

Undergraduate Research Symposium May 18, 2018 Mary Gates Hall

Online Proceedings

POSTER SESSION 1

MGH 258, Easel 189

11:00 AM to 1:00 PM

The Effect of Body Heat on Personal Ambient Air Temperature Measurements During Physical Activity

*Gabino Abarca, Senior, Public Health-Global Health
McNair Scholar*

Mentor: June Spector, Environmental & Occupational Health Sciences and Medicine

Heat exposure is associated with several potential adverse heat health effects, including heat-related illness and kidney injury, for example in outdoor workers who labor in the heat. In order to best study and prevent these heat effects, accurate assessment of personal ambient heat exposure is needed. An iButton® is a small coin-shaped data-logging thermometer that can measure and log ambient temperature and humidity. The iButton® has been adapted for use in environmental human health research as an easy to use and affordable data logger for personal ambient temperature measurements. The degree to which personal iButton® measurements are influenced by heat generated by the human body, for example during physical exertion, or solar radiation (iButtons® are contained in stainless steel cases), has not been systematically described. The goal of this study is to quantify the radiated body heat from subjects exercising in both direct sunlight and shaded settings. We recruited a sample of 15 adults, who participated in sessions that were roughly two hours long, with half of the time in a shaded area followed by the same activities in direct sun. Ambient temperature was recorded using iButtons® attached to participants as well as two, five, and ten feet away. Data collected will be analyzed using R 3.4.2 data analysis software. Preliminary results suggest that monitors attached to the participant capture the release of some body heat but that this dissipates by two feet away from the participant. Preliminary results also suggest that, while monitors positioned facing the sun capture some of the effects of solar radiation, monitors in the shade or facing away from the sun do not. This information is expected to contribute to the interpretation of personal ambient temperature measurements used in future studies that examine the relationship between heat exposure and health effects.

POSTER SESSION 1

MGH 258, Easel 183

11:00 AM to 1:00 PM

Assessment of Two Different Quantum Dots on Proliferating and Differentiating Human Neural Progenitor Cells In Vitro for Potential Developmental Neurotoxicity

Elijah Marsh Jung, Senior, Biology (Molecular, Cellular & Developmental)

Joan Lee, Junior, Public Health-Global Health

Youjun Suh, Senior, Biochemistry

Mentor: Sungwoo Hong, Environmental & Occupational Health Sciences

Mentor: Elaine Faustman, Environmental & Occupational Health Sciences, Institute for Risk Analysis and Risk Communication

Nanoparticles have begun to see wide adoption in commercial application particularly in fields relating to medicine and electrical engineering. While their small size provides unique properties furthering advancements in these fields, their effect on human health are still relatively unknown. As their use increases, their potential to cause health effects will need to be considered. This is especially true for workers who manufacture engineered nanomaterials (ENM) and who use them in commercial products. Quantum dots (QD) are an emerging application of nanoparticles using their semiconductor properties for optical imaging. To determine the effects of QD on human neural development, an in-vitro model was established using an adverse outcome pathway (AOP) framework. This methodology provided observations of QD on their ability to alter cellular morphology and developmental endpoints. H9 human neural progenitor cells were cultured and maintained in vitro for up to 21 days in 96 well plates. Normal morphology and biochemistry was characterized under both proliferating and differentiating conditions. H9 cells were then treated with two different QD, Qtracker non-targeted QD and Qdot ITK carboxyl QD, under the concentrations of 0, 2.5, 5, 10, 20, and 40 nM at culture day in vitro (DIV) 1, 6, 13, and 20. After 24 hours of exposures, cell viability was evaluated for dose-response relationships using LDH and MTT assays. H9 cultures undergoing proliferation saw resistance towards Qtracker non-targeted QDs, however cell viability declines with increasing concentrations of ITK carboxyl QDs. H9 with differentiation media under DIV 1 and 6 saw declin-

ing cell viability towards accumulative exposure to Qtracker non-targeted QDs. Nonetheless, H9 cells with differentiation media showed resistance towards ITK carboxyl QDs. In the future, we will investigate the resistance of our cells to various Q dots by assessing dosimetry of QD uptake under our test conditions.

POSTER SESSION 2

Commons East, Easel 76

1:00 PM to 2:30 PM

Baseline Characterization of Testes Co-Culture System

Youjun Suh, Senior, Biochemistry

Elijah Marsh Jung, Senior, Biology (Molecular, Cellular & Developmental)

UW Honors Program

Joan Lee, Junior, Public Health-Global Health

Mentor: Elaine Faustman, Environmental & Occupational Health Sciences, Institute for Risk Analysis and Risk Communication

Mentor: Sung Woo Hong, Environmental & Occupational Health Sciences

Mentor: Collin White, DEOHS

Mentor: Ji Hyun Lee, DEOH

As demand for accurate evaluation of chemicals and their potential to cause effects on humans increases, we have designed in vitro methods to facilitate such assessments. However, models of in vitro cell cultures are limited in their approach typically using a monoculture of limited cell types. Using a 3 dimensional organotypic approach to culture multiple cell types more reflective of a functioning organ can provide a framework for evaluating systematic interactions minimizing the need for in vivo assessment. We utilized a novel organotypic, in vitro model of testicular development that mirrors the development shown by in vivo studies. Our baseline study isolated testes that were harvested from C57BL/6 male mice on post-natal day 9. Testicular co-cultures were maintained for up to 16 days in 24 well plates with data collections occurring at days in vitro (DIV) 3, 7, and 16. Plates were stained with antibodies providing markers that correspond to morphological features of specific cell type markers. Five different primary antibodies were used as markers to visualize the change in cell populations. Markers for Sertoli cells corresponded to Vimentin, Leydig cells with 3b-HSD, Germ cells to DAZL and c-kit, and proliferation with PCNA. For each well, we used an immunofluorescence dual staining technique with Scanning Laser Image Cytometry (iCys) to observe the percentage of cell types and to determine change in phenotypic distribution of the cell population. From our preliminary observations, we were able to observe changes in Sertoli cells, Leydig cells and Germ cells throughout testicular development. Future studies would further develop and confirm the population and morphological changes observed

with the testicular cell cultures. With a baseline model created, this in vitro model can be used to test the lethality of chemicals without the need of in vivo alternatives.

SESSION 20

BIOMARKERS AND DIAGNOSTICS

Session Moderator: Paul Yager, Bioengineering

MGH 389

3:30 PM to 5:15 PM

* Note: Titles in order of presentation.

Refining Oral Swabs as a Non-Invasive Tuberculosis Diagnostic

Rita Noor Olson, Senior, Microbiology

Mentor: Gerard Cangelosi, Environmental and Occupational Health Sciences

Mentor: Rachel Wood, Department of Environmental & Occupational Health Sciences

Conventional diagnostics for tuberculosis use the patient's sputum, mucus from the lower airways, as the primary sample type. Collecting sputum can be difficult and hazardous and for some patients, especially pediatric cases, sputum must be induced through an invasive procedure. An alternative to sputum samples is oral swabs, which have been used to detect the presence of Mycobacterium tuberculosis DNA in clinical trials. However, the current threshold of oral swabs is not yet sufficient for widespread implementation. In a new phase of assessing the viability of oral swab samples, nine different swab types were compared to improve DNA yield and subsequent qPCR readouts. The comparison was based on qPCR Cq values after spiking the swabs with mTB DNA, and analysis of each swab's suitability for culturing. Earlier results suggest that oral swabs are a feasible alternative to sputum samples when using swabs that are heavily "flocked", meaning that they have many small fiber particles deposited across a surface. Moreover, samples performed with dissolvable swabs may be better suited for further molecular and microbiological analysis.

POSTER SESSION 3

MGH 241, Easel 132

2:30 PM to 4:00 PM

Exploring the Oral Microbiota by Region of the Mouth in Healthy Controls and Tuberculosis Patients

Divya Naidoo, Junior, Pre-Sciences

Mentor: Gerard Cangelosi, Environmental and Occupational Health Sciences

Mentor: Rachel Wood, Department of Environmental & Occupational Health Sciences

Oral swab analysis (OSA) is a possible alternative sample type for tuberculosis diagnostics. It has been observed that tongue swabs contain greater amounts of *Mycobacterium tuberculosis* DNA than cheek swabs ($p < 0.0001$). This project investigates whether other oral microbiota follow the same pattern by analyzing oral swabs from healthy controls and TB patients from South Africa. To determine the oral bacterial load, the first step was to optimize a universal bacterial PCR protocol that could be used on real samples. The Com1/769R primer set that amplifies the 16s region of prokaryotic DNA (Dorn-In, 2017) was selected after a literature review and tests with DNA from 5 different bacterial species at known amounts to compare sensitivity. Next, samples were collected from 5 healthy volunteers from Seattle. They provided two swabs each, one from their tongue and one from their cheek, which we then analyzed by PCR. Oral swab samples from South Africa were also analyzed by PCR. We found that the tongue swabs contained greater amounts of total bacterial DNA than the cheek swabs. For the healthy Seattle volunteers, the tongue swabs had an average of 294 times more DNA than the cheek swabs. For the South African samples, the tongue swabs had an average of 13.2 times more DNA than the cheek swabs. Our results suggest that there may be more prokaryotic DNA on the tongue than cheek. Our next steps are to test additional samples from South African subjects. This research could impact the development of future diagnostic tests that detect pathogens within the oral cavity.

POSTER SESSION 3

Commons West, Easel 16

2:30 PM to 4:00 PM

Food Waste and Disposal: Identifying Barriers through a School Waste Audit

Samara Danielle (Sam) Kleinfinger, Junior, Environmental Health, Environmental Studies

Mentor: Tania Busch Isaksen, DEOHS

Mentor: Nicole Errett, Environmental and Occupational Health Sciences

The United States recycles or composts only a quarter of its waste, with the remainder being buried in landfills. Organics, paper, and plastics, which can be recycled and composted, comprise the three largest categories of landfilled materials. It has been demonstrated that familiarizing youth with proper waste disposal methods increases landfill diversion at school and home, resulting in behaviors that continue through adulthood. In this study, I used a solid waste audit to characterize the waste disposal of a large Washington middle school. Initially, I used a survey and focus group to better understand the research needs of this particular school. Targeted goals and specific research questions were then derived from these data, which had been collected from the school's green team. Fifteen students from the green team completed the survey,

while seven participated in a focus group to further articulate their goals and aims. The overall goals were to reduce the school's total amount of food waste and improve sorting efficiencies. Research questions included: how much waste is improperly disposed of, which materials are misplaced most, and how well do teachers sort their waste compared to students. I then conducted the audit during Spring quarter. Waste was collected from three student lunch periods as well as the teacher's lounge and was then sorted into disposal categories, examined for contamination, and weighed. Data analysis included identifying barriers to disposal. In collaboration with the school's green team, I then used my findings to help inform the creation of more targeted and efficient interventions. Participation in the waste audit's design and analysis helped create a more meaningful experience for students to reflect on their own disposal habits. Hopefully, this research may act as a guide or template so other schools may incorporate waste audits into their own recycling or composting programs.

POSTER SESSION 3

Commons West, Easel 31

2:30 PM to 4:00 PM

Transcending Barriers: How Migrant Student Engagement Can Benefit Agricultural Health and Safety

Arthur (Art) Aguilar, Junior, Public Health-Global Health
Mentor: Vanessa Galaviz, DEOHS

Despite the progress made in the health and safety of farm workers and their families, barriers still exist for optimal health and safety, particularly for hard-to-reach populations. Barriers include, but are not limited to, geographic and linguistic isolation which complicates efforts to provide resources such as health and safety education. This project seeks to strengthen farm worker knowledge about occupational exposures and risks, with specific emphasis on pesticides, by engaging and educating children of farm workers (hereafter referred to as migrant children). Engaging migrant children in the form of an interactive class activity would allow them to walk away with increased understanding, thus, translational capacity to improve awareness for their families. The rationale behind an interactive class activity is to understand their experiences and the types of exposures and risks faced by themselves and their family. Additionally, the goal is to introduce these critical concepts to younger members in a community, so they can pursue opportunities that have positive impacts for the health and safety of their family. Pre- and post-assessments will measure change in knowledge and allow for an understanding of the interactive class activity effectiveness in knowledge gained. By engaging and educating migrant students there is opportunity to increase health and safety literacy and capacity to respond to issues among hard-to-reach farm worker populations.

POSTER SESSION 3

Commons West, Easel 14

2:30 PM to 4:00 PM

Communicating Health Risks: Analyzing Ethnic News Media from Vietnamese, Spanish, and Somali Speaking Communities during the Heat and Air Quality Events in King County, Washington

Kim Anh (Kim) Tran, Junior, Environmental Studies, Public Health-Global Health

Mentor: Nicole Errett, Environmental and Occupational Health Sciences

Mentor: Tania Busch Isaksen, DEOHS

King County is home to many non-English speaking populations. Language and cultural barriers must be accounted for in risk communication materials to limit adverse health risks following extreme weather events. During the summer of 2017, Washington experienced a heat wave and increase in air pollution from wildfires. In King County, exposure to extreme heat has been shown to increase risk of all cause mortality by 10%, while poor air quality contributes to 16% of all deaths worldwide.. Despite the growing evidence about communication with non-English populations during extreme weather events, research has yet to examine ethnic media outlets' approaches to communicating health risks. In Washington state, Spanish, Vietnamese, and Somali are the top 5 non-English languages spoken. In response, we sought to assess if and how local ethnic newspapers disseminated risk communication information during the 2017 heat wave and air quality event. We used descriptive statistics to compare the number of ethnic news media messages to the Seattle Times, the predominant newspaper in King County, and assessed the messages communicated based on air quality day. The research findings show that non-English newspapers included fewer risk communication messages, including on days with air quality levels known to increase health risks. On the other hand, the Seattle Times included messages about health prevention methods, daily weather forecasts, and informational articles on the cause of the events. Our findings suggest that there is a need to improve risk communication during wildfire and heat events to populations with limited English proficiency in King County. We recommend engaging members of non-English speaking communities to identify the best communication methods, and work with newspapers serving these communities to promote awareness during future extreme weather events.

POSTER SESSION 3

Commons West, Easel 32

2:30 PM to 4:00 PM

Preventing Heat-Related Illness in Agriculture: An Educational Toolkit for Trainers

Jannah Asirah Amaly, Junior, Public Health-Global Health

Mentor: Vanessa Galaviz, DEOHS

Mentor: Jennifer Krenz, Environmental and Occupational Health

HRI is a serious health risk for agricultural workers that can result from prolonged exposure and overexertion in hot temperatures. The four different types of HRI in increasing severity are heat rash, heat cramps, heat exhaustion, and heat stroke. These conditions are preventable using simple approaches such as proper hydration and limiting one's time in the sun. To better equip agricultural supervisors and safety trainers responsible for conveying heat safety to their workers, I collaborated with members of University of Washington's Pacific Northwest Agricultural Safety & Health (PNASH) Center to develop a modular flipbook that will serve as training material, covering the following topics: symptoms, risks, and treatments, clothing, hydration, and other preventative measures; as well as where and how to cool off in the home and community. I helped advance this project by taking the lead on the production of the flipbook content under the direction of the Research Scientist. I collated images, researched instructional designs, and developed prototypes of the flipbook, as well as edited the written instructions for facilitators. The educational activities in the book consists of an interactive portion, guiding questions for the facilitator to pose to the group, role-play scenarios, take-home points to reinforce the educational messages, and 3-dimensional educational charts. Modules also contain a "Knowledge Evaluation" section with questions for the facilitator to ask participants both before and after the activities to gauge their change in knowledge and to ensure the material has been understood. As a final product, the flipbook will be a portable and convenient resource for safety trainers to use in the field. My goal is to ultimately make this flipbook accessible to the general public, as it is not only a great safety resource for agricultural workers, but also for their families and surrounding community.

POSTER SESSION 4

Balcony, Easel 92

4:00 PM to 6:00 PM

Modeling the Hepatic Ontogeny of Epigenetic Gene Expression in HepaRG Cells

Yasmin Kaori Everson, Senior, Environmental Health

Mentor: Julia Cui, Environmental and Occupational Health Sciences

Mentor: Joseph Dempsey, Environmental and Occupational Health Sciences

During development, profound changes occur in the liver from a hematopoietic organ to a major organ for xenobi-

otic biotransformation. Developmental regulation of epigenetic modifiers, such as factors involved in DNA methylation and histone modifications, may modify the transcriptional output of drug-metabolizing enzymes. The goal of this study was to develop an in vitro model to recapitulate human liver development using HepaRG cells and implement this model in investigating the developmental regulation of epigenetic modifiers and drug-metabolizing enzymes. Previously, HepaRG cells were maintained in proliferative phase and were tested up to 14 days at fully differentiated stage. In this study, we collected cells during differentiation from 0 to 14 days, and isolated total RNA for RT-qPCR of epigenetic modifiers and drug-metabolizing enzymes. Among a panel of 92 epigenetic genes, there were 52 genes differentially regulated at earlier time points as compared to the fully differentiated stage. Most of these epigenetic modifiers increased during differentiation and reached a plateau either at Day 7 or Day 14; whereas the class II major histocompatibility complex transactivator (CIITA), which has an acetyltransferase domain, gradually decreased during differentiation. The adult hepatocyte marker albumin gradually increased and reached a peak at Day 7, which decreased thereafter. The mRNA of the fetal-specific drug-metabolizing enzyme cytochrome P450 3A7 (CYP3A7) was high between Day 2 and Day 3 but decreased rapidly thereafter. Conversely, CYP3A4 mRNA was increased till Day 7, when peak mRNA levels were observed, and decreased thereafter. In conclusion, the present study is a first step in establishing a working model to investigate human liver development in vitro.

POSTER SESSION 4

Balcony, Easel 90

4:00 PM to 6:00 PM

Adaptation of the Bag-Mediated Filtration System for Bacterial Indicator and Pathogenic Bacteria Capture and Concentration

Camila Valdebenito, Junior, Biology (Molecular, Cellular & Developmental)

Mentor: John Scott Meschke, Environmental & Occupational Health Sciences

Mentor: Nicola Beck, DEOHS

Monitoring bacterial indicators and pathogens in environmental waters via filtration is critical in preventing the spread of water-borne diseases. Despite the many filtration methods described in the literature, few have demonstrated applicability to multiple target agents and are adapted for collection of water samples in rural or remote areas. Recently, a novel system for collection and concentration of samples in remote settings was designed and validated. This bag-mediated filtration system (BMFS), originally developed for environmental surveillance of poliovirus, collects water-based samples in a bag then gravity filters the sample through a capsule filter,

which can be easily transported back to the lab. The objective of my project is to adapt and validate the BMFS for capture and concentration of bacterial indicators and pathogens found in environmental waters. To evaluate the adaptability of the BMFS method to bacterial targets, I inoculated a series of 10L of tap water, with either *E. coli*, enterococcus, or an attenuated vaccine strain of *S. typhi* (Ty21a). I filtered the water through a ViroCap™ filter, then eluted the filter using 0.1% tween-20. Recovery rates were quantified using qPCR, defined substrate MPN methods, and spread plating techniques. Results were expressed as percentage of the bacterial inoculum recovered in the eluates. My next steps will be to filter various environmental water types obtained near the University of Washington, in order to assess the effect of water quality on BMFS performance. The experimental design will mimic the procedure described above, except we will be innoculating environmental water instead of tap water. It is our hope that the results of this study demonstrate the potential for environmental surveillance of multiple bacterial targets in remote settings where site location, climate, and road conditions make sample-site access challenging.

POSTER SESSION 4

Commons East, Easel 46

4:00 PM to 6:00 PM

Environmental Justice in Washington State

Maedot A. Yidenk, Senior, Microbiology

Mentor: Lianne Sheppard, Env & Occ Health Sciences; Biostatistics

Studies have shown disparities in environmental justice as manifest by the variation of air pollution exposure based on race and socioeconomic status (SES) on a national scale. This study focused on determining whether a similar association exists in Washington. I hypothesized that in areas where people of color are the majority residents, exposure to air pollution as measured in PM_{2.5}, will be higher. I also hypothesized that there would be a higher average concentration of PM_{2.5} in low-income areas. PM_{2.5} are solid particles with a diameter smaller than 2.5 micrometers. In this cross-sectional study, I used linear regression and analysis of variance to evaluate my hypotheses where I treated income as a continuous variable and majority residency by race as a categorical variable. Overall, I found a negative association between PM_{2.5} and income, which supports the hypothesis. I estimated that PM_{2.5} is expected to decrease by 0.03 $\mu\text{g}/\text{m}^3$ on average per 10,000 dollars increase in income [95% CI: -0.032, -0.02.8]. I controlled for urbanity and education. When I considered effect modification, I found that the negative association was not present in rural areas. In descriptive analyses, I considered four different racial categories (White, Black, Latinos, and Asian). I observed that for areas where black people are the majority, the PM_{2.5} exposure is the highest, with a mean

of 6.86 $\mu\text{g}/\text{m}^3$ [95% CI: 6.82, 6.87], with a maximum of 7.65 $\mu\text{g}/\text{m}^3$ and a minimum of 6.06 $\mu\text{g}/\text{m}^3$. In comparison, in areas where white people are the majority population, the mean was 6.12 $\mu\text{g}/\text{m}^3$ [95% CI: 6.11, 6.13], with a maximum of 8.12 $\mu\text{g}/\text{m}^3$ and a minimum of 3.54 $\mu\text{g}/\text{m}^3$. I conclude that there is a disparity in air quality based on ethnicity and SES in Washington State. Directly or indirectly, environmental injustice contributes a greater risk of health issues and diseases.

POSTER SESSION 4

Balcony, Easel 89

4:00 PM to 6:00 PM

Recovery of Poliovirus from Primary Sludge

Yarrow Linden, Senior, Environmental Health

Mentor: John Scott Meschke, Environmental & Occupational Health Sciences

Mentor: Christine Fagnant, Environmental & Occupational Health Sciences

In some locations (e.g., refugee camps or rural areas in countries like Haiti), fecal waste systems are not water-based, but resemble a pit latrine, for which current sampling and processing methods were not designed. The fecal waste is collected and held, often under poor infrastructure. The fecal sludge management process is important to consider for environmental surveillance, as this may be the only source for screening. This surveillance is important for the continued eradication of poliovirus (PV). The goal of my research was to develop a method for PV extraction from sludge in pit latrines for use in locations with concentrated fecal waste. Towards this goal, I modified two existing protocols for extraction of various viruses from different solid media and applied them to evaluate virus recovery from primary sludge. The first (M1) was originally designed to recover hepatitis A from oysters, while the second (M2) was originally developed to extract somatic coliphages from sewage sludge. The modified methods were evaluated using primary sludge collected from a wastewater treatment plant in Seattle and spiked with the vaccine strain of poliovirus type 1 (PV1). Virus recovery was assessed by a plaque assay. M1 involved acid adsorption followed by glycine elution, threonine elution, skimmed milk flocculation, vertrel and threonine extractions. For M2, liquid and solid fractions were initially separated by centrifugation. Processing of the solid fraction involved beef extract elution, sonication, skimmed milk flocculation, and vertrel extraction, while the liquid fraction was concentrated using membrane filtration and beef extract elution. The preliminary data indicate that M1 may have a higher recovery, and therefore be the preferred method for use during environmental surveillance of poliovirus. Future work should include additional replicates and optimization of the preferred method.

POSTER SESSION 4

Balcony, Easel 88

4:00 PM to 6:00 PM

Transmission Pathways of Maternal and Neonatal Tetanus

*Sophie Claire Morse, Senior, Anthropology, Microbiology
UW Honors Program*

Mentor: John Scott Meschke, Environmental & Occupational Health Sciences

In the past few years, tetanus has caused many cases of the disease, according to the World Health Organization, specifically 34019 newborns died in 2015 from Maternal and Neonatal Tetanus (MNT). Many studies have been done on its transmission pathways and its effect on patients. This project explored research in academic journals and other professional publications with keywords such as “maternal and neonatal tetanus” and “transmission.” We explored existing transmission pathways of tetanus, as well as current recommendations for these specific cases, such as proper wound care regarding umbilical cord cutting and vaccination of pregnant mothers. Such recommendations would be to prevent illness and death, as well as diminishing the risk of infant demise. It was found that tetanus is endemic in certain regions. Its contamination can be prevented through proper wound care, vaccination, and proper umbilical cord practices. This review used the disciplinary lenses of microbiology, clinical medicine, epidemiology, and public health. There is overwhelming evidence pointing to the recommendation to vaccinate in the circumstances of a wound or as a form of prevention. This review will suggest implementation in regards to vaccination and prevention options to accompany current healthcare provider efforts to further assist their endeavors in eliminating the specific presentation of MNT in the remaining 15 countries where it remains a public health threat, such as Mali or Pakistan.

POSTER SESSION 4

Balcony, Easel 87

4:00 PM to 6:00 PM

Environmental Detection of Poliovirus Utilizing Reverse Transcription Recombinase Polymerase Amplification (RT-RPA)

Karissa Crawford, Senior, Microbiology

Mentor: John Scott Meschke, Environmental & Occupational Health Sciences

The last clinical case of wild poliovirus type 2 (WPV2) was in 1999, with eradication declared in 2015. However, the type 2 vaccine strain is responsible for nearly 90% circulating vaccine-derived polio virus (cVDPV2) cases. Poliovirus (PV) is spread in areas with low immunization rates, poor

sanitation and resource limited settings. Environmental monitoring of PV is a useful complement to ongoing disease-based clinical surveillance. Current PV detection methods, such as plaque assays and real time quantitative polymerase chain reaction (RT-qPCR), are costly, time consuming, and require specialized facilities. Reverse transcription recombinase polymerase amplification (RT-RPA) is an isothermal molecular amplification technique that has been adapted for PV2 detection. The high sensitivity and specificity of this method could provide a tool for rapid PV2 screening of environmental concentrates. To simulate testing of environmental samples, we collected and concentrated raw sewage from a local wastewater treatment plant using ViroCap filters. We performed secondary concentration using one of two methods, skimmed milk flocculation (SMF) and polyethylene glycol (PEG) precipitation. We added known amounts of PV2 to each concentrated sample, and compared detection results between RT-RPA, RT-qPCR and plaque assays to determine any matrix associated inhibition. RT-RPA detects target RNA with a change in fluorescence (mV). A change >200 mV indicates a positive reading for PV2. I conducted experiments with varying initial PV2 concentrations, and this resulted in positive detection by RPA in 46% of SMF and 88% of PEG samples. All RT-qPCR and plaque assay results yielded positive PV2 recovery. These preliminary findings suggest inhibition in SMF samples; further testing is required to determine the cause of inhibition. Future research should include the addition of PV2 directly into wastewater samples prior to concentration, to determine recovery with RPA. This is important for the successful implementation of RT-RPA with environmental samples.