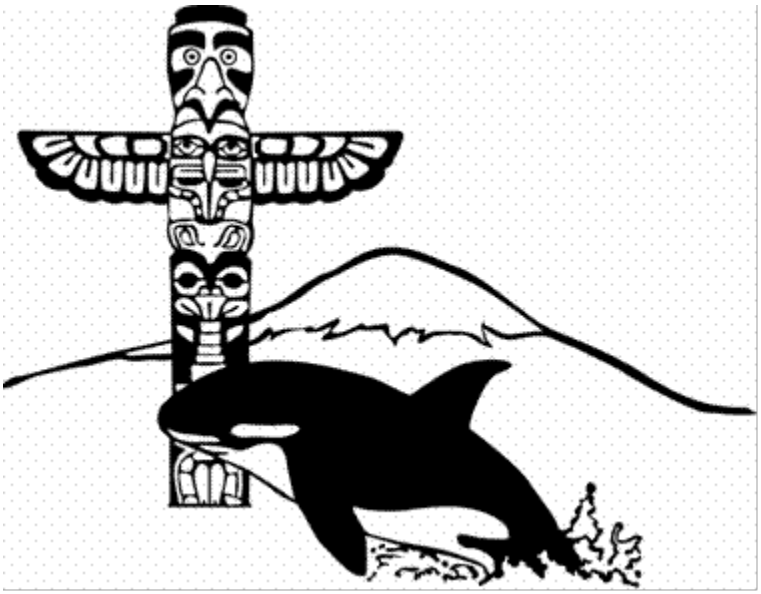
Program and Abstracts

13th Annual Meeting of the Pacific Northwest Chapter of the Society of Environmental Toxicology and Chemistry (PNW-SETAC)

The Next Decade of Research in Environmental Toxicology

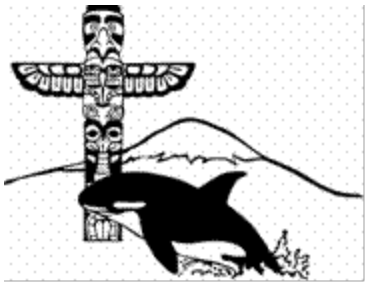
Fort Worden Conference Center, Building 204
Port Townsend, WA
April 15 - 17, 2004



PNW-SETAC ANNUAL MEETING

April 15 to 17, 2004

Meeting Program



PNW-SETAC
Chapter Meeting Program

Thursday, April 15, 2004

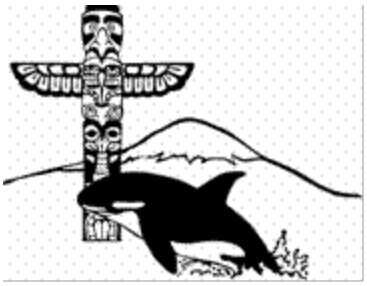
8:30 AM - 5:30 PM	**Conference Check-in, Building 204**
8:00 AM - 12:00 PM	Short Course 1: "*Modeling Exposure: Deterministic, Probabilistic, Spatial, and Population-Level Approaches*"
3:30 PM - 5:30 PM	Short Course 2: Career panel discussion/seminar
3:30 PM - 7:30 PM	**On-site Housing Check-in, Building 204** (The Commons/Dining Hall after 5:30 PM)
5:30 PM - 8:30 PM	Reception/Buffet & PNW-SETAC Board Meeting, The Commons/Dining Hall

Friday, April 16, 2004

7:30 AM - 8:15 AM	**Conference Check-in and Poster Setup, Building 204**
8:15 AM - 9:00 AM	Welcome address
9:00 AM - 12:00 PM	Platform sessions with 20 min break for refreshments and poster viewing
12:00 PM - 1:30 PM	Buffet Lunch, The Commons/Dining Hall
1:30 PM - 5:00 PM	Platform sessions with 30 min break for refreshments and poster viewing
5:00 PM - 6:00 PM	Poster Social, Building 204
6:00 PM - 8:30 PM	BEACH BBQ! Food and Drinks at the Fort Worden Kitchen Shelter

Saturday, April 17, 2004

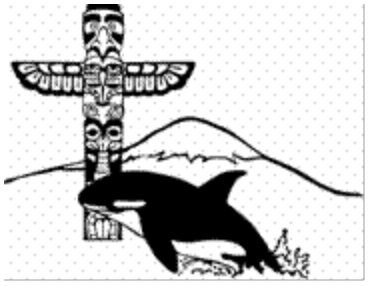
7:30 AM - 8:30 AM	PNW-SETAC Business Meeting and Breakfast, Building 204 (All welcome!)
8:30 AM - 8:40 AM	Student Award Presentation
8:40 AM - 12:15 PM	Special Session: "*Critical Body Burdens and Modes of Action*"



Thank You for

PNW-SETAC
Chapter Meeting Sponsorship

Student Awards and Travel Grants	Battelle Marine Sciences Laboratory
	Dinnel Marine Resources
	Exponent, Inc.
	Germano & Associates, Ltd.
	Hart Crowser, Inc.
Short Course Materials and Room	North American SETAC
A/V System	Institute of Environmental Toxicology and Chemistry, Huxley College, Western Washington University
Thursday Morning Break	Avocet Consulting
Thursday Reception	Battelle Marine Sciences Laboratory
	Exponent, Inc.
Friday Morning Break	NW Aquatic Sciences
Friday Lunch	ENTRIX, Inc.
Friday Afternoon Break	SAIC
Friday Night Barbecue	Hart Crowser, Inc.
Saturday Morning Break	Germano & Associates, Ltd.
Fish Sculptures	John Hicks

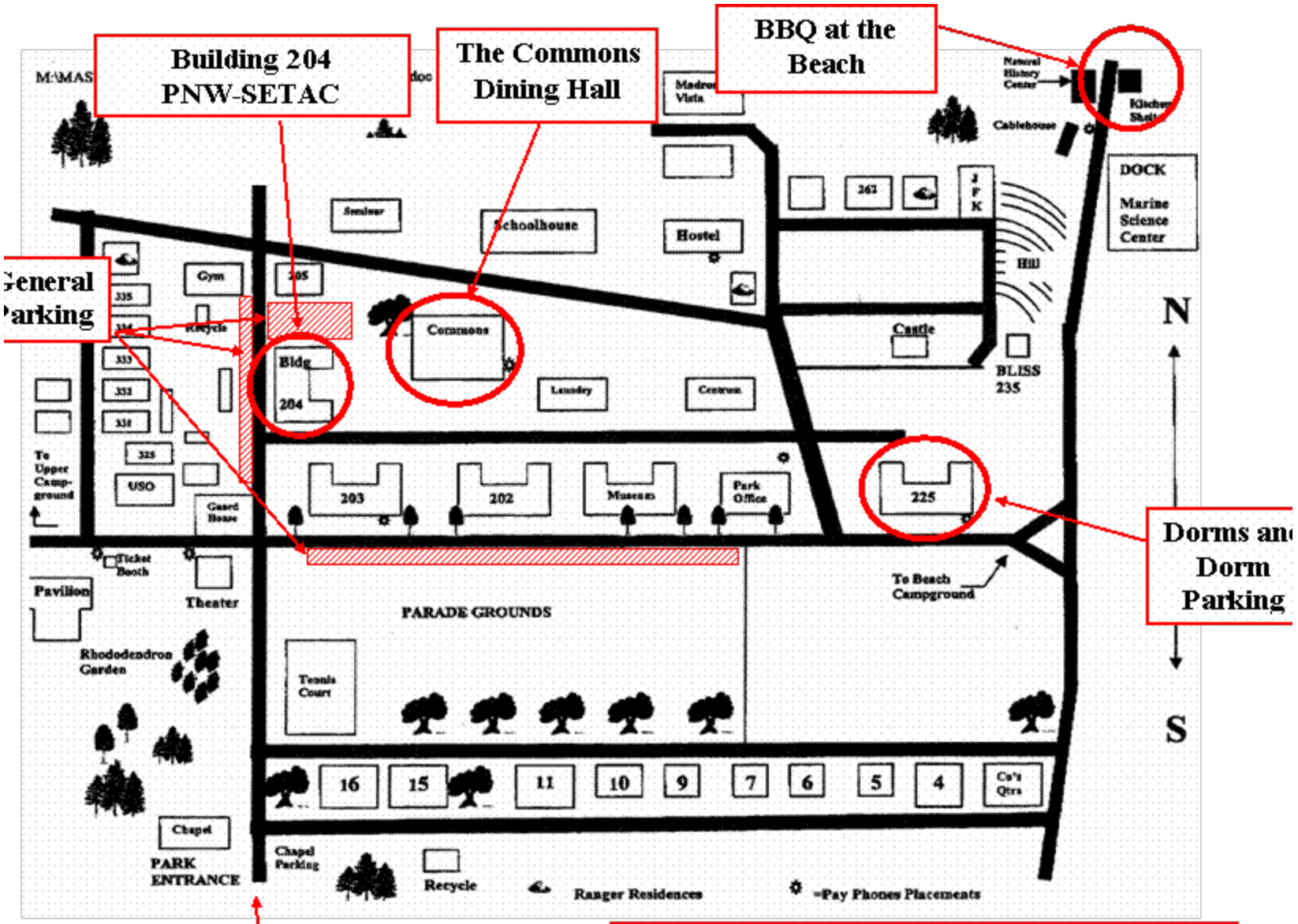


PNW-SETAC

Acknowledgements

Thanks to all of the following who volunteered their time to make this meeting possible:

Conference Organization:	Jack Word, MEC Analytical Systems, Inc. – Weston Solutions, Inc. Bill Gardiner, MEC Analytical Systems, Inc. – Weston Solutions, Inc. Angie Obery, Oregon Department of Environmental Quality Taku Fuji, Hart Crowser, Inc. Bruce Hope, Oregon Department of Environmental Quality Jim Meador, NOAA Fisheries April Markiewicz, Western Washington University
Abstract Review:	Jack Word, MEC Analytical Systems, Inc. – Weston Solutions, Inc. Bill Gardiner, MEC Analytical Systems, Inc. – Weston Solutions, Inc. Bridget Gregg, MEC Analytical Systems, Inc. – Weston Solutions, Inc.
Meeting Program:	Bill Gardiner, MEC Analytical Systems, Inc. – Weston Solutions, Inc. Cindy Word, MEC Analytical Systems, Inc. – Weston Solutions, Inc. Bridget Gregg, MEC Analytical Systems, Inc. – Weston Solutions, Inc.
Meeting Registration:	Angie Obery, Oregon Department of Environmental Quality Cindy Word, MEC Analytical Systems, Inc. – Weston Solutions, Inc.
Volunteer Coordinators:	Kristen Lawrence, AMEC Earth & Environmental, Inc. Heather Brunelle, AMEC Earth & Environmental, Inc.
Sponsorships:	Jack Word, MEC Analytical Systems, Inc. – Weston Solutions, Inc.
Student Awards:	Patty Toccalino, Oregon Health & Science University
Session Chairs:	Bill Gardiner, MEC Analytical Systems, Inc. – Weston Solutions, Inc. Nancy Musgrove, Management of Environmental Resources
Special Session:	Jim Meador, NOAA Fisheries
Useful Advice:	Nancy Kohn, Battelle Marine Sciences Laboratory Angie Obery, Oregon Department of Environmental Quality
Student Award Judges:	Nancy Kohn, Battelle Marine Sciences Laboratory Taku Fuji, Hart Crowser Ruth M. Harper-Arabie, Western Washington University Lisa Marko, Western Washington University
Student Funding Reviews:	Doug Henderson, King County Department of Natural Resources



For everything you need to know about Port Townsend, visit http://www.ptguide.com
For additional information about Fort Worden State Park, visit http://www.olympus.net/ftworden/

Pacific Northwest Chapter
Society of Environmental Toxicology and Chemistry
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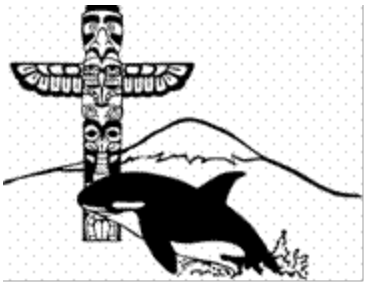
13th Annual Meeting

Schedule of Platform and Poster Presentations

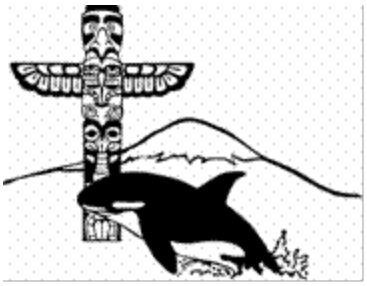


PNW-SETAC Annual Meeting
Platform Presentation Schedule
Friday, April 16, 2004

Chair: William (Bill) Gardiner, MEC Analytical Systems, Inc. – Weston Solutions, Inc.

Friday Morning:

9:00	Robert Johnston, Space and Warfare Systems Center	Cooperative Watershed Studies to Develop Water Cleanup Plans for Sinclair and Dyes Inlets, Washington
9:20	Chris Grue, University of Washington	Active Ingredient, Formulation, and Tank Mixes: What Should Be Regulated?
9:40	Zachary Guerette, University of Washington	Brain Cholinesterase Inhibition in Juvenile Rainbow Trout Exposed to Carbaryl--Benchmark Concentrations for Active Ingredient vs. Formulated Products
10:00	Paul Herschberger, USGS - BRD	Ichthyophoniasis: an Emerging Disease of Chinook salmon (*Oncorhynchus tshawytscha*) in the Yukon River
10:20	**Break/Poster Viewing**	
10:40	Judy Nedoff, Jenks Consultants	How Many PCB Congeners are Really Needed to Estimate Totals
11:00	Jennifer McIntyre, University of Washington	Mercury and Organochlorines in the Fishes of Lake Washington
11:20	Peggy Myre, Exa Data & Mapping Services	Investigation of Screening Values for Sediment Targeted for Wetland Restoration in San Francisco Bay
11:40	Robert Killin, Oregon State University	Anthropogenic Semi-Volatile Organic Compounds Measured at Cheeka Peak Observatory During Spring 2002
12:00	**Lunch**	

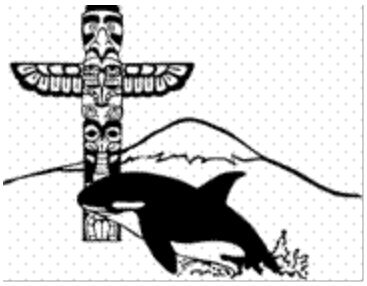


PNW-SETAC Annual Meeting
Platform Presentation Schedule
Friday, April 16, 2004

Chair: Nancy Musgrove, Management of Environmental Resources

Friday Afternoon:

1:30	Taku Fuji, Hart Crowser, Inc.	Comparison of the Response of the Ten-Day Midge and Amphipod Freshwater Sediment Bioassay Tests Versus the 20-Day Midge and 28-Day Amphipod Freshwater Sediment Bioassay Tests
1:50	Julann Spromberg, NOAA	Relating Chronic Toxicity Responses to Population-Level Effects: Modeling Effects on Wild Salmon Populations
2:10	Shirley Morales, Shannon Point Marine Center	Comparative Toxicity of Naphthalene to Early Life Stages of the Pacific Herring and Green Sea Urchin
2:30	Jeffrey Ward, Battelle Marine Sciences Laboratory	Marine Amphipod Challenge Testing: an Approach to Address Sublethal Effects
2:50	Vaishnavi Sarathy, Oregon Health and Sciences University	Copper Complexation by Ligands in Wastewater Effluent Versus Surface Waters
3:10	**Break/Poster Viewing**	
3:40	Audrey Colnar, Western Washington University	Regional Risk Assessment of the European Green Crab, *Carcinus maenas* in the Cherry Point Washington Region
4:00	Andrew Deines, Western Washington University	Modeling Non-Indigenous Species Introduction Using a Patch-Dynamics Approach
4:20	Eric Van Genderen, Parametrix, Inc.	Influence of Acute Copper Exposure on Whole-Body Sodium Levels in Larval Fathead Minnows
4:40	Mariana Tamayo, University of Washington	Non Target Impacts of B.t.i. Treatments to Control Mosquito Vectors of West Nile Virus-Implications for Aquatic Habitat, Salmonid, and Sage Grouse in Washington State
5:00	**Poster Social**	



PNW-SETAC Annual Meeting
Platform Presentation Schedule
Saturday, April 17, 2004

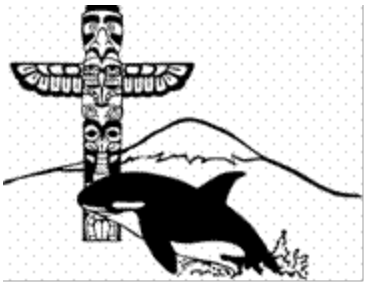
Chairs: Jim Meador, NOAA Fisheries

Guest Speaker

8:40	Wayne Landis, Western Washington University	My Most Annoying Things: NOEC, Reference Sites and the Misunderstanding of Man's Place in Nature

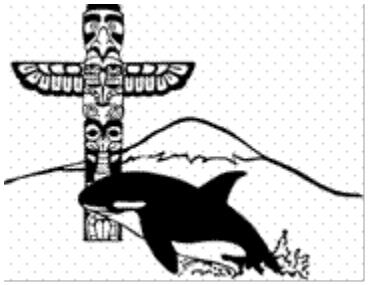
Critical Body Burdens and Modes of Action

9:00	Jim Meador, NOAA Fisheries	Overview of the Tissue Residue Approach for Toxicity Assessment
9:20	Jim Meador, NOAA Fisheries	Assessing Toxicity with the Tissue Residue Approach: Analysis with Tributyltin
9:40	Nancy Beckvar, NOAA OR&R	Evaluation of Four Methods for Assessing Risk Associated with Mercury and DDT Tissue Residues in Whole Fish
10:00	Burt Shepard, US Environmental Protection Agency	An Evaluation of Uncertainties Associated with Tissue Screening Concentrations Used to Assess Ecological Risks from Bioaccumulated Chemicals in Aquatic Biota
10:20	**Break**	
10:40	Mike Salazar, Applied Biomonitoring	Using Field Experiments to Determine Critical Body Residue Relationships and Mode of Action
11:00	Jack Word, MEC Analytical, Inc. – Weston Solutions, Inc.	Toxic Modes of Action and Mutualistic Responses of Test Organisms
11:20	Jeff Steevens, US Army Engineer Research and Development Center	Probabilistic Tissue Residue Benchmarks for Fish and Invertebrates
11:50	Jim Meador, Moderator	Panel Discussion
12:30	**End of Meeting**	



PNW-SETAC Annual Meeting
Poster Presentations

1. Margaret Dutch, Washington Department of Ecology	Chemical Contamination in the San Juan Islands, Eastern Strait of Juan De Fuca, and Admiralty Inlet
2. Margaret Dutch, Washington Department of Ecology	The Sediment Quality Triad Index in Puget Sound
3. Ed Long, Washington Department of Ecology	Sediment toxicity in the San Juan Islands, Eastern Strait of Juan De Fuca, and Admiralty Inlet
4. Jack Anderson, Battelle Marine Sciences Laboratory	Identification of Contaminated Sediments in Puget Sound by use of a Biomarker Assay
5. Nancy Kohn, Battelle Marine Sciences Laboratory	Sediment Metals Verification Study for Sinclair and Dyes Inlet Study
6. Stephen Quinnell, Washington Department of Fish and Wildlife	Use of Acoustic Tagging to Study Home Range and Migration of English Sole in Puget Sound: Application to Management of Contaminated Sediments
7. Stanley Howell, Western Washington University	Evaluation of Biodiesel (Soy Methyl-Ester) Toxicity to Soils Using the Lumbricid Earthworm *Eisenia fetida*
8. Frieda Taub, University of Washington	Closed Ecological Systems
9. Richard Caldwell, Northwestern Aquatic Sciences	The Toxicity of Free Cyanide to Yellow Rock Crab (*Cancer irroratus*) First Stage Zoeas
10. Goro Kushima, Western Washington University	Application of Life Cycle Assessment to Waste Water Treatment Systems in a Petrochemical Industry
11. Paul Dinnel, Shannon Point Marine Center	Toxicity of a Proposed Ballast Water Biocide, Seakleen, to Embryo and Larval Stages of Pacific Herring (*Clupea pallasi)*
12. Kerrie Serben, Viszon SciTec Inc.	The Influence of Water Hardness on Uranium Toxicity to Freshwater Organisms
13. Eric Larsen, Western Washington University	Applications of Respirometry for Determination of Toxicity to Activated Sludge and Implication for Secondary Wastes Water Treatment Plants
14. Mike Salazar, Applied Biomonitoring	Using Copper CBRs As A First Step Toward Determining Mode of Action in Bivalves



PNW-SETAC Annual Meeting
Poster Presentations

15. Catherine Curran, University of Washington	Toxicity of Rodeo and Arsenal Tank Mixes to Juvenile Rainbow Trout
16. Catherine Curran, University of Washington	Avoidance of Herbicide Tank Mixes by Juvenile Salmonids
17. Walter Major, University of Washington	Exposure of Salmonids to Carbaryl Following Application to Control Burrowing Shrimp
18. Kerensa King, University of Washington	Effects of Aquatic Herbicides on Smoltification in Juvenile Coho Salmon
19. Patricia Toccalino, Oregon Health & Science University	Assessing the Occurrence of Pesticides and VOCs in Ground Water in a Human-Health Context
20. Carrie Smith, Parametrix, Inc.	Distribution of Soil Bioavailability Parameters in Europe
21. Ruth Harper-Arabie, Western Washington University	Interactions of Microbial Exopolysaccharides with Plutonium in the Subsurface Environment
22. Wayne Landis, Western Washington University	General Conceptual Model for the Regional and Landscape Risk Assessment for Invasive Species
23. Angela Schuler, Western Washington University	Cherry Point Regional Risk Assessment: A risk Management Tool
24. Amanda Seebach, Western Washington University	Uncertainty Reduction through Nearshore Habitat Mapping of Cherry Point, Washington
25. Guilherme Lutufo, US Army Engineer Research and Development Center	Bioaccumulation and Critical Body Residues of Explosive Compounds in Aquatic Invertebrates and Fish
26. Sarah Brace, Puget Sound Action Team	What's Known about Polybrominated Biphenylethers in Puget Sound?
27. Luke Ackerman, Oregon State University	Polybrominated Diphenyl Ether Residue Analysis Method for Fish Tissues from Remote High Elevations Ecosystems
28. Richard Jack, King County Water & Land Resources	Evaluation of Endocrine Disrupters in Ambient Waters of King County Washington

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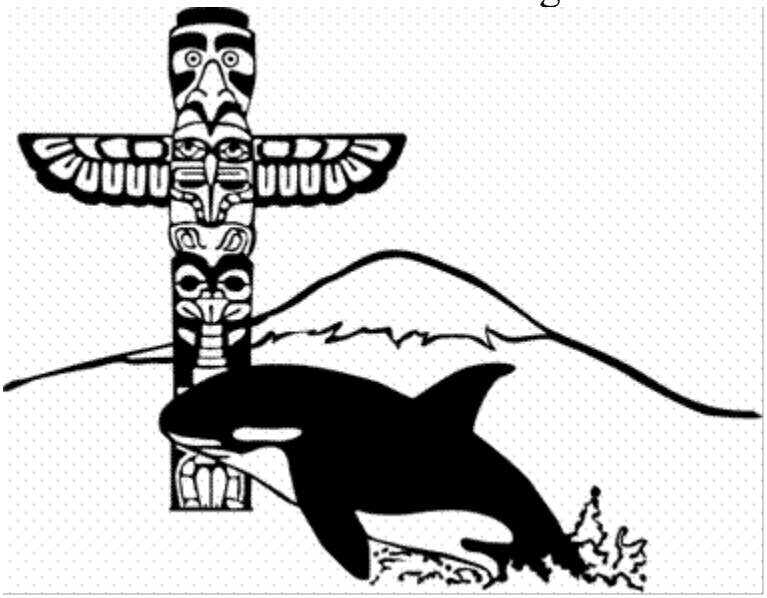

Platform Presentation Abstracts

ACTIVE INGREDIENTS, FORMULATIONS, AND TANK MIXES: WHAT SHOULD BE REGULATED?

Grue, CE[1*], Curran, CA[1], Cabarrus, JL, Grassley, JM[1], Gregg, BC[1], Gardner, SC[2], [1]Washington Cooperative Fish and Wildlife Research Unit, School of Aquatic and Fishery Sciences, University of Washington, Seattle WA; [2]Centro de Investigaciones Biologicas del Noroeste, La Paz, BCS

The assumption that within "end products", active ingredients represent the greatest hazard to non-target species and therefore should be the primary, if not sole, focus of pesticide regulations is not always valid. Type of formulation, impurities, inert ingredients, and adjuvants are all important in assessing hazards and risks associated with pesticide use. After all, it is the "end product" that is released into the environment. However, this is not adequately reflected in the existing regulatory frameworks within FIFRA and TSCA. Inconsistency in the regulatory classification of chemicals within tank mixes other than active ingredients - and therefore how these chemicals are treated within and between FIFRA and TSCA - is a concern. We briefly review the evidence indicating formulations and tank mixes can be significantly more toxic than active ingredients and that a new regulatory strategy is needed. Results of recent studies on the toxicity of formulations of aquatic herbicides (Rodeo®, ARSENAL® Herbicide and ARSENAL® AC), surfactants (R-11®, LI 700®, HASTEN®, and AGRI-DEX®), and tank mixes to juvenile rainbow trout (*Oncorhynchus mykiss*) will be highlighted. The hazards "end products" pose to non-targets needs to be assessed and the information made available to federal and state regulators and pesticide applicators.

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ANTHROPOGENIC SEMI-VOLATILE ORGANIC COMPOUNDS MEASURED AT CHEEKA PEAK OBSERVATORY DURING SPRING 2002.

Killin, RK*, Hauser, CD, Wilson, GR, and Simonich, SL, Oregon State University, Corvallis, OR.

Ambient high-volume (hi-vol) air samples were collected between March 15th and May 30th, 2002, at Cheeka Peak Observatory (CPO), located on the tip of the Olympic Peninsula, Washington State. This sampling campaign was in conjunction with the 2002 Inter-Trans Continental Transport and Chemical Transformation (ITCT 2K2) Campaign and the Photochemical Ozone Budget of the Eastern North Pacific Atmosphere (PHOBEA2) experiment. The goals of ITCT 2K2 and PHOBEA2 are to understand the long-range transport of ozone and aerosols and the impact that this intercontinental transport has on regional climate and air-quality. This research, which includes some of the first measurements of anthropogenic semi-volatile organic compounds (SOCs) in the Pacific Northwest, measured the levels of polycyclic aromatic hydrocarbons (PAHs) and various current-use and historical-use pesticides at CPO. The PAHs were measured in a range of 0.480-4.49 ng/m^3, which is comparable to other remote sites. Six pesticides (hexachlorobenzene, dacthal, chlorothalonil, heptachlor, trans-nonachlor, and cis-nonachlor) were also measured at various concentrations (0.104-57.0 pg/m^3) and found to be comparable to other remote sites. The potential sources of these compounds at CPO were determined using diagnostic ratios of their concentrations, back trajectories calculated using HYSPLIT4, local meteorological conditions, and U.S. pesticide use data. Gas-phase / particle-phase partitioning was explored, with strong correlation to temperature found with endosulfan I. Additional data is needed to confirm the sources of anthropogenic SOCs to CPO, either as a result of regional sources or the result of Trans-Pacific long-range transport.

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BRAIN CHOLINESTERASE INHIBITION IN JUVENILE RAINBOW TROUT EXPOSED TO CARBARYL – BENCHMARK CONCENTRATIONS FOR ACTIVE INGREDIENT VS FORMULATED PRODUCTS

Curran, CA, Black, TJ, Granger, BJ, Guerrette, ZN*, Ireland, KL, Marchesini, EA, Schneider, RC, Spardlin, CE, Sternberg, MR, Grassley, JM, Cabarrus, JL, Major, WW III, Grue, CE, School of Aquatic and Fishery Sciences, University of Washington, Seattle, WA.

A variety of pesticides have been detected in surface waters within agricultural and urban areas. In most cases, their biological significance (singly or in combination) to aquatic communities is not known. In the Pacific Northwest, the potential impact of low levels of pesticides in surface waters on efforts to enhance or restore habitats for juvenile and adult salmon is of particular concern. Several of the most commonly reported pesticides are cholinesterase-inhibitors and carbaryl (1-naphthyl N-methylcarbamate; trade name Sevin®) is the most commonly reported carbamate. Acetylcholinesterase (AChE) activity in brain tissue is the primary biomarker in fish and wildlife for diagnosing exposure to, and mortality from, pesticides that are cholinesterase-inhibitors. The enzyme is critical for normal nerve function and therefore reductions in enzyme activity may result in changes in physiology and behaviors essential for survival and reproduction. Studies by our laboratory and others suggest that "end products" may be significantly more toxic than active ingredients (ai) alone. Our objectives are: (1) determine dose response curves and threshold concentrations (benchmarks) for AChE inhibition in juvenile rainbow trout (*Oncorhynchus mykiss*) exposed to carbaryl as active ingredient and two formulated products (Sevin®) for 96-h, (2) determine differences in inhibition between soluble and membrane-bound forms of the enzyme, and (3) compare these values to concentrations reported in surface waters. Preliminary results suggest that benchmarks are ca. 15 ppb ai for the formulated products and technical grade material with inhibition similar between the soluble and membrane-bound forms.

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COMPARATIVE TOXICITY OF NAPHTHALENE TO EARLY LIFE STAGES OF THE PACIFIC HERRING AND GREEN SEA URCHIN

Morales, SA, Shannon Point Marine Center, Anacortes, WA

The Pacific herring (*Clupea pallasi*) is a valuable economic and forage fish species. Cherry Point, WA stocks of Pacific herring have suffered a 90% decline over the last thirty years, and it is possible that industrial effluents may have contributed some level of stress to already environmentally stressed populations. Development of Pacific herring embryo and larval toxicity tests for use in Whole Effluent Toxicity (WET) testing programs have been underway at the Shannon Point Marine Center in Anacortes, WA to provide additional bioassay tools for testing effluents. Previous testing during protocol development focused on the toxicity of selected reference toxicants (KCl, SDS and copper) and additional metals (cadmium, silver, zinc). Testing of naphthalene, the first organic compound to be tested in our laboratory, required special consideration of its volatility and limited solubility in seawater. Parallel tests of naphthalene using a 96-hour green sea urchin (*Strongylocentrotus droebachiensis*) embryo test provided data that can be used to assess the relative sensitivity of herring and sea urchin test protocols. Results will be presented for the following test endpoints: herring 18-day embryo development (normal survival, heart rate, embryo movements, time to 50% hatch, larval length at hatch), herring 10-day larval survival and growth (LC50, larval dry weight), and sea urchin development (normal survival).

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COMPARISON OF THE RESPONSE OF THE TEN-DAY MIDGE AND AMPHIPOD FRESHWATER SEDIMENT BIOASSAY TESTS VERSUS THE 20-DAY MIDGE AND 28-DAY AMPHIPOD FRESHWATER SEDIMENT BIOASSAY TESTS

Fuji, TT*[1], Kroeger, KA[1], Irissarri, G[2] and Sherman, T[3]. 1: Hart Crowser, Portland, OR. 2: Northwestern Aquatic Sciences, Newport, OR. 3. US Army Corps of Engineers, Portland, OR.

Sediment bioassay testing has been advocated for use in sediment and dredge material characterization studies and ecological risk assessments in the Pacific Northwest. Historically, the bioassays recommended for use in freshwater systems were limited to the 10-day amphipod (*Hyalella azteca*) survival test and the 10-day midge (*Chironomus tentans*) survival and growth bioassay. Recently, longer-term tests have been developed for both of these test organisms, although there as been very limited use of these longer-term tests in the Pacific Northwest. This work compares the results of the ten-day midge and amphipod tests and 20-day midge and 28-day amphipod tests conducted concurrently on 19 test sediments and two reference sediments collected from the Lower Willamette River in Portland, Oregon. The results of the bioassay tests, analytical chemistry data for test and reference sediments, physical characteristics of the test and reference sediments, and potentially confounding factors will be presented and discussed.

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COOPERATIVE WATERSHED STUDIES TO DEVELOP WATER CLEANUP PLANS FOR SINCLAIR AND DYES INLETS, WASHINGTON

Johnston, R.K.[1*], Sherrell, G.M.[2], Leisle, D.[2], Kendra, W.[3], Lawrence, S.[3], Rylko, M.[4], Turvey, M.[4], May, C.W.[5], Miller, M.[5], Richter, K.E.[1], Ostrom, T.[6], Mecham, M.[7], Tucker, D.[8], Zimny, J[9] , [1]Space and Naval Warfare Systems Center, [2]Puget Sound Naval Shipyard & Intermediate Maintenance Facility, [3]Washington Department of Ecology, [4]Environmental Protection Agency Region X , [5]Pacific Northwest National Laboratory, [6]Suquamish Tribe, [7]City of Bremerton, [8]Kitsap County Surface and Storm Water Management, [9]Kitsap County Health District

Sinclair and Dyes Inlets, Washington were listed on the 1998 303(d) list of impaired waters because of fecal coliform (FC) contamination in the marine waters and metals and organic contaminants in bottom sediments and fish tissues. The Puget Sound Naval Shipyard & Intermediate Maintenance Facility, Department of Ecology, U.S. Environmental Protection Agency and local stakeholders are working together on Project ENVVEST (an acronym for ENVironmental InVESTment) to address contamination issues and develop water cleanup plans for the watershed. Significant progress has been made on the FC Total Maximum Daily Load (TMDL) study for Sinclair and Dyes Inlets, which has benefited from the collaboration and cooperation of many stakeholders within the watershed. Currently, the FC model verification sampling for the TMDL study is being planned and storm water flow monitoring is being initiated for representative storm water outfalls within the study area. Considerable progress has been made on modeling the watershed and receiving waters within the Inlet. The modeling studies have directly contributed to the recent reclassification of Northern Dyes Inlet (about 1500 acres) from “prohibited” to “conditionally approved” for shellfishing by the Washington State Department of Health. Further progress has also been achieved for storm event sampling of streams, watershed monitoring, and conducting a sediment metals verification study. Additional studies are being conducted to evaluate contamination accumulation in the food chain of Sinclair and Dyes Inlets by analyzing species of fish and invertebrates from Sinclair Inlet and reference areas in the Puget Sound for metals, polychlorinated biphenyls, and pesticides.

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COPPER COMPLEXATION BY LIGANDS IN WASTEWATER EFFLUENT VERSUS SURFACE WATERS

Sarathy, V*[1] and Allen, HE[2], 1. Oregon Graduate Institute, OHSU, Beaverton, OR, 2. University of Delaware, Newark, Delaware.

Metal complexation by wastewater treatment plant (WWTP) effluents is important in understanding bioavailability and potential metal toxicity. Models such as WHAM or BLM do not independently consider WWTP organic matter from that of natural organic matter (NOM). We characterized organic matter from surface waters and a wastewater treatment plant and studied their copper complexing properties using a copper ion-selective electrode. NOM and organic matter from WWTP have ligands with similar binding constants, but the copper binding of WWTP effluents is not well predicted by WHAM especially at low copper concentrations (below 10^{-6}M). Experimental data also showed that the concentration of strong ligands at low Cu concentrations were about 15 times higher for the WWTP DOM than for the DOM sampled from the Edisto River, South Carolina. This could be due to the presence of sulfide in wastewater effluents that binds strongly to copper, but which is not considered by WHAM. However the consideration of sulfide alone does not fully explain the deviation of experimental data from WHAM predictions. This suggests that non-humic ligands that probably come from proteins and other biological macromolecules, which account for the strong copper binding, may be present. Hence organic matter from WWTP may need to be considered as an alternative set of ligands to humics in toxicity and speciation predicting models.

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HOW MANY PCB CONGENERS ARE REALLY NEEDED TO ESTIMATE TOTALS?

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Samples of crayfish and several resident fish species (sculpin, smallmouth bass, crappie, carp and bullhead) collected from Portland Harbor to support ecological and human health risk assessments (ERA and HHRA) were analyzed for PCB congeners. Average total PCB concentrations showed the PCB concentration pattern: carp > bass > sculpin > bullhead > crappie > crayfish, but there was considerable overlap in the concentration ranges. The contribution of each individual congener to the total PCB concentration was evaluated to develop a subset of the congener list that is representative of those present in tissue from resident species. The summed concentration of the 12 dioxin-like congeners ranged from 2 to 9 percent of the total PCB concentration. The summed concentration of the 18 congeners recommended in the NOAA analytical method ranged from 44 to 62 percent of the total PCB concentration for the various species. The 25 congeners that comprise the combined list ranged from 45 to 64 percent of the total PCB concentration for each species. These results demonstrate that if only these congeners are analyzed, they would account for approximately half of the expected total PCB concentration. Ranked by concentration, the top 30 congeners in each of the six species account for over 80 percent of the total PCBs. To include the dioxin-like and NOAA congeners requires an additional 12 to 16 congeners, depending on the species. To be inclusive of the top 30, the 12 dioxin-like, and the NOAA method congeners in all six species requires inclusion of 62 congeners. Two NOAA congeners (8 and 18) are detected at very low concentrations in the six species and do not affect the total. The resulting 60-congener list represents 82 to 89 percent of the total PCB concentration.

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ICHTHYOPHONIASIS: AN EMERGING DISEASE OF CHINOOK SALMON (*ONCORHYNCHUS TSHAWYTSCHA*) IN THE YUKON RIVER

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Ichthyophonus sp. Is a highly pathogenic protozoan parasite, primarily of marine and estuarine fishes, that has been associated with massive epidemics in populations of wild fishes over the past century. From 1999-2003, a total of 3,189 adult Chinook salmon (*Oncorhynchus tshawytscha*) from the Yukon River drainage in Alaksa, USA and Yukon Territory, Canada were examined for *Ichthyophonus*. Overall infection prevalence in Yukon River fish peaked at ~45%. Mean infection prevalence from 1999 to 2003 was significantly higher in females in the mainstem Yukon River, but infection prevalence in males increased significantly over this period. Clinical signs of ichthyophoniasis were lowest at the mouth of the Yukon River (~10%), but increased to 29% at the middle of the Yukon River, and 22% at the upper Tanana River. As fish approached the upper reaches of Alaska, the prevalence of infection dropped significantly to <15% and the prevalence of clinical disease dropped to ~12% on the Yukon River and <10% on the Chena and Salcha Rivers, presumably due to pre-spawning mortality of diseased fish. Position of fish within the run did not affect the overall prevalence of infection or disease, but eh severity of disease was greater in late run fish exposed to higher water temperatures during their in-river migration.

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INFLUENCE OF ACUTE COPPER EXPOSURE ON WHOLE-BODY SODIUM LEVELS IN LARVAL FATHEAD MINNOWS

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Surface waters of low hardness (≤ 40 mg/L as $CaCO_3$) may inherently present significant stress to aquatic biota. For example, freshwater fish must continually regulate internal concentrations of ions (i.e., Na^+, K^+, Cl^-) in order to survive. As ambient concentrations of these ions decrease, the concentration gradient between the plasma and the environment increases. Since metals, such as copper, inhibit ionoregulation, the increased energy requirement to counter passive diffusive losses in soft water may translate into increased sensitivity to metal exposure. We developed a method to determine whole-body sodium concentrations of larval fathead minnows as a physiological indicator of health. This method was used to characterize net rates of sodium flux from fish exposed to copper in the presence of varying levels of hardness and alkalinity. Organisms exposed to copper at acutely lethal levels had significantly less whole-body sodium than control fish by 6 hours following test initiation. By 12 hours, sodium flux had diminished and whole body sodium remained relatively constant at low levels until mortality occurred. Hardness dependent rates of sodium loss showed evidence of saturation-type kinetics. Competitive inhibition of copper by hardness cations was demonstrated above hardness of 20 mg/L as $CaCO_3$, when alkalinity was held constant. However, rates of whole-body sodium loss decreased as alkalinity increased from 10 to 100 mg/L as $CaCO_3$ (constant hardness), which was attributed to carbonate complexation. Consequently, current hardness-based water quality regulation for copper may be accurate only when alkalinity is co-varied with hardness. Further characterization of the mechanism of metal toxicity in soft water, and the resulting physiologic response in larval fish, may better explain observed sensitivity in this environment.

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INVESTIGATION OF SCREENING VALUES FOR SEDIMENT TARGETED FOR WETLAND RESTORATION IN SAN FRANCISCO BAY

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The San Francisco Regional Water Quality Control Board issued revised sediment screening guidelines in 2000 for beneficial reuse of dredged material for wetland restoration. Two sets of guidelines exist for wetland surface and underlying foundation sediments. The guidelines default either to ambient concentrations or to national guidelines, with no information on the performance when applied to Bay sediments. The first goal of this project was to measure how well these guidelines, as well as other existing regional and national guidelines, predict actual biological toxicity. Because wetland surface material will have direct exposure to organisms, surface material guidelines should avoid incorrect prediction of toxic sediments as non-toxic (false negatives). In contrast, the goal for foundation material is to minimize placement of sediment suitable for critical surface material, and thus avoid prediction of non-toxic sediments as toxic (false positives). The database generated and used for the project included 337 amphipod (*Ampelisca abdita*, *Eohaustorius estuarius*) bioassays. Results of the performance analyses for surface material showed that, while optimal target rates were nearly achieved by most of the national guideline sets, very few samples were predicted to be suitable for surface material, restricting the existing guidelines' practical utility. The existing foundation guidelines did not perform well. Because of the substantial overlap in concentration distributions between the toxic and non-toxic populations, we investigated methods to optimize the existing values. First, we adapted the Receiver Operating Characteristic (ROC) approach, both to find chemicals that most accurately predict toxicity (toxicity drivers) and to optimize the value of specific guidelines. Finally we developed site-specific values using the Floating Percentile (FP) method that can simultaneously optimize false negative and false positive error rates. Significant improvements in accurately predicting toxicity for both surface and foundation material were gained with the site-specific FP guidelines.

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MARINE AMPHIPOD CHALLENGE TESTING: AN APPROACH TO ADDRESS SUBLETHAL EFFECTS

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Acute, 10-day bioassays with marine or estuarine amphipods are commonly used to evaluate sediment quality, and are often a required component of ecological assessment programs conducted under federal or state guidance. Amphipod bioassays are generally good at predicting adverse effects associated with moderately or highly contaminated sediment, but are less useful when low-level contamination is present that exerts a sublethal rather than acutely toxic effect. In an attempt to increase the sensitivity of standard amphipod tests, toxicologists have included sublethal endpoints (e.g., reburial), or have extended the exposure duration to enable assessments of growth and reproductive endpoints. A challenge test represents an alternative approach to assess sublethal effects by evaluating the health and viability of test organisms after exposure to sediment. Marine amphipods (*Rhepoxynius abronius)* were exposed to uncontaminated and moderately contaminated sediment for the standard 10-day period; then, surviving amphipods were recovered and "challenged" in a 4-day reference toxicant exposure to cadmium. The dose response observed in the challenged amphipods was compared with the response observed in unchallenged amphipods that were subjected to a similar reference toxicant exposure. The results of the experiments suggest this technique shows promise as a tool to evaluate sublethal effects that may not be detectable in standard amphipod bioassays.

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MERCURY AND ORGANOCHLORINES IN THE FISHES OF LAKE WASHINGTON

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Elevated levels of organochlorines (up to 2.2 ppm ΣPCB) and moderately elevated levels of mercury (up to 0.6 ppm THg) were recently found in resident fishes of Lake Washington, a large urban lake in the Puget Sound basin of Washington State. Historical comparisons show that mercury levels in Lake Washington remain essentially unchanged since the mid-1970s. Regional comparisons show that organochlorine levels are average. Possible sources of contamination will be discussed. Lipids are shown to be correlative with contaminant levels in some cases, but not causative. Seasonal patterns in contaminant levels of forage fishes are explained by patterns in growth and diet

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MODELING NON-INDIGENOUS SPECIES INTRODUCTIONS USING A PATCH-DYNAMICS APPROACH

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The importance of spatial relationships in population biology, ecology and risk assessment is increasingly clear. The application of patch models to the investigation of interspecific competition has been mostly limited to meta-population models which do not have resolution of population structure at local patch levels. The goal of this work was to develop an iterative model concerning affects of toxicity and competition on a three-patch system to aid in the development of testable hypothesis at the patch scale. I adapt the patch model of Spromberg et al (1998) to include a second competitive species. The connectivity of patches was modeled using diffusion-reaction and competition using Lotka-Volterra equations. Four major predictions resulted from simulation. 1) Competition changes the population structure of distinct outcomes and the frequency of these outcomes. 2) Changes in population structure are possible in patches where the second species is not present. These changes may occur without altering the patch of the introduction. 3) Competition increases the variability in the population of the first species, making it more susceptible to random events. 4) High competitive ability does not increase the establishment potential of an introduced species.

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NON-TARGET IMPACTS OF *B.t.i.* TREATMENTS TO CONTROL MOSQUITO VECTORS OF WEST NILE VIRUS – IMPLICATIONS FOR AQUATIC HABITATS, SALMONIDS AND SAGE GROUSE IN WASHINGTON STATE

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Wetlands, particularly those in urban areas, are often the target for mosquito control to reduce the nuisance to surrounding residents. In response to potential loss of human lives associated with the rapid spread of mosquito-borne West Nile virus, local governments are being pressured to control mosquitoes. The bacterial larvicide, *Bacillus thuringiensis israelensis* (*B.t.i.*), is commonly used to control mosquito larvae because of its low direct toxicity to vertebrate species. Although vertebrates are not expected to suffer direct adverse effects, there is greater potential for indirect effects from food web disruption. The non-target effects of *B.t.i.* are poorly understood impeding the development of mosquito control strategies compatible with mandates to enhance and/or protect threatened species and their habitats. In Washington State, efforts to protect human lives may result in losses of invertebrates important to juvenile salmonids (e.g., listed salmon and trout species) for which habitat enhancement activities are underway. The recent listing of sage grouse (*Centrocercus urophasianus*) likely will further justify the control of mosquitoes within wetlands and moist habitats within its range, as sage grouse are susceptible to the virus. We review the existing information on the non-target effects of *B.t.i* and their implications to salmonids and sage grouse in Washington State. Ongoing studies on the non-target effects of *B.t.i.* on invertebrates important to juvenile salmonids on the Franz Lake NWR will be described as well as efforts to develop an effective research strategy to determine the impacts of *B.t.i* to salmonids and sage grouse in the State.

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REGIONAL RISK ASSESSMENT OF THE EUROPEAN GREEN CRAB, *CARCINUS MAENAS*, IN THE CHERRY POINT, WASHINGTON REGION

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Risk assessments determine the probability of adverse effects of stressors upon socially valued ecological structures or functions. Non-indigenous species are now receiving more attention as a new category of stressor. Previous attempts at assessing the risk of invasion by non-indigenous species primarily consider the risk of introduction, which is merely the exposure of the stressor to the habitat. The subsequent effects to selected biological endpoints are not considered, resulting in an incomplete analysis. Furthermore, the previous studies are more qualitative than quantitative and have yet to fully consider the scale and complexity associated with invasion by non–indigenous species. Applying the Relative Risk Model developed by Landis and Wiegers (1997), I performed the first quantitative, regional risk assessment of non-indigenous species in the Cherry Point region of Washington state with the European green crab (*Carcinus maenas*) as a model species. This risk assessment estimates the risk of invasion by a non-indigenous species and the risk of effects to selected biological endpoints within the Cherry Point region. Critical considerations in performing a risk assessment of invasion by non-indigenous species include: 1)the life history of the organism and 2) the small and large scale dynamics of the system that influence the habitat suitability and patch dynamics of the organism, which in turn affect the success of invasion. Much uncertainty still remains concerning these dynamics and therefore, this risk assessment will help to identify areas of research that can alleviate this uncertainty, making regional risk assessments of invasion by non-indigenous species a more standard process.

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RELATING CHRONIC TOXICITY RESPONSES TO POPULATION-LEVEL EFFECTS: MODELING EFFECTS ON WILD SALMON POPULATIONS.

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Standard laboratory toxicity tests assess the physiological responses of individual organisms to exposure to toxic substances under controlled conditions. Time and space restrictions often prohibit the assessment of population-level responses to a toxic substance. Compounds effecting various toxicity endpoints, such as growth, fecundity, behavior or immune function, alter different demographic traits and produce different impacts on the population. Chronic effects of immune suppression, reproductive impairment, and growth reduction were examined using life history models for Chinook salmon (*Oncorhynchus tshawytscha*), and coho salmon (*O. kisutch*). Immune suppression acted through reductions in age-specific survival. Output from the chronic reproductive impact models lowered the population growth rate (λ) to a lesser extent than impacts to survival. A 10% reduction in fecundity, survival of adults during the spawning migration, and spawning behavior of adults, all produced a similar λ for the respective species, but different sensitivity matrices and stable age distributions. Growth reduction models incorporating effects to both survival and reproduction produce greater effects than each would individually. Similar changes in λ often produced very different changes to the age distribution. Sensitivity and elasticity analyses show that, for Chinook and coho salmon, changes to the first-year survival rate produce the greatest per unit effect on λ. The reduction of age 1 survival by 10% for 5 years can lead to a population that is more than 10% below its potential abundance in 20 years. These results indicate the importance of linking toxicity measurement endpoints to the demographic traits that they affect, and help generate toxicity tests that are more relevant for the species. Further models will be conducted for sockeye salmon (*O. nerka*), and English sole (*Pleuronectes vetulus*). Life history modeling provides a useful tool to develop testable hypotheses regarding impacts on specific populations as well as to conduct comparisons between populations.

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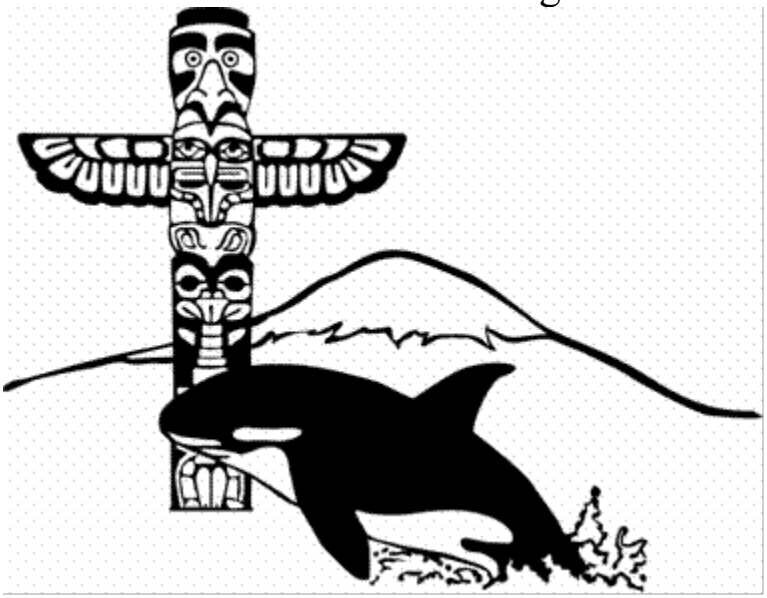

Special Session Abstracts

AN EVALUATION OF UNCERTAINTIES ASSOCIATED WITH TISSUE SCREENING CONCENTRATIONS USED TO ASSESS ECOLOGICAL RISKS FROM BIOACCUMULATED CHEMICALS IN AQUATIC BIOTA

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Tissue screening concentrations (TSCs), the product of a water quality criterion and a bioconcentration factor, have been in use for several years as an ecological risk assessment screening tool to identify chemicals in aquatic biota tissues which are elevated to concentrations that may pose unacceptable ecological risk to the aquatic biota themselves. To confirm the utility of TSC values, over 4000 residue-effects records from the literature have been compiled and compared to the TSC values, with at least one literature record available for 133 of the 180 chemicals for which TSCs exist. Nearly 1000 of the literature records are for field exposed biota. The availability of this literature has permitted detailed analysis of the utility of and uncertainties associated with TSCs. For both laboratory measured and field observed adverse effects, TSC values are lower than approximately 94% of the residues associated with toxicity. For residues measured in field exposed biota that exceed TSC values, the literature indicates that toxicity occurs at lower tissue residues relative to concentrations bioaccumulated by laboratory test species in single chemical toxicity tests with observed adverse effects, possibly due to independent joint action of multiple chemicals found at low concentrations in the environment. Other findings are that risks from metals may be overestimated by some of the existing TSC values; that aquatic plants as a group are more tolerant of elevated contaminant residues than are animals; and that the overall sensitivity of invertebrates and fish to most contaminants are similar. By using receiver operating characteristic (ROC) curve statistics, false positive and false negative error rates can be quantified for any TSC where sufficient effects and no effects literature exists to generate an ROC curve. The overall accuracy of any residue concentration proposed as a TSC can also be quantified.

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ASSESSING TOXICITY WITH THE TISSUE RESIDUE APPROACH: ANALYSIS WITH TRIBUTYLTIN

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Lethal and sublethal toxicity values based on tributyltin (TBT) exposure concentrations display a very large range in values. Mortality responses for TBT in water exposures are reported from 0.1 ng/mL to over 200 ng/mL, a range of about 2000 fold. Most of this variability is due to differences in the uptake and elimination kinetics between species; however some of the values underestimate the toxic response because of insufficient time for exposure. Sublethal responses to water exposure are also highly variable ranging from the low (0.01 ng/mL) to high (0.5 ng/mL) parts per trillion. The most common sublethal endpoints measured are growth inhibition, shell chambering in oysters, histological and behavioral abnormalities, and imposex in prosobranch gastropods. Although TBT toxicity may occur from several modes of action, the mortality response and growth inhibition appear to occur at relatively constant whole-body tissue concentrations for a variety of species. The lethal whole-body tissue concentration of TBT affecting 50% of the individuals (LR_{50}) exhibits low variation, occurring at approximately 48 μg/g (166 nmol/g dw) in a variety of species. An examination of several studies shows that the lowest observed effect tissue residue (LOER) associated with impaired growth for several species is approximately 3 μg/g (10.4 nmol/g dw). Additional information on tissue residues associated with reproductive impairment indicate response residues in the 0.5 μg/g (10.4 nmol/g dw) range. Based on these data, the tissue residue approach appears to work very well for TBT.

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EVALUATION OF FOUR METHODS FOR ASSESSING RISK ASSOCIATED WITH MERCURY AND DDT TISSUE RESIDUES IN WHOLE FISH.

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Attempts by a number of investigators to use tissue residue concentrations in ecological risk assessments have increased with the emergence of several published tissue-residue compilations and databases. However, methods for assessing risks associated with tissue contaminant concentrations are not well developed and vary greatly among investigators. Water quality criteria and sediment quality guidelines, which have been thoroughly examined and critiqued, may provide suitable models for the development of tissue-residue guidelines. We selected mercury and DDT to evaluate methods because they are persistent, bioaccumulative and toxic contaminants routinely detected in fish and the environment. Data for deriving protective tissue residue concentrations are limited to existing publications that usually were done for other purposes. We developed guidelines for screening potential papers, with the goal of retaining as many studies as possible, while eliminating uncertain or low quality data. A set of decision rules was formulated to provide guidance for extracting data from the screened papers in a consistent manner. Paired no-effect and low-effect whole-body residue concentrations in fish were identified from the published literature. Four analytical approaches of increasing complexity were evaluated for use in deriving tissue residue guidelines. The approaches ranged from the empirical to probabilistic. Ranked-based, percentile, Threshold Effect Level (TEL) and cumulative distribution function (CDF) approaches were evaluated using the same datasets for both mercury and DDT. The advantages and limitations of each approach will be discussed.

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MY MOST ANNOYING THIINGS: NOECS, REFERENCE SITES AND THE MISUNDERSTANDING OF MAN'S PLACE IN NATURE

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After having done environmental toxicology for twenty five years I am still disappointed, even annoyed but many things. My top three most annoying things currently are: the use of NOECs as data, reference sites as real objects, and the misunderstanding of man's place in nature (with homage to T. H. Huxley) First, given the drawbacks of NOECs and LOECs and the recent work of Moore and Caux (1997) and Stephenson et al (2001) there are adequate methods for using curve fitting as the preferred method of the analysis of concentration-response relationships. Second, reference sites are symptomatic of a misunderstanding of the dynamics and structure of ecological systems. Gradients exist in a variety of directions and should be accounted for in any field analysis. A gradient design will provide the necessary information for establishing cause-effect in a Weight of Evidence approach. Finally, there still exists a dichotomy where humans are seen as separate from the natural world. T. H. Huxley was among the first to articulate the falsehood of this separation. Mankind is but one part of the world, deeply interacting with it, there being no natural or artificial except in a human centric sense. Assuming that a minimally-impacted site or condition is attainable underestimates the impact of our species, natural events and species interactions no matter the cultural context. If successful long-term management of ecological systems is attainable, it will require a fundamental and deep understanding of the nature of these systems and mankind's integral relationships within it.

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PROBABILISTIC TISSUE RESIDUE BENCHMARKS FOR FISH AND INVERTEBRATES.

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From the fundamental perspective, an internal dose of a contaminant is a much better surrogate for estimating the dose at the toxicological site of action than the external or the ambient dose because it precludes the effects of various external factors that modify toxicity. For this reason, many risk assessment and management practitioners have been eager to develop and apply tissue residue-based toxicity benchmarks (TRBs). However, the traditional TRBs have the limitation of being single point estimates based on the critical body residue (CBR) associated with the most sensitive toxicity endpoint of the most sensitive species and, therefore, they tend to be overly conservative. To solve this problem, we have developed a methodology that establishes benchmarks as distributions as a replacement for the current approach of single point estimates. The methodology is based on the species sensitivity distribution (SSD) approach that captures the toxicological sensitivity to a specific chemical for a range of species to generate a statistical distribution. This distribution is expressed as a cumulative distribution function (CDF) that is composed of effect concentrations obtained during toxicity testing (LCxs, LOELs, or NOELs) and cumulative probability. The number of data points used to construct each curve depends on the number of species tested and specific toxicological endpoint availability. Emphasis is placed on ecologically-relevant effects (e.g., survival, growth, or reproduction), which can be liked to significant consequences at the population level. Benchmark distributions have the advantage of allowing the user to accurately match benchmarks with the probabilities of an adverse effect occurring. To date, we have applied the methodology to derive benchmarks for a number of contaminants including PCBs and mercury. The results of the benchmark development will include discussion of the derivation process in terms of data needs, analysis, shortcomings, comments, and utility.

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TOXIC MODES OF ACTION AND MUTALISTIC RESPONSES OF TEST ORGANISMS

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Responses of test organisms to complex mixtures of chemical contaminants and water or sediment conditions that may influence test organism survival can be additive, less than additive, antagonistic or synergistic in nature. In the most common case the contribution of toxicants with the same or similar mode of toxic action (e.g., acute responses to PAH compounds) have been shown to be additive. A summation of the total number of toxic units associated with PAH compounds is additive and the resultant toxicity of the combined compounds increases with additional complexity of the mixture. This apparent additivity is not observed with the more carcinogenic PAH compounds. In fact, it has been demonstrated by a 'top down approach' that the carcinogenic potential of benzo-a-pyrene decreases as the complexity of the PAH mixture increases. Additionally, it has been determined that complex mixtures of chemicals with different modes of toxic action often show greater effects than would be predicted based on the combined toxic units of the individual components. A survey of literature demonstrates varying modes of toxic action; how to evaluate potential combined interactions of chemicals with different modes of action will be presented and discussed in order to stimulate further research into the area of research on toxic modes of action and how best to predict combined toxicity of various contaminant classes.

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USING FIELD EXPERIMENTS TO DETERMINE CRITICAL BODY RESIDUE RELATIONSHIPS AND MODE OF ACTION

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There is a tendency among many of those working on aquatic toxicity problems to foster and perpetuate the myth that definitive answers can only be provided by laboratory experiments. Although these expectations might be applied to developing tissue residue effects data and identifying mode of action, the combination of Acontrolled@ field experiments, tissue chemistry and biomarkers can be used in combination with more traditional approaches to provide important insights into what actually happens under environmentally realistic conditions. Appreciation of these field approaches has been enhanced by the development of ecotoxicology as a science and refinement of ecological risk assessment protocols which have placed equal emphasis on characterizing exposure and effects. Moreover, consensus-based, standardized protocols have been developed for a number of these field approaches. Recently, we have shown how caged bivalves have been used to predict critical copper body residues for effects on survival, growth, and reproduction using field data. This represents the first step in establishing more defined links between tissue chemistry and effects using field data. We have also shown that endocrine disruption, based on a vitellin assay in caged and indigenous bivalve populations, has been linked to chemicals associated with effluents from both municipal treatment facilities and pulp and paper mills. More importantly, it is possible to use this biomarker to distinguish between estrogenic and androgenic effects and establish mode of action even before the chemical associated with those effects has been identified. Relationships that have been established using ecological risk assessment-based monitoring demonstrate how field experiments help characterize exposure and effects and establish causality. Examples will be provided to demonstrate how this approach can be used to enhance traditional monitoring and assessment approaches.

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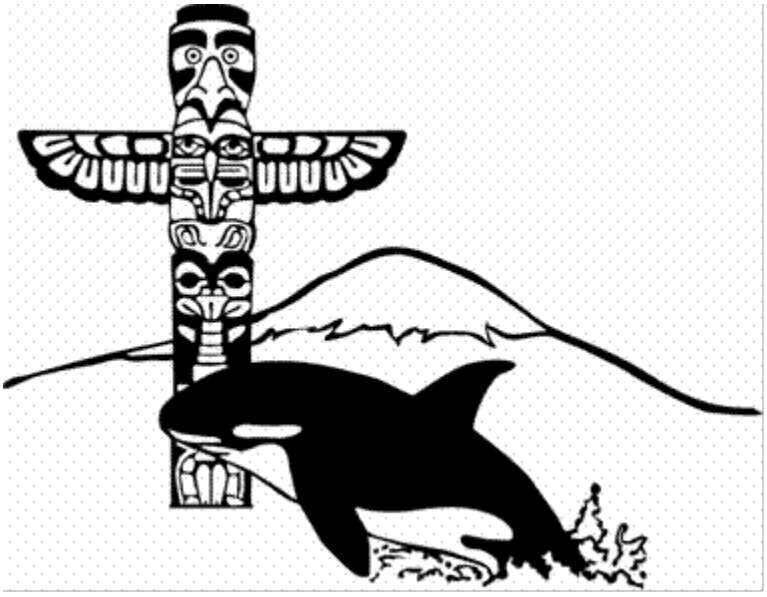

Poster Presentation Abstracts

APPLICATION OF LIFE CYCLE ASSESSMENT TO WASTEWATER TREATMENT SYSTEMS IN A PETROCHEMICAL INDUSTRY

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Life Cycle Assessment (LCA) is an integrated step-wise technique to evaluate the environmental aspects and potential impacts associated with a product, process or service. It has been used extensively by manufacturing industries to reduce ecological toxicity, resource consumption, adverse impacts to environment and to human, as well as to assess productivity and efficiencies in the manufacturing process or to improve confidence of stakeholders. Previous studies have been conducted that focus on utilizing LCA approach to evaluate the environmental impacts of the product, process or service and without necessarily using eco-toxicological parameters. The purpose of this study will be to use the LCA method to evaluate several wastewater treatment processes at a chemical industry to compare potential impacts by using eco-toxicological parameters. Similar to Ecological Risk Assessment, LCA also has a framework of steps or stages that consist of the following: 1) goal definition & scoping, 2) inventory analysis, 3) impact assessment, and 4) interpretation. LCA therefore enables the quantitative evaluation and comparison of a variety of processes and productions based on various types of data collected along the life cycle of the product, process or service. These results can be used in turn to reduce manufacturing and wastewater treatment inefficiencies, as well as significantly decrease potential impacts to the environment. Due to the large variety of products and processes that are manufactured, there are numerous types of wastewaters generated, depending on the types of treatments used in the manufacturing process. The objective of this study will be to 1) create an LCA model that will include eco-toxicological parameters and 2) compare the data from the wastewater treatment processes of the industry with and without a recycling component. The results of this study will demonstrate the applicability of LCA to quantitatively evaluate eco-toxicological parameters in wastewater treatment processes.

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APPLICATIONS OF RESPIROMETRY FOR DETERMINATION OF TOXICITY TO ACTIVATED SLUDGE AND IMPLICATIONS FOR 2^O WASTEWATER TREATMENT PLANTS

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Wastewater treatment plants (WWTP) often utilize secondary (2^o) aerobic treatment to remove nutrients and dissolved organic compounds from the waste stream. This is achieved via the metabolic activity of activated sludge. Respirometry is a useful tool for quantifying the metabolism of the microbial biomass in terms of the oxygen uptake rate measured as the change of mg O_2 / L biomass / hr. Respirometry can give important information regarding potentially toxic influents and aid in WWTP optimization. This work uses the Arthur Respirometer to quantify the potential toxicity of triazol, an anticorrosion agent often found in aircraft deicing agents, to activated sludge in terms of an inhibitory concentration (IC_{xx}). Currently, the precision of the Arthur Respirometer has been verified and positive controls (anhydrous $CuSO_4$, EtOH and Phenol), which verify IC_x calculations, are being prepared. Various concentrations of triazol will be administered to the activated sludge. If triazol elicits a toxic response, a dose/ response range that gives an IC_{20}-IC_{50} will be determined and run multiple times to give statistical validity and the slope of the dose/ response curve.

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ASSESSING THE OCCURRENCE OF PESTICIDES AND VOCS IN GROUND WATER IN A HUMAN-HEALTH CONTEXT

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A State-scale pilot effort was conducted to enhance the understanding of the potential human-health importance of ground-water-quality data collected as part of the U.S. Geological Survey's National Water-Quality Assessment Program. Ground-water samples collected from 30 public-supply, 82 domestic, and 108 monitoring wells in New Jersey were analyzed for 97 pesticides and 85 volatile organic compounds (VOCs). For 32 of the 98 compounds detected, no U.S. Environmental Protection Agency (USEPA) drinking-water standards or guidelines exist. However, new Health-Based Screening Levels (HBSLs) were calculated for 12 of these 32 compounds using USEPA toxicity values and USEPA Office of Water methodologies. The new HBSLs increased the number of detected compounds with human-health benchmarks from 66 to 78 (of 98), thereby improving the basis for determining the potential importance of the water-quality data to human health. Measured concentrations of regulated and unregulated compounds were compared to Maximum Contaminant Levels (MCLs) and HBSLs, respectively. Benchmark Quotients (BQs), defined as ratios of measured concentrations to MCLs or HBSLs, were calculated for compounds detected in each well type. Compounds were identified as being of potential human-health concern if maximum detected concentrations were within a factor of 10 of the associated MCL or HBSL (that is, BQmax $\geq$ 0.1) in any well type. Most (78 of 98) pesticides and VOCs were detected at concentrations well below these levels (BQmax < 0.1). However, BQmax was greater than or equal to 0.1 (range, 0.1 - 3,000) for 6 pesticides and 14 VOCs. Of these 20 compounds, 4 compounds (dieldrin, 1,2-dibromoethane, tetrachloroethylene, and trichloroethylene) (1) had measured concentrations that met or exceeded MCLs or HBSLs and (2) were detected in more than 10 percent of samples collected from raw ground water used as sources of drinking water (public-supply and (or) domestic wells), and therefore are the most relevant to human health.

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AVOIDANCE OF HERBICIDE TANK MIXES BY JUVENILE SALMONIDS

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Herbicides are frequently used to control exotic aquatic plants but utilization has been hampered by concerns over non-target toxicity of active ingredients. Adjuvants, chemicals added to tank mixes to make active ingredients more effective, can be orders of magnitude more toxic than the herbicidal ingredients. This is particularly true for the surfactant R-11®, a commonly used surfactant with the aquatic herbicide Rodeo® (ai: glyphosate). Data on avoidance of tank mixes by salmonids are lacking for herbicides and surfactants with existing aquatic labels (Rodeo + R-11) and those being considered for use (ARSENAL® + HASTEN®). The ability of non-targets to avoid tank mixes needs to be assessed. Avoidance tests were conducted in a 'Y' maze. Thirty juvenile rainbow trout (*Oncorhynchus mykiss*) were placed in the base and given 30 min to acclimate. Following acclimation, fish were exposed to the chemical gradient for an hour, after which the location of fish recorded. A previous test with clean freshwater indicated that the trout did not prefer one side of the apparatus to the other. Positive control tests with copper were also conducted to ensure avoidance could be determined using the protocol. Maximum label rates for aerial application of Rodeo (20 gal/ac) and ARSENAL (30 gal/ac) with the surfactants were used to determine tank mixes. Test concentrations selected were 0.1x, 1x and 10x the median lethal concentrations (LC50) for the surfactants. Five trials were conducted for each concentration. Chi-square analyses were used to determine preference or avoidance. Significant avoidance was observed at the LC50 for R-11 with Rodeo and the LC50 and 1x and 10x concentration of the ARSENAL + HASTEN tank mix. Mortality at the 10x concentration of Rodeo + R-11 may have affected the results.

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BIOACCUMULATION AND CRITICAL BODY RESIDUES OF EXPLOSIVE COMPOUNDS IN AQUATIC INVERTEBRATES AND FISH

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In an effort to better understand the potential environmental risks associated with underwater ordnance we evaluated the toxicity, bioaccumulation and critical body residue (CBR) of the explosives TNT, RDX and HMX, and the TNT metabolites 2ADNT and 2,4DANT using the amphipods *Leptocheirus plumulosus* (LP), *Eohaustorius estuarius* (EE), *Rhepoxynius abronius* (RA), the midge *Chironomus tentans* (CT), the worm *Tubifex tubifex* (TT) and the minnow *Cyprinodon variegatus* (CV). TNT was efficiently biotransformed in all species; therefore bioaccumulation is expressed as the sum molar concentration of TNT, ADNTs and DANTs (SumTNT). For CV, the uptake and elimination rates were much higher for TNT than for RDX, resulting in steady-state for SumTNT within hours. The BCF for TNT, 2ADNT, 2,4DANT, RDX and HMX determined for CV and CT ranged from 0.4 to 6.3 confirming their low bioaccumulative potential, expected due to their low hydrophobicity. Body residues are expressed as nmol/kg and represent the LOEC. For CV, the CBR was higher for 2ADNT (268) and DANT (284) than for SumTNT (19), suggesting much higher potency for TNT in this species. For CT, CBRs were similar for SumTNT (20) and 2ADNT (17) and 2,4DANT (41), suggesting equipotency. The toxicity of TNT to marine amphipods was compared using spiked sandy sediments. The CBR for SumTNT was 13 for LP, 34 for EE, and 4 for RA, indicating similar sensitivity across related of species. For RDX, the CBR was 40 for CV but no mortality of EE occurred at a higher body burden (150). No mortality of CV at a HMX body burden of 4 in saturated water. CBRs for explosives were over an order of magnitude lower than lethal residues associated with nonspecific (narcotic) modes of toxic action. Bioaccumulation of TNT was also determined using radioactivity as a surrogate for parent and metabolite compounds, CBRs for ^{14}C-labeled molecules were approximately 10-100 times higher than those for SumTNT. The toxicological roles of the large fraction of TNT that enters the organism and transforms to non-identified, solvent extractable or tissue-bound compounds remain unknown.

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CHEMICAL CONTAMINATION IN THE SAN JUAN ISLANDS, EASTERN STRAIT OF JUAN DE FUCA, AND ADMIRALTY INLET

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The quality of sediment is monitored by the Department of Ecology annually as a component of the Puget Sound Ambient Monitoring Program (PSAMP). The primary objectives of this monitoring program are to quantify the spatial extent and geographic patterns in degraded sediment quality. Using the Sediment Quality Triad approach, data are evaluated on the chemical contamination and toxicity of sediments along with analyses of the composition of the resident infaunal benthos. During June of 2002 and 2003, Ecology collected 81 samples in the bays and inlets of the San Juan Islands, eastern Strait of Juan de Fuca, and Admiralty Inlet. A stratified-random method was used to identify sampling locations to ensure a lack of bias. Included in the analyses, was a quantitation of 118 potentially toxic chemicals. The results of the chemical analyses were compiled from the two survey periods and merged to identify spatial patterns and spatial extent of chemical contamination in these three regions. Comparisons between chemical concentrations and Washington State standards showed that the majority of the samples were not contaminated. However, some samples were contaminated with mixtures of chemicals, the composition of which varied from site to site.

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CHERRY POINT REGIONAL RISK ASSESSMENT: A RISK MANAGEMENT TOOL

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An interactive risk management tool was developed based upon the regional ecological risk assessment conducted by Hart Hayes and Landis (2003) for the Cherry Point region in northern Whatcom County, WA. The regional ecological risk assessment for the Cherry Point region determined the following: 1) the major sources of risk are commercial and recreational vessel traffic, upland urban and agricultural land use and shoreline recreational activities, 2) the biological endpoints most at risk are the great blue heron and juvenile Dungeness crab, 3) the habitats most at risk are the sandy inter-tidal, eelgrass and macro-algae habitats, and 4) the sub-regions where the greatest risk occur were Lummi Bay, Drayton Harbor and Cherry Point. The purpose of the risk management tool is to allow risk and natural resource managers to assess the outcomes of adjusting the habitat and stressor rank values and the exposure and effects filter values used to quantify the relative risk in sub-regions, relative contribution of risk from sources and the risk to assessment endpoints within the Cherry Point region. The results of the rank-value adjustments can be displayed and compared to the original relative risk scores. The tool will be useful for testing alternative management scenarios regarding ecological risk in the Cherry Point region.

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CLOSED ECOLOGICAL SYSTEMS

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Both freshwater and marine ecosystems consisting of phytoplankton, zooplankton, and microbial populations have been maintained without atmospheric exchange for greater than a month, and in some cases, years. Freshwater systems were initiated with a chemically defined algal media and a commercial "instant freshwater," Kent Cichlid salts. This mixed medium was more successful in sustaining *Daphnia magna* populations than the chemically defined medium (T82) plus $NaHCO_3$. We are now investigating the role of trace metals, buffers, and alternative sources of carbon. These ecosystems displayed the expected ecological interactions of a phytoplankton bloom followed by a grazer bloom accompanied by phytoplankton depletion, and a longer period of reduced populations of algae and *Daphnia*. *Daphnia* populations were enumerated by digital photography. Marine systems were initiated with nutrient enriched seawater, mixed phytoplankton and the copepod *Tigriopus. Tigriopus* populations have been maintained for more than a year in a glass carboy sealed with a rubber stopper, and for more than 4 years in 700 ml tissue-culture-flasks. Modifications of these systems are being used as Undergraduate Research Projects. Closed Ecological Systems can demonstrate the degree to which an ecosystem can maintain an environment capable of supporting aerobic organisms, without subsidy from external ecosystems. In addition to displaying the properties of ecosystems, closed systems can be used to study the effects of toxic chemicals with volatile forms.

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DISTRIBUTION OF SOIL BIOAVAILABILITY PARAMETERS IN EUROPE

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As part of an effort to develop a predictive model of bioavailability and toxicity of copper to terrestrial organisms, 19 soils from 9 European countries were collected in 2002 and analyzed for parameters important in controlling bioavailability. Although these soils were found to have a wide range of values for each parameter, it was unknown if they were fully representative of the soil types found throughout Europe. Therefore, the primary objective of this research was to determine if the soils collected in 2002 were inclusive of the natural range of European soil bioavailability parameters, as was required for the development of a valuable regional model. The following steps were taken to fulfill this objective. First, spatially explicit data describing European soil bioavailability parameters were obtained. The most attractive datasets covered a large area and contained information on the spatial distribution of some, if not all, of the following soil properties: pH, clay-silt-sand content, carbon and nitrogen content, calcium carbonate content, metals concentrations, and soil saturation. If data were distinguished by depth, the top 20-30 cm of the soil profiles were summarized to correlate with the depth of the soils collected in 2002. Second, the statistical distributions of each parameter were defined by determining the distribution type, and calculating the median, 90th and 10th percentiles. Third, the distributions of the European soil bioavailability parameters were compared with the soils collected in 2002. In general, the collected soils were in close agreement with the natural distributions of several soil parameters. These results suggest that the soils collected in 2002 are representative of the natural range of European soil conditions and are appropriate for use in model development.

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EFFECTS OF AQUATIC HERBICIDES ON SMOLTIFICATION IN JUVENILE COHO SALMON

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Utilization of herbicides to control aquatic weeds has been hampered by court injunctions directed at the non-target toxicity of active herbicidal ingredients. Adequate data on non-target toxicity of aquatic herbicides to aquatic resources are lacking, threatening the permitting process and success of control strategies. Declines in several species/stocks of salmon and efforts to restore these populations heighten concerns. Our objective was to determine if label application rates for three commonly used aquatic herbicides impair smoltification in juvenile Pacific salmon, using coho (*Oncorhynchus kisutch*) as a model. The herbicides were Sonar®AS (active ingredient: fluridone; 10, 90 ppb), REWARD® (active ingredient: diquat dibromide; 0.34, 1.37 ppm), and RENOVATE® (active ingredient:triclopyr; 0.75, 2.50 ppm). Fish were exposed to the herbicides or negative control for 96 h under static conditions and then transferred directly into flowing seawater for 14 days. Five fish per tank were sacrificed after exposure to the chemicals, and after 1, 7 and 14 d in seawater. Endpoints were survival, body weight and fork length, muscle water content, hepatosomatic index, plasma sodium and chloride concentrations, gill ATPase, and gill histology. Actual concentrations were similar to nominal with the exception of fluridone (1, 10 ppb) due to a calculation error. All fish survived the chemical exposure and, with few exceptions, endpoints before and after the seawater exposure were similar to controls. Results suggest the herbicides are unlikely to affect seawater adaptation in free-living juvenile Pacific salmon.

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EVALUATION OF BIODIESEL (SOY METHYL-ESTER) TOXICITY TO SOILS USING THE LUMBRICID EARTHWORM *EISENIA FETIDA*

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The development of alternative fuels has gained momentum, particularly in the last decade, due to rising petroleum costs and a society gradually becoming more environmentally conscientious. Although this new stock of fuels may provide an immediate alternative to petroleum products, the full ramifications of their use should be examined and scrutinized in order to increase their viability as "alternatives". Biodiesel is one such alternative fuel comprised of methyl- or ethyl- esters. Soy methyl-ester is the most prevalent type of biodiesel available for commercial use; it is produced primarily by the National Biodiesel Board and is broadly distributed. Prior tests indicate biodiesel to be significantly less toxic than petroleum-derived diesel, but biodiesel ecotoxicity alone has not been studied extensively. The establishment of this toxicological ranking is important if ever biodiesel is to be widely used. Earthworm bioassays can provide an effective, methodically homogenous evaluation of a substance's toxicity. This assay will employ methods indicated by ASTM 2000, E1676-97. Two trials performed in triplicate will ensure adequate statistical power in producing a concentration-response curve for earthworm (*Eisenia fetida*) mortality to soy methyl-ester biodiesel.

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EVALUATION OF ENDOCRINE DISRUPTERS IN AMBIENT WATERS OF KING COUNTY WASHINGTON

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Recently, there has been an increased public concern regarding the potential ecological effects associated with exposure to chemicals classified as "endocrine disrupters". In part, this concern originated from the finding that exposure to some synthetic chemicals in the environment was associated with adverse reproductive and developmental effects in fish and wildlife. These chemicals are often pervasive and widely distributed in the environment. Discharge of sewage effluent has been identified as a source of some compounds classified as endocrine disrupters. King County, in Washington State, currently operates two sewage treatment facilities serving the greater Seattle Metropolitan area with a combined average annual discharge of 200 MGD into Puget Sound. Washington Department of Fish and Wildlife and NOAA recently found that male fish collected in Elliott Bay, an urban bay in Puget Sound adjacent to Seattle, had greater vitellogenin levels than fish collected from other urban areas the Sound. As such, endocrine disrupters are an emerging concern for King County. As a result, the county has initiated a pilot monitoring program to analyze a select number of chemicals classified as endocrine disrupters in fresh and marine waters. Both traditional chemical analysis methods and the Enzyme-Linked Immunosorbent Assay (ELISA) were used to analyze water samples collected from lakes, streams and the marine environment throughout King County. In general, most compounds analyzed were not detected, or were present at very low levels. Data and sampling and analysis methods from this recent monitoring program will be presented.

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EXPOSURE OF SALMONIDS TO CARBARYL FOLLOWING APPLICATIONS TO CONTROL BURROWING SHRIMP

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Efforts to restrict the use of carbaryl to control burrowing shrimp in Washington State have been in part driven by concerns over effects on salmonids documented in laboratory studies, and not actual exposure during operational applications. Data on the actual exposure of salmonids to carbaryl are lacking, threatening the permitting process and control operations. We studied the use of treated areas by salmonids. Use of the water column above oyster beds by salmonids was determined on the first daylight high tide at each of three oyster beds (10-25 ac) preceeding two carbaryl spray events (2 and 14 July 2003), and during each of the three subsequent daylight high tides (ca. 6, 30 and 54 h posttreatment). Sampling methods included use of a two-boat trawl net and gill nets. Juvenile chinook were the only salmonid captured before and after treatment. Concentrations of carbaryl before each of the two spray events were < 1 ppb (ND, below detection limits); concentrations after spray ranged from ND to 11.3 ppb. Brain AChE assays and chemical analyses of stomach contents are underway. Results to date suggest (1) juvenile chinook are the only salmonid within the water column above oyster beds before and after carbaryl applications, (2) juvenile chinook are exposed to low levels of carbaryl and its breakdown product 1-naphthol, and (3) concentrations of both chemicals are 2-3 orders of magnitude below levels lethal to chinook ($LC50_{carbaryl}$ = 2,400 ppb, $LC50_{1\text{-}naphthol}$ = 1,400 ppb [rainbow trout]). Results of brain AChE assays will be presented.

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GENERAL CONCEPTUAL MODEL FOR THE REGIONAL AND LANDSCAPE RISK ASSESSMENT FOR INVASIVE SPECIES

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Most studies of the risk assessment of invasive or non-indigenous species have focused on hazard and exposure, not on risk as currently understood or at a regional scale. At a regional scale there are always multiple other stressors, a variety of environmental gradients, spatial heterogeneity and a wide variety of valued characteristics. These factors make estimating risk a daunting but achievable task. We use two approaches to address the estimate of risks due to invasive species. First we use simple three patch models as developed by Spromberg et al to examine the types of interactions between a native species, the invasive and the presence of a toxicant or other stressor. Second, in order to estimate regional risk due to invasive species at a regional scale we use spatially explicit conceptual models based upon the source-stressor-habitat-impact relative risk model as formulated by Landis and Wiegers These conceptual models for invasive species have been applied to Cherry Point, Washington and to the northern part of the Chesapeake Bay. Our efforts have demonstrated a disconnect between the data needs for estimating regional scale risks available data. Modeling have demonstrated that data on dynamics of the system are critical in understanding the probabilities of effect and where even in simple three patch models. The process of developing conceptual models for the two regions has demonstrated that little or no data on the dynamics of the valued species are available, and typically concurrent data on stressors also are not available. The result is that uncertainty in the risk estimates is high. Our efforts do point to specific research and monitoring requirements that will reduce uncertainty and make the risk assessment of invasives routine.

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IDENTIFICATION OF CONTAMINATED SEDIMENTS IN PUGET SOUND BY USE OF A BIOMARKER ASSAY

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A biomarker assay based on a human reporter gene system (EPA method 4425), used on over 1300 NOAA samples from all coasts, has proven effective in understanding the distribution of contaminated sediments in Puget Sound. Sediment collections (100 from each region) and subsequent analyses in northern (1997), central (1998) and southern (1999) Puget Sound were conducted by the Washington Department of Ecology (WDOE) and the National Oceanic and Atmospheric Administration (NOAA). Samples were analyzed for chemical contaminants, and evaluated for toxicity by amphipod toxicity, sea urchin fertilization, MicroTox, and Method 4425. In addition, the WDOE analyzed the benthic community structure in these 300 samples. The mean EPA Method 4425 responses from sediment extracts (n = 100 in each region) increased from 11.1 to 37.5 to 52.8 µg/g benzo[a]pyrene equivalents (B[a]PEq), as station locations moved from strata in the north to those in the south. There were 33 central and 22 southern stations in the Sound, where B[a]PEq values were above the 32 µg/g threshold where biological effects are possible. Stations with 60 µg/g B[a]PEq, where degraded benthic communities have been described, were 19 and 14 in the central and south Sound, respectively. Method 4425 results were highly correlated (r2 = 0.7-0.87) with subsequent analyses of total PAHs. The only 4 high level stations in the north were in Everett Harbor, and were later found to contain dioxins. The best concordance for biological tests was between Method 4425 and pore water effects on sea urchin fertilization. While Method 4425 identified 55 stations as potentially toxic, and 33 stations as likely degraded, not one sample produced < 80% survival in toxicity tests with the amphipod, *Ampelisca abdita*. As compared to other harbors and coastal regions evaluated in NOAA investigations, the majority of Puget Sound contained surficial sediments of relatively high quality.

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INTERACTIONS OF MICROBIAL EXOPOLYSACCHARIDES WITH PLUTONIUM IN THE SUBSURFACE ENVIRONMENT.

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Metals such as plutonium and uranium can be present in the environment in association with organic ligands. Among the organic ligands that may be associated with these metals are low molecular weight hydroxycarboxylic acids potentially of microbial origin and microbially produced exopolysaccharides, a higher-molecular weight fraction. These complex organic ligands can bind to and form stable metal-organic species which may be more soluble and, as a result, more mobile. The formation of these metal-organic species, therefore, may promote migration of metal species in the subsurface environment. Several organic complexants have been investigated in this research, including citric acid, and galacturonic acid, which can be used as simple model ligands. In addition the complexation of Pu and U to exopolysaccharides isolated from three species of environmentally relevant bacteria were investigated. Metal-organic ligand species were prepared at $Pu(IV)_T$ concentrations ranging from 10^{-10}-10^{-12} M and U(VI) concentrations ranging from 10^{-4} to 10^{-6} M. The stability constants were determined using an ion-exchange/ligand competition technique in conjunction with FITEQL, a computer program that can be used to determine "best-fit" equilibrium constants for reactions that are postulated to explain the experimental data. The methods used in this research to investigate metal-ion binding to complex organic ligands represent novel techniques that have been developed in our laboratories. The results thus far indicate that both Pu and U will be strongly complexed by model and naturally occurring organic ligands in the environment and should be considered in an assessment of the fate and transport, and in risk assessment models of metals such as Pu and U.

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POLYBROMINATED DIPHENYL ETHER RESIDUE ANALYSIS METHOD FOR FISH TISSUES FROM REMOTE, HIGH ELEVATION ECOSYSTEMS

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Polybrominated Diphenyl Ethers (PBDEs) are a class of compounds used as flame retardants in synthetic materials, and have shown persistent, bioaccumulative, and toxic properties. These compounds have been measured in urban and rural areas around the globe at increasing levels of concentration over the past 20 years. A method was developed for congener specific analysis of PBDEs and other anthropogenic semi-volatile organic compounds (SVOCs) in fish tissues from remote, high elevation ecosystems. A benchtop gas chromatographic mass spectrometry (GC-MS) method, optimized for molecular ion sensitivity of each PBDE congener, was developed. This method utilized isotope labeled recovery standards for more accurate quantitation in environmental extracts. Fish tissues were cold homogenized, dried and ground, and solvent extracted. Percent lipid was determined and lipids were removed from the extract by size –exclusion chromatography. Sample extracts underwent further purification techniques including silica adsorption chromatography. Extracts were reduced to < 300 μL, spiked with isotope labeled internal standards, and analyzed by both electron impact (EI) and electron capture negative ionization (ECNI) GC-MS. This fish contaminant residue method achieved good recoveries for ~100 SVOCs (including PBDEs, PCBs, organochlorines, and PAHs) permitting analysis of fish tissues from 'pristine' ecosystems for low level contaminant concentration determinations.

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SEDIMENT METALS VERIFICATION STUDY FOR SINCLAIR AND DYES INLETS, WASHINGTON

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This study was conducted to address metal contaminants listed on the State of Washington's 1998 303(d) list of impaired waters, specifically those waters located in Sinclair and Dyes Inlets near Bremerton, Washington. Hydrodynamic modeling has shown significant water exchange and sediment transport between Sinclair and Dyes Inlets. The primary objectives of the study were to document the current sediment metal concentrations in Sinclair and Dyes Inlets and evaluate whether the sediment concentrations exceed sediment quality criteria for metals. Significant cleanup and source control activities have been conducted in the Inlets since the data supporting the 1998 303(d) listings were collected. Another objective was to provide metals data to support contaminant loading and transport modeling throughout Sinclair and Dyes Inlets. A total of 160 sediment samples from Sinclair Inlet, Dyes Inlet, Port Orchard Passage, and Rich Passage were screened for copper, lead, and zinc using X-Ray Fluorescence (XRF). The XRF results for these metals correlate well with standard analytical method (ICP-MS) results. About 40 samples (25%) were selected for confirmatory metals analysis by ICP-MS. Most of the confirmatory samples were located in Sinclair Inlet, where metal concentrations were historically highest. The study outcomes are expected to help prioritize management actions if sediment remains a source of sediment quality impairment.

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SEDIMENT TOXICITY IN THE SAN JUAN ISLANDS, EASTERN STRAIT OF JUAN DE FUCA, AND ADMIRALTY INLET

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The quality of sediment is monitored by the Department of Ecology annually as a component of the Puget Sound Ambient Monitoring Program (PSAMP). The primary objectives of this monitoring program are to quantify the spatial extent and geographic patterns in degraded sediment quality. Using the Sediment Quality Triad approach, data are evaluated on the chemical contamination and toxicity of sediments along with analyses of the composition of the resident infaunal benthos. During June of 2002 and 2003, Ecology collected 81 samples in the bays and inlets of the San Juan Islands, eastern Strait of Juan de Fuca, and Admiralty Inlet. A stratified-random method was used to identify sampling locations to ensure a lack of bias. Included in the analyses, was a battery of four laboratory toxicity tests. Results from the two survey periods have been compiled and merged. A small minority of samples was toxic in all four tests and there was relatively little overlap or agreement in results among the four tests. These observations suggest that the quality of sediments in these three regions was comparable to that of some of the least degraded regions previously studied by this program in Puget Sound.

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THE INFLUENCE OF WATER HARDNESS ON URANIUM TOXICITY TO FRESHWATER ORGANISMS

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The influence of water hardness on the toxicity of uranium to six freshwater species was investigated in waters of low alkalinity (5-14 mg/L as $CaCO_3$) and various hardnesses (5 to 252 mg/L as $CaCO_3$). Tests included 7-d survival and growth tests with fathead minnows (*Pimephales promelas*), 96-h acute lethality and 30- and 31-d embryo/alevin tests with rainbow trout (*Oncorhynchus mykiss*), 3-brood survival and reproduction tests with *Ceriodaphnia dubia*, 14-d water-only survival and growth tests with *Hyalella azteca*, 72-h growth inhibition tests with *Selenastrum capricornutum*, and 7-d static growth inhibition testswith *Lemna minor*. Environment Canada test methods were used with modifications to the recommended reconstituted water recipes in order to achieve low alkalinity and Ca/Mg molar ratios of 1:1 (or 3:1 for *H. azteca*). In general, *H. azteca*, *C. dubia*, and *S. capricornutum* were the most sensitive species to uranium with LC/IC50 estimates (based on mortality, reproduction, and growth inhibition, respectively) that ranged between 0.017 and 0.34 mg U/L. Rainbow trout alevins were the next most sensitive, with EC50 estimates (based on alevin viability) between 0.46 and 0.64 mg U/L. Survival was more sensitive than growth in the 7-d tests with fathead minnows, giving 7-d LC50 estimates of 1.5 to 2.0 mg U/L. At 96-h, survival of fathead minnows yielded LC50 estimates that were almost half that of rainbow trout fry (1.8 to 2.1 mg U/L compared to 3.8 to 4.2 mg U/L). *L. minor* was the least sensitive species tested, with IC50 estimates starting at 7.4 mg U/L (frond increase). Toxicity remained relatively constant with increasing hardness for most species. *H. azteca* was the exception, where the 14-d LC50 estimates in soft water (0.017 mg U/L) was 20 times less than that in the hardest water (0.34 mg U/L).

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THE SEDIMENT QUALITY TRIAD INDEX IN PUGET SOUND

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From 1997-1999, sediments were collected from 300 locations throughout Puget Sound as part of a joint monitoring program conducted by the Washington State Department of Ecology and the National Oceanic and Atmospheric Administration. Analyses were performed to quantify concentrations of potentially toxic chemicals, responses in laboratory toxicity tests, and the structure of benthic infauna communities in sediments. A Sediment Quality Triad Index was generated from these three data sets, and was used to classify and quantify sediment quality in six monitoring regions and five strata, and for the entire Puget Sound study area. While the majority of the Puget Sound study area had sediments of high quality (68%), 31% displayed some degree of degradation, while 1% was of lowest quality. Typically, the most highly degraded sediments are found in urban harbor areas (especially near the urban centers of Everett, Seattle, Tacoma, and Bremerton), near river mouths, and along shallow, nearshore areas. While small in area, these locations historically have represented and provided critical habitat for many species of marine biota, and degradation of sediments in these areas may have a disproportionate impact on the health of Puget Sound species. Results were also compared with data from estuaries and marine bays around the U.S. Relative to these locations, Puget Sound sediments were ranked among the least contaminated and toxic.

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THE TOXICITY OF FREE CYANIDE TO YELLOW ROCK CRAB (*CANCER IRRORATUS*) FIRST STAGE ZOEAE

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The U.S. Environmental Protection Agency (U.S. EPA) national water quality criterion for cyanide in salt water is driven by zoeal toxicity data for the yellow rock crab, *Cancer irroratus*. The reported LC50 for *C. irroratus* is six times lower than for any other marine species, and from 14 to 31 times lower than recent larval toxicity data for four other species of *Cancer* crab. This discrepancy prompted a reexamination of *C. irroratus* cyanide toxicity as part of a larger study that is re-evaluating the national Ambient Water Quality Criteria (AWQC) for cyanide. Flow-through cyanide acute toxicity tests were undertaken with the first stage zoeae of *C. irroratus*. Free cyanide, measured by the microdiffusion method, was measured daily in all test concentrations. Based on the tests reported here, species mean acute toxicity values for *C. irroratus* may be approximately 10 times higher than that presently used in the U.S. EPA water quality criteria document. If further confirmed experimentally, use of these revised data may suggest that the current AWQC for marine species is overly protective of the acute toxicity of cyanide to marine organisms.

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TOXICITY OF A PROPOSED BALLAST WATER BIOCIDE, SEAKLEEN®, TO EMBRYO AND LARVAL STAGES OF PACIFIC HERRING, *CLUPEA PALLASI*

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Widespread transport of exotic marine organisms in ship ballast water has led to development of a variety of proposed methods to treat ballast water prior to discharge. One chemical treatment proposed for use as a biocide is SeaKleen®, which is a mixture, by weight, of menadione sodium bisulfate and menadione. Experiments to test the efficacy of using SeaKleen as a ballast water biocide have been proposed for the Puget Sound region, with discharge of treated ballast water into Sound waters. However, concerns about SeaKleen's toxicity to local fauna, especially to early life stages of the Pacific herring (*Clupea pallasi*), resulted in bioassay testing of SeaKleen with embryo and early larval stages of herring. Development of Pacific herring embryo and larval testing protocols designed for use in Whole Effluent Toxicity (WET) testing programs has been underway at the Shannon Point Marine Center in Anacortes, WA to provide additional biological tools for testing effluents. Tests of SeaKleen toxicity represent the first application of the new herring test protocols developed at Shannon Point. SeaKleen toxicity test results will be presented for the following test endpoints: 18-day embryo development (normal survival, heart rate, embryo movements, time to 50% hatch, larval length at hatch), 10-day larval survival (LC50), and 10-day larval survival and growth (LC50, larval dry weight).

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TOXICITY OF RODEO® AND ARSENAL® TANK MIXESTO JUVENILE RAINBOW TROUT

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Efforts to utilize herbicides in aquatic environments have been hampered by concerns over the potential non-target effects of active ingredients. However, adjuvants (e.g., surfactants) can represent the most toxic component of herbicide tank mixes. Data on the relative toxicity of tank mixes (formulated herbicide product + adjuvants + carrier) to salmonids are lacking for herbicides with existing aquatic labels (e.g., Rodeo®) and those being considered for aquatic use (e.g., ARSENAL®). As an initial step in evaluating the toxicity of tank mixes of Rodeo® (ai=glyphosate) and ARSENAL (ai=imazypyr), we determined 96-h static LC50s for three formulated products: Rodeo 53.8% ai (782 ppm); ARSENAL Herbicide, 28.7% ai, (77,716 ppm); and ARSENAL AC, 53.1% ai, (22,305 ppm) using juvenile rainbow trout (*Oncorhynchus mykiss*, ca. 0.65 g). We then determined LC50s for tank mixes expressed as ppm surfactant for Rodeo with the surfactants R-11® (5.4 ppm) and LI 700® (23 ppm) and ARSENAL Herbicide with the surfactants HASTEN® (113 ppm) and AGRI-DEX® (479 ppm), and compared these values with LC50s for the surfactants alone (R-11: 6.0; LI 700: 17; HASTEN: 74, AGRI-DEX: 271 ppm). Results suggest (1) all three herbicide formulations have relatively low toxicity to juvenile rainbow trout, but their LC50s differ by 2 orders of magnitude, (2) toxicity of the tank mixes is driven by the surfactants and varies by >2 orders of magnitude, and (3) depending on the surfactant selected and its percentage of the tank mix, surfactants may pose a greater hazard to non-target species than the formulations tested.

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UNCERTAINTY REDUCTION THROUGH NEARSHORE HABITAT MAPPING OF CHERRY POINT, WASHINGTON

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This study will reduce uncertainty and refine risk predictions regarding eelgrass (*Zostera marina*) and macroalgae habitats that were identified in the Cherry Point regional ecological risk assessment (Hart Hayes, 2002). The risk assessment predictions, generated using the Relative Risk Model (Landis and Wiegers, 1997; Wiegers et al., 1998), identified the Cherry Point region as an area of high relative risk, and the majority of relative risk was predicted to occur in sandy, intertidal eelgrass and macroalgae habitats. Cherry Point, in northern Whatcom County, Washington has been recommended as an aquatic reserve. The area is ecologically important as habitat for a variety of species including Pacific herring and Dungeness crab. The focus of this study is to identify, quantify, and map the distribution of eelgrass and macroalgae habitats within the Cherry Point region during the summer of 2000. Eelgrass and associated macroalgae vegetation will be classified and mapped at a scale of 1:4800 based on full, true, color aerial photographs. The aerial photographs have high spatial resolution and will be scanned and rectified to digital ortho data for on-screen delineation of target features. Habitat and vegetation units of approximately 1000 m^2 will be delineated based on units that appear to be homogeneous with respect to color and texture. Intertidal landcover classifications will be developed and will include dense eelgrass, sparse eelgrass, green algae, sparse green algae, brown algae, and sand, gravel, rock cobble, and boulder substrates. In order to quantify the effects of parameter uncertainty regarding eelgrass and macroalgae habitats I will re-assign Monte Carlo input distributions to derive new probability distributions of possible risk estimates (Hart Hayes, 2002). Upon completion, this study will provide site and resource managers, stakeholders, and regulatory agencies with useful information that will assist in management decisions of the site.

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USE OF ACOUSTIC TAGGING TO STUDY HOME RANGE AND MIGRATION OF ENGLISH SOLE (*PLEURONECTES VETULUS*) IN PUGET SOUND: APPLICATION TO MANAGEMENT OF CONTAMINATED SEDIMENTS.

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English sole are suitable as a sentinel species for studies on effects of contaminants because they are broadly distributed in benthic habitats along the Pacific Coast where both juveniles and adults can contact contaminated sediment. Tagging studies have shown that, with the exception of a winter spawning migration, adult English sole demonstrate site fidelity: tending to remain on discrete feeding grounds most of the year. Significant correlations between liver disease (and other adverse health effects) in English sole and chemical contaminants in the sediments of their capture sites further support the hypothesis of high site fidelity. However, more complete information is needed on home range and habitat utilization to adequately characterize relationships between sediment contaminant exposure and fish health. A two-year study was initiated in the summer of 2003 to document movements by adult English sole tagged with acoustic transmitters. Twenty fish trawled from Eagle Harbor, a small, contaminated bay, were implanted with transmitters and released at the site of capture. Stationary receivers, equipped with omni-directional hydrophones, were deployed to monitor fish moving in and out of the harbor. Individual fish movements within the harbor were actively tracked from a small boat using a portable receiver with both directional and omni-directional hydrophones. Ten fish (50%) left the bay and did not return (9 of the 10 left within two weeks of release). Fish that stayed in the bay were generally found near the area of capture. Another set of tagged fish will be released in April 2004 and monitored through the feeding season. These data will help refine estimates of home range, habitat use, and migration timing of English sole so we can better understand their exposure to contaminated sediments.

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USING COPPER CBRs AS A FIRST STEP TOWARD DETERMINING MODE OF ACTION IN BIVALVES

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The development and refinement of the ecological risk assessment paradigm has helped place equal emphasis on characterizing exposure and effects. This has also led to an increased appreciation for characterizing and understanding processes associated with bioaccumulation, critical body residues, and mode of toxic action. All three are essential to establishing links between exposure and effects. We have attempted to draw attention to this concept by developing the exposure-dose-response triad and suggested that the internal dose was more directly related to effects than external exposure from chemicals in water and sediment. We have also used caged bivalves to establish many of these relationships, including those for copper. It is important to note, however, that whole body CBRs only represent the first step toward establishing critical links with critical residues at receptors where effects begin to occur. Recently, we conducted caged bivalve studies that included measuring copper concentrations in individual tissues such as the gill and digestive gland to estimate exposure from water and food, respectively. While there are no perfect monitoring and assessment tools, there is both theoretical and empirical evidence that relationships between whole body tissue residues and effects provide a number of advantages over similar relationships for water and sediment. However, just as water, sediment, and tissue relationships have served as surrogates, measuring chemical burdens in individual tissues also serves as a surrogate. Ultimately, these chemical burdens must be linked with specific receptors and a specific mode of action. Currently, a biotic ligand model is being developed for bivalves using basic elements of a model developed for the fish gill. Examples will be provided to demonstrate how this approach can be used for copper and bivalves.

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WHAT'S KNOWN ABOUT POLYBROMINATED DIPHENYLETHERS IN PUGET SOUND?

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Currently little is known of the distribution of polybrominated biphenylethers (PBDEs or flame retardants) in Puget Sound. One recent study reports elevated levels of these compounds in human breast mild in the Puget Sound area, but no studies have examine the occurrence of PBDEs in the Sound's marine environment. PBDEs are similar to PCBs in their molecular structure as well as their lipid-binding properties, are presumed to pass through the food chain accumulating in the tissue of higher trophic organisms. This presentation will provide an overview of what is known about these compounds in the environment, review what research is going on in other regions of the country, and describe the Puget Sound Ambient Monitoring Program's (PSMAP) approach to identifying where these compounds may be accumulating in the Puget Sound marine environment.

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