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Poster Session

Kentucky Water Resources Research Institute, University of Kentucky

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Poster Session

1. Advancing Prediction of Headwater Flow Permanence and Stream Expansion and Contraction Using a Process-Based Hydrologic Model

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Streamflow in headwater systems supports both ecosystem functioning and water quality across stream networks by facilitating hydrologic connectivity between upstream and downstream watershed compartments. Additionally, knowledge of streamflow permanence characteristics in headwaters is particularly important for their federal protection. However, characterization of the frequency, magnitude, and duration of flow in headwater streams remains limited – largely due to the scarcity of both monitoring efforts and the development of process-based models to understand streamflow in these types of systems. Recent advancements in process-based, semi-distributed hydrologic models show promise for characterizing streamflow in headwater systems where data collection may be difficult, but this is largely untested at catchment scales. The objectives of this study were to: (1) develop and test an approach for simulating the frequency, magnitude, and duration of headwater streamflow with a process-based, semi-distributed hydrologic model and (2) apply model outputs to map the spatiotemporal dynamics of headwater expansion and contraction throughout the stream network.

We modified and applied a highly resolved hydrologic model (Dynamic TOPMODEL) to a 1-km2 headwater network in University of Kentucky's Robinson Forest, located in the Appalachian region of Kentucky. We evaluated model performance using discharge data at the watershed outlet, reach-scale flow-state sensor data, and observed headwater extent collected from field reconnaissance. The model framework performed well at simulating flow across the watershed uplands and characterized important stream dynamics within reaches at a high spatiotemporal resolution. The model successfully estimated the probability of streamflow permanence at the reach scale and simulated network scale dynamics of streamflow expansion and contraction. This study underscores the potential for watershed-scale, process-based hydrologic models to characterize headwater streamflow dynamics in systems throughout the eastern United States.

2. Developing ANN Model for Predicting Lake Michigan E.Coli Counts

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During beach goers season, Indiana Department of Environmental Management (IDEM) conducts regular beach sampling at different beaches located in the southern tip of Lake Michigan. Using the test results, beach advisory is made. As this procedure requires time to get E.Coli results, efforts were made in the past to develop E.Coli prediction model using other information to develop an instantaneous decision. In this research work, efforts are made to develop a functional approximation model using Artificial Neural Network model to predict E.Coli. For accomplishing this model development, sampling was conducted in five beaches of Lake Michigan during 2019. During this sampling work, along with water sample for E.Coli, other variables such as total dissolved solids, total dissolved solids, pH, Electrical conductivity, water temperature, beach conditions such as pet counts, beach visitor counts, wave conditions, other meteorological parameters such as humidity, rainfall, temperature, flow observations in creeks draining to southern end of Lake Michigan. After consolidating the data, using different training algorithms, ANN models were developed by dividing the data into training, testing and validation data sets. E.Coli outputs were predicted as classified outputs by dividing the E.Coli into 4 categories.

Category 1:	0 to 125 –	Safe
Category 2:	126 -235 –	Advisory – still safe
Category 3:	236- 799 –	Unsafe
Category 4:	> 799 –	Highly unsafe

Overall, the results indicate 65 % of predicting ability.

Acknowledgement:

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3. Examining Long Term Trends in Rainfall and Stream Flow at Upper Wabash River Basin Using Self Organizing Map

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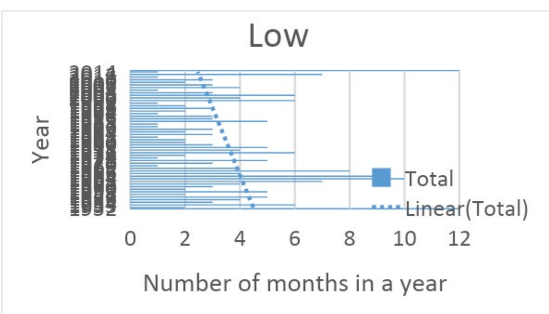
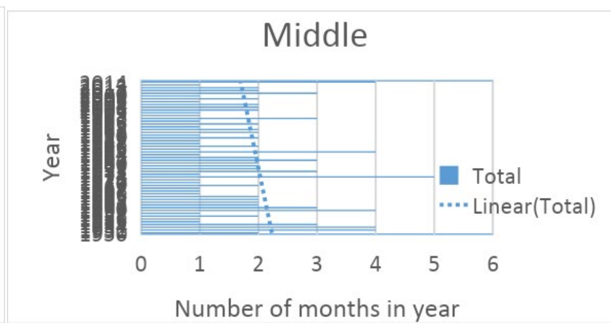
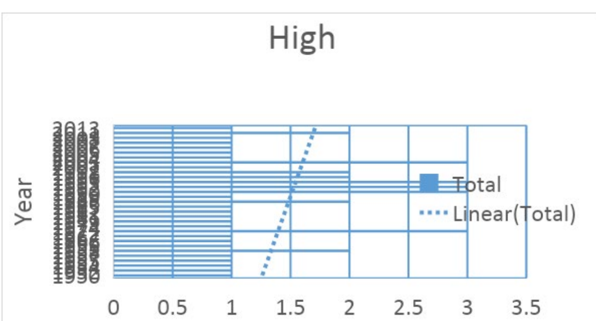
Self Organizing Maps (SOM) is an artificial intelligence technique which is popularly used to cluster similar data during data mining. This tool is also used in the past by researchers to study the long term trends in hydrologic variables. SOM models function differently than the regular ANN models, which are popularly used as a functional approximation tool. The SOM model uses a predefined topology set by users and clusters the data to different neurons during training process. It functions as an effective clustering algorithm. Data associated with each neuron are similar in characteristics. By grouping adjacent neurons, regional data can be grouped into different categories to examine trends. In this study, Upper Wabash River Basin in Indiana was considered. This watershed is located in the central Indiana region. Wabash river is a major tributary to Mississippi-Missouri river system. In stream flow analysis, 12 USGS flow observation stations located in this watershed were considered. These stations cover several tributaries and main stream. To make the data consistent, monthly data from 1950 to 2015 were considered. In a similar way, for the rainfall analysis, monthly rainfall data for 5 counties were used in another trial to group months into different clusters using SOM. Both these results were examined together to study the long term trends. For examining the trends, number of months for each cluster in each year were captured for Low flow, Medium flow and High flow categories. Low flow magnitudes show a declining trend in the recent past, but the high flow clusters shows an increasing trend.







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4. Investigating Water and Sediment Transport Processes with High-Resolution Sensor Measurements and Hysteresis Analysis in the Cane Run Royal Spring Basin, Kentucky, USA

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During storm events, a large amount of water and sediment is mobilized. One way to analyze the behaviors is to use hysteresis. Hysteresis analysis has been used extensively to investigate the behaviors of nutrients and sediments in storm events in surface streams. Discharge vs. sediment concentration hysteresis can give insight into how sediment is distributed throughout the watershed catchment, while electrical conductance vs. sediment concentration tells a story about the deposition or reuptake of sediment. Less research has been carried out in karst settings, which can be of great benefit to understand the characteristics of sediment transport in underground fractures and caves. Recent developments of high-resolution sensors also make it possible to collect a wide variety of water quality parameters to be monitored more feasible than previously. Electrical conductivity sensing provides an approach to water origin tracing, and turbidity sensing provides a surrogate to estimate sediment pulses through hydrologic systems.

Our goal is to gain an understanding of hydrologic transfers in karst basins, especially the transfer of water and sediment analyzed via hysteresis, and to use this information for developing a predictive model of daily water and sediment loads daylighting at a karst spring. This study is investigating a karst spring located in the inner bluegrass area of central Kentucky, which is known for its high karst potential and mature karst development. The Royal Spring basin is located between the city of Lexington and the city of Georgetown and functions as the primary drinking water source for Georgetown while draining the Cane Run Royal Spring basin. Several high-resolution sensors have been deployed at the spring and across the basin for the past 10 years and have collected data in 10-minute and 15-minute intervals. Flow data has been measured and calculated with a Marsh-McBirney 201-D velocimeter or obtained by downloading the data from the corresponding United States Geological Survey gage station. Sensor data was collected with a YSI 6920 V2-2 Multiparameter sonde, a YSI 6600 V2 Multiparameter sonde, and two YSI 600 OMS V2 Optical Monitoring sondes.

This data made it possible to learn about the transport mechanisms in the aquifer and determine the differences between subsurface sediment transport and surface sediment transport in the Cane Run Royal Spring basin. During storm events, water and sediment are transferred from the surface stream network to the karst aquifer and primary cave via numerous swallets in the creekbed. The hydraulic forces produce a fast-response water discharge peak at Royal Spring followed by the delivery of surface water and sediment at the spring 6 to 18 hours later. Water-sediment hysteresis analyses show counterclockwise behavior, which is less widely reported in hysteresis studies, but based on further analyses of data reported by others appears to be a consistent behavior of karst aquifers. Turbidity-conductivity hysteresis suggests either sediment is resuspended in the cave during the initial pressure pulse or the concentration of sediment in water entering the karst cave via the swallets is varying pronouncedly across the event. Un-mixing analyses are ongoing to estimate which process is controlling for this system. Comparison of karst basins across five continents is ongoing. The development of predictive water transfer and sediment load models is also ongoing.

5. Blue Water Farms: Edge-of-Field Water Quality Monitoring of Nutrient and Sediment Loss from No-Till Corn and Soybean Fields in the Lower Cumberland River Watershed

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Nutrient (nitrogen and phosphorus) and sediment derived from urban construction as well as food production activities are leading contaminants resulting in stream and river impairment in Kentucky. While agricultural producers commonly employ best management practices (e.g., crop rotation, cover crops, no-till, etc.) to mitigate nutrient and sediment losses to retain nutrients in-field, studies evaluating the efficacy of best management practices on the reduction of nutrient and sediment in agricultural runoff are limited in western Kentucky. To further understand the relationships between agronomic practices and water quality, researchers from the University of Kentucky have collaborated with the U.S. Department of Agriculture Natural Resources Conservation Service, Kentucky Soybean Promotion Board, Kentucky Agriculture Development Board, and Kentucky row-crop producers to conduct edge-of-field water quality monitoring in the lower Green River watershed. This project is part of a national effort to evaluate the efficacy of best management practices and assist the agricultural community in making informed nutrient management decisions.

Watershed analysis was conducted in the lower Cumberland River watershed to identify 12 watersheds within no-till corn/soybean rotation fields. LIDAR and survey data were used to subdivide the fields into 6 control watersheds and 6 treatment watersheds ranging in size from 3 to 10 acres. In each watershed, a monitoring station was instrumented with a flume, ultrasonic flow sensor, flow meter, rain gauges (manual and tipping bucket) and a composite water sampler.

Year-round sampling of surface water runoff from no-till corn/soybean fields to monitor changes in nutrient and sediment loads is in progress. Data will be collected over an 8-year period, including 2 years of baseline data and 6 years of control/treatment data. In the 12 paired watersheds, the control watersheds will be planted with a wheat cover crop, while the treatment watersheds will be planted with a cereal rye cover crop after the calibration period.

The results of this project are expected to help agricultural producers implement best management practices that reduce erosion and improve on-farm nutrient retention in Kentucky. Monitoring data are also expected to be utilized for modeling nutrient and sediment losses under differing farm-management practices. If effective best management practices are implemented on a larger scale, this project may guide efforts to mitigate sediment pollution and downstream eutrophication (e.g., the Northern Gulf of Mexico Hypoxic Zone).

6. Determination of Microcystin Cyanobacterial Toxins in Kentucky Lakes by Diffusive Gradients in Thin Films*

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Numerous public health advisories have been issued at several KY lakes and reservoirs due to elevated cyanotoxin concentrations, particularly microcystins, which are cyclic peptide hepatoxins produced by several cyanobacterial genera including primarily *Anabaena*, *Microcystis*, and *Planktothrix* (Fig. 1).

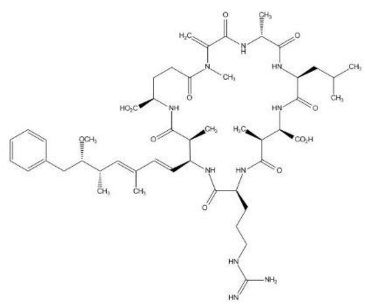
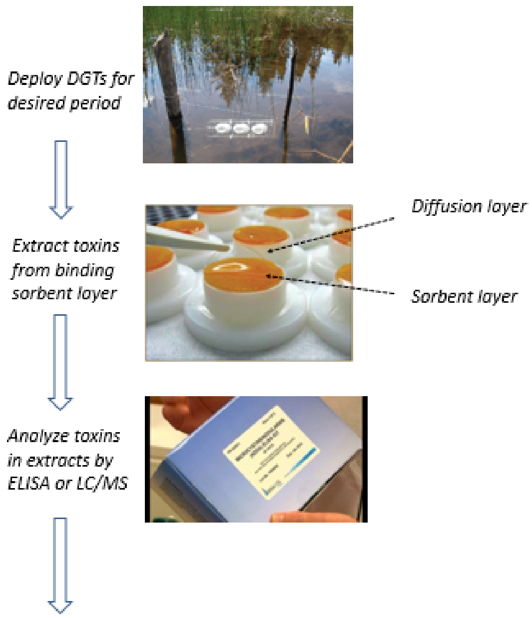


Figure 1. Structure of microcystin-LR, the most commonly detected cyanotoxin in KY lakes.

The traditional approach for collecting water samples for cyanotoxin determination is point-in-time grab sampling, which is useful for determining total cyanotoxin concentrations at a particular point-in-time, but on the downside, grab sampling is not able to determine cyanotoxin concentrations over a longer period without intensive sampling, which makes grab sampling too impractical and expensive for making accurate risk assessments.

To overcome the shortcoming of grab sampling, passive samplers have been developed which typically contain a sorbent that continuously accumulates toxins in water during the sampler deployment period, which is typically several weeks. The most common type of passive sampler for detecting cyanotoxins in water is the "Solid Phase Adsorption Toxin Tracking" (SPATT) sampler which contains sorbent in a mesh bag which accumulates cyanotoxins, which can then be extracted and analyzed with concentrations expressed on a mass sorbent basis. The main benefit of SPATT sampling is its ability to detect presence/absence of cyanotoxins in the water, but has the shortcoming that cyanotoxin concentrations in the sorbent are not related to concentrations in water, which makes results difficult to interpret for health advisory purposes.

Another type of passive sampler, referred to as Diffusive Gradients in Thin Films (DGT), overcomes the weaknesses of grab and SPATT sampling by including a diffusion layer of known thickness over a sorbent layer which allows cyanotoxin concentration in the water to be related to cyanotoxin mass in the sorbent layer by Fick's First Law of Diffusion (Fig. 2) (D'Angelo, 2019. Development and evaluation of a sensitive Diffusive Gradients in Thin-Films (DGT) method for determining microcystin-LR concentrations in freshwater and seawater. Harmful Algae, 89:101688).



$$\text{Toxin concentration in water} = \frac{\text{Toxin mass in sorbent layer x Thickness of diffusion layer}}{\text{Area of sampler x Time x Diffusion coefficient in diffusion layer}}$$

Figure 2. Overview of the Diffusive Gradients in This Films passive sampler method which will be used to determine microcystin concentrations in eight KY lakes during the summer 2021 harmful algal bloom season.

The main objective of this study was to determine microcystin concentrations in eight KY lakes using DGT samplers and enzyme linked immunosorbent assay (ELISA) analysis and to relate microcystin concentrations to water quality properties during the summer 2021 harmful algal bloom season. Another goal is to train undergraduate environmental scientists in limnological research and provide them opportunities to present results at scientific conferences sponsored by KWRRI and University of Kentucky Office of Undergraduate Research.

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7. Environmental Conditions on the Lower Ohio River with Comments on Phytoplankton Assemblages

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The Ohio River has had a long history of water quality problems and is considered one of the top 10 most polluted rivers in North America. The river has experienced severe noxious cyanobacteria blooms in recent years, particularly in upstream regions from Huntington, WV, and Cincinnati OH, sometimes extending downstream of Louisville, KY. As part of a larger study to predict potential causes of toxic cyanobacteria blooms on the Ohio River, this study describes water chemistry and phytoplankton community dynamics at two shoreline sampling sites on the lower river; one at Paducah, KY, immediately downstream from the mouth of the Tennessee River and one directly across the Ohio at Brookport, IL. The study area represents the downstream end of the 1575 km long Ohio River, the width of which can exceed 3.2 km during flooded conditions.

Water samples for nutrient analyses were collected every 16 or 32 days in coordination with the long-term monitoring program at Hancock Biological Station (HBS) from January 2017 through October 2020. Physicochemical parameters (temperature, dissolved oxygen, turbidity, pH, ORP, and specific conductance) were measured using a YSI multiparameter sonde at the 1 m depth. Ammonium, nitrate+nitrite-N, total nitrogen, soluble reactive phosphate, total phosphorus, sulfate, chloride, silica, calcium, and chlorophyll a were analyzed using APHA and U.S. EPA methods. Phytoplankton samples were identified to genus using a Zeiss Axioplan microscope (400X mag, Palmer-Maloney cell). Analysis of variance (ANOVA) was carried out on water quality data using SYSTAT 13.2.01 for Windows (2017); additional analyses of phytoplankton data (e.g., PCA, CCA) were carried out using XLSTAT (2021).

Temperature, dissolved oxygen, and pH patterns showed no difference between the two sites. All other variables were significantly different between the Brookport and Paducah sites (2-factor (site, season) ANOVA, p values ≤ 0.05). Specific conductance, turbidity, nitrate+nitrite, total nitrogen, total phosphorus, sulfate, chloride, and calcium ions were significantly higher at Brookport than at Paducah. Soluble reactive phosphate was significantly higher at Brookport during spring; ammonium was higher only during fall with much variation in both parameters during other seasons. Silica was significantly higher at Brookport during all seasons except summer. Chlorophyll α also was significantly higher at Brookport during summer and fall than at Paducah. We attribute the differences to represent downstream accrual of ions and nutrients from upstream sources in the mainstem Ohio River at Brookport and the "cleaner" inputs of more dilute inflow from the Tennessee River just upstream of the Paducah site.

Three major algal classes, Bacillariophyceae, Chlorophyceae, and Cyanobacteria, represented the dominant taxa at both sites. Representatives of other classes, the Euglenophyceae, Cryptophyceae, and Dinophyceae, and Ochrophyceae were all < 5% of the total. A total of 81 genera were collected at Paducah and 77 genera at Brookport. Similarity index (SI) (e.g., coefficient of community) between Paducah and Brookport was 0.93 indicating the two sites were very similar in composition. The diatoms (Bacillariophyceae) far outnumber (as % of total cells) all other taxa at both sites (ranging 60%−80% at Paducah and highest during winter; and 60%−70% at Brookport and highest during spring). The Chlorophyceae were the next most numerous class ranging from 10%–20% of the total cells at both Brookport and Paducah and fairly consistently represented throughout the year. Cyanobacteria ranged from 5%–22% and were highest during summer and winter at Brookport and 5%–25% of the total at Paducah and highest during summer. The top five genera with highest relative abundances at Paducah were *Aulicoseira* (37.1%), *Navicula* (9.2%), "coccoid" chlorophyceans (11.4%), *Cryptomonas* (11.4%), and *Microcystis* (6%). The top five genera with highest relative abundances at Brookport were *Aulicoseira* (34.3%), *Cyclotella* (7.1%), *Synedra* (12.1%), "coccoid" chlorophyceans (10.6%), and *Pseudanabaena* (9.8%).

Temperature was the most important driver of each of the abundances of major taxa. While the water chemistry between the two sites was significantly different, phytoplankton assemblages were quite similar. Further exploratory analyses of potential physicochemical drivers of phytoplankton abundance or community structure did not reveal significant trends.

In conclusion, when the Brookport and Paducah phytoplankton assemblages are viewed within the construct of ecological "functional groups", the most common groups typically are representative if the lower reaches of very large rivers (turbid, slow flow, high light at the surface, lower light penetration at depth). At no time during the study period were the Cyanobacteria or other potential HABs taxa found to be either dominant components or in bloom conditions in this area of the Ohio River. We attribute this to the fact that pooling (stagnation) does not occur here, nor are nutrients in concentrations high enough to support the exponential growth of cells to reach bloom conditions. This study presents updated algal and water chemistry information for the lower Ohio River.

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8. Restoring Kentucky Streams Containing the Threatened Arrow Darter

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A mitigation sponsor, Ecosystem Investment Partners (EIP), partnering with Kentucky-based engineering and construction firms, Beaver Creek Hydrology (BCH) and Stream Restoration Specialists (SRS), has achieved a milestone in the environmental restoration sector in the state of Kentucky. EIP's 930-acre mitigation bank in Breathitt County, Kentucky, has successfully restored 800 linear feet of habitat specific to the federally threatened Kentucky arrow darter (*Etheostoma spilotum*) and received an Outstanding State Resource Water (OSRW) designation from the Kentucky Division of Water (KDOW). The reach was nominated in 2018 and officially listed as an OSRW on January 3rd, 2020. This designation has not yet been granted to any other private mitigation bank projects in the state.

The restored reach is part of the North Fork of the Kentucky River Stream Mitigation Bank located in the Frozen Creek watershed near Campton, Kentucky. The Mitigation Bank has restored 107,540 linear feet of previously degraded stream. The project area was previously used for farming, grazing, and logging but is now comprised of over 20 miles of restored and permanently protected headwater streams.

To facilitate the recovery of the Kentucky arrow darter within the Graham Branch watershed, the project used Natural Channel Design methods to replicate their ideal habitat. Design features included increased run/glide habitat, shaded pools, and establishment of native riparian vegetation that is typically found in conjunction with stable populations of the darter (i.e. eastern hemlock and rhododendron). The effects of upstream restoration have also contributed to the success of the arrow darter through the significant reduction of sediment and increased water retention.

Field sampling efforts were led by Mike Floyd, PhD., with the Kentucky Ecological Field Office of the U.S. Fish and Wildlife Service (USFWS). Three surveys have been conducted to monitor the population of the Kentucky arrow darters throughout the restoration project. A pre-construction survey on November 17, 2016 initially identified four adult Kentucky arrow darters in the downstream reaches of Graham Branch. A second survey was conducted a year after construction on December 6th, 2018 and identified a total of 5 Kentucky arrow darter individuals. The most recent sampling event took place on August 24, 2020 and identified eighteen Kentucky arrow darters as well as two individuals that were observed but not captured. Both post-construction surveys have shown the migration of the arrow darter further upstream than the initial survey in 2016. Figure 1 shows the size-frequency histogram for the eighteen individuals captured during the 2020 sampling event. These data demonstrate the presence of two distinct age classes (juvenile and adult) on the site indicating successful reproduction and recruitment in 2019 and 2020. The increase in the total number of individuals and the proliferation of juvenile individuals, as well as the expanded range of the fish, suggest that the restoration project has successfully improved arrow darter habitat on the site.

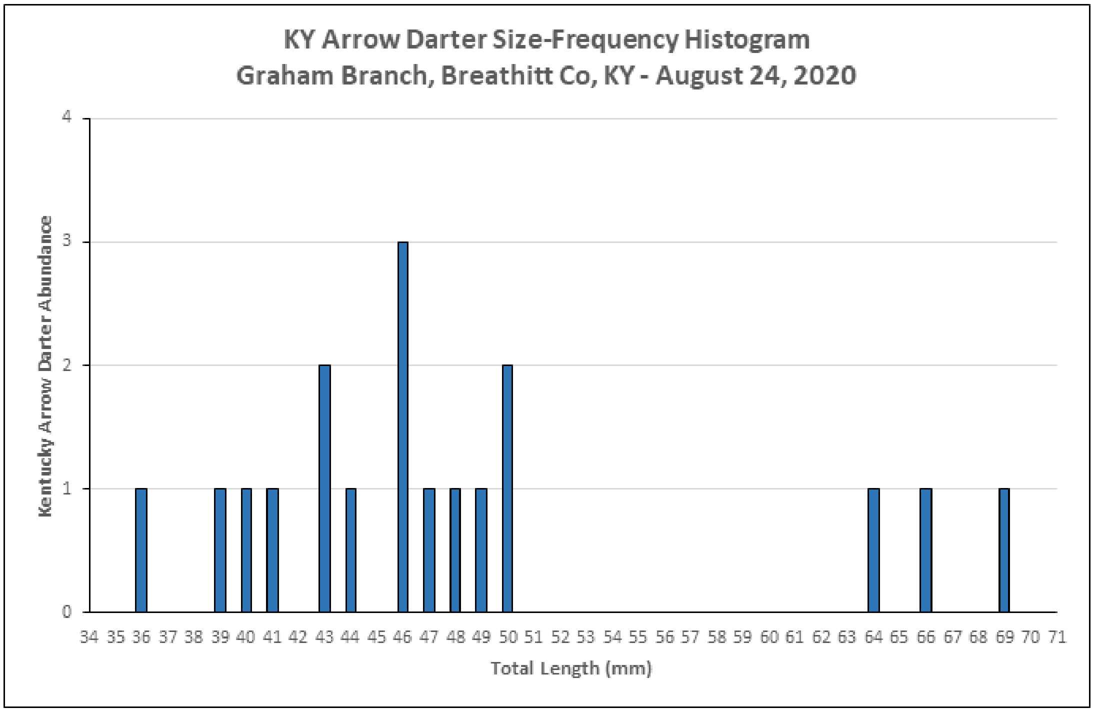

Figure 1. Size-frequency histogram for Kentucky arrow darters collected in Graham Branch during the 2020 sampling event three years after construction.

9. Comparisons of Conductivity and Chloride Concentrations in the Upper Ohio River Valley During Summer and Winter Months

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During hot and dry summers, streams are affected by high water temperature that may lead to low flow conditions that could increase conductivity concentrations and stress stream organisms. During the winter the use of road salts can increase chloride and conductivity concentrations in streams through runoff of salts from impervious surfaces. We collected water chemistry data weekly throughout the Upper Ohio River Valley near Wheeling, WV over the last two years and examined summer and winter trends in conductivity and chloride. We defined summer as June 1st through September 30th and winter as December 1st through February 29th. In the winter of 2020 (December 2020 to February 2021) 90% of the sites had significantly higher chloride levels compared to the winter of 2019 (December 2019 to February 2020). Conductivity concentrations in 2020 were significantly higher at 57% of the sites than in 2019. During the summer of 2020 chloride levels were significantly higher at 50% of the sites compared to the summer 2019. Conductivity concentrations at 58% of the sites were significantly higher in 2020 than in 2019. If these patterns continue, we should expect to see higher conductivity and chloride concentrations becoming more common.

10. Development and Optimization of Green Polymer and Solvent-Based Ultrafiltration Membranes for Water Treatment Applications

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Polymeric membranes are commonly utilized for water treatment applications and are fabricated via phase inversion due to the ease in altering the permeance and selectivity; nonsolvent phase-induced separation (NIPS) is regarded as the dominant phase inversion method. However, one critical drawback is the use of petroleum-based solvents that pose significant hazards to human health and the environment. Moreover, the nature of traditional solvents increases the overall environmental impact of membrane technology, including the large-scale generation of membrane fabrication wastewater that typically undergoes minimal treatment to remove solvents. As such, regulations on solvent use have recently increased and further motivate the need to develop polymeric membranes with green properties (i.e., non-hazardous, recyclable, bio-derived, and/or biodegradable characteristics).

Among potential green solvents, Methyl-5-(dimethylamino)-2-methyl-5-oxopentanoate (Rhodiasolv® PolarClean) and γ-valerolactone (GVL) are considered ideal candidates for membrane fabrication due to their intrinsic properties. The sustainability of membranes can be further enhanced by using recycled polymer materials, namely polyethylene terephthalate (PET) due to its excellent thermal and chemical resistance properties. Moreover, integration of PET into membrane fabrication could create a high-value niche application for PET recycling and reduce plastic pollution.

In this study, PET-PolarClean-GVL ultrafiltration membranes were fabricated via NIPS and compared to other polymeric membranes. In addition to measuring water flux and BSA rejection, evaporation time was altered during NIPS to study its effect on these parameters; membrane characterization was conducted using FTIR and SEM microscopy to analyze the morphology. The end-products of this study are ultrafiltration membranes for water treatment applications with adequate performance parameters and green properties to reduce environmental impacts, as well as guidance for the fabrication and optimization of green polymeric membranes.

11. Municipal Water Quality Concerns and Rebuilding Trust in a Rural Community

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Introduction: A community engaged research approach was used to explore customer perceptions of municipal water quality, accessibility, and affordability in a rural Appalachian county with a complex history of environmental and socioeconomic challenges.

Methods: Academic researchers and community members collaborated to develop and administer a survey instrument that included a health survey and seven open-ended questions. A stratified random sample of 73 water district customers responded. Interview transcripts were iteratively coded and analyzed for emergent themes using NVivo 12 (QRS International) software.

Results: The data suggested respondents generally perceived water quality as poor and only 12% used their tap water for drinking. Many felt they had insufficient knowledge regarding tap water and used a wide range of news and social media sources to access water-related information. Themes of historically-rooted distrust of the water district and local leadership emerged as did perceived inequities for customers.

Implications for Public Health: Opportunities to rebuild public trust through stakeholder engagement and multidirectional risk communication are explored.

12. Application of a Water Treatment Inspired Technique on a 3D Support for Air Filtration

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The Covid 19 pandemic has led to growing demands for personal protective equipment (PPE) to effectively control the spread of the virus. Facemasks are an effective defense against aerosols containing pathogenic bacteria and viruses such as Sars-Cov-2. Membrane filters have been used extensively in face masks to remove these microbes from the air. These filters are usually designed for single-use due to inadequate and laborious cleaning/decontamination techniques. This work attempts to make a breathable antiviral face mask by immobilizing silver nanoparticles (AgNPs), which could suppress bacterial and viral activity on a cellulose acetate (CA) membrane filter required for mask production. AgNp was chemically immobilized by attaching a polymerized epoxy, glycidyl methacrylate (GMA) to CA, allowing for more functionalization of the CA/GMA copolymer. Cysteamine was then combined with the CA/GMA complex, providing thiol groups that immobilized AgNP's on the membrane surface. FTIR analysis confirmed the successful polymerization of the monoGMA, while electron microscopy and X-ray energy dispersive spectroscopy was used to verify the presence of silver on the CA membrane surface. The resulting membrane filters are quite thin and require support for use in mask production. We have used a 3D printed support to ensure strong membrane filters for mask production in this work. An airflow test was carried out on the unmodified CA membrane on a 3D support to ensure breathability. A high airflow resistance was observed through the membrane at pressures up to 10psi. This was hypothesized to be due to small pore sizes inherent in biobased membranes. Therefore, polyethylene glycol (PEG), an organic chemical known to form pores in membranes, was introduced in the dope solutions, and subsequent increases in pore sizes and air permeability were observed.

13. Comparison of Leaf Litter Bag and Environmental DNA in Detection of Salamanders in Maywoods Environmental and Educational Laboratory

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Environmental DNA (eDNA) utilizes DNA released from aquatic organisms into the environment to detect their presence and provides an effective, non-invasive method to survey organisms in an efficient manner. While the majority of early eDNA studies focused on single species presence/absence, a number of more recent studies suggest a relationship between eDNA levels and biomass. This observation has generated considerable interest in the relationship between traditional sampling methods and eDNA, considering the potential benefits of eDNA in enhancing the ease of organism detection. We will survey the salamander community of Maywoods Environmental and Educational Laboratory (Garrard and Rockcastle County, KY) using both traditional (leaf litter bags) and molecular (eDNA) assessment methods. Briefly, leaf litter bags will be placed at 3 m intervals at each sampling location. Water samples will be collected concomitantly below the lowest leaf bag at each site on each sampling day and filtered in the lab within 24 hours. eDNA will be extracted following established laboratory protocols and quantitative PCR will be used to detect salamander DNA. Preliminary data indicates detection of Eurycea cirrigera (southern two lined salamander) in leaf litter bags in two of three sites but in all three sites using eDNA. The final results should both provide interesting insight into the relationship between traditional and novel methods of amphibian detection and useful data concerning the species present in Maywoods Environmental and Educational Laboratory.

14. Reusable Polymeric Sorbents and their Applications in Water Remediation

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Decades of use of per- and polyfluoroalkyl substances (PFAS) in a multitude of consumer and industry-based products have led to a devastating amount of soil and water contamination. The chemical and thermal stability of PFAS have proved them to be an especially daunting challenge from an environmental remediation standpoint. Presently, the only full-scale water treatment separates via sorption and uses non-selective materials such as activated carbon (AC) or mineral media which are extremely difficult and/or costly to regenerate. Developing effective and renewable remediation technologies that lead to clean and safe drinking water sources are therefore a vital part of current research efforts. Research focused on selective adsorption is becoming a more practical route for capture and removal from contaminated water systems.

This work seeks to develop and assess various polymeric and nanocomposite adsorbents that have affinity towards two of the most commonly detected PFAS, perfluorooctanoic acid (PFOA) and perfluorosulfonic acid (PFOS). Two routes of sorption were explored: (1) contaminant binding and removal via magnetic decantation of functionalized polymer composites; (2) contaminant binding through flocculation with functionalized thermo-responsive polymers. Polymers are synthesized via free radical polymerization reactions with (1) amine functionalized cationic monomers with crosslinker N,N'-methylenebisacrylamide (NMBA) (2) N-isopropylacrylamide (NIPAAm) and various cationic and/or fluorinated comonomers

In some instances, composite systems were created with the inclusion iron oxide nanoparticles during the synthesis process. Two types of binding systems and removal are examined depending on polymer structure (i.e. crosslinked or linear). Binding studies are conducted by subjecting 2.5 mg/mL of each sorbent to 500 ppb of aqueous PFAS for up to 24 h at room temperature for crosslinked systems and 1 h at 50 ºC for linear systems. Cationic crosslinked polymers showed high affinity for PFOA (>80%) and PFOS (>90%) across a range of aqueous pH (4 – 10). Linear polymers that included both cationic and fluorinated monomers showed improved flocculation and contaminant removal as compared to those systems with isolated functionality. Both routes of treatment show promising results for future application as water remediation materials.

15. Investigating Plant-Soil Processes and Nitrate Seasonality Using High Resolution Sensors and Stable Isotope Measurements

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Nitrate generated and leached from soils is a contributor to the total export of nitrogen in agricultural watersheds and can lead to water quality problems in downstream rivers, lakes and estuaries. Controls on the seasonal nitrate pattern and its delivery to water bodies has been hypothesized to be dominated by soil-plant processes in some systems, such as the Ohio River Basin. For example, a hypothesized seasonal model suggests that the dormant winter season (Dec-Mar) results in plants that are relatively inactive and allow most of the nitrate to be leached through the soil rather than assimilated via biochemical processing. A spring planting season (April-June) supplies crops with ammonium nitrate fertilizers that can runoff or follow groundwater pathways that lead to high nitrate loads. A growing-harvesting season in late summer and fall (July-Nov) results in plants utilizing nutrients for growth and retaining the nitrate resulting in low instream nitrate loads. Further, the warm temperature seasons with dry soils due to high evapotranspiration demand results in a low transport rate of water and nutrients. Despite recent hypotheses and advances in knowledge gained about the nitrogen cycle, the biogeochemical and hydrologic controls on nitrate delivery in temperate water bodies remains un-resolved and under-studied.

Our research goal is to use high-resolution nitrate sensors and discrete samples of nitrate (NO_{3-}) and its isotopic signature $\delta^{15}N_{NO3}$ and $\delta^{18}O_{NO3}$ to further test hypotheses and gain new knowledge of nitrogen seasonality in temperate river systems. To do so, we use a multi-year high resolution (15 minute) submersible ultraviolet nitrate analyzer measurements and a multi-year dataset of $\delta^{15}N_{NO3}$ and $\delta^{18}O_{NO3}$ collected from agricultural basins in the inner Bluegrass region of central Kentucky USA. We carry out time series analyses and load analyses to investigate biogeochemical and hydrologic controls on nitrate transfer.

Time series analyses of nitrate sensors shows 5 mg/L N peaks in January-February and 2 mg/L N lows in July-August. Analyses of $\delta^{18}O_{NO3}$ shows a similar trend to that of nitrate concentration with peaks of 4‰ in July-August and lows of 0‰ in January-February. Time series analyses shows $\delta^{15}N_{NO3}$ peaks around 9‰ in September-October and $\delta^{15}N_{NO3}$ low periods of 6‰ in March. The nitrogen isotopic signature peaks occur when nitrate delivery to the stream is low and soils are dry, allowing the enriched denitrified nitrate to be slowly leached from soil into the watershed. Statistical analysis of seasonal trends are ongoing.

Results allow further discussion of nitrate processes in soils of watersheds. The spatial variability of soil properties produces conditions where nitrate can be at different stages of its oxidation-reduction potential and therefore differs in both the amount of available nitrate as well as its isotopic signature. In an ammonium-limited system the nitrate generated via mineralization-nitrification shows little fractionation and instead the nitrate will have a similar isotope signature towards that of the organic nitrogen source. This process is observed in the March low periods of the multi-year isotope datasets. Denitrification shows a distinct fractionation of both $\delta^{15}N_{NO3}$ and $\delta^{18}O_{NO3}$ that tends to increase the isotope signature due to preferential removal of the lighter isotope. This process is observed in the isotope peaks in the fall period. In the present agricultural soil system, we expect these peaks and low nitrate isotope results reflect a lag time in the system. For example, nitrate sensor lows and highs occur in July-August and January-February, but the corresponding shifts are not seen in isotope data until a one or two months later. We attribute the reason to the fact that nitrate generated via mineralization-nitrification in loose, aerated soils can be quickly transported to a river system and displays an immediate response in water as compared to nitrate that is tied up into tight and compact soil microsites. Soils with a low porosity creates the environmental conditions that limit oxygen and favor anaerobic microbes such as those that carry out denitrification. These microsites allow for partial nitrate removal but also makes the physical process of nitrate leaching more difficult (i.e. slower) than those sites that favor aerobic mineralized nitrate. The extended transport time of this pool of denitrified nitrate results in a lagged effect of the nitrate response in stream.

Future work includes further data analysis and a process-based numerical model to elucidate the fate and transport of nitrate in streams. To do so, we plan to expand the numerical modelling previously performed and focused on nitrate source and transfer in the basin. The model will be expanded to include stable isotope subroutines for soil nitrate $\delta^{15}N_{NO3}$ and $\delta^{18}O_{NO3}$.

16. Development and Validation of qPCR Assays for use in eDNA Detection of *Ambystoma* Species

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In the past decade environmental DNA (eDNA) has become firmly established as an effective method for detecting the presence of organisms of research and conservation interest and promises to greatly increase the ease, efficacy, and scope of ecological studies. Salamanders of the family Ambystomatidae are large, fossorial species; adults are rarely encountered aboveground outside of their brief reproductive season. Larvae develop rapidly, in ephemeral pools or streams, often with multiple species coexisting in a single habitat. A number of Ambystomatid species are of conservation interest in various portions of their range. We developed species-specific qPCR assays for *Ambystoma barbouri*, *Ambystoma jeffersonianum*, and *Ambystoma opacum* and tested them in silico, in vitro, and in situ. Tissue tests confirm specificity of primers among these three species and *A. maculatum* as well as the frequently sympatric *Notophthalmus viridescens.* To validate assays in situ larvae surveys were conducted and water samples collected from a number of sites in central and Eastern Kentucky. Initial eDNA results indicate accurate determination of species assemblage via molecular methods. These assays provide an effective means of determining species present in particular habitats rapidly and definitively and therefore offer to increase the ease of range delineation and spawning habitat studies.

17. Is Chloride Driving Specific Conductance in Streams in the Upper Ohio River Valley?

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The salinization of freshwaters is increasingly recognized as a global problem, resulting in the degradation of freshwater ecosystem health and biodiversity. Salts and metals washed in from urbanized landscapes are a primary cause, but extraction industries, mining and agriculture, as well as failing sewage infrastructure, illicit discharges, eroding stream banks and other sources all contribute to the problem. Chloride (Cl) concentrations over 30 mg/L can negatively impact sensitive aquatic taxa, and specific conductivity (SPC) can cause impairment of aquatic biodiversity at concentrations well below 500 µS/cm. We sought to explore the relationship between Cl and SPC in streams in the Upper Ohio River Valley using a data set extending back to 2018. We used 21 sampling locations distributed throughout 4 tributaries to the Ohio River near Wheeling, WV, including 2 sampling sites on the Ohio River. We found that mean Cl at our sampling sites ranged from ~7 mg/L up to 269 mg/L, while mean SPC ranged from 302 µS/cm to 2,432 µS/cm. Correlation strength (R^2) of SPC and Cl ranged from >0.01 to 0.82 (mean = 0.32), with some of the lowest R^2 values occurring at sites with the lowest or highest SPC and Cl concentrations. Using each site's average Cl and the Cl to SPC regression equation, we estimated that on average Cl accounted for between 1 and 16% of the SPC at our sampling sites, suggesting that ions other than Cl constitute a majority of the SPC in these streams. However, across all sites, mean Cl and mean SPC were highly correlated ($R^2 = 0.79$), indicating that Cl concentration is predictive of the SPC in local streams. Overall our data indicates that 1) many tributaries are likely impaired (at least in part) to high concentrations of salts and metals, 2) tributaries to the Ohio River are often higher in Cl and SPC than the Ohio River itself, and 3) these tributaries are negatively impacting water quality in the Ohio River by increasing the concentrations of salts and metals in the Ohio River.

18. The Use of Electrical Resistivity Tomography for Delineating Ridgetop Wetland Hydrogeology in the Daniel Boone National Forest in Eastern Kentucky*

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Isolated ridgetop perched aquifer-wetland systems in the Daniel Boone National Forest are important environments that locally store groundwater at high elevation. During the winter and early spring, water is collected and stored above a thick, impermeable clay layer. In the spring and summer, these systems provide drought resilience for the upland forest ecosystems by supplying stored water to local forest vegetation, surface water pools that breed native amphibians, groundwater springs, and adjacent lowland ecosystems.

Previous work done by Eastern Kentucky University has shown general trends in the geologic stratigraphy and hydraulic conductivity of these groundwater-surface water systems. That said, the different variables that lay beneath the wetlands themselves including the shape of clay layers and underlying bedrock topography provide significant challenge for estimating the amount of groundwater that is stored, the rate it drains, and whether the water remains on the ridge or flows to lowlands. Electrical resistivity tomography (ERT) provides a low-impact instrument for imaging underlying geology, alongside the monitoring of groundwater levels and sediment coring at these sites. With ERT, we expect to be able to learn more about the hydrogeologic features/pathways that stored groundwater takes from these perched aquifer systems towards lower elevations. Our observations may have important implications for predicting how drought might affect the forests and stream environments of Eastern Kentucky and for creating accurate water budgets for these kinds of water systems.

In the summer of 2021, ERT was used at multiple ridgetop wetlands, and resulting transect models showed the shallow subsurface geometry (Figure 1). Cross-sections show that the geometry of impermeable clay layers are complex and vary widely in size. Clay layers often have a bowl-shape that captures subsurface water that forms the primary surface water feature when groundwater levels are high. Other common features found in the subsurface impermeable layer included subsurface "depressions" where the clay layer forms a bowl shape that is not always expressed in the surface topography and breaks in the clay layer. Subsurface depressions are of differing size and location in different wetlands. In some cases, this depression is upslope from the wetlands itself, which could mean that this feature acts like a water tower or a pocket of groundwater that forest vegetation may utilize. Finally, breaks in the clay layer serve as leakage points that may allow groundwater to slowly seep to lowlands, making these systems and important source of water to valley streams.

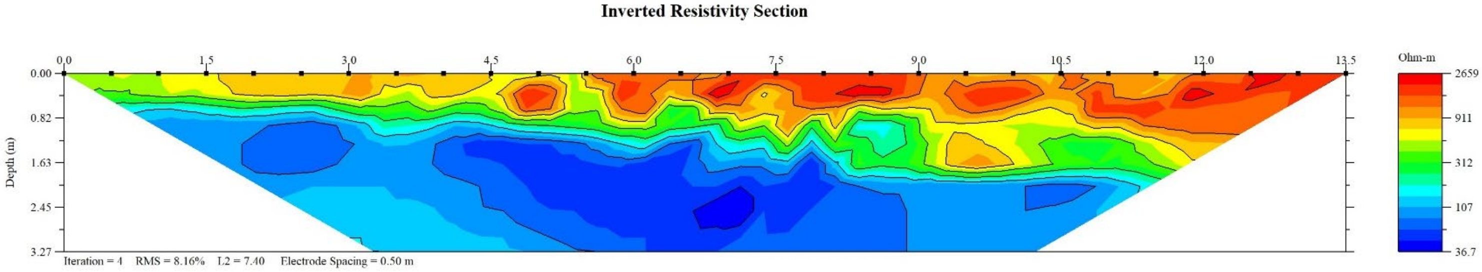

Figure 1. A transect model of a ridgetop wetland using Electrical Resistivity Tomography. The low resistivity layer (blue) represents the impermeable clay common to all of these systems. In this case the clay layer dips deeper below the land surface to create a variable aquifer geometry.

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