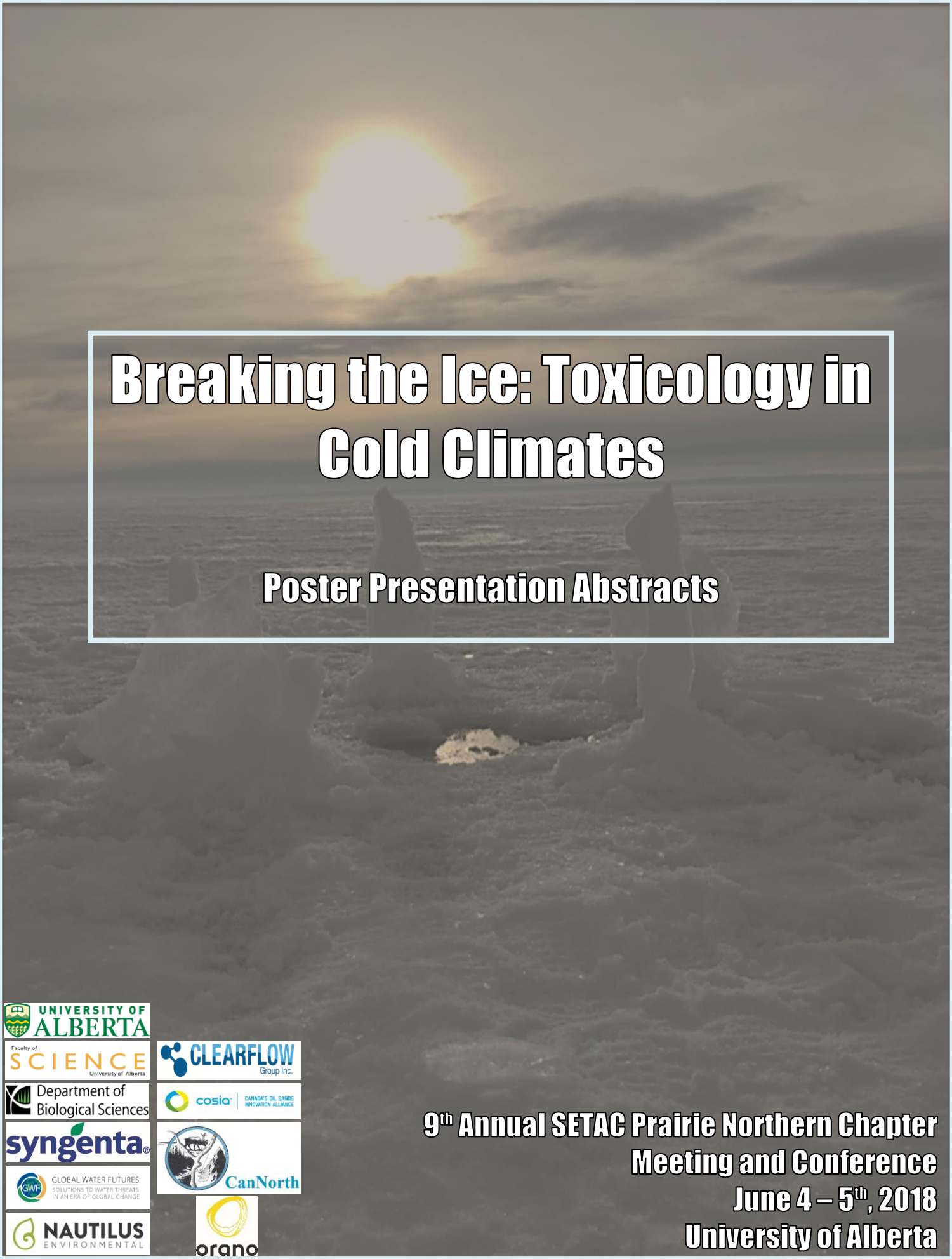

Breaking the Ice: Toxicology in Cold Climates

Poster Presentation Abstracts

9th Annual SETAC Prairie Northern Chapter Meeting and Conference
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9th Annual Chapter Meeting of SETAC Prairie-Northern

"Breaking the Ice: Toxicology in Cold Climates"

Poster Presentation Abstracts – In Alphabetical Order

1. Assessing the Effects of Diluted Bitumen to Invertebrate Community Structure and Abundance in Mesocosms

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There is increasing public concern around the impact of accidental oil spills on sensitive freshwater habitats from expanding pipeline infrastructure in North America. Diluted bitumen (dilbit) is of specific concern as it differs greatly in its physical and chemical properties relative to conventional crude oils. Despite the risk of pipeline spills, little is known regarding the effects of dilbit on freshwater organisms. In our pilot study, we assessed the effects of a dilbit spill on zooplankton taxa and abundance in three microcosms at low (1:100,000 v/v dilbit:water) and high concentrations (1:1,000 v/v dilbit:water) of dilbit and contrasted with effects observed in an untreated control microcosm. A total of nine species of zooplankton were observed across all microcosms at 11 days post dilbit addition. Community density decreased with increasing dilbit concentration; densities relative to the control tank (14.5 organisms per L) were 64.8% and 21.8% in the low and high tanks, respectively. Holopedium glacialis was the dominant zooplankton species in the control tank and was observed to disappear almost entirely from the high treatment. The results of this study will inform in a large-scale in-lake mesocosm experiment at the IISD-Experimental Lakes Area in summer-fall 2018, with invertebrate community data contrasted with effects on phytoplankton, benthic invertebrates, and fish.

2. Genetic Deletion of Soluble Epoxide Hydrolase Protects Cardiac Mitochondria from LPS-Induced Toxicity

Victor Samokhvalov[1], Kristi L Jamieson[1], Ahmed M. Darwesh[1], Tomoko Endo[1,2], Matthew Edin[3], Darryl C. Zeldin[3] and John M. Seubert[1,4*]
University of Alberta

Lipopolysaccharide (LPS) is a bacterial wall endotoxin producing many pathophysiological conditions including myocardial inflammation leading to cardiotoxicity. Arachidonic acid is a polyunsaturated fatty acid that can be

metabolized to cardioprotective epoxyeicosatrienoic acids (EETs) by cytochrome P450 epoxygenases. These metabolites are subsequently hydrolyzed to less bioactive dihydroxyeicosatrienoic acids (DHET) by soluble epoxide hydrolase (sEH). EETs are known to trigger a wide range of pathways protecting cellular structures, reducing cell death and promoting anti-inflammatory reactions in various cell types. We have recently demonstrated that EETs protect rat neonatal cardiomyocytes against LPS-induced cytotoxicity. Increased exposure to LPS as an environmental toxin can have a negative impact on the heart resulting in cardiovascular complications. The goal of this study is to investigate whether inhibition of sEH, genetically or pharmacologically, will influence mitochondrial function following LPS exposure.

3. A Simple Behavioural Test of Memory in Fish

Dean, Rachel. L [1], May, Zacnicte [1], Morrill, Adam, [1] Kline, David. I [2], Tresguerres, Martin [2], Hamilton, Trevor. J [1, 3]
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The Novel Object Recognition Test, a simple one-trial test of memory, can be used to determine whether fish are capable of remembering a previously viewed object. Fish are first presented with two identical objects and then following a specific length of time, are presented with one previously viewed (familiar) object as well as one newly added (novel) object. Greater exploration, indicated by a disproportionate time spent near either object or in each corresponding zone, signifies a preference for that particular object, and a recognition for the familiar object. Expanding on our previous research demonstrating object recognition memory in zebrafish, we used this test to determine that memory is retained for a maximum of five minutes and is enhanced by 50 mg/L of nicotine for a period of up to 15 minutes in zebrafish. Additionally, administration of 10 mg/L of a dopamine D_1-receptor agonist (SKF38393) induced a previously absent capability in Caribbean Damselfish to remember the familiar object following a 10-minute retention interval. This should be considered for researchers examining toxicological changes in cognitive ability of fishes.

4. Differential Toxicity Associated with Temporally Different Flowback and Produced Water Samples from a Horizontally Hydraulic Fractured Well in Multiple Freshwater Toxicological Model Species.

Folkerts, Erik J., Blewett, Tamzin, A., Delompré, Perrine, and Goss, Greg G.
University of Alberta

In North America, horizontal hydraulic fracturing (HHF) is an emerging industrial practice used to extract oil and natural gas reserves. The relatively little toxicological research on potential HFF environmental impacts has necessitated toxicological characterization of the wastewater by-product associated to HFF processes: flowback and produced water (FPW).

We characterized the toxicity of FPW samples collected from three different times after opening of a horizontally fractured well. We determined, through lethal concentration analyses, a modal toxic response, where earlier collected samples during well production elicited greatest toxicity, while samples collected mid-production demonstrated a decrease in toxicity. Then as sampling time was extended, toxicity was shown to increase again. Comparatively, *Daphnia magna* and zebrafish embryos displayed highest collective toxic responses amongst species tested, where as rainbow trout (both embryonic and juvenile forms) were more tolerant.
Our results suggest that when FPW releases occur, stage of FPW is a significant characteristic to consider when performing risk assessments and developing remediation efforts.

5. The Effects of Developmental Exposure to Raw and Ozonated Oil Sand Process-Affected Water (OSPW) Exposure on Anti-predator Behavioural Responses in Adult Zebrafish (*Danio rerio)*

Brittaney Frank[1], Danielle Philibert[1], Keith Tierney[1]
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Oil sand process-affected water (OSPW) is a chemically complex mixture produced as a by-product of oil sands extractions in mining operations in Northern Alberta. OSPW is a mixture of organic and inorganic constituents that has been associated with a wide range of sublethal and lethal effects in aquatic species. A potential remediation strategy for OSPW is ozonation. Ozone treatment degrades the organic component and is a potential strategy to decrease the toxicity associated with OSPW exposure. Though many studies have been conducted on the effects of OSPW exposure on early life stages of fishes, to date very few studies have been done to determine the potential persisting effects of OSPW exposure on later life stages. In our study, zebrafish were exposed to raw and ozonated OSPW from 0-7 days post fertilization. At approximately 2 years of age fish were subjected to a behavior test looking at the response of an individual to a computer simulated visual predator, the Red Tiger Oscar. Exposure to a visual predator affected the time the fish spent in the central and distal zones of the tank across treatment groups, but distance travelled, maximum velocity, and time spent moving were unaffected. Complex multisensory behaviors in fishes, like predator evasion, can provide insight into potential impacts of early life exposure of fishes to complex mixtures like OSPW.

6. Toxicity of Aqueous L-Selenomethionine Exposure to Early Life-Stages of *Pimephales promelas*

Gerhart, Allyson[1]; Lane, Taylor[1]; Hecker, Markus[2,3]; and Janz, David[3,4]

University of Saskatchewan

Previous studies investigating embryotoxic effects of selenium (Se) have consisted of maternal dietary exposures, yolk microinjections, and more recently aqueous embryo exposures. However, research investigating the effects of aqueous embryo exposures to Se on fish species native to North America has received little attention. Using fathead minnow (*Pimephales promelas*) as a model test organism, we sought to evaluate the effects of elevated selenomethionine (SeMet), in solution, on developing embryos exposed during early life stages. Newly fertilized embryos were exposed for 6 days to graded concentrations of Se (30 to 65,610 µg Se/L) as SeMet. Survival, hatchability, days to hatch, as well as the frequency and severity of deformities (total and type) were quantified. Whole-body Se concentrations were determined in swim-up fry following the 6 day exposures. Selenomethionine exposure significantly decreased survival (LC_{50} = 17,763 µg/L), increased the incidence and severity of deformities, and led to bioaccumulation of Se in fry, although clear dose-dependent effects on deformities and bioaccumulation were not apparent. Overall, the present study suggests that fathead minnow embryos are less sensitive to aqueous SeMet exposure than zebrafish or medaka.

7. Microplastic Concentrations in Calgary Stormwater Runoff

Groeneveld, Tianna[1]; Loutan, Alyssa[1]; Molenaar, Danielle[1]; Bujaczek, Taylor[1]; Kolter, Sheldon[1]; Moon, Sarah[1]; Arnold, Victoria[2]; Ruecker, Norma J[2]; Ross, Matthew S[1].
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Microplastics have recently come into the spotlight as a major pollutant in the hydrosphere. They have been found in seawater and are believed to predominantly originate from terrestrial sources. Urban rivers have been identified as being contaminated with microplastics which are transported to the marine environment. This study investigates microplastic concentrations in stormwater runoff from the City of Calgary. Stormwater runoff occurs when water flows through urban environments and is unable to soak into the soil, therefore transporting airborne and ground level contaminants to freshwater bodies. Samples were collected from 15 sites throughout Calgary between May-September 2017 under baseflow and rain event conditions. The samples were spiked with an internal standard, filtered, dried, and underwent a peroxide digestion to remove organic materials. Samples were sieved into 5 size fractions and vacuum filtered onto gridded filter paper to collect the microplastics within each fraction. After extraction, each sample underwent visual microscopy to determine quantity and type of microplastics present. Microplastics were found in all samples, and preliminary data suggests differences in microplastic concentrations between rainfall and baseflow events. This work represents one of the first studies on the occurrence of microplastics in stormwater runoff and will provide a baseline for future monitoring and mitigation studies.

8. Developmental Toxicity of the Hydraulic Fracturing Flowback and Produced Waters to Early Life Stages of Zebrafish (Danio rerio)

He, Yuhe[1]; Sun, Chenxin[2]; Zhang, Yifeng[2]; Folkerts, Erik J.[1]; Martin, Jonathan W.[2]; Goss, Greg G[1]

University of Alberta

Hydraulic fracturing (HF) has emerged as a major recovery method of unconventional oil and gas reservoirs and concerns have been raised regarding the environmental impact of releases of Flowback and Produced Water (FPW) to aquatic ecosystems. To investigate potential effects of HF-FPW on fish embryo development, HF-FPW samples were collected from two different wells and the organic fractions were isolated from both aqueous and particle phases to eliminate the confounding effects of high salinity. Each organic extract was characterized by non-target analysis with HPLC-Orbitrap-MS, with targeted analysis for polycyclic aromatic hydrocarbons provided as markers of petroleum-affected water. The organic profiles differed between samples, including PAHs and alkyl PAHs, and major substances identified by non-target analysis included polyethylene glycols, alkyl ethoxylates, octylphenol ethoxylates, and other high molecular weight (C_{49-79}) ethylene oxide polymeric material. Zebrafish embryos were exposed to various concentrations of FPW organic extracts to investigate acute (7-day) and developmental toxicity in early life stages. The acute toxicity (LD_{50}) of the extracted FPW fractions ranged from 2.8× to 26× the original organic content. Each extracted FPW fraction significantly increased spinal malformation, pericardial edema, and delayed hatch in exposed embryos and altered the expression of a suite of target genes related to biotransformation, oxidative stress, and endocrine-mediation in developing zebrafish embryos. These results provide novel information on the variation of organic profiles and developmental toxicity among different sources and fractions of HF-FPWs.

9. Physiological effects of mitigation of chemical flocculation toxicity with a proprietary mitigation agent.

Alexander M. Clifford, Edyta Jasinska, G. Hanna, Greg G. Goss
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Industrial operations such as mining, road building and aggregate washing result in high concentrations of suspended particles (Total Suspended Solids; TSS) in effluent waters. Tailing/settling ponds promote sedimentation of TSS and prevent adverse side effects in aquatic organisms prior to discharge; however, use of chemical coagulants/flocculants is known to improve the efficacy of TSS sedimentation. These chemical flocculants themselves have strong adverse biological effects including gill

damage within 4 days at very low concentrations (0.3-0.5 mg/L). Current practices are to secondarily treat flocculated water with a proprietary mitigation agent (MA), which appears to reduce toxicity of discharged water; however, this has yet to be empirically demonstrated. Given that exposure to the flocculent causes gill damage, we hypothesized that the mode of toxicity was *via* hypoxemia. We exposed fingerling trout *(Oncorhynchus mykiss*) to either flocculant (0.5 mg/L) MA (0.75 mg/L) or a mixture of the two and measured O_2 consumption over a 48h period. Furthermore, we excised gill for light microcopy (LM) and plasma, brain and liver tissue for analysis of tissue fuel (glucose/glycogen, ATP, phosphocreatine) and metabolites (lactate) to confirm the mode of flocculent toxicity and verify that mitigation causes no adverse sub-lethal effects. LM confirmed flocculent gill adhesion and damage in the flocculent exposure grout. When exposed to flocculant alone, trout presented ~50% reduction in O_2 consumption compared to both pre-exposure conditions and pairwise MA alone and in combination with flocculent. Our results demonstrate the mode of toxicity of flocculant exposure and further demonstrate the efficacy of the MA.

10. Transport of Selenite by Human Red Blood Cells in the Presence of Arsenite

Javed, Warda[1], Kaur, Gurnit[2], Swanlund, Diane[1], Capicio, Michelle[1], Cordat, Emmanuelle[1], Leslie, Elaine[1,2]
University of Alberta

Over 200 million people worldwide are exposed to the proven human carcinogen arsenic, due to contaminated drinking water. Animal studies have shown that arsenic and the essential trace element selenium can undergo mutual detoxification through the formation of the seleno-bis(S-glutathionyl) arsinium ion $[(GS)_2AsSe]^-$ which undergoes biliary excretion, resulting in fecal elimination of both compounds. $[(GS)_2AsSe]^-$ is also formed in animal red blood cells (RBCs), resulting in the sequestration of arsenic and selenium. In human RBCs (hRBCs), the influence of selenium on arsenic and arsenic on selenium accumulation is largely unknown. In rat RBCs, Se^{IV} uptake is inhibited by 4,4'-diisothiocyano-2,2'-stilbenedisulfonic acid (DIDS), suggesting uptake is mediated by the erythrocyte anion-exchanger 1 (AE1, or Band 3). Preliminary experiments with radioactive $^{73}As^{III}$ and Se^{IV} suggested that Se^{IV} increased As^{III} accumulation within hRBCs. This led us to hypothesize that the presence of As^{III} would increase radioactive $^{75}Se^{IV}$ uptake into human RBCs by means of Band 3. Uptake was quantified using $^{75}Se^{IV}$ transport assays ± As^{III} ± DIDS. Se^{IV} accumulation in hRBCs was inhibited by ~90% in the presence of DIDS (50 μM). Counter to our hypothesis, As^{III} had little effect on Se^{IV} accumulation. This may be due to a faster rate of Se^{IV} uptake by hRBCs than As^{III}, making As^{III} uptake the limiting step in detoxification.

11. Influence of Selenium on Arsenic Uptake and Efflux by Human Hepatoma Cells and Primary Hepatocytes

Kaur, Gurnit[1], Ponomarenko, Olena[2], Summers, Kelly[2,3], Dolgova, Nataliya[2], Antipova, Olga[4], Douglas, Donna[5], Kneteman, Norman[5], Pickering, Ingrid[2,3], George, Graham[2,3], Leslie, Elaine [1,6]
University of Alberta

Millions of people worldwide are exposed to the proven human carcinogen arsenic at unacceptable levels. Animal models show that selenium and arsenic are mutually protective; increasing the biliary excretion of each other. However, the influence of selenium on human hepatic handling of arsenic is poorly understood. We hypothesized that selenium would increase the uptake and efflux of arsenite (As^{III}) in HepG2 cells and hepatocytes. To test this, we studied the influence of selenite (Se^{IV}) and selenide (Se^{II-}), on (i) arsenic uptake by suspended HepG2 cells and human hepatocytes, and on (ii) arsenic efflux from sandwich-cultured human hepatocytes (SCHH). After SCHH were treated with $^{73}As^{III}$ (± Se^{IV}/Se^{II-}), efflux across basolateral and apical surfaces was measured. Unexpectedly, SCHH biliary efflux of ^{73}As in the presence of Se^{IV} was reduced by 11 to 100% (n=5); basolateral efflux was reduced by 20 to 47%. Uptake of $^{73}As^{III}$ by suspended HepG2 and human hepatocytes was also inhibited by Se^{IV}, but increased by Se^{II-}. X-ray fluorescence imaging of HepG2 cells also suggested that As^{III}+Se^{II-} accumulation was 3-4 times higher than As^{III}+Se^{IV}. These results are consistent with the rapid reduction of Se^{IV} to Se^{II-} in erythrocytes; Se^{II-} being the main form transported to the liver.

12. Microplastic Contamination of the Canadian Archipelago

Kolter, S., Moon, S., Furze, M., Pieńkowski, A., Ross, M.
MacEwan University

Microplastics (MPs) are considered to be synthetic polymeric material <5mm in size and originate from either primary sources, those manufactured to be <5mm in size, or from the breakdown of larger pieces. MP pollution has been recorded in marine sediments in many of Earth's oceans and has been recently quantified in Arctic and Greenlandic sea ice and Greenlandic sea way sediments.
This study aims to map MP concentrations in the Canadian Archipelago to investigate their sources, migratory patterns and sinks. Eight samples were collected from Frobisher Bay, near Iqaluit, NU, during a 2017 summer expedition aboard the CCGS Amundsen ice breaker. MPs were isolated from sediments and organic materials by a combination of filtration, density floatation, and peroxide oxidation. Microplastics were identified by visual microscopy and specific MPs of interest were selected for spectral analysis, which may enable identification of polymer types.
This study has found the presence of MP contamination within the Canadian Arctic Archipelago at comparable concentrations to other published results on seafloor sediments. This is one of the first studies to report MPs in marine sediments from the Canadian Arctic Archipelago and enables us to begin mapping concentration levels in these areas. With further sampling, specific polymer identification, and ocean current mapping we may begin to realize point sources and migratory patterns of MPs.

13. Recovery of Macrophytes, Algae, and Periphyton Following Herbicide Exposure: A Case Study with Atrazine

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Atrazine is used in agriculture to control broadleaf weeds and acts through inhibiting the photosystem II protein reversibly. In the field, atrazine exposure is typically a pulsed event; a rapid increase of concentration due to runoff event and followed by a decline over several days. Pulsed characteristic and reversible inhibition allow primary producers to recover from effects related to this exposure. Therefore, understanding the recovery ability of macrophyte, algae, and periphyton from atrazine exposure is important to determine if atrazine in environment would have long-term effect on plants. A critical literature review was performed to examine the strength and relevance of studies looking at recovery from atrazine based. A total of 11 studies from peer-reviewed journal were obtained in the search and each paper scored based on a rubric for strength of methods. The exercise resulted in scores of 7 – 14 out of maximum score of 15. Of these, 96% reported no observed effect concentrations (NOEC) at or greater than environmental concentrations of atrazine. Majority of tested endpoints were not statistically different from the control following recovery periods (ranging from 2 hours to 14 days). Therefore, pulsed atrazine exposures in agroecosystems are not likely to lead to long-term effects in aquatic ecosystem.

14. Zebrafish Exposed to Elevated CO_2 Demonstrate Increased Anxiety-like Behaviour in the Novel Tank Diving Test

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Carbon dioxide (CO_2) levels are on the rise due to anthropogenic (human generated) activity. There has been a recent surge in studies examining the impact of elevated CO_2 on aquatic organisms, but most have focused on marine and not on freshwater fish. In this study, we investigated the effects of 4-days of elevated CO_2 on zebrafish (*Danio rerio*) a freshwater fish, and quantified behavioural changes with an anxiety-like behaviour test and motion-tracking software. The 'novel tank diving test' involves placing an individual fish into an arena and measuring locomotion as well as time spent in the upper, middle and lower zones of the water column. Previous studies have shown that zebrafish exposed to anxiolytic (anxiety reducing) test drugs will spend more time in the upper zone, consistent with a natural anti-predation mechanism. We found that fish exposed to elevated CO_2 levels (3248 ± 442 μatm) spent more time in the lower zone of the arena compared to controls (454 ± 49 μatm). These results suggest that freshwater fish exposed to short-term high levels of CO_2 can alter anxiety-like behaviour.

15. Heavy Metals Concentrations in Surface Water and Sediment from Odidi Oil Field, Niger Delta, Nigeria

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Oil industry activities such as exploration, transportation, storage, disposal, as well as oil spills are sources of major contamination problems in Niger Delta, which have significant deleterious effects on aquatic organisms. The objective of this study was to evaluate the impact of crude oil spillage and production activities in Odidi oil fields. The sampling stations are Odidi Flow Station (OFS), Egwua11 Flow Station (EFS), Batan Flow Station (BFS), and Warri town (WT). The result from this study shows that the heavy metal concentrations in surface water ranged between 0.0144mg/l (WT) and 0.108mg/l (BFS), 0.0223mg/l (WT) and 0.0685mg/l (BFS), 0.0024mg/l (WT) and 0.0169mg/l (OFS), 0.0013mg/l (WT) and 0.0027mg/l (BFS) for Cadmium, Lead, Nickel and Vanadium respectively. The heavy metal concentrations in sediment samples also ranged between 0.0444mg/l (WT) and1.9289 mg/l (BFS), 1.0283 mg/l (WT) and 4.52645mg/l (BFS), 0.0398mg/l (WT) and 0.5680mg/l (OFS), 0.0018mg/l (WT) and 0.2487mg/l (EFS) for Cadmium, Lead, Nickel and Vanadium respectively. The decreasing trend of metals in both surface water and sediment were Pb>Cd>Ni>V. These results showed that heavy metals concentrations in sediments were high than that of water. Also, heavy metals concentrations in water and sediment were above the permissible limit by FEPA/DPR, USEPA and WHO. The areas sampled had high heavy metals concentrations that are likely to crude oil spillage and production activities. It could be concluded that the Odidi Oil Field is heavily polluted which could have negative impacts on aquatic organisms inhabit the area.

16. Cellular Uptake and Cytotoxicity of Photoluminescent Fe_3O_4 Nanoparticle/Si Nanocrystal Hybrids

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The design and fabrication of silicon (Si)-based multi-functional nanomaterials for biological and bio-medical applications, is an active area of research. Si-based nanomaterials are ideal materials to for biomedical imaging because their luminescent properties are highly tunable when size and surface properties are adjusted. As well, Si-based nanomaterials are generally considered biocompatible because of the low toxicity of silicon itself. Combining the luminescent characteristics of Si with the magnetic properties of Fe_3O_4 nanoparticles multiplies the options available for medical applications, including being used as a cancer therapeutic. For example, the targeted delivery and internalization of magnetic nanoparticles to tumour cells could destroy the cancerous cells alone using an external magnetic field to induce hyperthermia. However, the potential for iron-based nanoparticles to participate in unregulated Fenton reactions once inside a cell, could negate any benefit from the development of a fluorescent and magnetic nano-therapeutic. In the

current study, we explore some of the properties of these newly synthesized magnetofluorescent nano-hybrid nanoparticles, and evaluate their potential to be internalized by sentinel immune cells and activate their innate immune functions. We have also evaluated their capacity to trigger cellular oxidative stress and apoptosis with and without the application of an external magnetic field.

17. Prey Capture to Male Aggression: The Role of Ecologically Relevant Behaviours in the Assessment of Complex Petroleum-based Contaminants.

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Crude oil and its associated by-products are ubiquitous in the aquatic environment due to both natural and anthropogenic sources (i.e. oil seeps and rivers flowing over surface bitumen, and pipeline ruptures, grounded ships, storage tank leaks and tailing pond seepage, respectively). This diversity in sources gives rise to a large family of complex contaminant mixtures, including weathered and unweathered oil, unconventional oil, such as diluted bitumen (dilbit), and crude oil extraction-based mixtures, such as oil sands process water (OSPW). Historically, studies focused on lethality and cardiotoxicity; complex behaviours have been, for the most part, overlooked despite the merits of including these endpoints in toxicological studies. In this study, we compared various ecologically relevant behaviours (prey capture, male aggression, reaction to alarm odourant) of developmentally exposed fish (*Danio rerio* and *Cyprinodon variegatus variegatus*) across various contaminants. Exposure to oil-based contaminants did not impair outright function, but instead altered the variation in behavioral phenotypes present in the population of exposed fishes. Previous studies suggest cortisol can be associated with behavioural phenotypes, and that developmental cortisol levels may pre-determine the behavioural phenotypes found in a population of exposed fishes. Complex behaviours are sensitive sublethal endpoints that could be used in the risk assessment of contaminant mixtures. The inclusion of complex behaviours in toxicological studies brings ecological relevance to a biomarker dominated field.

18. An Examination of Feeding Behaviour by a Freshwater Snail (*Planorbella pilsbryi*) as a Mechanism for the Creation of Secondary Microplastic Fibres

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Microplastics constitute a considerable fraction of plastic contamination in aquatic ecosystems. Defined as being less than five millimetres in diameter, microplastics are further classified as primary (i.e. purposely manufactured to ≤ 5 mm) or secondary (i.e. weathered from a larger plastic item). Microplastic fibres often fragment from larger textiles (i.e. secondary particles from washing), have been ubiquitously detected in the environment. This study examined another possible mechanism of microfibre creation. Specifically, we hypothesized that freshwater

snails assist in the creation of secondary microplastic fibres via their abrasive, cyclic feeding mechanisms. To address this question, we exposed a pulmonate snail (*Planorbella pilsbryi*) to a 100% polyester textile for 85 days under controlled laboratory conditions. Synthetic fibre counts were significantly greater in experimental chambers containing snails compared to those without (one-way ANOVA day 15: $p= 0.010$; one-way ANOVA day 57: $p= 0.029$). Feeding regimen did not significantly impact the degree of fibre fragmentation by snails (low food versus high food) (Tukey HSD test, $p > 0.050$). While the ecological implications of microplastic exposure to biota remain primarily unknown, this study confirms the ingestion of synthetic fibres by gastropods and presents evidence for a unique mechanism by which microplastic fibres may fragment in freshwater systems.

19. Data Quality in Microplastic Research

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Microplastics are an emerging contaminant of environmental concern with a rapidly growing body of literature. Microplastic research is still in its infancy and recent publications are concerned about the quality and relevance of reported findings for exposure and effects data. To address this, we conducted a quantitative critical appraisal of the peer-reviewed literature. Our objective was to assess the quality of microplastic exposure studies using a scoring rubric, which ranked studies from weak to strong. The assessment will provide clarity on plastic type/source, experimental design/test conditions, ecological relevance of exposure densities, test organism, replication, control performance, and statistical analysis. We employed 15 distinct criteria in our assessments including: source/plastic verification, measured densities, number of concentrations tested, ecological relevance, source of test organism identified, standard protocol and test conditions followed, replication, statistical methods, control performance and raw values. We found that many studies are falling short of meeting these 15 criteria when conducting the experiment and are failing to report all of the necessary detail. Overall, we recommend future work in the field aim to meet these minimum standards in order to improve the overall quality of microplastic literature, and their ability to assess potential environmental risk.

20. Toxicity of Nano-enabled Azoxystrobin on Zebrafish Embryo

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The use of nanotechnology to enhance pesticide formulations holds the promise of reduced pesticide use, reduced mobility in soils and overall improvements in agricultural practices while simultaneously maintaining yields. However, toxicity of nano-enabled pesticides, including azoxystrobin, has not been well studied comparing to their bulk form counterparts. This study investigates both lethal and

sub-lethal endpoints in zebrafish embryos up to 120 hours post fertilization (hpf) under laboratory light or simulated sunlight. The median lethal concentration (LC50) of both nano-enabled and bulk form azoxystrobin are determined. Malformations, including pericardial edema, tail deformity, yolk sac edema, and spinal curvature are observed. The gene expression of stress related genes, including *catalase, cyp1a, gst, sod1* and *sod2*, and enzyme activities of oxidative stress related enzymes, such as Catalase, SOD, GSH and CYP1A, are measured and compared between nano and bulk azoxystrobin. The oxygen consumption of embryos is measured at 48, 72, 96 and 120 hpf. The results will provide important information on the toxicity of nano-enabled azoxystrobin under ecologically realistic conditions.