry.

Research Grant: USDA
Student Support: none

Assessing the effects of phthalate exposure on solubility phase transition during embryogenesis in zebrafish

Moira Roddy, Jin Yang, Meng Ma

Department of Comparative Biosciences, College of Veterinary Medicine, University of Illinois, Urbana, Illinois

Escalating levels of pollution are driving a rise in concern for environmental toxins. Phthalates have held the interest of researchers due to their common use as plasticizers and in a variety of personal care products. Phthalates can function as endocrine-disrupting chemicals, disrupt gene regulation, and have been shown to infiltrate placental tissues— all of which pose a risk to a developing fetus. It is known that phthalate exposure could result in adverse effects during development, however, the mechanisms are not fully understood yet. Our recent study reveals that the solubility phase transition is a novel gene regulatory mechanism important for the oocyte-to-embryo transition and early embryogenesis. Here, we assessed the effects of phthalate exposure on the solubility phase transition during zebrafish embryonic development. We used a phthalate mix composed based on the levels and types detected in pregnant women. We collected embryos via natural mating of wild-type breeding pairs and sorted into 5 treatment groups— vehicle control and 1, 10, 100, or 500µg/ml of phthalate mix. After exposure to treatment between the 1-cell and 1K-cell stage, embryos were either harvested for fractionation/RT-qPCR analysis or cultured in normal medium for 48 hours for morphological assessment. Our results reveal that embryos exposed to 100µg/ml and 500µg/ml phthalate mixture displayed morphological abnormalities when compared to control; however, molecular analysis revealed that the solubility phase transition was not affected by the treatment. Thus, exposure to the phthalate mixture causes adverse morphological effects during vertebrate embryonic development through mechanisms that are independent of solubility phase transition. This study highlights the need for further investigation to determine the factors mitigating the effects on embryo development.

Research Grant: NIH 1R01ES036194

Student Support: Office of Director, NIH, T35 OD011145

Emerging Reproductive Risks of Di-2-ethylhexyl terephthalate (DEHTP): Vascular Disruption in the Uterus

Rachel Roxas¹, Adarsh Ram¹, Athilakshmi Kannan¹, Genoa Warner², and Indrani C. Bagchi¹

¹Department of Comparative Biosciences, University of Illinois at Urbana-Champaign, Urbana, IL 61802, ²Department of Chemistry and Environmental Science, New Jersey Institute of Technology, Newark, NJ 07103

Phthalates are a class of synthetic chemicals commonly used in plastics and personal care products. Di-2-ethylhexyl terephthalate (DEHTP) is a newer phthalate substitute that has been increasingly incorporated into consumer products. Despite its growing use, the health risks associated with DEHTP exposure remain poorly understood. In particular, its effects on the uterus have not been studied, highlighting a significant knowledge gap. To investigate this, we examined uteri from adult CD-1 mice that were orally exposed to vehicle, 10 µg/kg, 100 µg/kg, or 100 mg/kg of DEHTP daily for 10 days. Hematoxylin and eosin staining of uterine sections from DEHTP-exposed and unexposed mice revealed altered vasculature upon DEHTP exposure. To confirm this, we performed immunofluorescence staining for CD31, a well-established marker of endothelial cells. While the 10 µg/kg dose showed no notable change in CD31 expression, uteri from mice exposed to 100 µg/kg exhibited reduced staining, and those treated with 100 mg/kg showed significantly increased CD31 levels, suggesting a dose-dependent dysregulation of uterine angiogenesis. Supporting this, qPCR analyses revealed altered expression of angiogenic factors such as Cx43 and Ccl2 in DEHTP-exposed uteri. Uterine blood vessels are essential for maintaining tissue homeostasis, and both insufficient and excessive angiogenesis can disrupt reproductive function. Our findings demonstrate that even short-term exposure to DEHTP disturbs the delicate vascular balance within the uterus. As environmental exposure to DEHTP continues to rise, these results highlight its potential reproductive risks and underscore the urgent need for further investigation in both animal models and human populations.

Student Support: Boehringer Ingelheim Veterinary Scholars Program, University of Illinois College of Veterinary Medicine

Assessing preservation of culturable anaerobic bacteria from cat feces using BIOME-Preserve™ collection tubes

Lara C. Skarbek, David A. Williams, and Patrick C. Barko

College of Veterinary Medicine, University of Illinois at Urbana-Champaign, Urbana, IL, United States (Skarbek), Department of Veterinary Clinical Medicine, University of Illinois at Urbana-Champaign, Urbana, IL, United States (Williams, Barko)

Anaerobic bacteria regulate intestinal health but investigating them is complicated by their sensitivity to oxygen after sample collection. An anaerobic collection system (BIOME-Preserve™, Anaerobic Systems) preserves viability of anaerobic bacteria in human feces. Herein we assessed whether BIOME-Preserve™ tubes preserve total anaerobic bacteria and *Bifidobacterium* in cat feces under different storage conditions. Feces from healthy cats were used to inoculate BIOME-Preserve™ tubes which were stored at room temperature (RT) for 24, 72, or 120 hours, or frozen at -20°C or -80°C for 7 days. Samples were plated on Brucella blood agar (BRU) and *Bifidobacterium* selective media (BSM) for enumeration of colony forming units (CFU). CFU/mL were compared among fresh feces and samples stored in BIOME-Preserve™. In both BRU and BSM, CFU/mL varied among storage conditions. For BRU samples, CFU/mL were increased in samples stored for 120 hours and -80°C compared with fresh feces and samples stored for 24 or 72 hours at RT. For samples plated on BSM, CFU/mL were increased in samples stored at RT for 72 and 120 hours compared with all other conditions. These preliminary findings indicate that BIOME-Preserve™ tubes can maintain viability of anaerobic bacteria and *Bifidobacterium* in cat feces, but their abundance may be affected by pre-analytic storage. The BIOME-Preserve™ collection system may be useful for studies of cat feces in which maintaining pre-analytic viability of anaerobic bacteria, but not their abundance, is important. Additional studies to replicate these findings and assess preservation of anaerobic diversity using 16S-rRNA sequencing are ongoing.

Research Grant: None

Student Support: Office of the Director, NIH, T35 OD011145

The role of UL47 nuclear export signal in Marek's disease virus pathogenesis in chickens

Authors: Glorianna Wright, Hafiz Sohaib Zafar, Keith Jarosinski

University of Illinois Urbana-Champaign College of Veterinary Medicine, Urbana, Illinois

The poultry industry has seen intensification of Marek's disease virus (MDV) and it does not seem to be halting its growth anytime soon. MDV enters the chicken through the respiratory tract and causes lymphocytic tumors and paralysis. While homologous avian herpesvirus vaccines have been used to reduce the effects of MDV, there is no substance that rids the virus from the environment or the bird completely. MDV matures only in the feather follicle epithelial cells (FFE) of chicken and studies showed that UL47 is essential for the virus to transmit between individuals. UL47 is also believed to transport mRNAs of the virus from the nucleus to the cytoplasm via the nuclear export signal (NES) of the FFEs. So what would happen if the UL47 NES was removed? To examine this, we deleted amino acids 418-432 of UL47 in MDV and tested it in chickens. Chickens (n =10) were inoculated with wildtype, UL47-null, and UL47_NES-null, and replication was monitored over 10 weeks. Another group of uninfected contact chickens (n=10) was housed with each virus to test transmission. After induction of MD in chickens, tumors were collected and used to produce Marek's disease chicken cell lines (MDCCs) for further studies. Following the completion of this study, we will gain a better understanding of how UL47, specifically when it is unable to translocate, affects replication, oncogenicity, and transmission in chickens. This information will help address future viral genes to target in vaccines or therapies for Marek's disease.

Student Support: Boehringer Ingelheim Veterinary Scholars Program, University of Illinois College of Veterinary Medicine, Tuskegee University College of Veterinary Medicine