



NOAA FISHERIES
NATIONAL MARINE FISHERIES SERVICE



Impacts of copper on the sensory biology and behavior of salmon

Nat Scholz, David Baldwin, Jenifer McIntyre¹, Jason Sandahl², Jim Meador, Dave Beauchamp¹, Jeff Jenkins², Tiffany Linbo, John Incardona, and Tracy Collier

NOAA Fisheries, Northwest Fisheries Science Center, Ecotoxicology and Environmental Fish Health Program

¹University of Washington, School of Aquatic and Fisheries Sciences

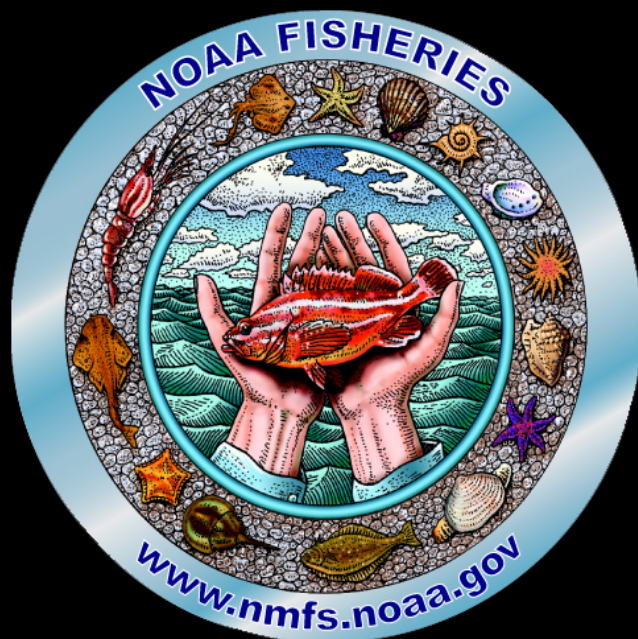
²Oregon State University, Department of Environmental and Molecular Toxicology



The NOAA Coastal Storms Program

www.csc.noaa.gov/csp/

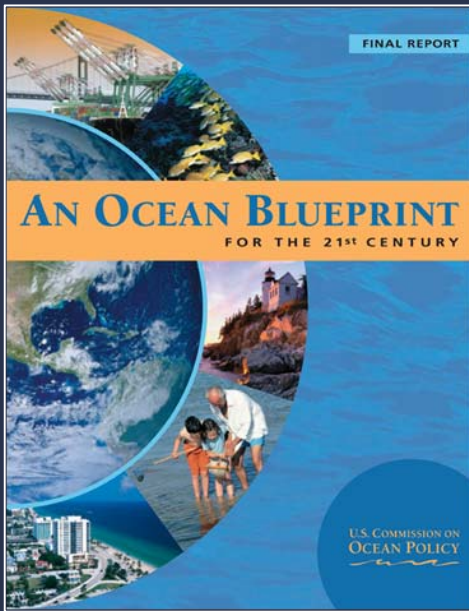
assessing the ecological impacts of storms



West Coast Salmon Recovery

*assessing the role of water quality as
a limiting factor for salmon conservation
and recovery*

Non-point source pollution

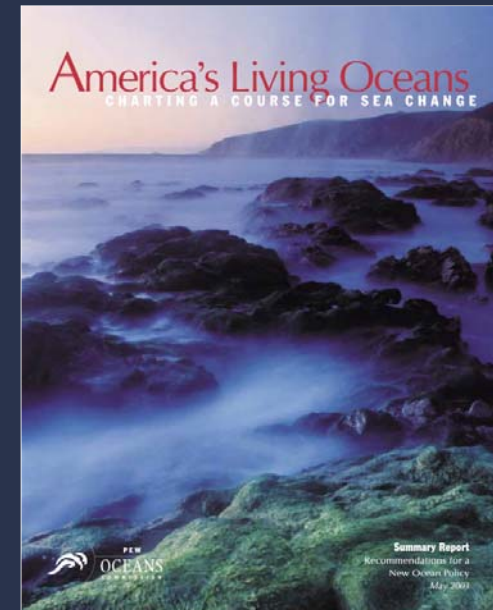


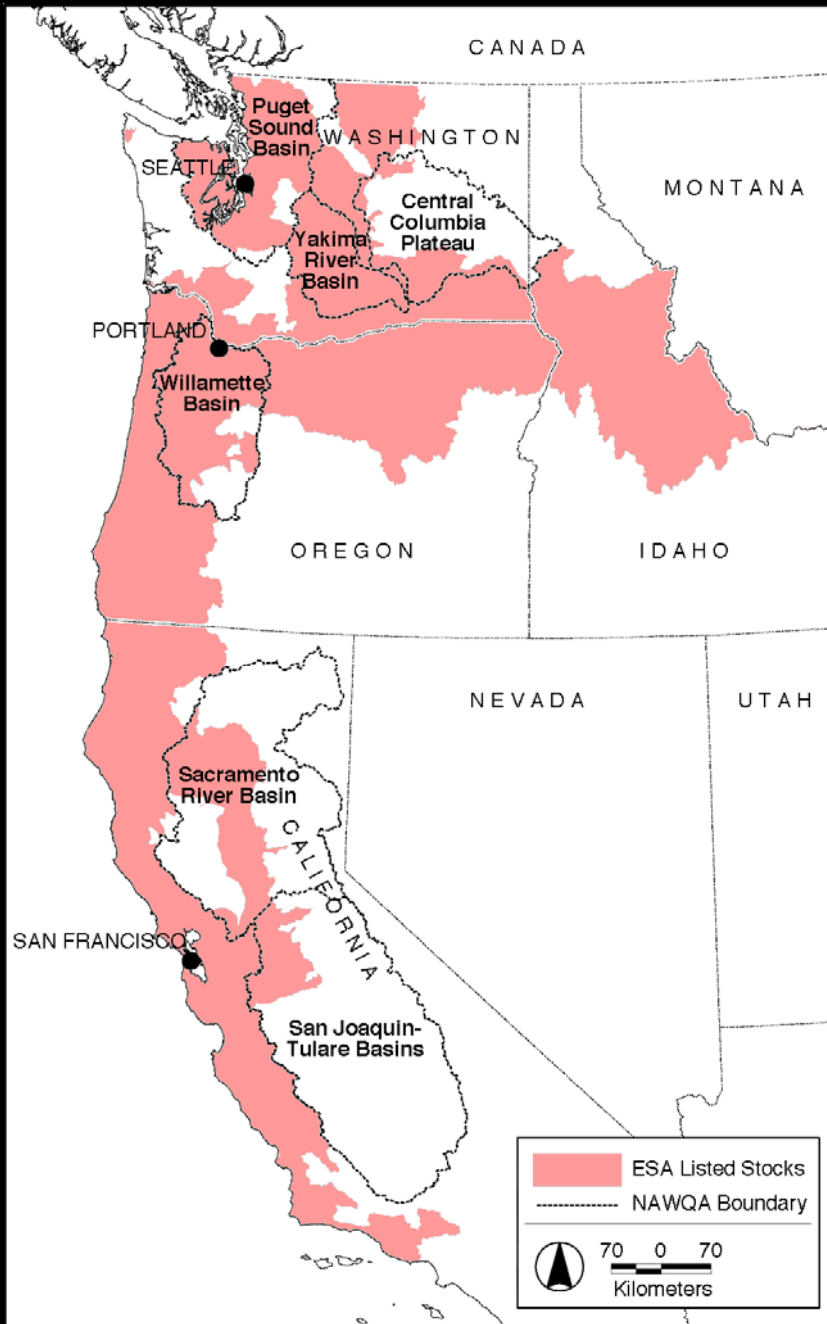
“Non-point source pollution occurs when rainfall and snowmelt wash pollutants... into our rivers and coastal waters... Our failure to manage the human activities that affect the nation’s oceans is compromising their ecological integrity, diminishing our ability to fully realize their potential, costing us jobs and revenue, threatening human health, and putting our future at risk”

- An Ocean Blueprint, Executive Summary

“Paved surfaces have created expressways for oil, grease, and toxic pollutants into coastal waters. Every eight months, nearly 11 million gallons of oil run off our streets and driveways into our waters - the equivalent of the Exxon Valdez oil spill”

- America’s Living Oceans, Executive Summary





Twenty six salmon and steelhead population segments, or **Evolutionarily Significant Units (ESUs)**, are currently listed as either threatened or endangered under the **U.S. Endangered Species Act (ESA)**.

ESA-listed species include:

- coho (*Oncorhynchus kisutch*)
- chinook (*O. tshawytscha*)
- chum (*O. keta*)
- sockeye (*O. nerka*)
- steelhead (*O. mykiss*)

Long-Term Ecosystem Response to the Exxon Valdez Oil Spill

Charles H. Peterson,^{1*} Stanley D. Rice,² Jeffrey W. Short,² Daniel Esler,³
James L. Bodkin,⁴ Brenda E. Ballachey,⁴ David B. Irons⁵

The ecosystem response to the 1989 spill of oil from the Exxon Valdez into Prince William Sound, Alaska, shows that current practices for assessing ecological risks of oil in the oceans and, by extension, other toxic sources should be changed. Previously, it was assumed that impacts to populations derive almost exclusively from acute mortality. However, in the Alaskan coastal ecosystem, unexpected persistence of toxic subsurface oil and chronic exposures, even at sublethal levels, have continued to affect wildlife. Delayed population reductions and cascades of indirect effects postponed recovery. Development of ecosystem-based toxicology is required to understand and ultimately predict chronic, delayed, and indirect long-term risks and impacts.

Before the Exxon Valdez oil spill, information available for constructing risk assessment models to predict ecologi-

Delays in recovery and emergence of long-term impacts are understood by bringing an ecosystem perspective to ecotoxicology

toxic hydrocarbons. Accordingly, mass mortalities of 1000 to 2800 sea otters (9) and unprecedented numbers of seabird deaths estimated at 250,000 (10) were documented during the days after the spill. An estimated 302 harbor seals, a short-haired marine mammal, were killed not by oiled pelage but likely from inhalation of toxic fumes leading to brain lesions, stress, and disorientation (2). Mass mortality also occurred among macroalgae and benthic invertebrates on oiled shores from a combination of chemical toxicity, smothering, and physical displacement from the habitat by pressurized wash-water applied after the spill (5, 7).

Table 1. Changing paradigms in oil ecotoxicology, moving from acute toxicity based on single species toward an ecosystem-based synthesis of short-term direct plus longer-term chronic, delayed, and indirect impacts.

Old paradigm

Oil effects occur solely through short-term (~ 4 day) exposure to water-soluble fraction (1- to 2-ringed aromatics dominate) through acute narcosis mortality at parts per million concentrations.

Emerging appreciation

Oil toxicity to fish

Long-term exposure of fish embryos to weathered oil (3- to 5-ringed PAHs) at ppb concentrations has population consequences through indirect effects on growth, deformities, and behavior with long-term consequences on mortality and reproduction.

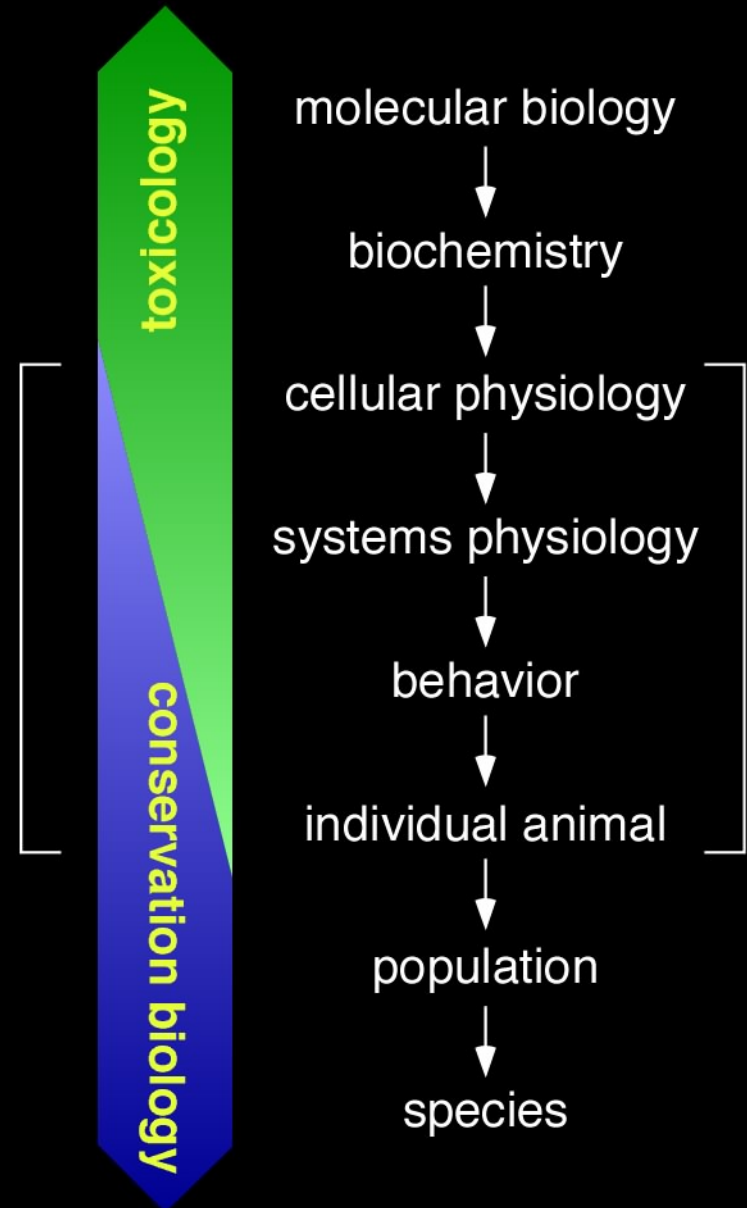
¹U.S. Department of Interior, Fish and Wildlife Service, 1011 East Tudor Road, Anchorage, AK 99503, USA.
²To whom correspondence should be addressed. E-mail: cpeters@email.unc.edu

experience high risk from floating oil (2, 6). Oil-
ing of fur or feathers causes loss of insulating
capacity and can lead to death from hypother-
mia, smothering, drowning, and ingestion of

shores where geomorphologic armoring by
boulders and cobbles inhibited disturbance
by waves (12). Some of this oil was similarly
trapped under mussel beds providing an

Evaluating the impacts of pollution on salmon health

The importance of inter-relationships between scales of biological complexity



DUMP NO WASTE

DRAINS TO LAKE



Automobiles as common sources of copper in runoff



Info-disruption: pollution and the transfer of chemical information between organisms

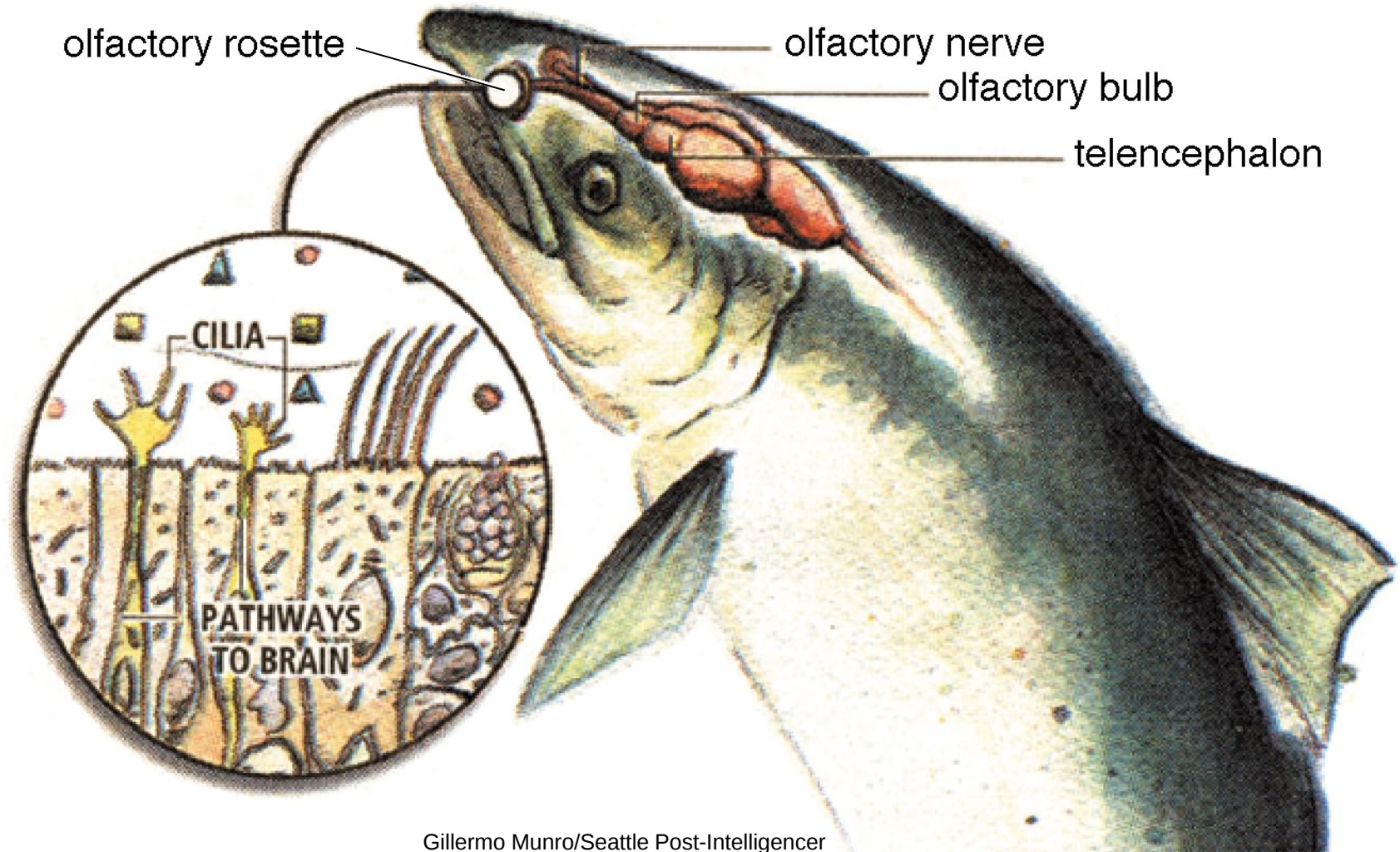
Miquel Lüring and Marten Scheffer

Aquatic Ecology & Water Quality Management Group, Department of Environmental Sciences, Wageningen University, PO Box 8080, 6700 DD Wageningen, the Netherlands

Many organisms use subtle chemical cues not only to find partners and food, but also to sense the presence of natural enemies and to avoid predation. As we discuss here, an increasing number of studies now show that low, non-toxic concentrations of chemicals, ranging from heavy metals and pesticides to seemingly harmless substances such as surfactants, can disrupt the transfer of chemical information, inducing maladaptive responses in both the signaller and the receiver. Similar to endocrine disruptors, these ‘info disruptors’ form a new class of chemical threats, which could have far-reaching implications for ecosystem functioning and conservation management.

Chemical information transfer between organisms is similar to the endocrine signalling system and there are ample data supporting the hypothesis that endocrine systems evolved from primordial exocrine (pheromonal) systems in unicellular organisms [2]. Thus, it is perhaps unsurprising that pollution can deleteriously affect information transfer at each step in the signalling pathway. Here, we discuss how chemical communication systems between organisms can be deregulated by chemical pollutants, and the implications of this deregulation. We refer to the widely chemically different substances that distort the nature and perception of the ‘smellscape’ in which organisms interact as ‘info disruptors’.

The salmon olfactory nervous system



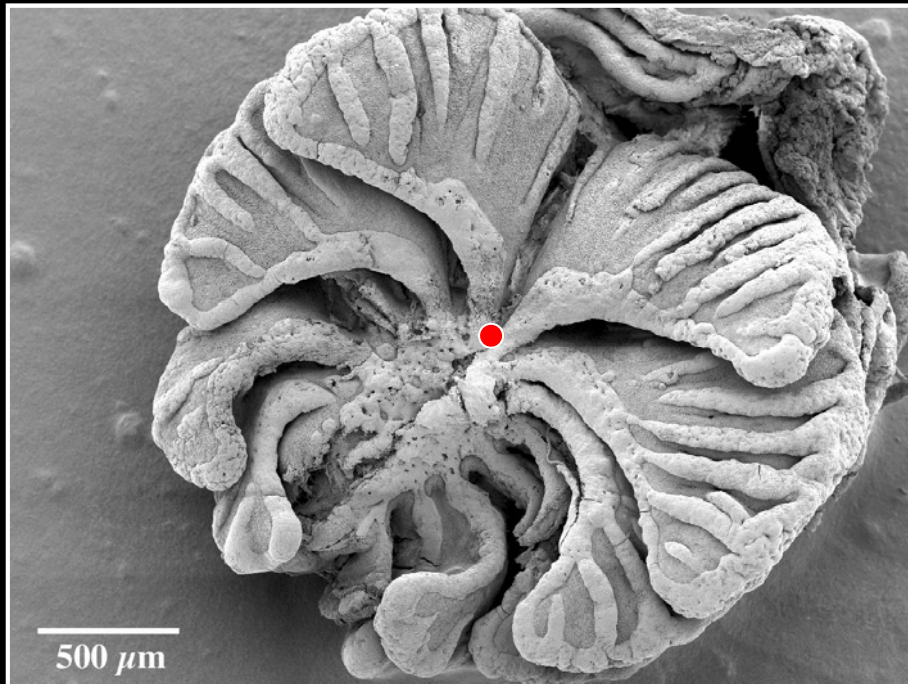
Gillermo Munro/Seattle Post-Intelligencer

Why focus on the salmon nose?

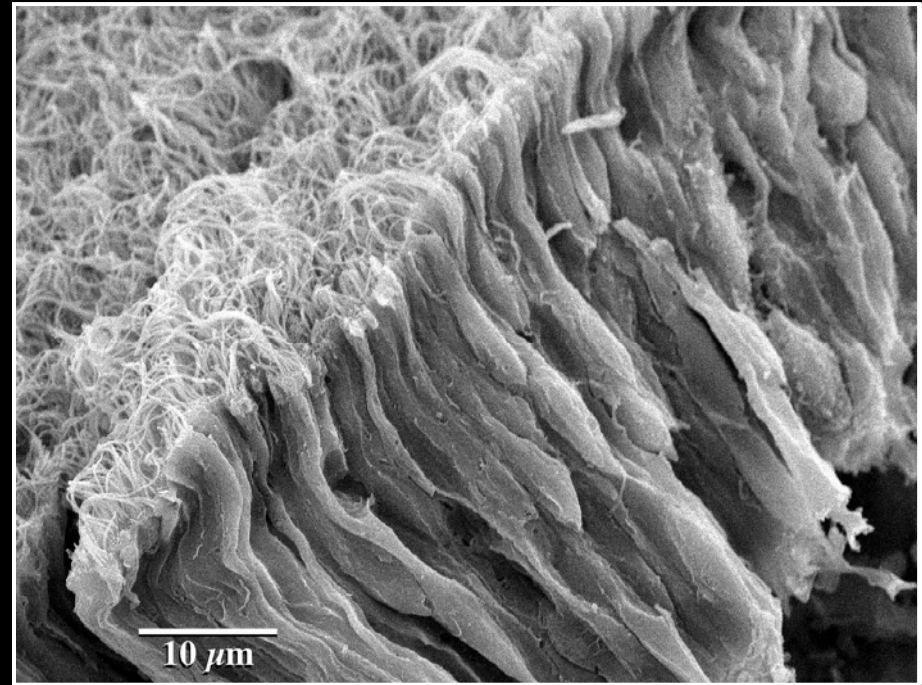
- The nose is directly exposed to pollutants.
- Receptor neurons are very sensitive.
- Basic biology is well understood.
- Neurophysiological recordings are straightforward.
- Olfaction is linked to survival and reproduction.
- Olfaction guides imprinting and homing behaviors.

The olfactory organ (rosette) of coho salmon

Age 0



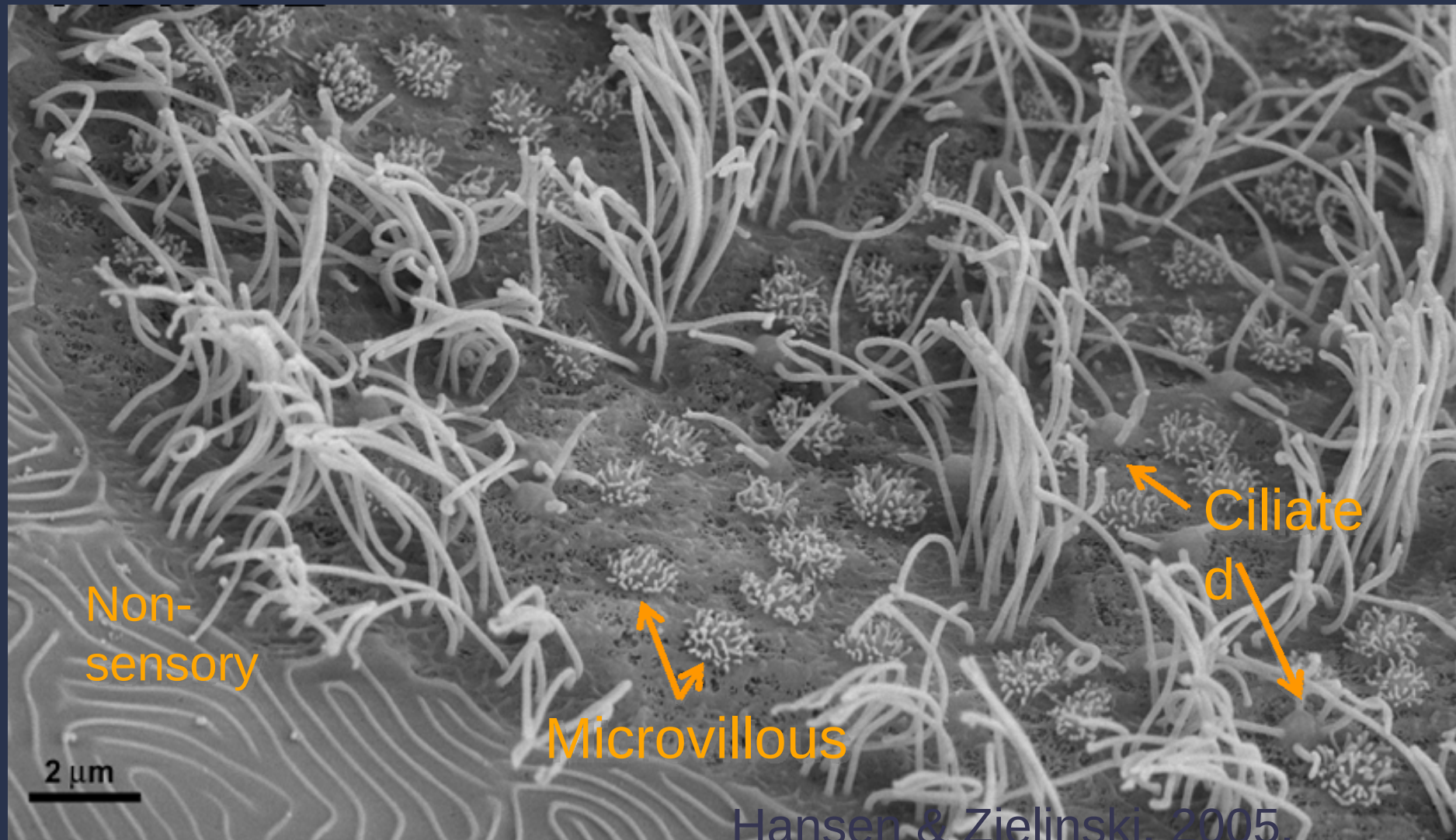
Age 1⁺



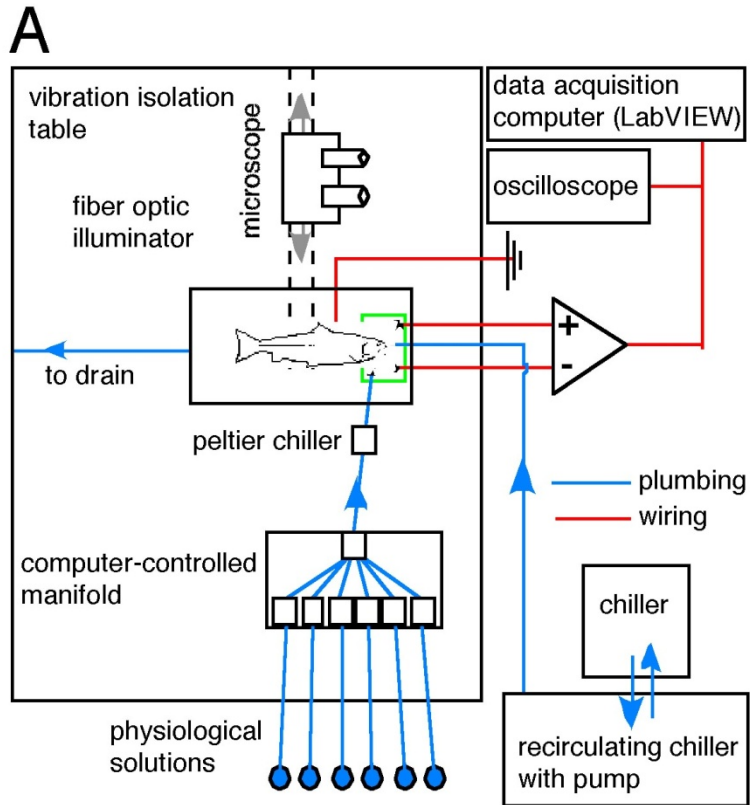
Scanning electron micrographs by Carla Stehr, NOAA Fisheries

● = electrode location for neurophysiological field potential recordings

Olfactory Sensory Neurons

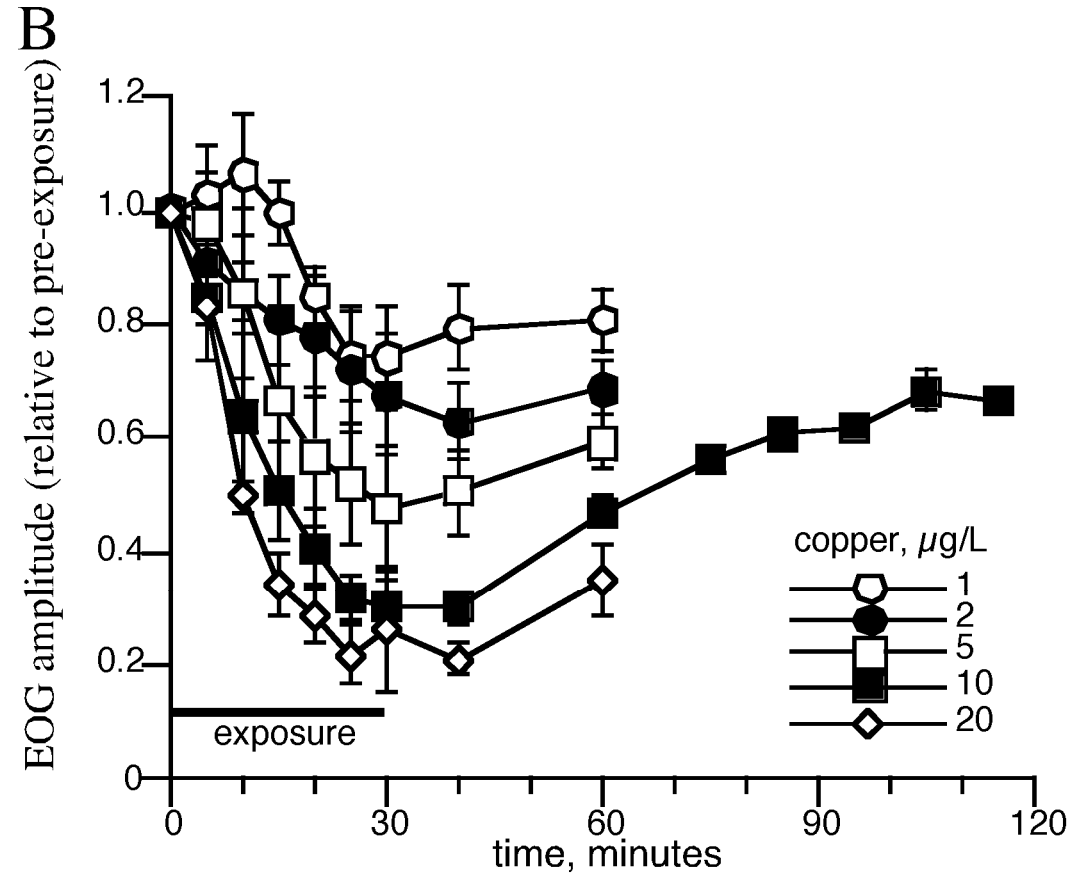
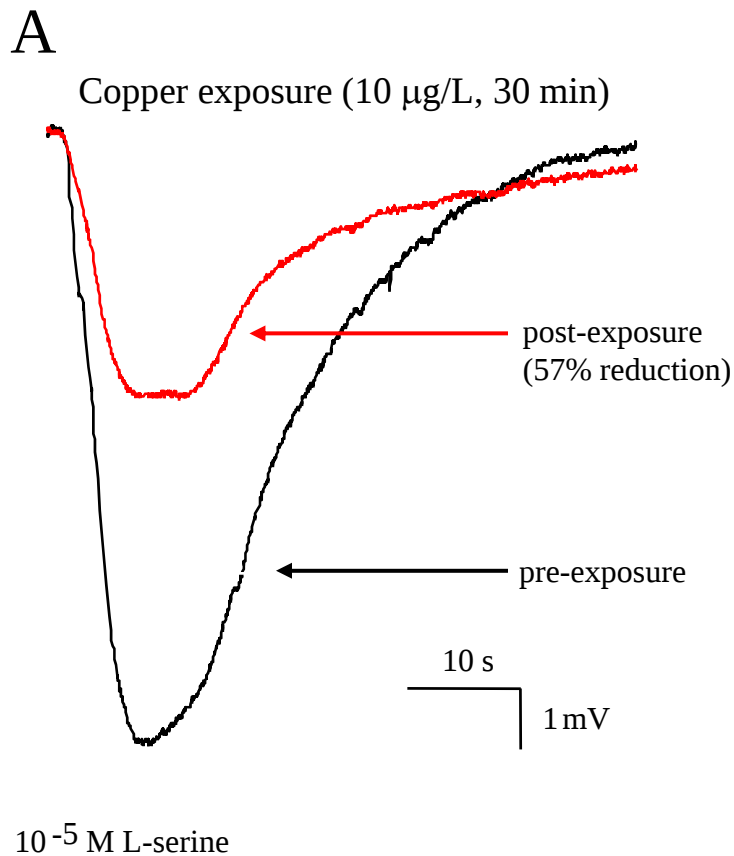


In vivo olfactory recordings



Baldwin, D.H. and Scholz, N.L. (2005). The electro-olfactogram: an *in vivo* measure of peripheral olfactory function and sublethal neurotoxicity in fish. In: *Techniques in Aquatic Toxicology, Volume 2*. GK Ostrander (ed.), CRC Press, Inc. Boca Raton, FL. pp. 257-276.

Sublethal copper neurotoxicity in juvenile coho (30 min exposures)

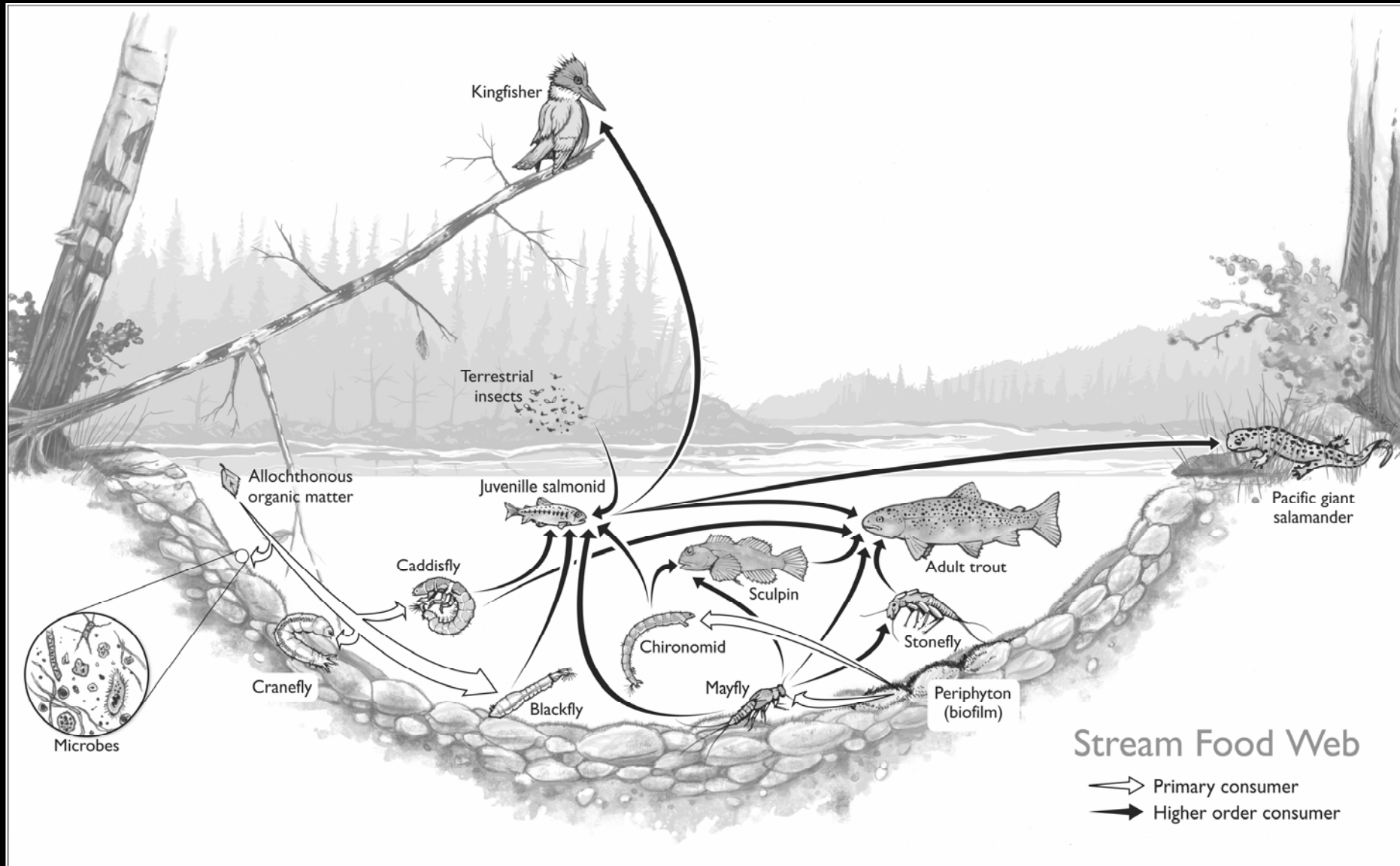


Conclusions:

- Copper-induced loss of olfactory function occurs very quickly (on a timescale of minutes)
- Copper is a general inhibitor - i.e., it knocks out sensory pathways that are responsive to different natural odorants

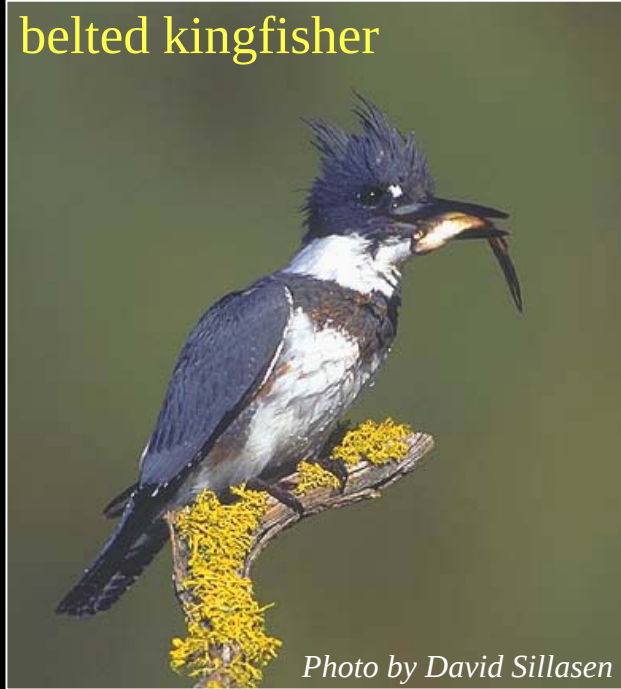
So what?

Salmon habitat - aquatic community structure



Natural predators on juvenile salmonids in the PNW

belted kingfisher



river otter



great blue heron

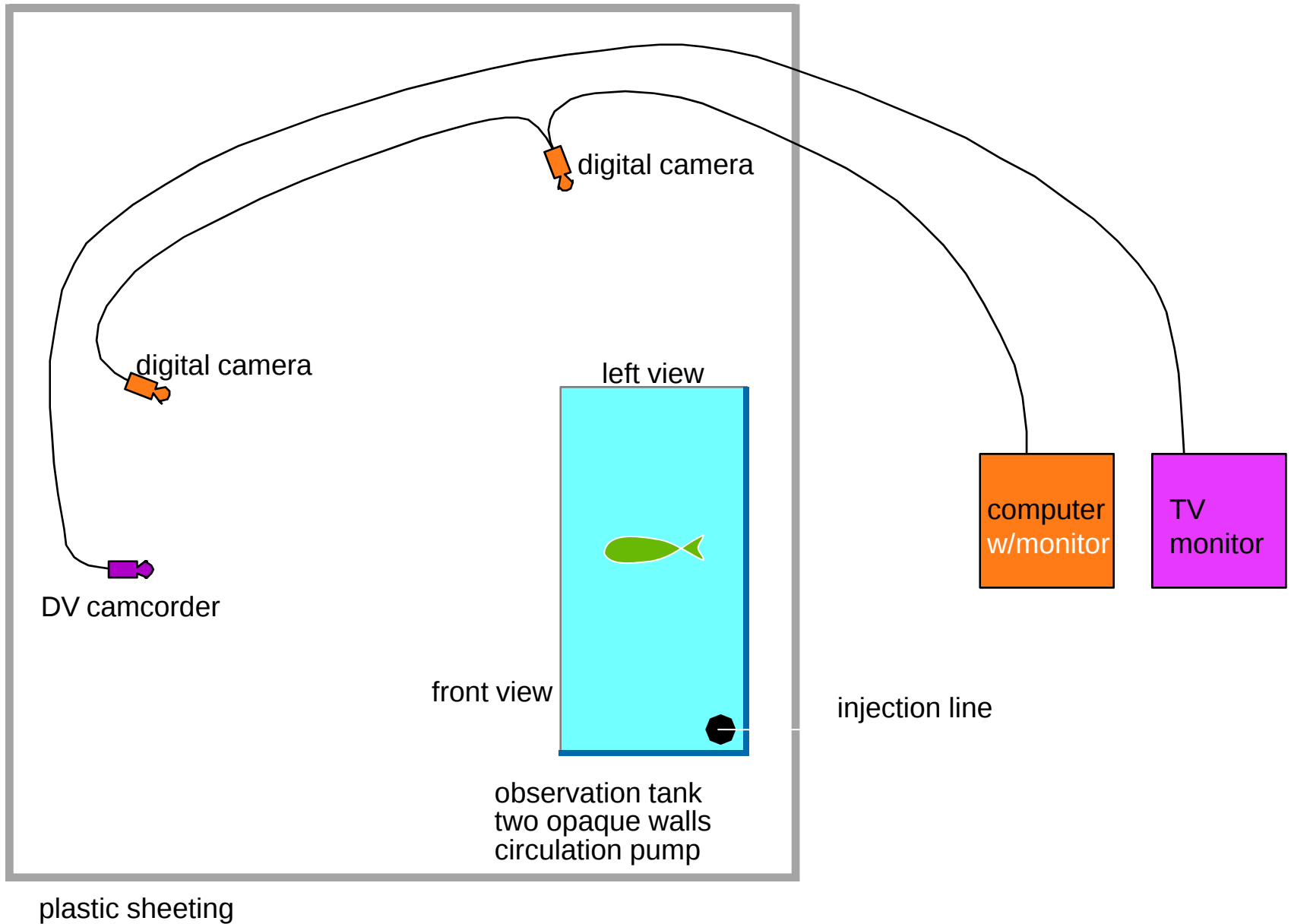


cutthroat trout





Computer-aided analysis of juvenile coho behaviors



Left Side View

before alarm odor



Front View

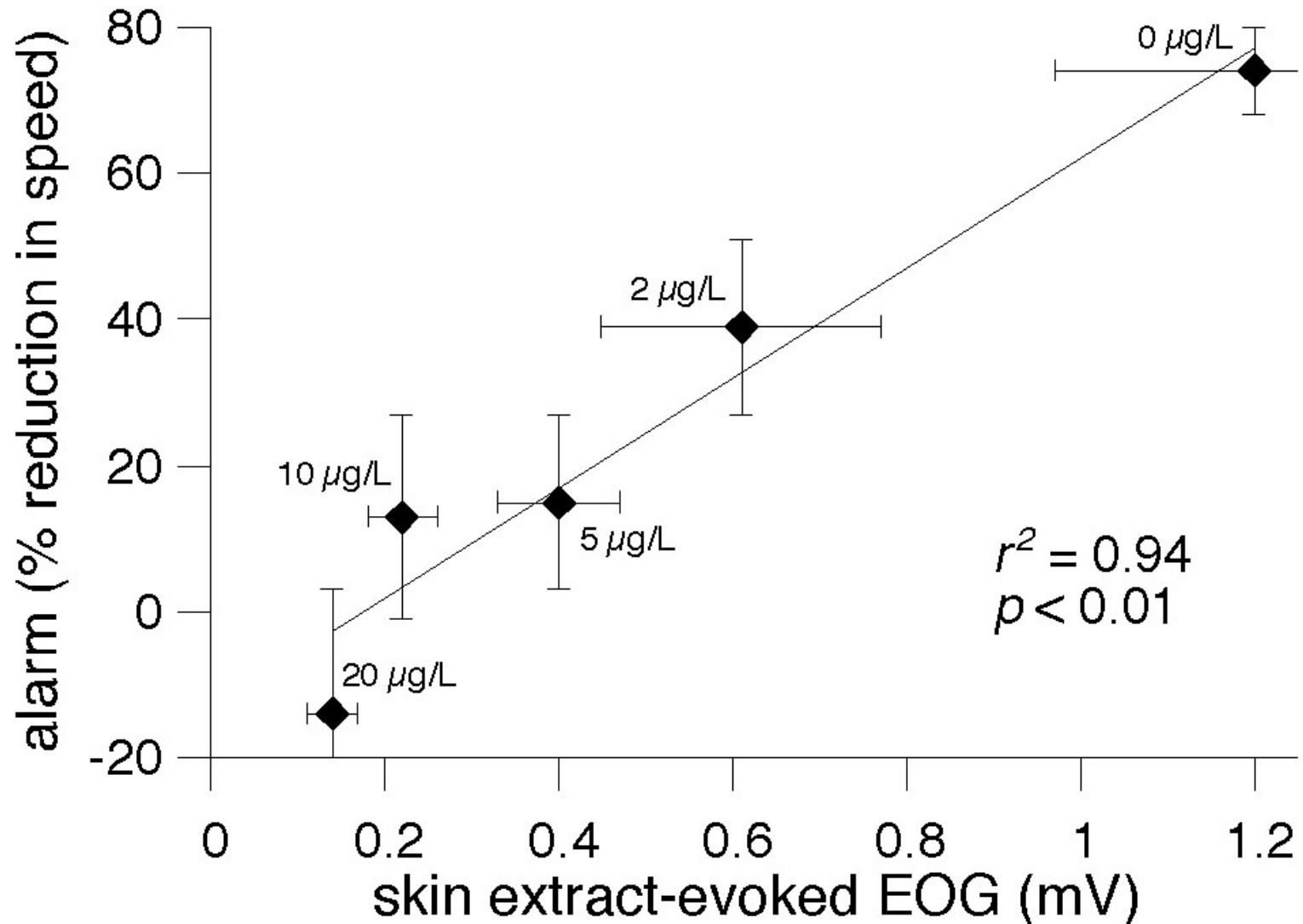
0 $\mu\text{g/L}$
copper
(3 hr)



10 $\mu\text{g/L}$
copper
(3 hr)



Relative impacts of dissolved copper on juvenile coho olfactory neurophysiology and predator-avoidance behavior



A Sensory System at the Interface between Urban Stormwater Runoff and Salmon Survival

JASON F. SANDAHL,[†]
DAVID H. BALDWIN,[‡]
JEFFREY J. JENKINS,[†] AND
NATHANIEL L. SCHOLZ^{*,‡}

Oregon State University, Department of Molecular and Environmental Toxicology, 333 Weniger Hall, Corvallis, Oregon 97331, and NOAA Fisheries, Northwest Fisheries Science Center, Ecotoxicology and Environmental Fish Health Program, 2725 Montlake Boulevard East, Seattle, Washington 98112

Motor vehicles are a major source of toxic contaminants such as copper, a metal that originates from vehicle exhaust and brake pad wear. Copper and other pollutants are deposited on roads and other impervious surfaces and then transported to aquatic habitats via stormwater runoff. In the western United States, exposure to non-point source pollutants such as copper is an emerging concern for many populations of threatened and endangered Pacific salmon (*Oncorhynchus* spp.) that spawn and rear in coastal watersheds and estuaries. To address this concern, we used conventional neurophysiological recordings to investigate the impact of ecologically relevant copper exposures (0–20 $\mu\text{g/L}$ for 3 h) on the olfactory system of juvenile coho salmon (*O. kisutch*). These recordings were combined with computer-assisted video analyses of behavior to evaluate the sensitivity and responsiveness of copper-exposed coho to a chemical predation cue (conspecific alarm pheromone). The sensory physiology and predator avoidance behaviors of juvenile coho were both significantly impaired by copper at concentrations as low as 2 $\mu\text{g/L}$. Therefore, copper-containing stormwater runoff from urban landscapes has the potential to cause chemosensory deprivation and increased predation mortality in exposed salmon.

ES&T's Best Papers of 2007

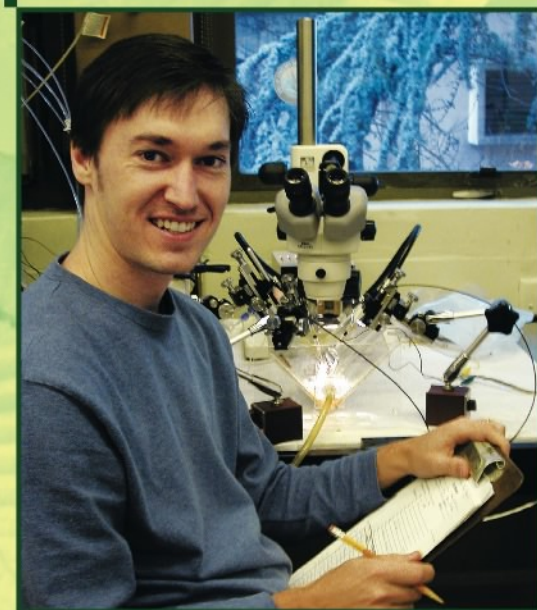
In 2007, *ES&T* published more than 1200 papers on a wide range of topics. But which papers were the top papers—the best of the year?

Second runner-up: Copper changes salmon behavior

"A Sensory System at the Interface between Urban Stormwater Runoff and Salmon Survival" by Jason F. Sandahl and Jeffrey J. Jenkins, Department of Molecular and Environmental Toxicology, Oregon State University; and David H. Baldwin and Nathaniel L. Scholz, NOAA Fisheries, Northwest Fisheries Science Center, Ecotoxicology and Environmental Fish Health Program, 2007, 41 (8), 2998–3004.

When Jason Sandahl started learning about salmon noses as a doctoral student at Oregon State University, he didn't know his research might help change the way toxicologists think about how contaminants affect animal health.

Sandahl grew up on a pear and cherry farm in Oregon's Hood River valley. "One concern in the Northwest is what contaminants may be doing to [declining] salmon populations," he says. In graduate school at Oregon State University, he began studying the effects of pesticides on salmon by using traditional toxicological approaches, which rely heavily on determining the levels that kill an animal.



Jason Sandahl probed fish noses and found that nerve damage from copper can spell big trouble for little salmon.

Conclusions:

- Copper-induced loss of olfactory function occurs very quickly (on a timescale of minutes)
- Copper is a general inhibitor - i.e., it knocks out sensory pathways that are responsive to different natural odorants
- Relative impacts of copper on olfaction and olfactory-mediated predator avoidance behaviors are closely correlated
- Thresholds for neurobehavioral toxicity are low (e.g., at or below 2 $\mu\text{g/L}$)

Cross-species extrapolations: hatchery-reared coho to wild-reared steelhead



LOCAL NEWS

Wednesday, May 9, 2007 - Page updated at 03:32 PM

Puget Sound steelhead declared "threatened"

By Warren Cornwall
Seattle Times environment reporter

First it was Puget Sound chinook and the bull trout. Then the resident orcas. Now Puget Sound steelhead have won a spot on a list no creature would want — the federal Endangered Species List.

The announcement yesterday by the federal National Marine Fisheries Service (NMFS) that wild Puget Sound and Hood Canal steelhead are "threatened" has been expected for more than a year. Nonetheless, it underscores the growing sense that something has gone haywire with Puget Sound's ecosystem.



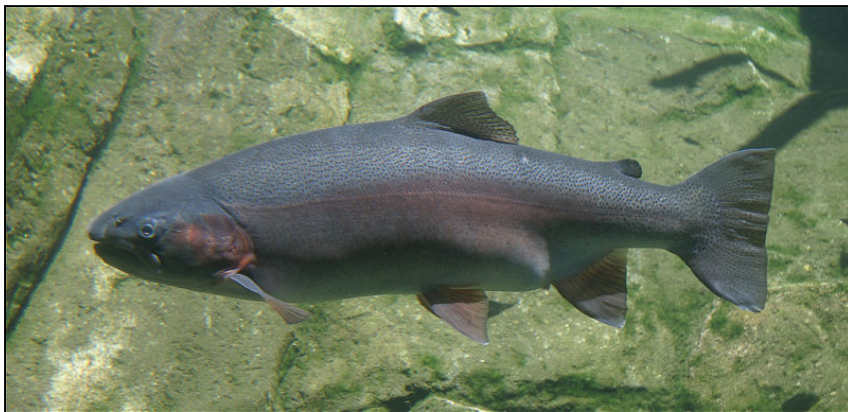
enlarge

MARK HARRISON / THE SEATTLE TIMES

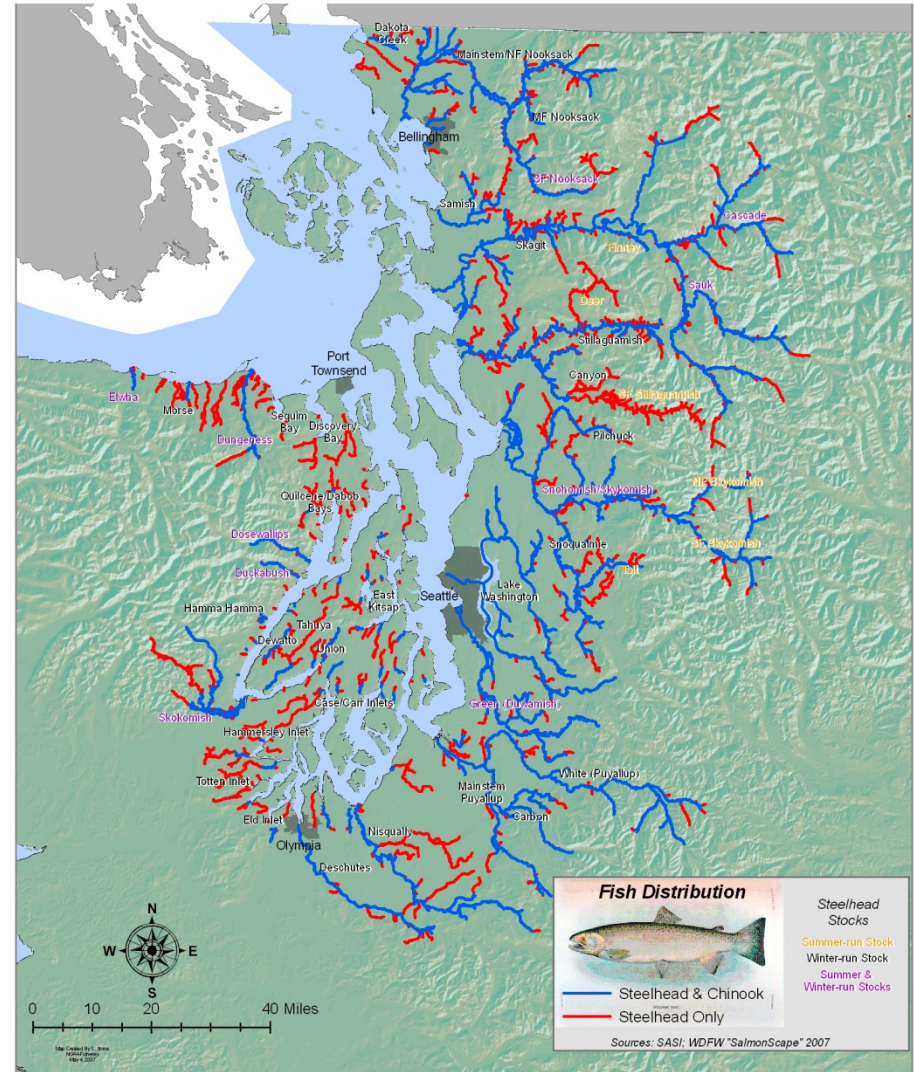
Steelhead migrate to the ocean and return upriver to spawn.

Related

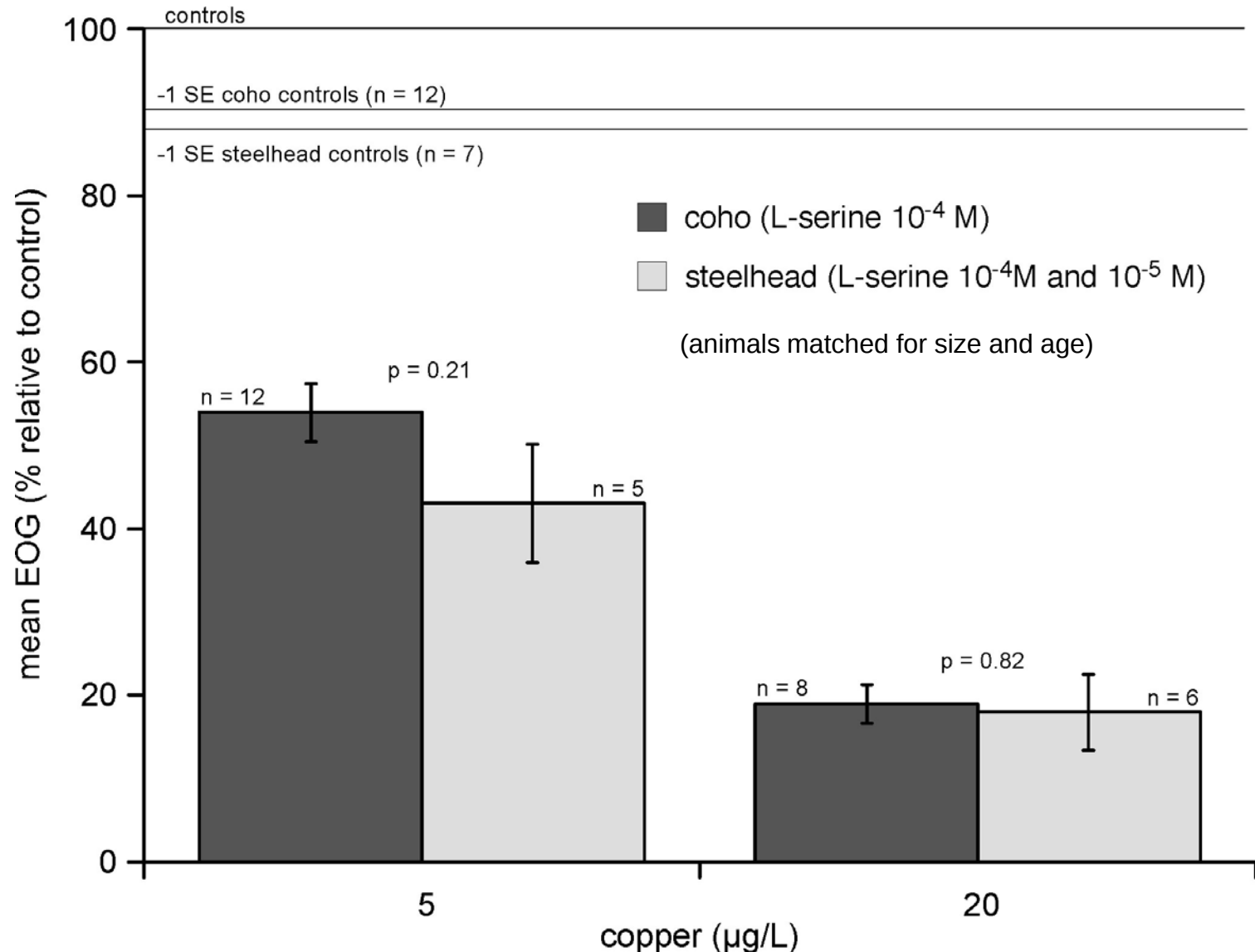
Ruckelshaus named to lead Puget



ESA-Listed Puget Sound Steelhead & Chinook



The neurotoxic impacts of copper are similar across different species and for fish raised in different freshwater environments



Coho data from Sandahl et al., 2007, *Environmental Science and Technology*, 31:2998-3004.
Steelhead data from Baldwin, Tatara, and Scholz, 200x, manuscript in preparation.

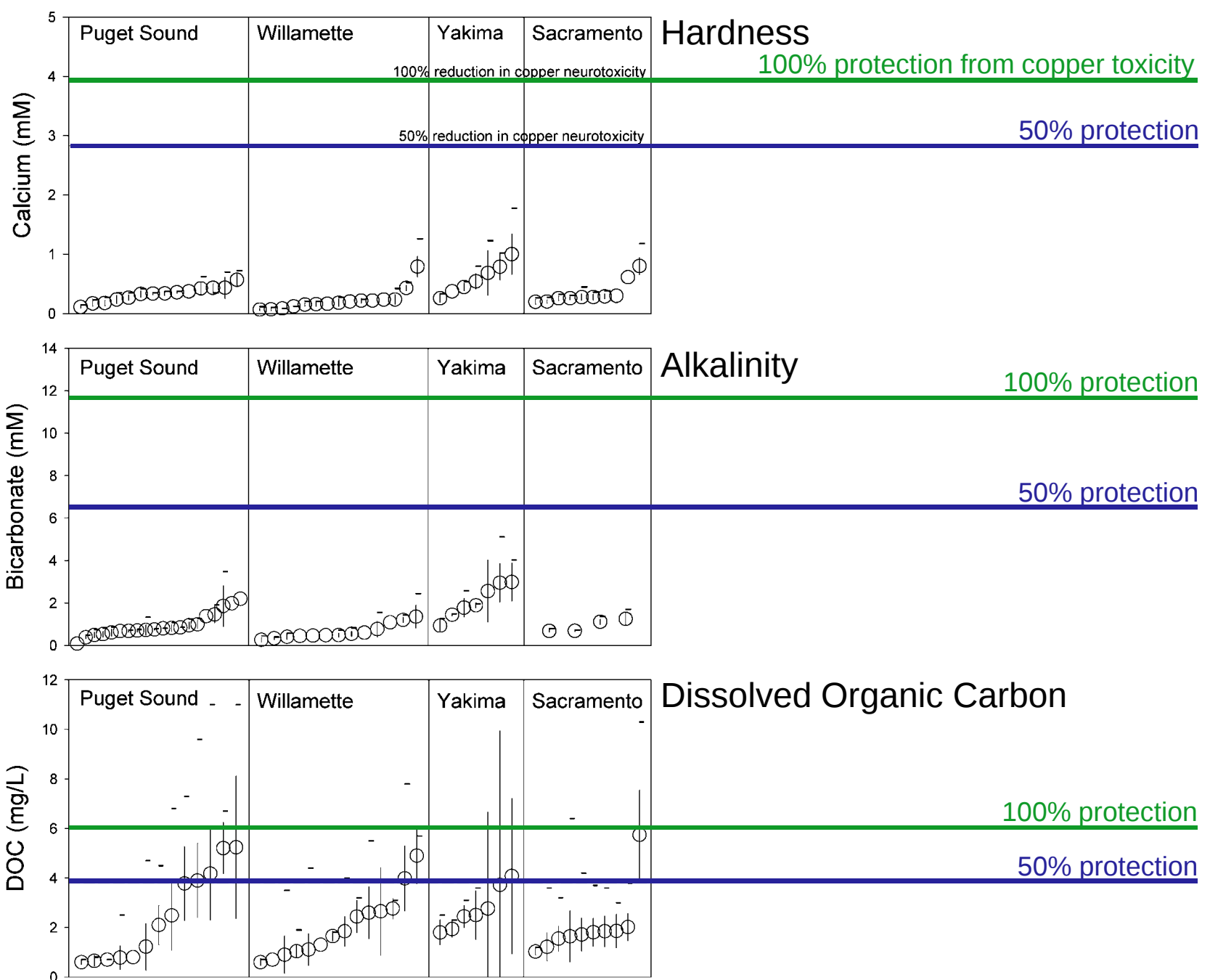
Conclusions:

- Copper-induced loss of olfactory function occurs very quickly (on a timescale of minutes)
- Copper is a general inhibitor - i.e., it knocks out sensory pathways that are responsive to different natural odorants
- Relative impacts of copper on olfaction and olfactory-mediated predator avoidance behaviors are closely correlated
- Thresholds for neurobehavioral toxicity are low (e.g., at or below 2 $\mu\text{g/L}$)
- Olfactory toxicity of copper appears to be similar across different species of salmon, steelhead, and trout
- Susceptibility of wild and hatchery-reared fish is similar

Jenifer McIntyre
EPA STAR Predoctoral Fellow

University of Washington
School of Aquatic and Fisheries Sciences
David Beauchamp





McIntyre, J., Baldwin, D.H., Meador, J.P., and Scholz, N.L. (2008). Chemosensory deprivation in juvenile coho salmon exposed to dissolved copper under varying water chemistry conditions. *Environmental Science and Technology*, 42:1352-1358.

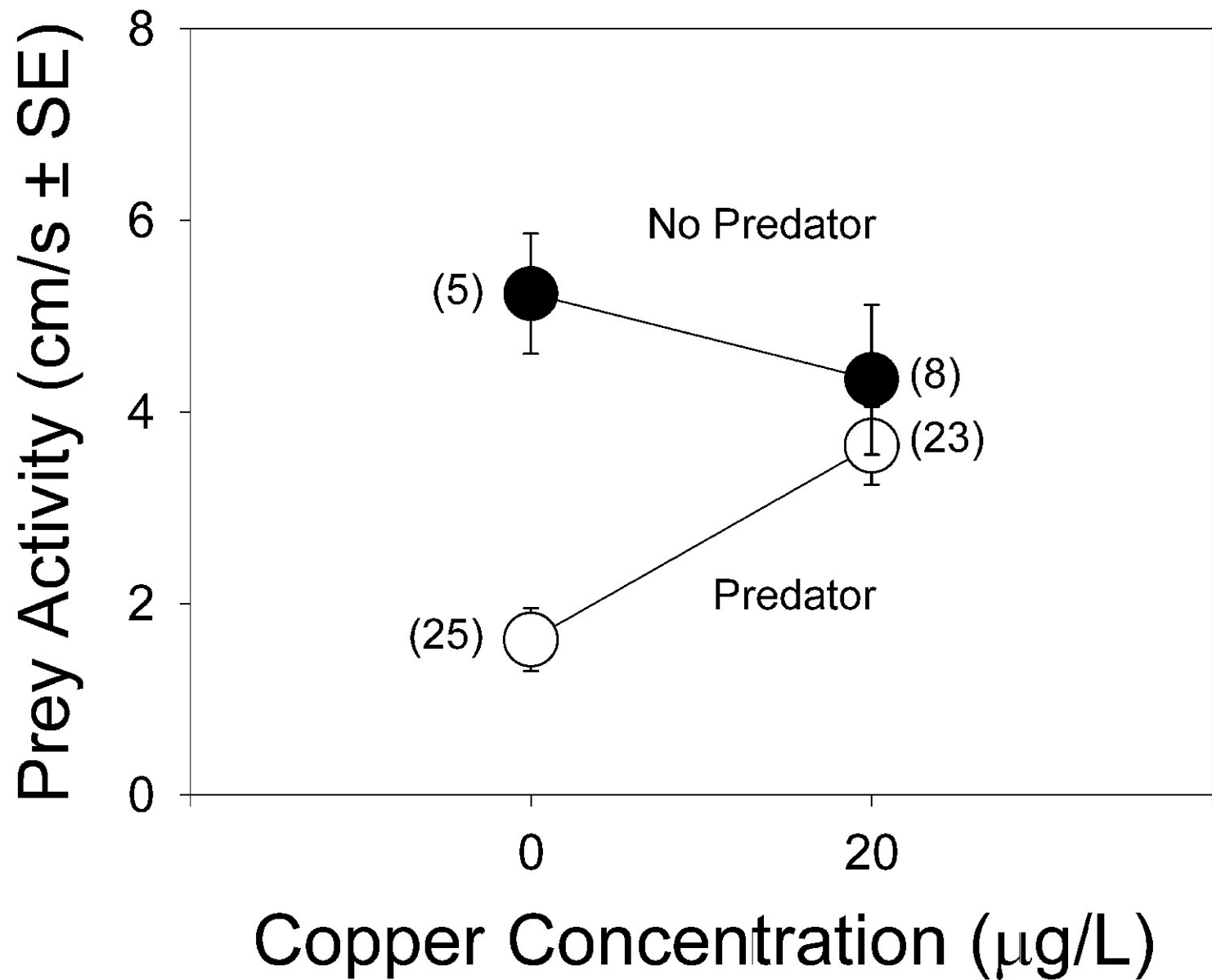
Conclusions:

- Copper-induced loss of olfactory function occurs very quickly (on a timescale of minutes)
- Copper is a general inhibitor - i.e., it knocks out sensory pathways that are responsive to different natural odorants
- Relative impacts of copper on olfaction and olfactory-mediated predator avoidance behaviors are closely correlated
- Thresholds for neurobehavioral toxicity are low (e.g., at or below 2 $\mu\text{g/L}$)
- Olfactory toxicity of copper appears to be similar across different species of salmon, steelhead, and trout
- Susceptibility of wild and hatchery-reared fish is similar
- Influence of water hardness and alkalinity are negligible across environmentally relevant ranges in western rivers
- Elevated DOC may provide some measure of protection against copper toxicity in certain stream systems

Predator-prey experiments at Big Beef Creek, WA

Spring 2007-2009





Latency to Capture Timeline



Prey treatment copper exposure (-3 h 15 min)

Predator acclimation
(-1 hr)

Prey acclimation (-15 min)

Introduce SE (0 s)

Lift prey chamber (30 s)

Lift predator
divider (50 s)

Per arena:

2 wild cutthroat predators

2 juvenile coho prey

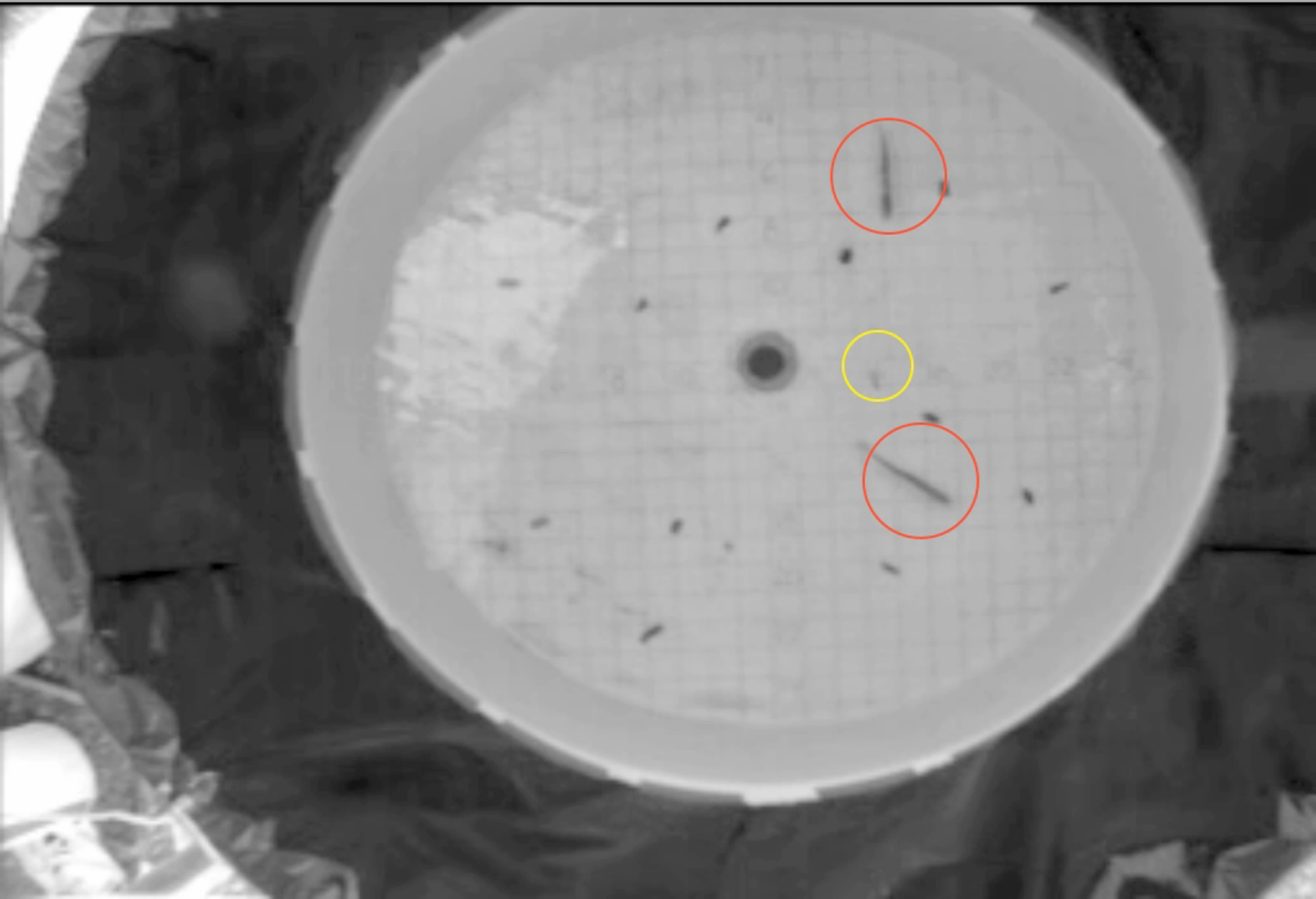
Skin extract (SE) stimulus

Threshold concentration (equivalent of 0.1 ug/L protein)

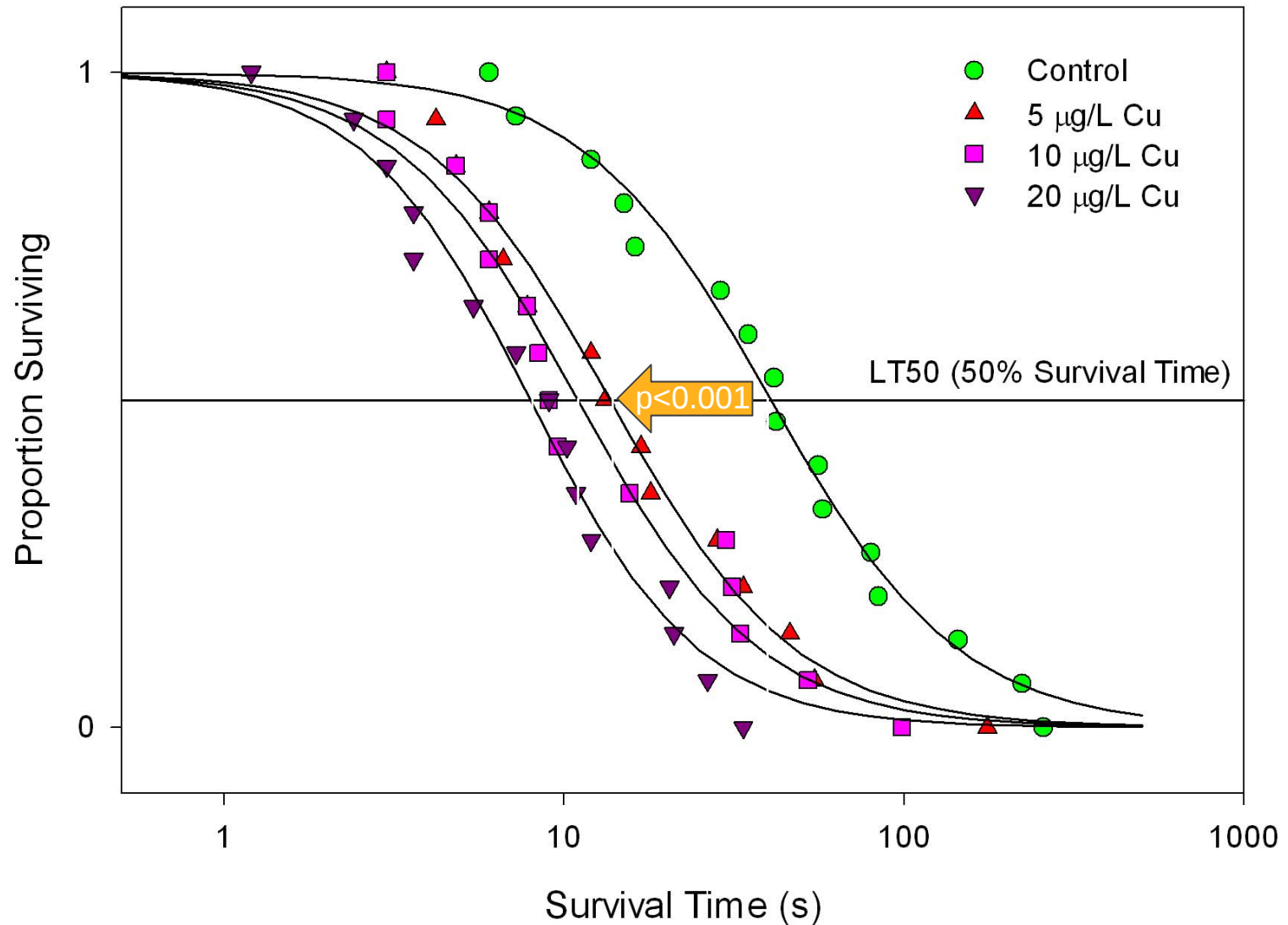


Remaining still avoids attack

prey: juvenile coho salmon
predators: two adult cutthroat trout



Copper Affects Survival with Predators

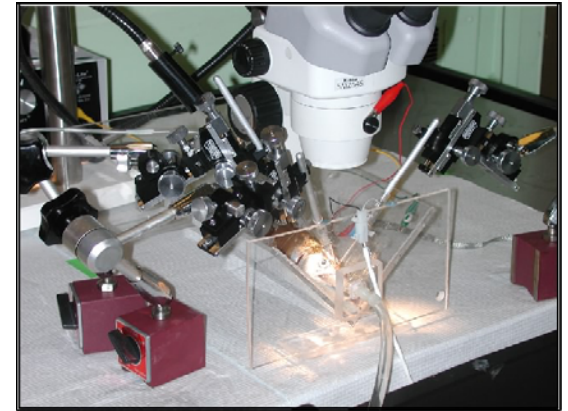


Conclusions continued:

- Copper-exposed juvenile salmon do not respond to sensory cues originating from upstream predators
- Salmon that are isolated from chemical predation signals suffer higher rates of mortality in predator-prey encounters

Task 1 - Direct *in vivo* measures of neurotoxicity in the salmon olfactory system.

- *Toxicity thresholds for dissolved copper*
- *Extent of neural injury and recovery*
- *Impacts on critical behaviors*

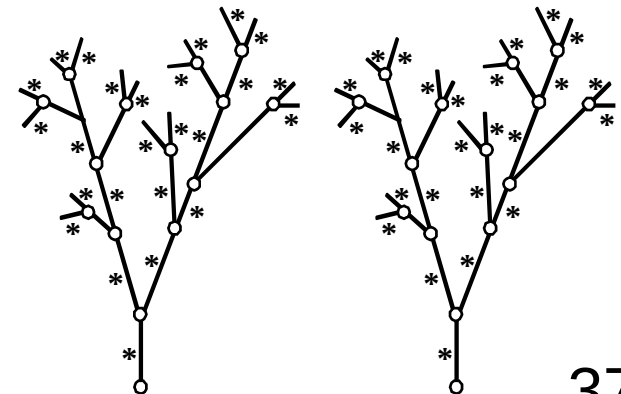


Task 2 - Quantifying the effects of copper on salmon survival and reproductive success.

- *Predator-induced mortality in juveniles*
- *Spawning behavior and fertilization success*

Task 3 - Modeling the impacts of copper on natural salmon populations.

- *Homing and straying in wild and hatchery fish*
- *Limits on the productivity of wild populations*



Effects of Mercury and Copper on the Olfactory Response in Rainbow Trout, *Salmo gairdneri*

TOSHIAKI J. HARA, Y. M. C. LAW, AND S. MACDONALD

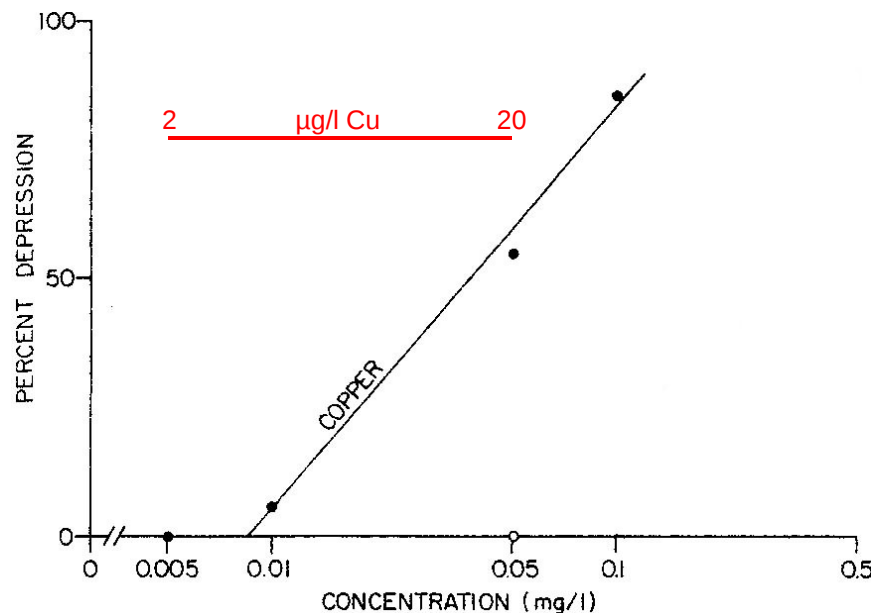
Department of the Environment, Fisheries and Marine Service, Freshwater Institute, Winnipeg, Man. R3T 2N6

HARA, T. J., Y. M. C. LAW, AND S. MACDONALD. 1976. Effects of mercury and copper on the olfactory response in rainbow trout, *Salmo gairdneri*. J. Fish. Res. Board Can. 33: 1568–1573.

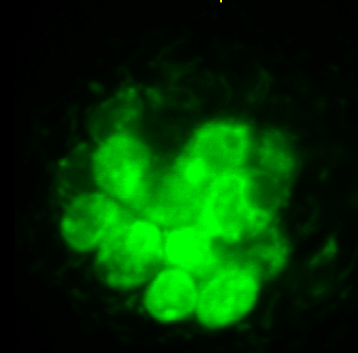
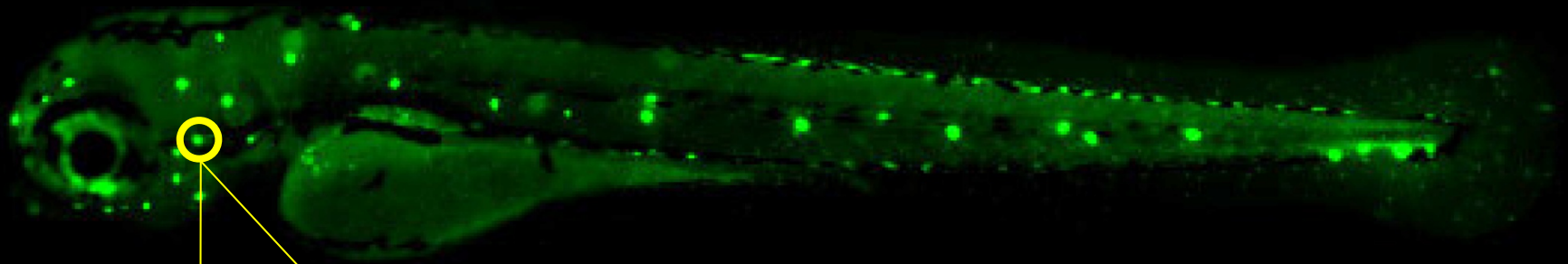
The effects of mercury and copper on the olfactory response of rainbow trout (*Salmo gairdneri*) were studied by perfusing HgCl_2 and CuSO_4 solutions through the olfactory organs while recording the olfactory bulbar electrical responses to the standard stimulant, L-serine.

The olfactory response was depressed during exposure to mercury and copper. The lowest concentrations of mercury and copper needed to cause appreciable effects within 2 h were estimated at 0.10 and 0.008 mg/liter, respectively. The depression increased with increase in the concentration and exposure time of the heavy metals. Recovery of the olfactory response was slower with higher concentrations and longer exposure.

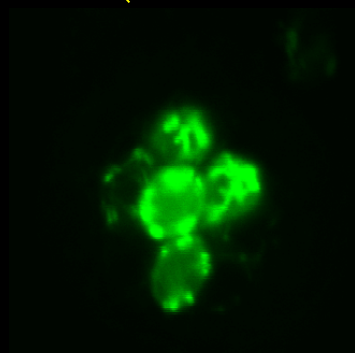
The method is rapid, accurate, and sensitive, and can be applied to other toxic chemicals and fish species.



DASPEI labeling of lateral line neuromasts in larval zebrafish (four days post-fertilization)



control



copper exposed
(3 hr)

