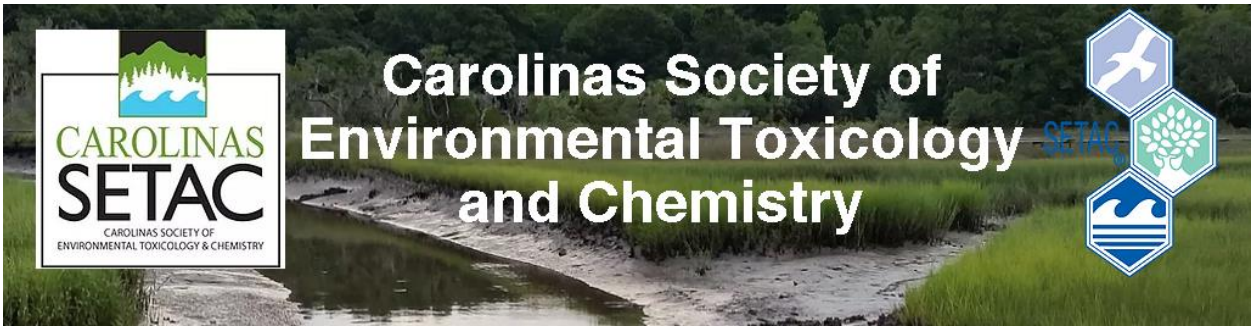

28th Annual Meeting

March 13th – 15th 2019

Fort Johnson Marine Science Complex

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CSETAC Meeting Program of Events

Wednesday March 13th

1:00 p.m. – 5:00 p.m. Harmful Algal Blooms and Associated Toxins Short Course

Location: *NOAA National Centers for Coastal Ocean Science - CCEHBR Auditorium*, 219 Fort Johnson Rd, Charleston, SC 29412

5:00 p.m. – 7:00 p.m. Opening Social Location: *Garage 75* 1175 Folly Rd., Charleston, SC 29412

Thursday March 14th

8:30 a.m. Welcome/Breakfast

Location: *SC Dept. of Natural Resources, Marine Resources Research Institute*, 217 Fort Johnson Rd, Charleston, SC 29412
Directions to MRRI: http://www.dnr.sc.gov/marine/mrri/directions.html

9:00 a.m. – 4:20 p.m. Platform Presentations (schedule next page)

12:00 p.m. – 1:20 p.m. Lunch (catered by Barbaritos on site)

6:00 p.m. – 8:00 p.m. Dinner (Oyster roast, Low Country Boil) Location: *Bowens Island Restaurant* (1870 Bowens Island Rd., Charleston, SC 29412)

Friday March 15th

8:30 a.m. Poster Set Up

Location: *SC Dept. of Natural Resources, Marine Resources Research Institute*, Outdoor Classroom - Fort Johnson Rd, Charleston, SC 29412

9:00 a.m. – 11:30 a.m. Poster Presentations (**l**ist on next page)

11:30 a.m. – 12:00 p.m. CSETAC Business Meeting

Platform Schedule

Time	Presenter(s)	Title
9:00	Catharine Parker	An Evaluation of Sediment Contamination in Headwater Tidal Creeks in the Southeast United States over a Twenty-Year Period in Relation to Coastal Development
9:20	Ed Wirth	
9:40	Cristina Sanders	Effects of Land Cover and Riparian Buffers on Cold-Water Fish Assemblages in Upper South Fork New River Headwaters
10:00	William McMahan	Lithologic Controls on Surface Water Chemistry and Benthic Macroinvertebrate Community Structure through Chemical Weathering
10:20	Break	
10:40	Michelle Pena-Ortiz	Longitudinal assessment of estrogenic activity along the New River
11:00	Bradley Baumgarner	Cellular fatty acid level regulates the effect of tolylfluanid on mitochondrial dysfunction and insulin sensitivity in C2C12 skeletal myotubes
11:20	Cindy Lee	Developmental origins of GABA genic and antioxidant transcriptome alterations in zebrafish exposed to 2, 2’, 3, 5’, 6 polychlorinated biphenyls (PCB-95) during embryogenesis
11:40	Ian Edhlund	Optimized Blood Volume Parameter for PBTK Modeling of PAH Exposure in Rainbow Trout
12:00	Lunch	
1:20	John Allran	Investigation of Crab Kill following Pyrethroid Application to NC Cotton Field: a Case Study in Inter-agency Cooperation in Pesticide Regulation & Protection of Agriculture & Aquatic Resources
1:40	Virginia Shervette	Mercury Bioaccumulation in Flathead Catfish
2:00	James Burke	Green Lakes from Space: Monitoring Algal Blooms in Charleston-area Stormwater Ponds
2:20	Sam Putnam	Detection and monitoring of harmful algal toxins in *Lyngbya wollei* in Lake Wateree South Carolina
2:40	Break	
3:00	Sarah Kell	Fate and Effects of Tire Wear Particles and Other Microplastics in Coastal Waterway
3:20	Allen Olmstead	

3:40		
4:00	Geoff Scott	Oceans and Human Health

Poster List

Presenter(s)	Title
Erik Andersson	Optimization of Sample Preparation Methods for NMR-based Metabolomics Analysis of Environmental Reef-Building Coral Samples
Danielle Beers	Comparing the impacts of ultraviolet light-enhanced toxicity of surface oil sheens on the survival, growth, and development of three different larval fish species, red drum (*Sciaenops ocellatus*), sheepshead minnows (*Cyprinodon variegatus*), and speckled seatrout (*Cynoscion nebulosus*)
Jordan Bralley	Behavioral effects of fluoxetine and sertraline and their photodegradants on southern toad (*Anaxyrus terrestris*) tadpoles
Wesley Brison	Sulfate Reducing Bacteria in Model Constructed Wetlands
Bryan Brooks	Influences of Nutrients and Salinity on Prymnesium parvum Elicited Sublethal Toxicity in Two Common Fish Models
Patrick Faught	Assessment of Biodegradation Potential of Diluted Bitumen in Seawater
Cassandra Horton	Bacterial Interactions with Coastal Pollutants and the Potential for Development of Antibiotic Resistance
Elena Legrand	Biomonitoring in the Great Lakes by untargeted metabolomics of dreissenid mussels
Julie Loewenstein	Characterization of the coral metabolome under different pH and temperature regimes, via 1H NMR
Edwina Mathis	Imposex induction in the Eastern mud snail: Investigation of organotins and other RXR endocrine disrupting chemicals in Charleston Harbor
Robert Podolsky	Effects of Personal Care Product Preservatives on the Larval Development and Growth of Sea Urchins (*Arbacia punctulata*)
Rose Rossell	Aromatic hydrocarbon emissions by marine cyanobacteria in the North Atlantic Ocean
Chelsea Woodruff	Assessment of Willingness to Pay for Coral-Safe Sunscreens

Abstracts

Platforms (in alphabetical order by presenter last name)

1) Investigation of Crab Kill following Pyrethroid Application to NC Cotton Field: a Case Study in Inter-agency Cooperation in Pesticide Regulation & Protection of Agriculture & Aquatic Resources. Allran J.W.

The Structural Pest Control and Pesticides Division of the NC Dept. of Agriculture and Consumer Services (NCDA & CS) is the state lead agency (SLA) for the EPA regarding pesticide regulation in NC. Pesticide Inspectors throughout the state conduct both routine and for cause inspections, in addition to investigations into complaints of potential violations of pesticide law. This presentation uses a recent investigation of a mortality event in which approximately 2000 blue crabs (*Callinectes sapidus*) died within 14 hours after a heavy rainfall followed an application of Fanfare 2EC (a.i. bifenthrin) to a cotton field adjacent to the canal in which the crab shedder ran his soft-shell crabbing operation. As part of the investigation, the pesticide inspector collected samples of vegetation and soil from the cotton field, in addition to sediment, water, and the blue crabs for analysis by the NCDA & CS laboratory. Sample analysis showed the presence of bifenthrin in all media, with the concentration decreasing with increasing distance from the application site. The label for Fanfare 2EC includes both advisory and mandatory statements about application near water, and Directions for Use includes statements regarding required buffer zones and the use of vegetative filter strips to mitigate risk to aquatic organisms. In addition to being the law, the label reflects the human health and environmental risk assessment and risk management processes, so that when the pesticide is applied according to the label, any risks should be negligible. However, when adverse effects occur, FIFRA provides a mechanism for dialogue between the SLA, EPA, and pesticide registrants to ensure label language and other risk mitigation measures are effective.

2) Cellular fatty acid level regulates the effect of tolylfluanid on mitochondrial dysfunction and insulin sensitivity in C2C12 skeletal myotubes. Davis AF, Thomas AA, Shorter KR, Brown SL, and Baumgarner BL

Previous research suggests that the endocrine disrupting chemical tolylfluanid (TF) may promote metabolic dysfunction and insulin resistance in humans. The potential impact of TF on skeletal muscle metabolism has yet to be fully investigated. The purpose of this study was to determine whether TF can promote insulin resistance and metabolic dysfunction in mammalian skeletal muscle cells. C2C12 murine skeletal myotubes were exposed to 1 ppm TF for 24 h. To examine the potential effect of cellular fatty acid levels on TF-dependent regulation of mitochondrial metabolism and insulin signaling, we treated skeletal myotubes with 0.25mM or 1.0mM oleic acid (OA) during TF exposure trials. Tolylfluanid (1e10 ppm) reduced lipid accumulation by approximately 20% in 0.25 and 1.0mM OA treated cells. The addition of 0.25mM OA completely inhibited the TF-dependent reduction in maximal mitochondrial oxygen consumption rate (OCR) while 1.0mM OA exacerbated the TF-dependent reduction in mitochondrial OCR. Exposing skeletal myotubes to 1 ppm TF promoted an 80% reduction in mitochondrial membrane potential, which was completely inhibited by 0.25mM OA and partially inhibited by1.0mM OA. The addition of 0.25mM OA promoted a TF-dependent increase in insulin-dependent P-Akt (Ser473). In contrast, the addition of 1.0mM OA promoted a significant reduction in insulin-dependent P-Akt (Ser473). Further, the addition of 1 ppm TF significantly

reduced insulin-dependent mTORC1 activity regardless of OA concentration. Finally, TF significantly reduced insulin-dependent protein synthesis in the 1mM OA treated cells only. Our results demonstrate that the effect of 1 ppm TF on mitochondrial function and insulin-dependent protein synthesis in skeletal myotubes was largely dependent upon cellular fatty acid levels.

3) Green Lakes from Space: Monitoring Algal Blooms in Charleston-area Stormwater Ponds. Burke J, Alpert S, Beckingham B, Ali K.A.

Stormwater ponds and lakes are engineered landscape features primarily built to mitigate flooding concerns due to increased surface runoff, but may also serve as an aesthetic anchor of a community. In coastal South Carolina, they primarily exist as wet detention ponds. There is concern that these ponds act as gateways for the transport of contaminants from developed landscapes to estuarine waters. Ecological and community-level health risks may also be present since ponds and lakes receiving stormwater inputs may act as temporary sinks of harmful organic contaminants and metals, and be stressed by eutrophication (nutrient-enrichment) leading to a proliferation of algae, which may include harmful algal blooms (HABs). The objective of this research is to understand whether water quality parameters in dynamic, engineered stormwater structures can be adequately monitored using optical remote sensing. Optical measures may include color, clarity, and visual appearance—these signatures may be relatable to priority water quality measures (chlorophyll-a/ total suspended solids). Both satellite and near-ground remote sensing platforms (e.g. unmanned aerial vehicles/drones) may be applied for monitoring stormwater ponds. Usefulness of these platforms requires benchmarking data to in-field and in-lab measured water quality parameters. This work addresses a significant research opportunity, as stormwater ponds are landscape features that are smaller and shallower than water bodies traditionally monitored by remote sensing. These ponds are numerous and typically lie on private property, which makes traditional monitoring difficult. These considerations make enabling water quality monitoring by advanced approaches (i.e. by remote sensing) essential. Initial results show strong correlations for chlorophyll-a and total suspended solids when models are formulated on a site-specific basis; however, complications arise when attempting to develop generic models. Considering this, future work aims to consider multiple parameters to better identify harmful algal blooms despite seasonal and location variation.

4) Optimized Blood Volume Parameter for PBTK Modeling of PAH Exposure in Rainbow Trout. Edhlund I and Lee C

We optimized the blood volume (Vblood) parameter in a Petri Net Physiologically Based Toxicokinetic (PBTK) model of blood-flow limited, waterborne fluoranthene exposure in rainbow trout. The optimized Vblood parameter is significantly larger than that assumed in prior research. To help explain the large Vblood parameter, we developed a series of PBTK models for other polyaromatic hydrocarbons (PAHs), which show a trend of increasing optimized Vblood with increasing log Kow. The Vblood parameter could be used in future PBTK models as a surrogate for complex diffusion limitation models.

5) Fate and Effects of Tire Wear Particles and Other Microplastics in Coastal Waterways Kell, S.E., Beckingham, B., Weinstein, J.E.

Studies conducted within the Charleston Harbor, SC watershed have revealed that >90% of total microplastics in sediments at some locations are tire wear particles (TWP) but little is known about the way by which they enter coastal waters and their potential effects on estuarine biota.

The objectives of the present study are to determine the pathways by which microplastic debris, including TWP, enter Charleston Harbor and to assess the residence times of TWP in the gut and gills of grass shrimp, Palaemonetes pugio. Pathways were assessed by sampling stormwater detention ponds (n=4) and adjacent receiving tidal waterbodies (at the discharge point, upstream 50 m, downstream 50 & 100 m). Residence times were quantified by exposing adult grass shrimp to different size classes (38-63, 63-150, 150-250 & 250-355 µm) of TWP at a concentration of 50,000 particles/L for three hours, then transferring the shrimp to TWP-free water. Acute toxicity was monitored during and after exposure. Preliminary results suggest that stormwater runoff and ponds are an important pathway for microplastics to adjacent waterways and that stormwater ponds may serve as a sink for microplastics. Abundances >8800 particles/kg ww, with TWP comprising 87% of the total, have been found in pond bottom sediment. Grass shrimp readily ingested and ventilated all sizes of TWP; no mortality was observed. Significant size dependent effects were seen in the mean gut retention time and number of TWP ingested and ventilated ($p<0.001$). TWP were retained in the digestive tract for a significantly longer time relative to natural prey items (average 1.6 hrs) and a non-digestible particle, activated charcoal (average 1.7 hrs) ($p<0.001$ and $p=0.002$, respectively). The results of this study have implications for stormwater management decisions and chronic effects to aquatic organisms that ingest TWP such as reduced energy assimilation, growth and fitness.

6) Lithologic Controls on Surface Water Chemistry and Benthic Macroinvertebrate Community Structure through Chemical Weathering. McMahan WG, Tuberty SR, Armstrong, WH, Gangloff MM

Anecdotal evidence from anglers have suggested that macroinvertebrate communities are quite variable across geologic regions of the Southern Appalachian Mts and beyond. Bedrock geochemistry can vary greatly across space due to local geologic history. Rocks from different origins contain process-specific minerals, with distinct chemical compositions. As rocks chemically weather at the surface, the composition and concentration of soluble weathering products in surface water varies based on the chemical properties of the regolith. This is hypothesized to influence the benthic macroinvertebrate communities due to variations in pH, conductivity, alkalinity, and concentrations of dissolved metal ions. An understanding of macroinvertebrate assemblage responses to these minerals and their distribution is necessary for effective management practices to determine streams of higher concern. The fishing industry also relies on the knowledge of local macroinvertebrate communities, so this information could be used to inform local fishing guide services and private recreational anglers. An exportable and freely modifiable software package is being developed to locate study areas for this or other similar projects. An ArcPy script locates areas with 3 geochemically disparate bedrock types in close proximity and uniform undeveloped land cover. It then calculates headwater streams and exports them to a MatLab script which conducts statistical analysis to locate elevation windows containing all three rock types. Once study sites are located, patterns in macroinvertebrate presence and rock type will be analyzed to determine correlation. Bedrock and streambed sediment samples will be collected from these monolithic drainages for chemical analysis. Basic water chemistry parameters will be measured in the field using a YSI multimeter, and samples will be collected for laboratory analysis of trace elements by Inductively Coupled Plasma Optical Emission Spectrophotometry (ICP-OES). Benthic macroinvertebrates will be collected using NCDEQ Qual-4 method for identification to species.

7) Olmstead, A.

8) An Evaluation of Sediment Contamination in Headwater Tidal Creeks in the Southeast United States over a Twenty-Year Period in Relation to Coastal Development. Parker C, Sanger D, and Wirth E

Salt marshes and the tidal creeks that drain them are essential ecosystems along the southeastern coastline as they provide nursery, feeding, and protective habitat for many coastal organisms. Furthermore, headwater tidal creeks are the primary link between estuarine and upland habitats, serving as conduits for runoff from surrounding uplands. This makes them useful as sentinel habitats, which provide early warning of potential harm and ideal systems to evaluate the effects of increasing coastal suburban and urban development on estuarine environmental quality. Estuarine sediments are often found to have concentrations of metals, polycyclic aromatic hydrocarbons (PAHs), pesticides, polychlorinated biphenyls (PCBs), and polybrominated diphenyl ethers (PBDEs) that are associated with human activity. Out of 43 headwater tidal creeks sampled between 1994 and 2006, 18 were sampled again in 2014/2015 for sediment chemical contamination. Creek watersheds were classified as forested, forested to suburban, suburban, or urban land use based on their percent impervious cover (IC) levels during several time points. Forested to suburban creeks are those that changed classifications over the last 20 years. Analyses of historical and current data resulted in significant relationships between IC and a number of sediment contaminant metrics (e.g., PAHs). In addition, 11 of the creeks sampled in 2014/2015 have paired data from 1994/1995, allowing for the analysis of change over the 20-year span. Overall, results indicate increased chemical contamination is occurring with increasing levels of development. These results expand our knowledge of how these systems respond to such development and can provide managers with information about how predicted human population growth along the coast may alter tidal creek health.

9) Longitudinal assessment of estrogenic activity along the New River. Pena-Ortiz M, Hawkins MB, Kraak MH, & Tuberty SR

Understanding exposure of endocrine disruptive compounds (EDCs) in aquatic ecosystems is becoming increasingly important as reports of fish feminization rise. EDCs, such as 17α-ethynylestradiol, originate from point and non-point source pollution, and make their way into aquatic ecosystems. Once in aquatic ecosystems, these compounds mimic natural estrogens by binding to estrogen receptors of fish and other wildlife, leading to disruptions in reproduction and development. One important mechanism of EDC action is modulation of activation of estrogen receptors (ERs). The identification of specific EDCs that interact with ERs is a complex issue due to the vast diversity of endocrine disrupting chemicals found in aquatic ecosystems. Since there are roughly 70,000 contaminants that remain unevaluated for EDC activity, detecting these compounds in aquatic ecosystems is crucial to understanding exposure concentrations in rivers, as well as the additive impacts of all downstream effluents. Moreover, EDC screening methods must be taken into account when conducting such assessments. Currently, the most common assays used to detect estrogenic activity screen samples based on the α-estrogen receptor found in humans. This approach may result in an underreporting of estrogenic contamination, particularly for non-mammalian aquatic species. Teleost fish express three ER subtypes (ERα, ERßa, and ERßb), with ligand binding profiles distinct from each other. Therefore, the TriFishER assay was created as an environmentally relevant method to evaluate estrogenic activity in water samples. Although estrogenic activity assessments are common,

there is a lack of understanding on additive impacts of EDC effluents from point and non-point sources. Furthermore, myriad rivers have been evaluated for EDCs using outdated assays, possibly resulting in inaccurate depictions of estrogenic activity. Therefore, the aim of this research is to utilize the TriFishER assay to assess additive longitudinal impacts of point and non-point sources of EDCs in a local watershed.

10) Detection and monitoring of harmful algal toxins in Lyngbya wollei in Lake Wateree South Carolina, Putnam, S., Smith, M., Metz, T., Scott, G., Ferry, J.

Harmful algal blooms are an increasing human health risk that is poorly understood; blooms can produce a wide array of toxic materials that are often difficult to detect. The harmful algae Lyngbya wollei colonized Lake Wateree (South Carolina) several years ago and has been spreading throughout favorable areas of the lake. Lake Wateree, located in Ridgeway South Carolina, is a source of water for both recreation and drinking for the surrounding communities. It is a man-made impoundment at the confluence of the Catawba and Wateree Rivers that joins in with the Santee watershed. Lyngbya wollei is a filamentous benthic algae that is known to produce several compounds that can have dermatological or neurotoxic effects. Here we report molecular analysis of Lyngbya wollei obtained from multiple sites on the lake from June 2018 until February 2019. Samples were collected both over time and over a variety of locations to give an overall estimate of presence and potential toxin accumulation in algal biomass over the course of the season. Samples were lyophilized, extracted, and then analyzed by hydrophilic interaction liquid chromatography/mass spectrometry. Initial results show the presence of several toxins in the Lyngbya wollei toxin (LWT) family, specifically LWTs 1, 4, 5, and 6 (all saxitoxin derivatives). Extraction and instrumental methods were developed for the analysis of this toxin and initial quantification was performed. Correlation between nutrients and other anthropogenic stressors and the LWTs is presented. An estimate of the total potential toxin inventory of the lake is presented for the 8 month sampling period.

11) Developmental origins of GABA genic and antioxidant transcriptome alterations in zebrafish exposed to 2, 2', 3, 5', 6 polychlorinated biphenyls (PCB-95) during embryogenesis. Ranasingha P and Lee CM

The central nervous system (CNS) is most sensitive to contaminant exposure during early development due to rapid cell proliferation, migration, axonal growth, and synaptogenesis. One of the chiral, non-dioxins like polychlorinated biphenyls (PCBs), known as 2, 2', 3, 5', 6 polychlorinated biphenyls (PCB-95), has been identified for its neurotoxic effects in some detail. However, the etiology of these toxic mechanisms remains unexplained. We hypothesized that γ-aminobutyric acid (GABA), one of the key components of CNS development, is a plausible target. Also, we suggest that neurotoxic effects can be characterized by increasing damage to brain cells, leading to cell death with the PCB-95 exposure. To characterize the effects of PCB - 95 on GABAgenic and antioxidant system, zebrafish embryos with intact chorions were exposed to different concentrations of PCB-95 (0.25, 0.5, 0.75 and 1ppm) with two controls (E2 solution and the 0.1 % DMSO). Gross morphological alterations, tissue up-take, neurotransmitter levels and gene expressions were determined. Results suggested that the embryonic exposure to PCB-95 induced alterations in morphology, neurotransmitter levels and the gene expressions. These findings provide support to reveal the developmental neurotoxic mechanisms of PCB-95 exposure during embryogenesis.

12) Effects of Land Cover and Riparian Buffers on Cold-Water Fish Assemblages in Upper South Fork New River Headwaters. Sanders CL and Tuberty SR

Riparian vegetation is an essential component of a stream ecosystem. Riparian buffers reduce runoff contamination, improve bank stability, and produce shading that regulates the water temperature for many organisms that can only thrive within specific temperature thresholds. Climate change combined with expanding urbanization and changes to land use pose a serious threat to many cold-water species as temperature increases during the summer months. Western North Carolina has many sensitive cold-water fish species to include brook (Salvelinus fontinalis), brown (Salmo trutta), and rainbow trout (Oncorhynchus mykiss). Maintaining the biodiversity of the cold-water fish species of the Southern Appalachian Mountains is not only vital to the stability of the ecosystem but also to the state of North Carolina. Fishing provides substantial revenue and job opportunities from trip expenses, fishing equipment, licensing, and guides My study focuses on effects of temperature and conductivity to the cold-water fish assemblages in seven headwater streams that comprise the Upper South Fork New River watershed. These streams vary widely in ability to support sensitive and endemic fish species ranging from extirpation to successful localized reproduction. The goals of this project are: 1) to determine ecological conditions and environmental variables critical to healthy rural and urbanized streams, and 2) to identify best management practices, remediation techniques, and sustainable technologies that can aid in maintaining or returning healthy fish habitat, Assemblages in each headwater stream will be obtained by electrofishing and regressed vs percent impervious or forested cover, riparian zone width, bank erosion hazard index (BEHI), temperature, discharge, and specific conductivity. Fish size and weight will be recorded to determine size-class composition metrics. Water chemistry data is recorded with Eureka water quality sensors that record temperature, specific conductivity, dissolved oxygen, and depth every 15 minutes.

13) Scott, GI

14) Mercury Bioaccumulation in Flathead Catfish; Shervette, V, Reed, LA

Flathead catfish *Pylodictis olivaris* is native to Gulf of Mexico drainages and is a popular large-bodied, long-lived, gamefish that commonly preys on other fishes. It was widely introduced to southeastern Atlantic slope drainages and now occurs in aquatic systems from Florida to New Jersey. Several studies have documented that coincident with the introduction of flatheads to southeastern rivers, declines occurred in the abundance of native centrarchids, bullhead catfish species, and madtom catfish species. Flathead catfish can directly impact native species through predation and indirectly through competition for food and habitat. Additionally, the popularity of flatheads as a food fish has associated environmental justice concerns for subsistence fishers, especially within the acidic blackwater river systems of South Carolina where water pH is significantly negatively correlated with Hg concentrations in fish tissue. Fishes from the Edisto River, a blackwater system, consistently have higher length-adjusted Hg levels compared to other SC rivers. Flathead catfish contain the third highest Hg levels of fishes collected from state waters, fresh and marine. South Carolina anglers that target large flathead catfish for consumption will freeze their catch and then consume it with family and friends over many days, potentially exposing themselves and others to prolonged intake of high levels of Hg (> 1.0 ppm wet weight). Documenting the relationship between Hg in flathead catfish muscle tissue and age,

sex, and size will be the first step in assessing the potential harm to which flatheads may expose subsistence fishers. Here, we present preliminary findings from this work.

15) <u>Wirth, EF</u>

<u>Posters (in alphabetical order by presenter last name)</u>

1) Optimization of Sample Preparation Methods for NMR-based Metabolomics Analysis of Environmental Reef-Building Coral Samples. Andersson ER, Day RD, Loewenstein JM, Woodley CM, and Schock TB

Coral reef ecosystems are of great economic and ecological importance, yet their status has been declining in recent decades due to culminating global and local stressors. Therefore, research efforts aimed at assessing reef health and furthering our understanding of coral biology are currently of great importance. The use of molecular "omics" tools has increasingly contributed towards this cause, including the more recent application of metabolomics to study corals. However, coral metabolomics is still a developing field that lacks standardized methods for the preparation of samples prior to analysis, and studies to date utilize a range of sample preparation protocols. The application of metabolomics to study reef-building corals may prove essential in the face of increasing environmental threats, and an optimized method for preparing coral samples for metabolomics analysis would benefit the field. Here, we evaluate three important steps during sample processing of stony corals: (i) metabolite extraction, (ii) metabolism preservation, and (iii) subsampling. We use these results to select an optimized workflow for environmental coral samples. Our results indicate that a modified Bligh and Dyer extraction is more reproducible across multiple coral species compared to methyl tert-butyl ether and methanol extractions, while a methanol extraction is superior for feature detection. Additionally, few differences were detected between spectra from frozen or lyophilized coral samples. Finally, extraction of entire coral nubbins increased feature detection, but decreased throughput and was more susceptible to subsampling error compared to a novel tissue powder subsampling method. Overall, we recommend the use of a modified Bligh and Dyer extraction, lyophilized samples, and the analysis of brushed tissue powder for the preparation of environmental coral samples for 1H NMR metabolomics.

2) Comparing the impacts of ultraviolet light-enhanced toxicity of surface oil sheens on the survival, growth, and development of three different larval fish species, red drum (*Sciaenops ocellatus*), sheepshead minnows (*Cyprinodon variegatus*), and speckled seatrout (*Cynoscion nebulosus*). Beers, D., DeLorenzo, M., Key, P., Watson, A., Sancho, G., Chung, K., Pisarski, E.

Oil spills are a common occurrence in today's world due to commercial and consumer endeavors and the widespread use of petroleum. Although large oil spills gain the attention of national news, such as the 2011 Deepwater Horizon disaster and the 1989 Exxon Valdez spill, smaller scale spills and coastal marine contaminations occur more frequently. Oil can enter coastal aquatic waterways on lesser scales through pipeline leakage, recreational boating, road runoff, port activity and atmospheric deposition. Petroleum, which is a naturally occurring substance, contains toxic chemicals called polycyclic aromatic hydrocarbons or PAHs. In the presence of

UV light, PAHs can be photomodified into more toxic compounds, leading to increased toxicity in aquatic organisms. PAHs cause a suite of physiological consequences particularly in the early stages of development due to increased vulnerability and less adapted methods of dealing with pollutants. Therefore, the goal of this study is to compare the potential impacts of the photoenhanced potency of oil slicks on the growth and development of three different ecologically important larval coastal fish species. After an acute 24-hour oil exposure, fish will be moved to clean seawater for a 7 to 30 day grow-out phase in which multiple survival, growth, and developmental sublethal endpoints will be examined. Red drum have already demonstrated a greater sensitivity with exposure to UV light and oil than sheepshead minnows and it hypothesized that speckled seatrout will be more sensitive than either the red drum or sheepshead minnows. Findings will be used to fill in data gaps for NOAA's Office of Response and Restoration to create more useful mitigation and restoration plans in the event of future oil spills.

3) Behavioral effects of fluoxetine and sertraline and their photodegradants on southern toad (*Anaxyrus terrestris*) tadpoles. Bralley, Jordan 1, Cory, Wendy 2, Welch, Allison M. 1

Pharmaceutical pollution is an emerging environmental concern, with a variety of mediations appearing in surface waters around the world. In the environment, exposure to UV radiation can transform these compounds into related molecules, which can be more toxic than the original compound. Despite increased attention to the effects of pharmaceutical pollution on aquatic life, very little is known about the ecotoxicology of pharmaceutical transformation products. Antidepressants, including the widely-prescribed selective serotonin reuptake inhibitors fluoxetine (Prozac) and sertraline (Zoloft), have been regularly detected in the environment and have been shown to cause a variety of behavioral changes in organisms ranging from mollusks to fish to tadpoles. We investigated the effects of these compounds and their UV-phototransformation products on the behavior of amphibian larvae, which are particularly vulnerable to aquatic pollution due to their permeable skin. Southern toad tadpoles were exposed to solutions of fluoxetine and sertraline, with or without phototransformation, and behavioral assays were conducted to examine startle response, aggregation behavior, and refuge use. These behaviors are relevant to sertraline's mode of action, similar to behavioral changes observed in other organisms exposed to similar antidepressants, and potentially important to tadpoles' vulnerability to predation. The results of this research will help us better evaluate the level of risk posed by these antidepressants in the aquatic environment.

4) Sulfate Reducing Bacteria in Model Constructed Wetlands. Brison, Wesley

With biogeochemical cycles being put into disarray heavy metals in sediment and aquatic locations are being released allowing for increased pollution and health concerns for the environment and public sector. Previous studies have shown that sulfides, produced by sulfate-reducing bacteria during the oxidation of organic matter, complex with divalent metal ions, which are precipitated and sequestered in the sediment layer. The objective of this project is to determine the best plant genus that allows for increased sulfate reducing bacteria and not methyl mercury, and to see if there is a correlation between sulfate, sulfide, and organic carbon. The experiment was conducted on 3 different plant genera; Canna, Pontederia, Carex, and a non-vegetated control using model constructed wetlands, each plant genus, and the control has 3 replicates. DNA was extracted from the sediment using MioBio Powersoil kit and then the relative abundance of Desulfovibrio vulgaris was determined by qPCR using primers for the

dsrA (dissimilartory sulfite reductase) gene as well as hgcA gene for Methylmercury producing bacteria.

5) Effects of Personal Care Product Preservatives on the Larval Development and Growth of Sea Urchins (*Arbacia punctulata*). Caruso JP, Podolsky RD

Personal care products (PCPs) comprise a wide variety of daily-use products that contain preservatives to prevent the growth of bacteria and mold. Parabens, the most commonly used preservatives, have been recently implicated in human health issues, causing PCP manufacturers to shift toward alternative preservatives in production and marketing. These compounds enter the ocean but their effects on marine organisms are not well known. This study aimed to test the lethal and sublethal effects of methylparaben (MP), the most widely used paraben, as well as two "safer" alternatives—2-phenoxyethanol (2-PE) and chlorphenesin (CPN)—on early development of the sea urchin Arbacia punctulata. Zygotes were added to stirred glass jars with five concentrations of 2-PE, CPN, and MP and allowed to develop in a 48-hour assay. Embryos were staged to record lethal effects, and larvae that had developed to the pluteus stage were measured using skeletal landmarks to estimate sublethal effects on growth. MP, CPN, and 2-PE showed lethal effects at log concentrations of 2, 2.5, and 3 ppm, respectively. Sublethal effects on skeletal growth were evident at log concentrations as low as 1.5 ppm and followed roughly the same order among compounds. Skeletal asymmetry also increased over the same concentrations. These effects on mortality and body size and shape could reduce success in the plankton for sea urchins and other marine invertebrates. Although environmental concentrations are generally lower than these harmful levels, more research is needed to understand effects of chronic exposure and the combined effects of multiple compounds or environmental stresses like UV and temperature.

6) Assessment of Biodegradation Potential of Diluted Bitumen in Seawater (Faught PW, Davis D, Swarthout RF)

Bitumen is a heavy crude oil produced from the Alberta tar sands in Canada, which must be diluted with lighter petroleum products before transport through pipelines or in rail cars to create diluted bitumen (dilbit). As Canada has intensified oil production in this region, additional pipelines have been built to deliver dilbit to coastal ports in Canada and refineries in the United States. Numerous inland dilbit spills have occurred and future marine spills are inevitable. Compared to other sources of crude, spills of dilbit can be more difficult to clean up, as evidenced by the five-year, $1 billion remediation of 900,000 gallons of dilbit spilled into the Kalamazoo river in 2010. Therefore, data regarding the environmental fate of dilbit are critical in preparing to respond to future spills. To assess the biodegradability of dilbit in seawater, aerobic microcosms containing 10 mL of a water accommodated fraction of dilbit, and 30 mL Atlantic Ocean seawater were prepared. Two separate conditions were tested: autoclaved water as a negative control, and native seawater. A polymerized chain reaction (PCR) was performed to confirm the presence of hydrocarbon-degrading bacteria. At specified time points replicate microcosms were extracted with methyl tert-butyl ether, and compound-specific biodegradation of hydrocarbons including n-alkanes and polycyclic aromatic hydrocarbons was determined by GC-MS and GC-FID. These results will be discussed relative to previous experiments using dilbit coated sand as a method for introducing the oil to the seawater. Quantifying the capacity for marine bacteria to degrade this emerging petroleum product will assist responsible agencies in designing appropriate response protocols for dilbit spills.

7) Influences of Nutrients and Salinity on Prymnesium parvum Elicited Sublethal Toxicity in Two Common Fish Models. Hill BN, Saari GN, Steele WB, Corrales J, Brooks BW.

The magnitude, frequency, and duration of harmful algal blooms (HABs) are increasing worldwide primarily due to climate change and anthropogenic activities. Prymnesium parvum is a euryhaline and eurythermal HAB forming species that has expanded throughout North America resulting in massive fish kills. Previous ecotoxicological work supported an understanding of conditions resulting in HABs and fish kills; however, the primary endpoint selected for these studies is primarily acute fish mortality. Additional adverse effects, including cellular and physiological alterations, are poorly understood during a P. parvum HAB. To begin to understanding sublethal responses, P. parvum cultures grown under varying nutrient and salinity conditions were used to examine antioxidant gene expression (gclc, gst, nrf2, sod), traditional biomarkers of oxidative stress (lipid peroxidation, DNA damage, total glutathione) and changes in photomotor behavioral response patterns in two common fish species, the fathead minnow (Pimephales promelas) and zebrafish (Danio rerio). Varying nutrient and salinity conditions influenced P. parvum related oxidative stress and fish behavioral responses and these effects were heightened by conditions nonoptimal for P. parvum growth. Such sublethal observations present important considerations for future assessment and management of P. parvum.

8) Bacterial Interactions with Coastal Pollutants and the Potential for Development of Antibiotic Resistance. Horton CL, Scott GI, DeLorenzo ME

The Southeastern coastal plain is the most rapidly urbanizing region in the U.S. Landscape changes which increase imperviousness lead to alterations in the hydrological cycle, increasing runoff of nonpoint source (NPS) pollution including nutrients, microbes and chemical contaminants such as trace metals. NPS runoff is discharged into coastal stormwater retention ponds to reduce loading into coastal ecosystems. In the SC coastal zone there are >21,594 retention ponds, which concentrate chemical contaminants, nutrients, and microbes. Previous studies in retention ponds indicate elevated levels of trace metals, in addition to the presence of clinically-relevant antimicrobial products and high levels of fecal coliform bacteria. Other investigations have shown significant association between trace metals contamination in surface waters and sediments with increased rates of antibiotic resistance (ABR). As a result, these ponds may pose a public health risk from drug-resistant microbes. Even in the absence of human pathogens, affected microbes may pass on resistance genes to harmful bacteria in nearby estuaries, where they may be transferred during flood conditions. With the increase in extreme rainfall events that accompanies climate change, such conditions will increase in frequency and severity. Interactions of trace metals commonly found in South Carolina ponds and estuaries (As, Cu, Zn), clinically-relevant pharmaceuticals (triclosan, ciprofloxacin, and oxytetracycline), and aquatic pathogenic bacteria (*Vibrio vulnificus* and *Enterococcus faecium*) will be discussed in terms of how they may enhance antibiotic resistance and underlying mechanisms for upregulation of resistance genes. Additionally, preliminary single-exposure data for *V. vulnificus* and E. faecium grown in the presence of antimicrobials or trace metals will be presented.

9) Biomonitoring in the Great Lakes by untargeted metabolomics of dreissenid mussels. Legrand E, Bearden DW, Casu F, Hagiwara KA, Jacob A, Johnson EW, Schock TB

Environmental metabolomics focuses on detecting system-wide biochemical changes in organisms in response to environmental exposure. As ubiquitous, sessile, filter-feeders that can bioaccumulate chemical contaminants, bivalves have been used as bioindicators in

ecotoxicology. In collaboration with the NOAA Mussel Watch Program, we evaluated variations in the metabolomes of the dreissenid mussel (Dreissena spp.) in the context of the Great Lakes restoration efforts using untargeted metabolomics with nuclear magnetic resonance (NMR)-based spectroscopy. Dreissenid mussels were caged and deployed along a suspected pollution gradient in the Maumee River (Lake Erie) and sampled four weeks later. Whole-body polar extracts were characterized using NMR spectroscopy. Multivariate statistical analysis differentiated up- and downstream sites. In depth analyses are ongoing to identify statistically significant features and pathways. The changes in the metabolome will be discussed in the context of the environmental exposure. Additionally, we will discuss the challenges of validating an organism as an environmental indicator of pollutant exposure using a sensitive metabolomics approach.

10) Characterization of the coral metabolome under different pH and temperature regimes, via 1H NMR. Loewenstein JM, Schock TB, Day RD, Woodley CM, Anderson PE

It is well established that anthropogenic activities are changing the physical and chemical properties of our oceans by increasing sea surface temperature and lowering oceanic pH. This is problematic for reef corals, which are extremely sensitive to environmental changes and exhibit decreased calcification and reproduction, and compromised competitive and immune abilities in response to stress. To this end, metabolomic measurements using 1H NMR provide an opportunity to determine the chemical response to a number of stress factors relevant to global climate change. The present study has several objectives: (1) To assess the change in the metabolome of Porites lobata and Pocillopora eydouxi colonies over a natural pH gradient and (2) To compare the coral metabolome of colonies exposed to different treatments of pH and temperature as well as the combined effect of these two treatments on inter- and intra- species variation of two major reef building species, Montipora capitata and Porites compressa. Coral tissue powder will be extracted for polar metabolites using chloroform, methanol, and water and analyzed on a 700 MHz 1H NMR. While metabolomics has been used widely in the pharmaceutical and human health industries, it has had little application addressing issues of coral health. Previous studies have demonstrated 1H NMR as a powerful tool for profiling the coral metabolome however this will be the first study to examine the coral metabolome as it is affected by climate change related issues, using NMR. This work will lay the foundation for future research exploring different species responses and answering fundamental questions about how our reef systems will change as global climate change continues to alter their physical and chemical environment.

11) Imposex induction in the Eastern mud snail: Investigation of organotins and other RXR endocrine disrupting chemicals in Charleston Harbor. Mathis, ES and Spyropoulos, DD

Endocrine disrupting chemicals especially those that act as ligands that bind and signal through the retinoid X receptor are a major concern throughout the food web because of their varied effects on development, reproduction, neurobiology and immunology. In gastropods, such EDCs can induce imposex. Imposex occurs when females express male characteristics such as a penis, vas deferens, or convolution of the oviduct. The eastern mud snail, Tritia obsoleta, is a dominant intertidal, benthic marine snail along the eastern United States coastline and prevalent within Charleston Harbor. Organotins, like tributyltin (TBT), are well-studied EDCs that have been shown to act as ligands to RXR and induce imposex in gastropods. Organotins are also legacy

compounds that persist in sediments. The dredging of Charleston Harbor creates the possibility of such legacy compounds being resuspended into the water column. Mud snails can be considered a sentinel species and imposex a potential biomarker for EDC exposure to multiple organisms. We first sought to determine RXR isoform sequences specific to T. obsoleta and use those sequences to monitor RXR isoform expression by qPCR and study how RXR isoform expression related to males, females and imposex adults in known contaminated and reference sites within the Charleston Harbor. By measuring isoform induction and field sampling, this study will establish pre-dredging levels of imposex and legacy chemicals within the harbor to serve as a baseline for later comparisons. We also seek to establish a toxicological assay that relates timing and dosage of TBT exposure to similar patterns of RXR isoform expression in T. obsoleta. Such an assay could be used to monitor the molecular and physical expression of imposex and assess TBT (and others) contribution to the process. Thus, defining a time-course of RXR isoform induction related to imposex will allow for more streamlined contaminant testing in the future.

12) Aromatic hydrocarbon emissions by marine cyanobacteria in the North Atlantic Ocean. Rossell R, Swarthout R, Trott I, Hunt S, Wyland W

Atmospheric emissions of aromatic hydrocarbons (AH), including benzene, toluene, ethylbenzene, and xylenes, can negatively impact human health through their effects on the oxidative capacity of the atmosphere and secondary organic aerosol (SOA) formation. While the number of reported measurements of AH mixing ratios and emissions in urban environments is substantial and has demonstrated the importance of anthropogenic sources, there have been relatively few measurements conducted in remote locations where biogenic sources may dominate. Recent, albeit sparse, evidence has suggested that marine phytoplankton have the ability to produce measurable quantities of AH, particularly toluene. Here we discuss the results of atmospheric hydrocarbon measurements over the remote North Atlantic Ocean during the May 2017 Phosphorus, Hydrocarbons, and Transcriptomics cruise aboard the R/V Neil Armstrong. 160 whole air canister samples were collected along a transect through the North Atlantic from Woods Hole, MA to Bermuda and back with 24 hour stops at nine stations encompassing different cyanobacterial populations. At each station, a diurnal time series of samples was collected. Canister samples were analyzed on a five-detector gas chromatography system for over 80 individual hydrocarbons including various biogenic and aromatic compounds. Initial correlations showed elevated concentrations of AH in warm, oligotrophic waters, as well as during a bloom of cyanobacteria. The compound trends will be discussed in relation to source signatures, cyanobacterial cell counts, and air mass back trajectories. These data will expand information on the spatial and temporal variability of trace gases associated with these globally important photosynthetic cyanobacteria and increase our understanding the role of marine biogenic AH emissions of the oxidative capacity of the remote atmosphere and potential marine SOA formation.

13) Assessment of Willingness to Pay for Coral-Safe Sunscreens. Woodruff, Chelsea; Burnett, Wesley; Blose, Julia; Hansen, David; Shuler, Andrew.

Thousands of tons of synthetic sunscreens enter our oceans each year, carrying with them coral-toxic chemicals. Coral reefs function as hubs of marine biodiversity and shoreline protection. Mineral-based coral-safe sunscreens are relatively new to the market. Rising awareness of reef-safe products over their synthetic counterparts is expected to have a positive impact on demand.

Need exists to examine potential consumers and their willingness to pay (WTP) for products such as coral-safe sunscreen. Three aspects of consumer attitudes are of interest: the nature and extent of environmental concerns, extent to which these concerns might result in consumer behavior, and the extent of WTP. This project will use an online survey to reach members of the Charleston community and beyond by asking referenda, close-ended, and Likert-scale questions about these products and about individuals' attitudes towards the environment. The method of analysis will be through the framework of contingent valuation method (CVM) to directly determine a consumer's WTP. The outcome of this project will provide a data set of consumers' attitudes and perspectives towards coral-safe products. This will also evaluate the efficacy of consumer knowledge about environmental issues, such as this one.