Effects of Micro and Nanoplastics across Biological Levels of Organization

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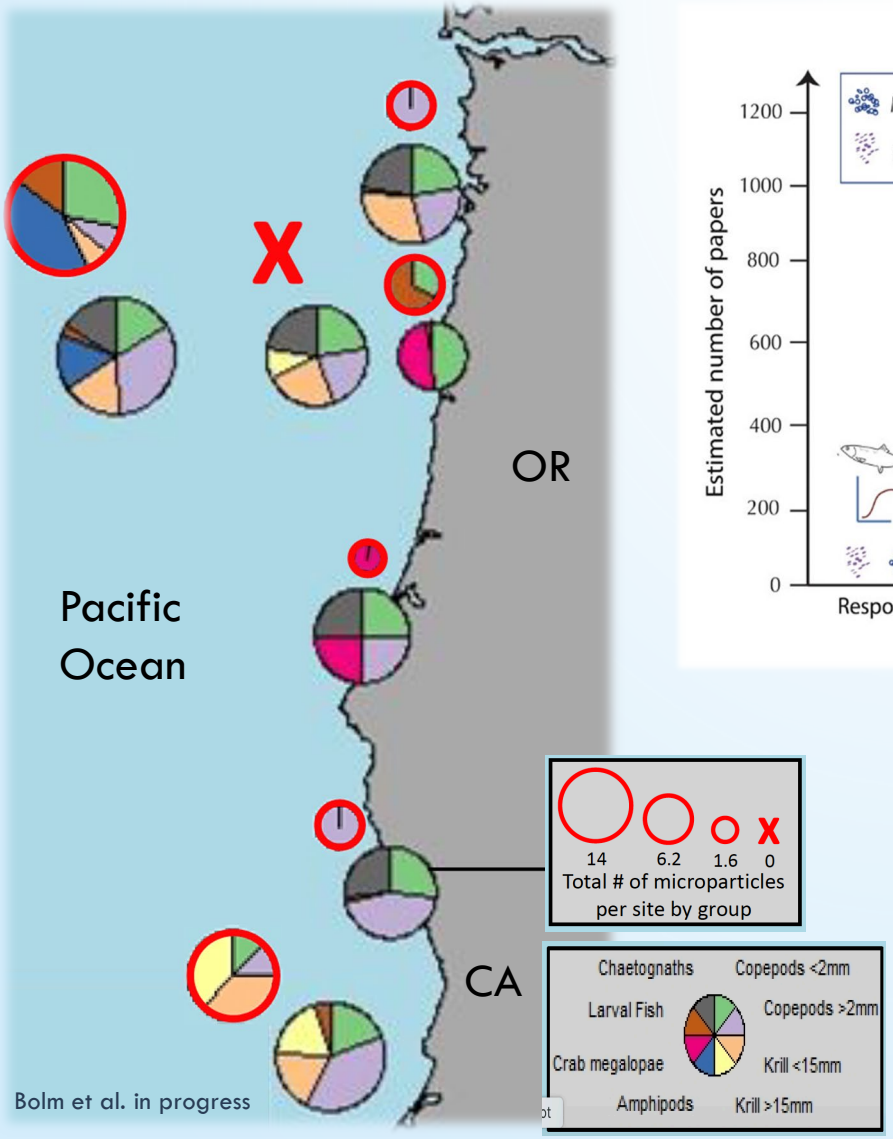
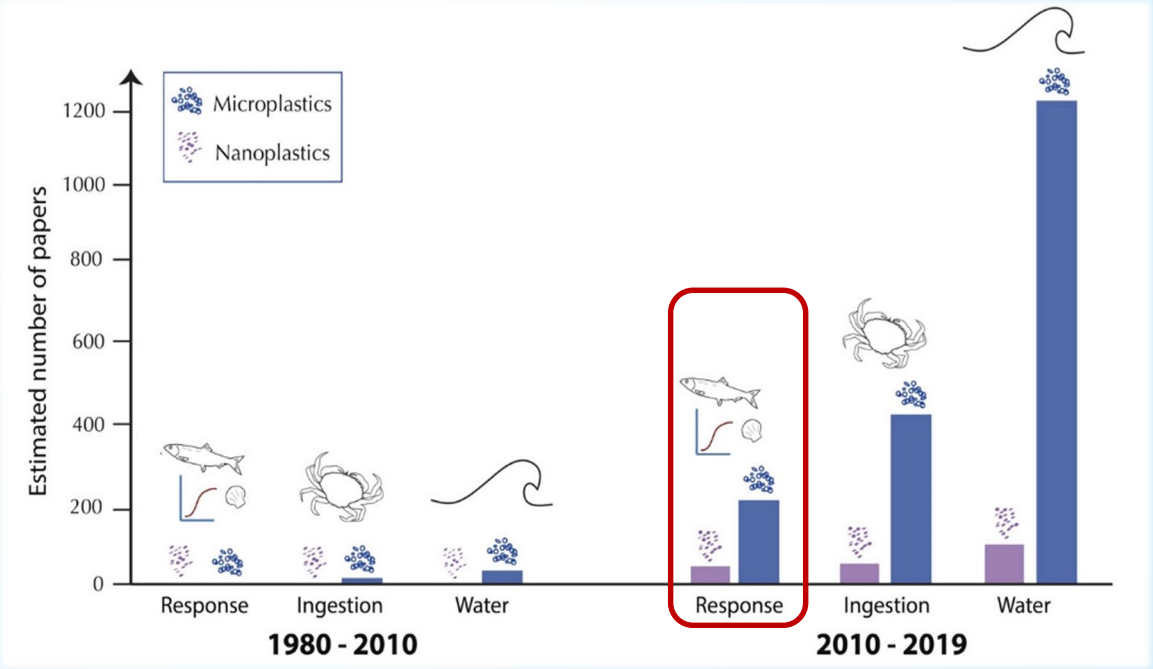
Lands acknowledgment: Work performed on land historically occupied by people of the Kalapuya and Chepenefa, as well as the Waccamaw and Cape Fear (https://native-land.ca/)

- Background
- Effects Across the Biological Hierarchy
- Moving towards AOPs for Plastics?
- Pacific Northwest Consortium on Plastics Research
- Moving Forward


Photo credit: Anna Bolm

Research on plastics …





Granek, Brander, Holland 2020, *Lim Oce Lttrs*

Found across marine, freshwater, and terrestrial organisms.

Still far fewer papers on effects than occurrence

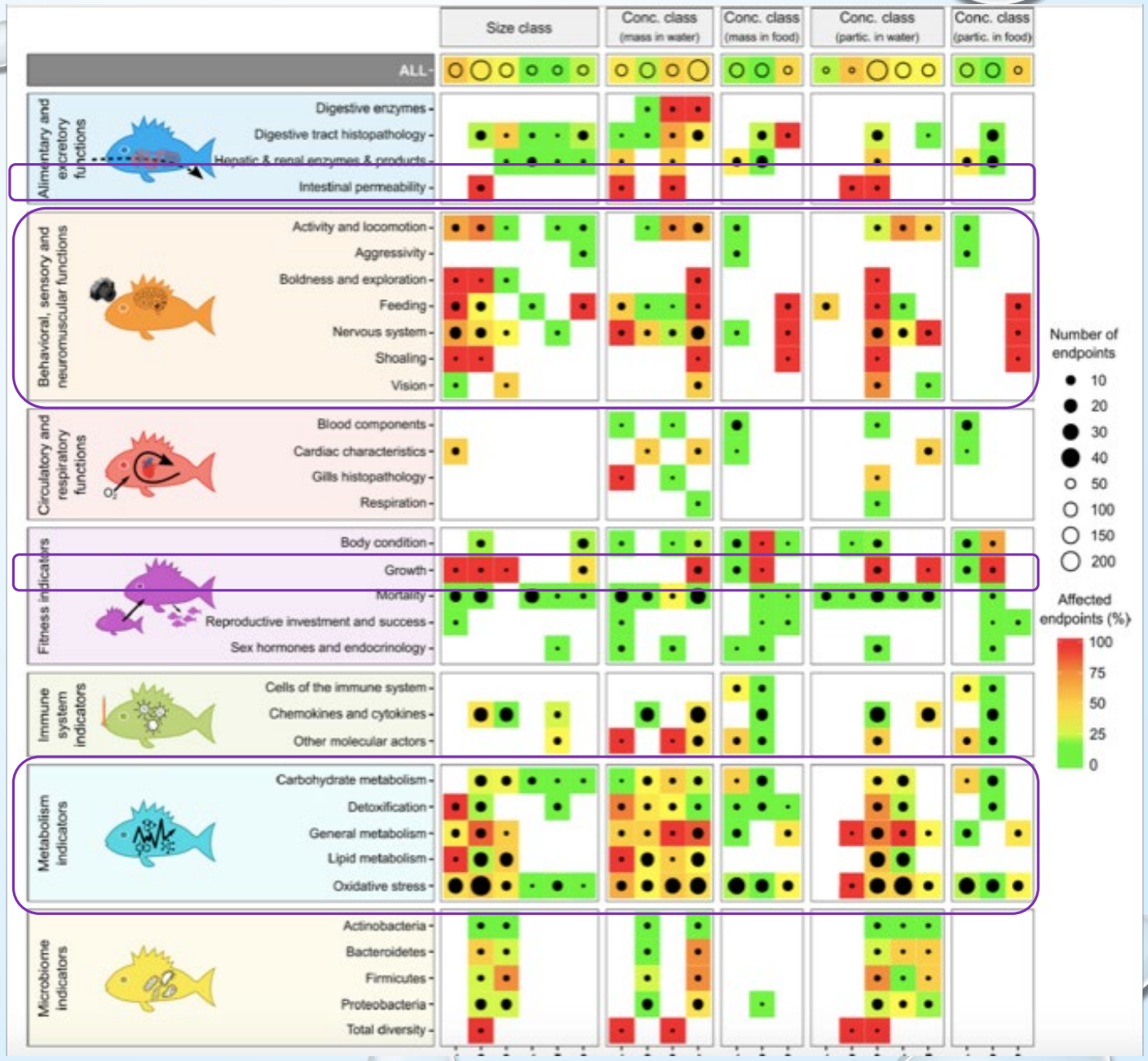
Toxicity…

- Size and shape matter
  - Common aquatic test organisms (e.g. zebrafish, grass shrimp) respond more negatively to fibers or irregularly shaped fragments than spheres
  - Can also affect propensity to accumulate or to be ingested
  - Smaller particles can be translocated to internal organs (e.g. liver)
- Polymers across sizes and shapes can cause oxidative stress
  - Upon exposure to microplastics cells may produce reactive oxygen species that can cause structural damage
- Associated pollutants influence toxicity in some cases
  - Recent work in fish showed no leaching of phthalates from ingested plastic pellets over 120 hrs (Joseph et al. in prep)
  - However, the large surface area to volume ratio of nanomaterials increases can increase adsorption capabilities

Jacob et al. 2020, Bucci et al. 2020, Choi et al. 2018, Qiao et al. 2019, Ziajahromi et al 2017, Koelmans et al. 2015, Velzeboer et al. 2014

Effects Summary

Responses such as **behavioral changes**, and **generation of reactive oxygen species,** as well as **intestinal permeability and growth** stand out in a recent **meta-analysis** of fish studies.



Jacob et al. 2020

Centropristis striata

- Commercial / recreational fishery east coast of U.S.
- Average life span of 8-12 years
- Early life stages in estuaries and coastal habitats
- Bottom dwellers, opportunistic predator (invertebrates, small fishes)
- Cultured at University of North Carolina at Wilmington

https://www.mass.gov/service-details/learn-about-black-sea-bass

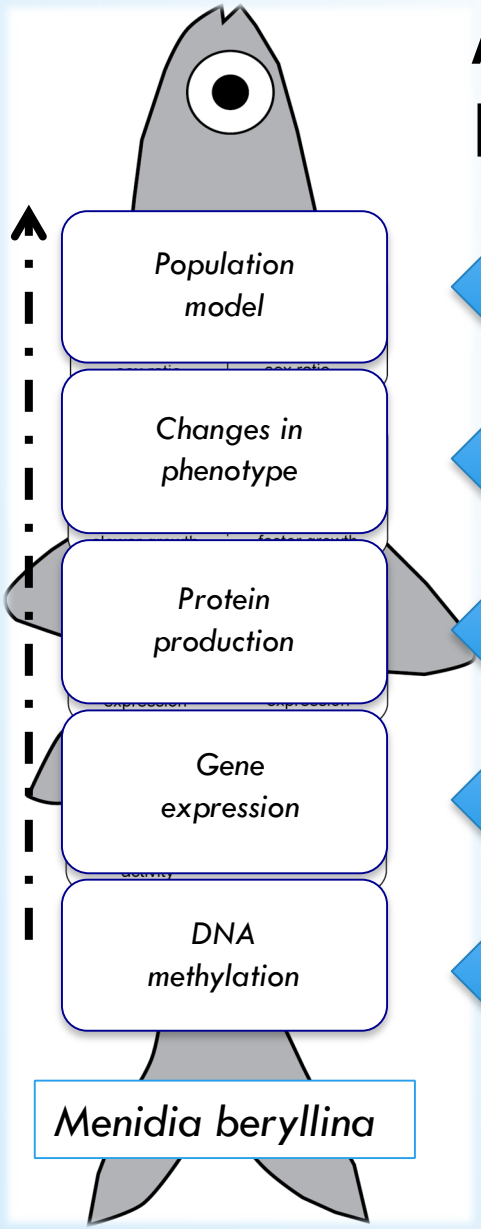
Menidia beryllina

- Important species in N. American estuaries
- Short generation time, spawn readily
- Sensitive to stressors within and across generations

Aquaculturenurseryfarms.com

Brander et al., 2012a, b, *Ecotoxicology*, *Env Tox Chem*; DeCourten and Brander 2017, *Sci Rpts*
Brander et al., 2013, *PLoS One,* DeGroot and Brander, 2014, *Aq Toxicology*; Frank et al. 2019 *Aq Toxicology,* DeCourten et al. 2020 *ES&T*

Menidia beryllina…
Responses across scales to CECs



This is projected to reduce population size

Linked with reduced fecundity

Lead to changes in protein expression

Small changes in gene expression

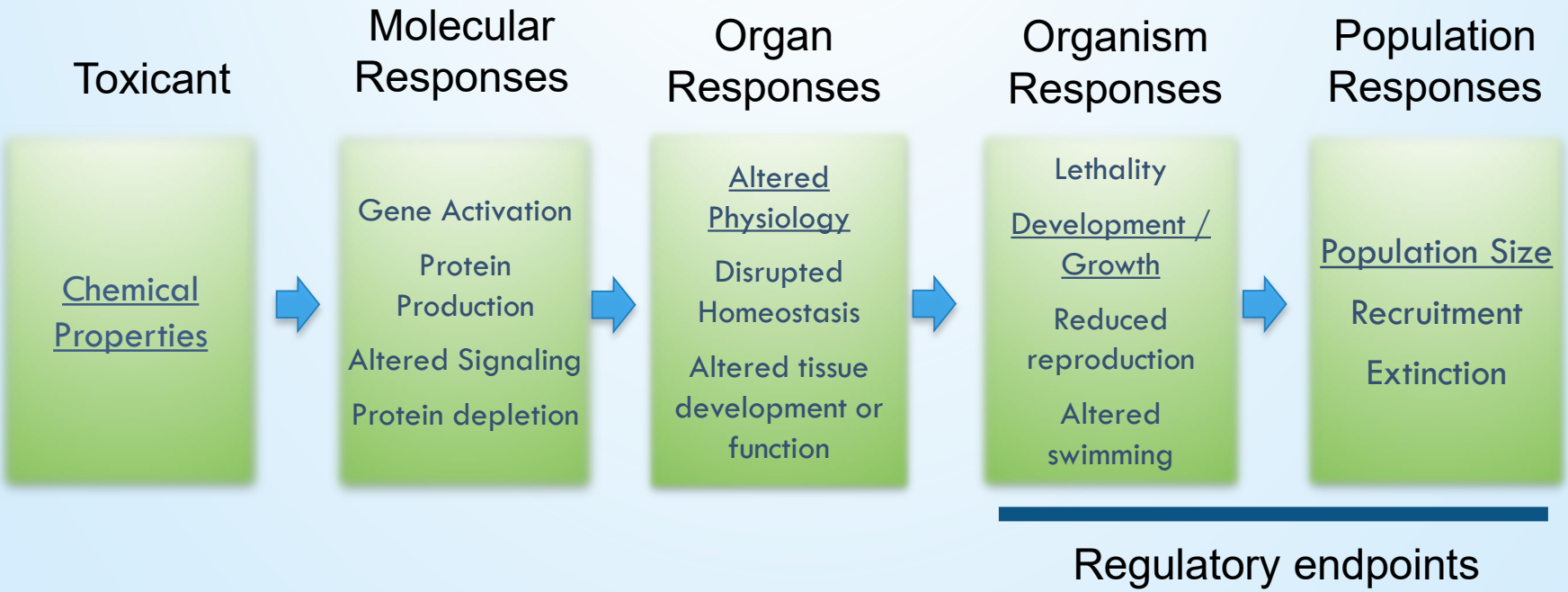
DNA methylation in promoter / gene body

Brander et al., 2012a, b, *Ecotoxicology, Env Tox Chem; Aquatic Tox*
Brander et al., 2013, *PLoS One*, DeGroot and Brander, 2014, *Aquatic Tox*
DeCourten and Brander 2017, *Nat Sci Rpts*, Frank, Brander et al. 2019, *Aquatic Tox*
Major et al. 2020 Frntrs Mar Sci, DeCourten et al. 2020, Env. Sci. Tech.

Jeffries, Brander et al., 2015, *Env Sci & Pollution Research*
Brander et al., 2016, *Aquatic Toxicology*
Cole, Brander et al., 2016, *Archives of Environmental Contamination and Toxicology*
White, Cole, Cherr, Connon, and Brander, 2017, *Environmental Science and Technology*

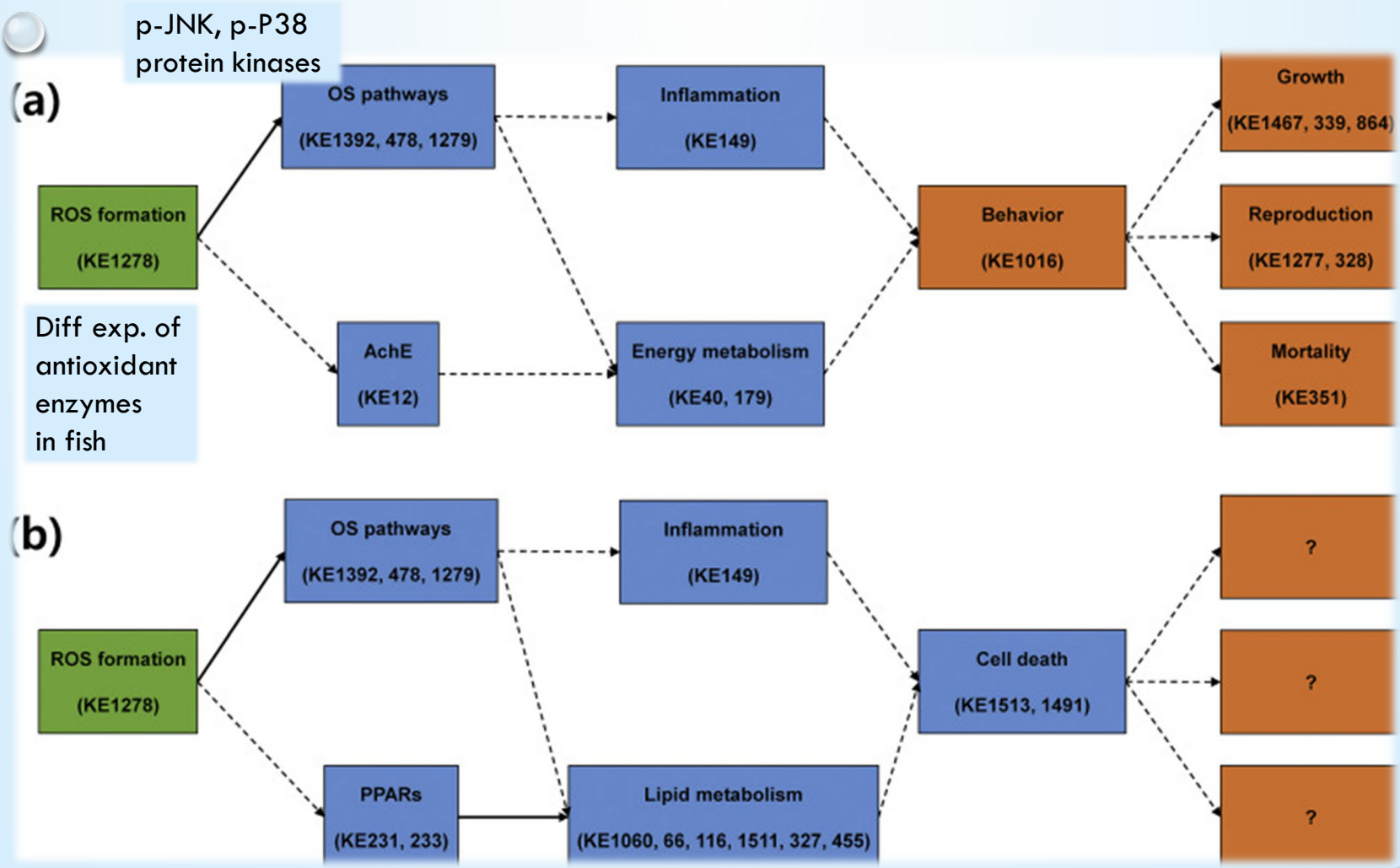
Assessing impacts across levels of organization …

The challenge in applying AOPs to microplastics (and eventually nanoplastics) is that an AOP is defined as a series of key events that begins with a molecular initiating event. Microplastics are a diverse group of contaminants that don't have an easily defined MIE and instead appear to induce a more general oxidative stress response.



Ankley et al. 2010, Ankley and Edwards 2018, Yong et al. 2020

Putative AOP for microplastics …



Jeong and Choi 2019, Jeong et al. 2016, Choi et al. 2020

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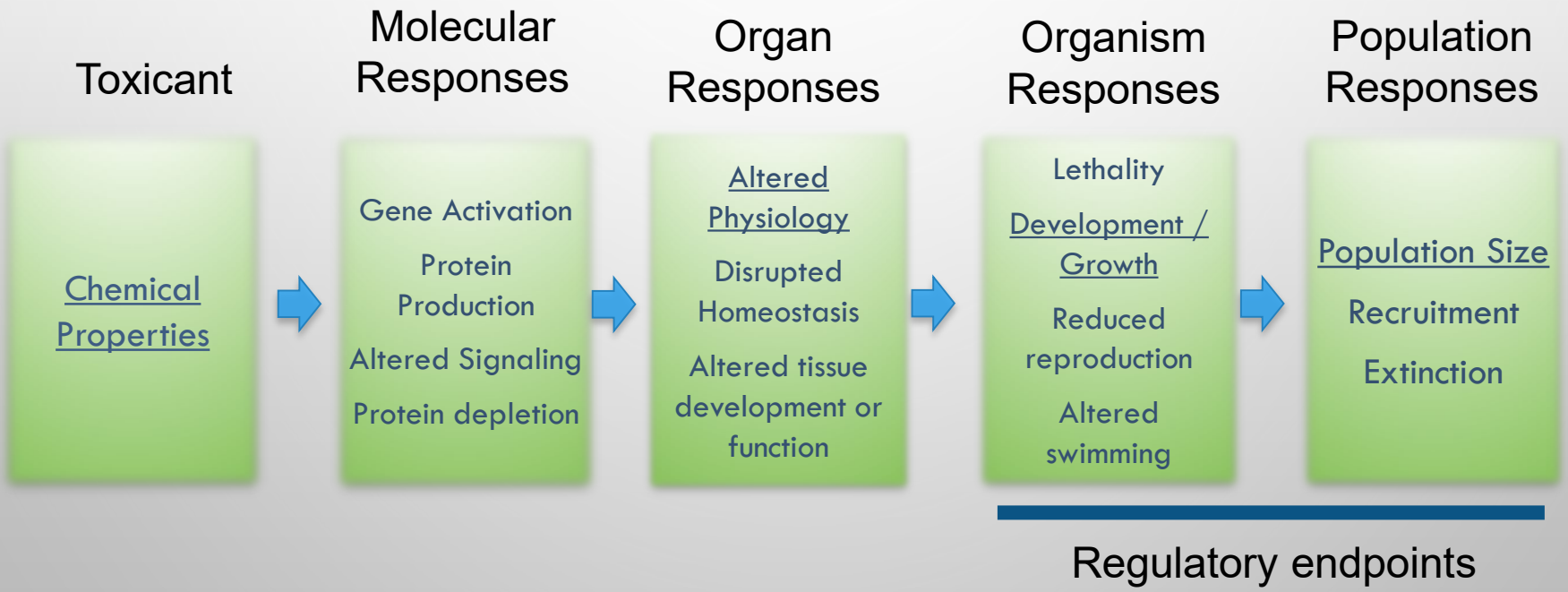
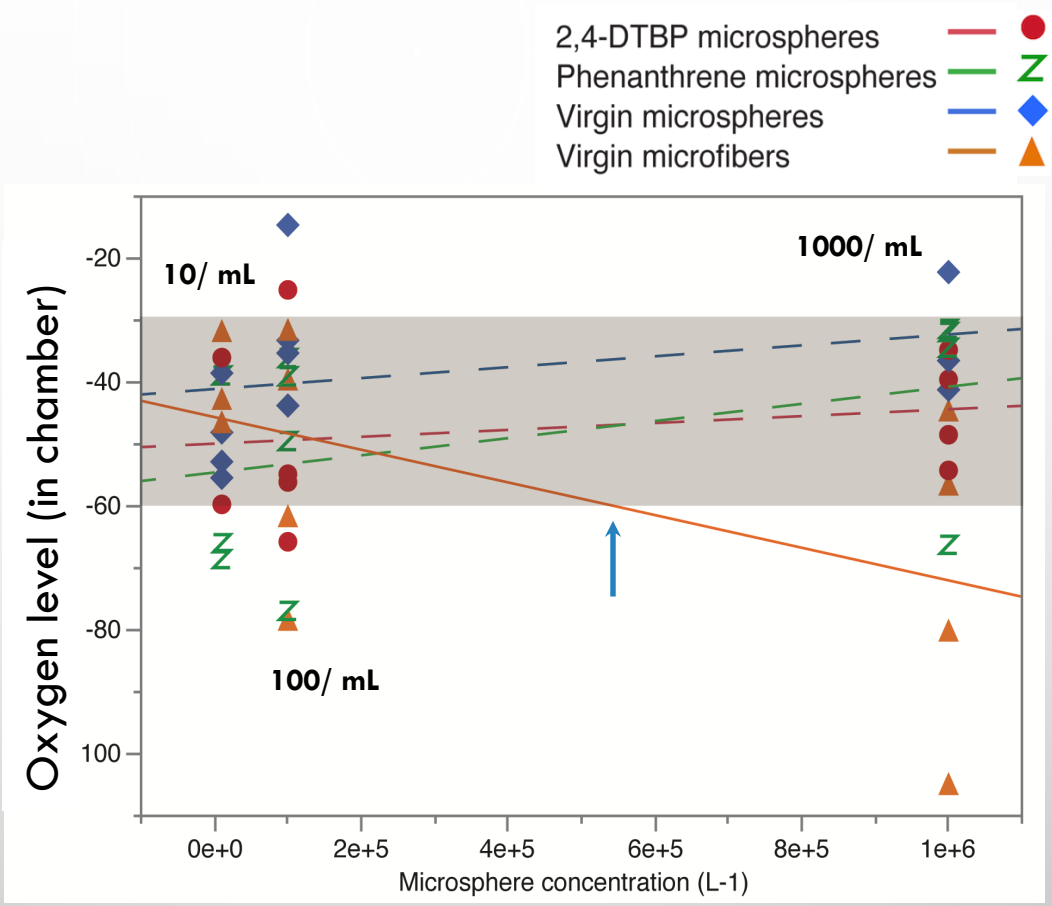


Ankley et al. 2010, Ankley and Edwards 2018, Yong et al. 2020

Shape specific effects on respiration …



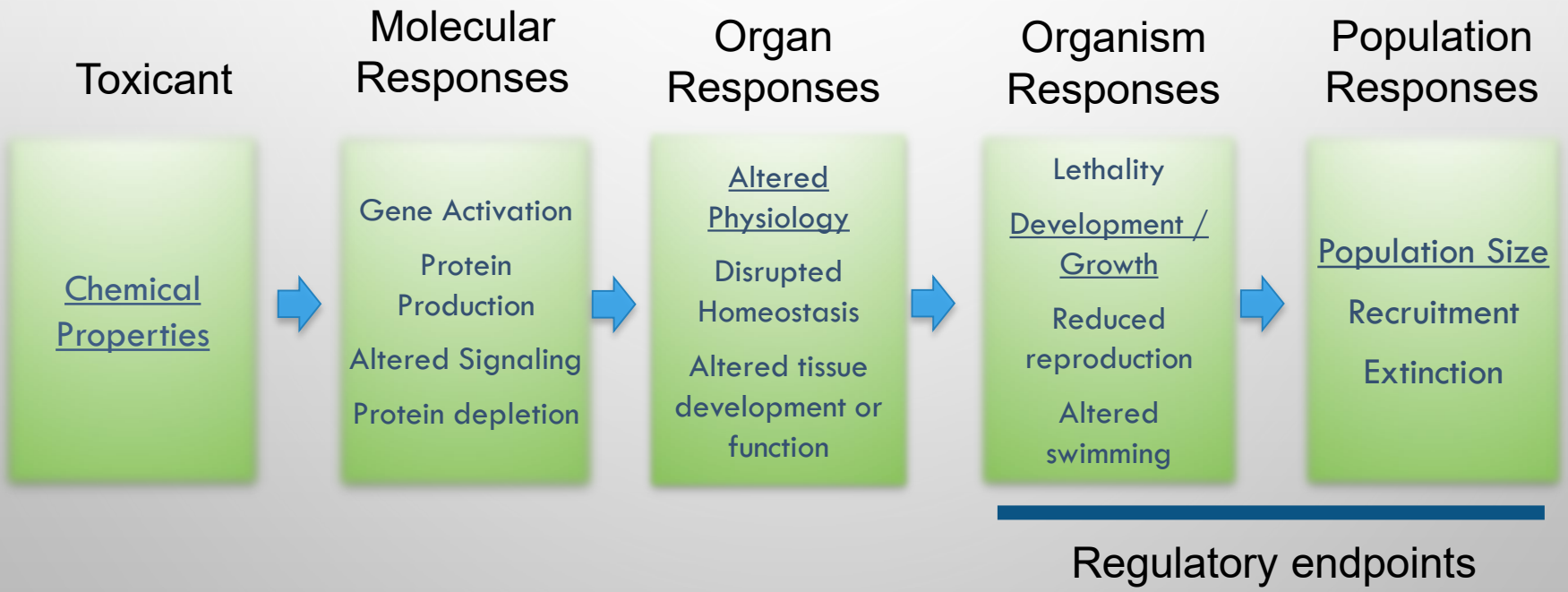
Linear Regression $p < 0.05$

C. *striata* exposed to virgin microfibers exhibited a significant increase in oxygen consumption (n=4)

Stienbarger et al. to be submitted to *ET&C*

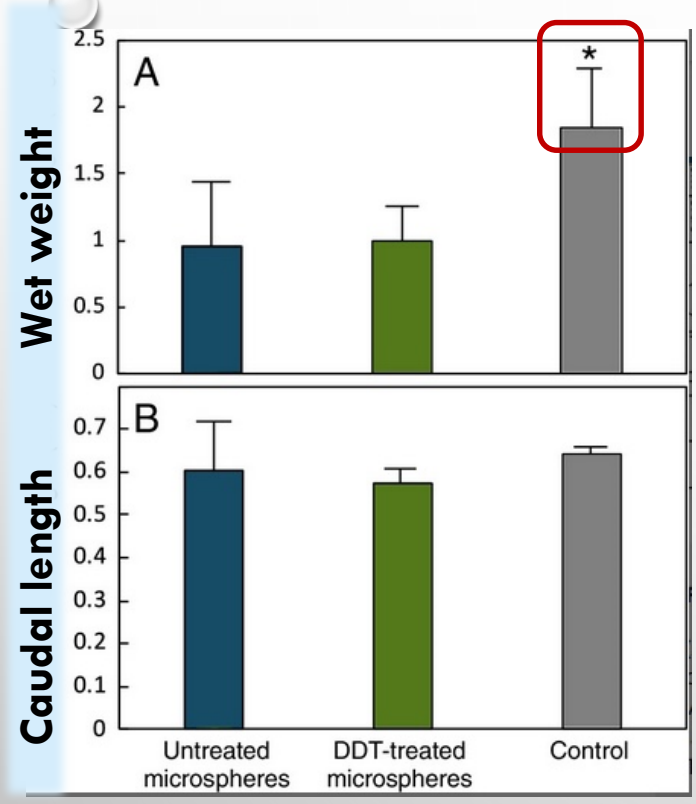
Assessing risk …

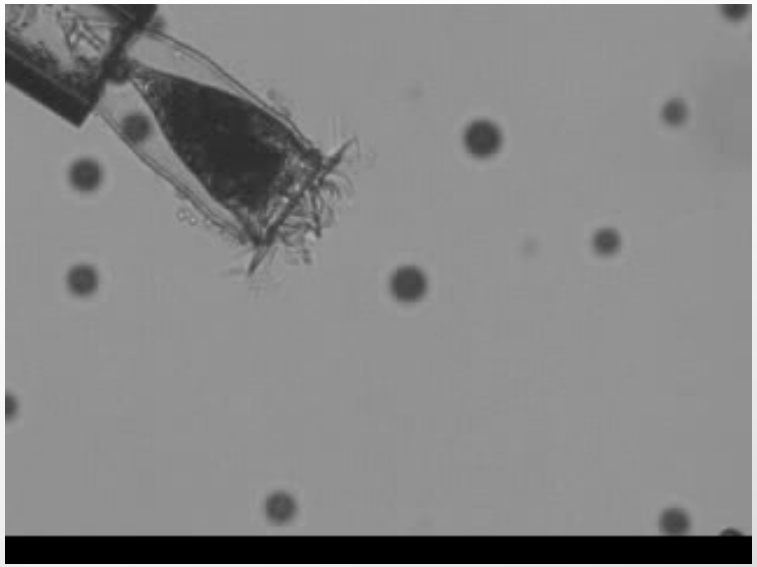
The challenge in applying AOPs to microplastics (and eventually nanoplastics) is that an AOP is defined as a series of key events that begins with a molecular initiating event. Microplastics are a diverse group of contaminants that don't have an easily defined MIE and instead appear to induce a more general oxidative stress response.



Ankley et al. 2010, Ankley and Edwards 2018, Yong et al. 2020

Early life stage effects, trophic transfer…





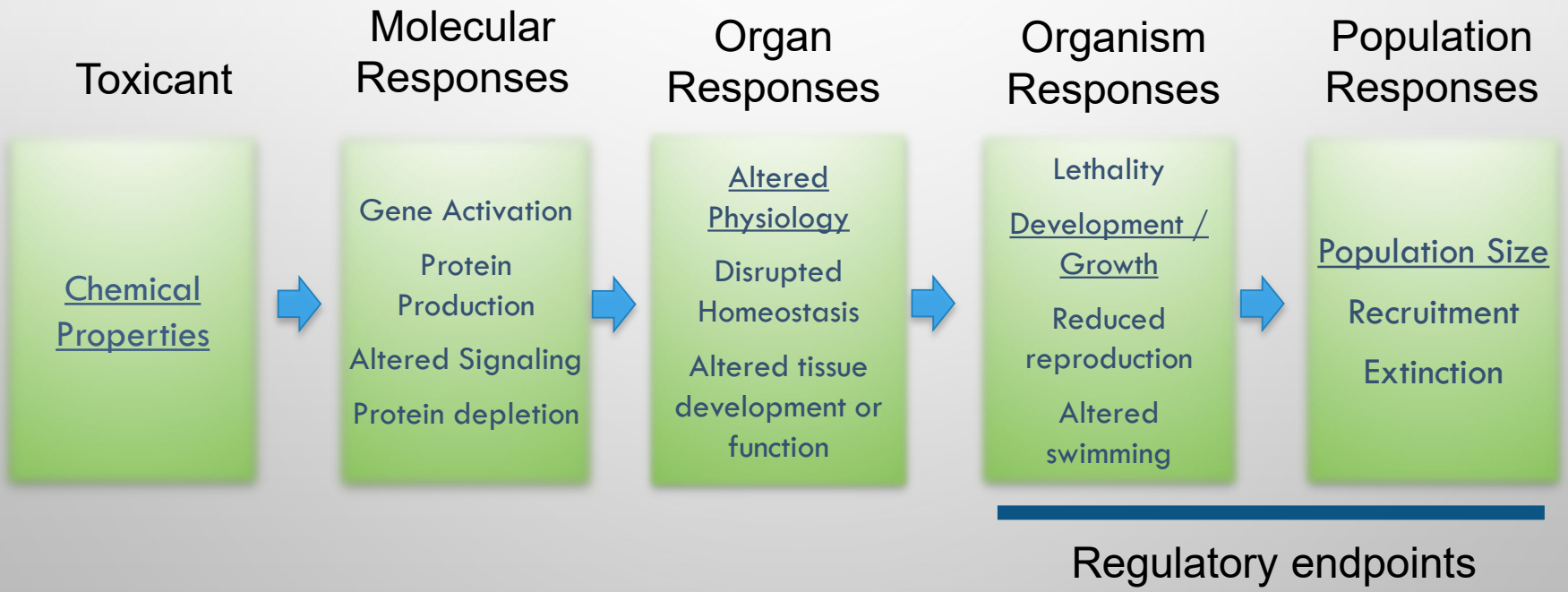
Compared larval *Menidia beryllina* fed virgin or DDT-contaminated plastics from water or ciliate prey for 2 hr period

More microplastics ingested via prey, mass of fish consuming10-20 μm LDPE plastics from water for 2 hrs ($5x10^5$/mL) after two weeks being reared in clean water was lower compared to controls.

Athey et al 2020, *Lim Oce Letters*

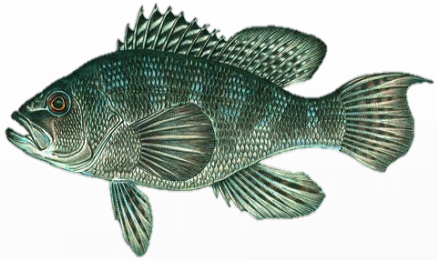
Assessing impacts across levels of organization …

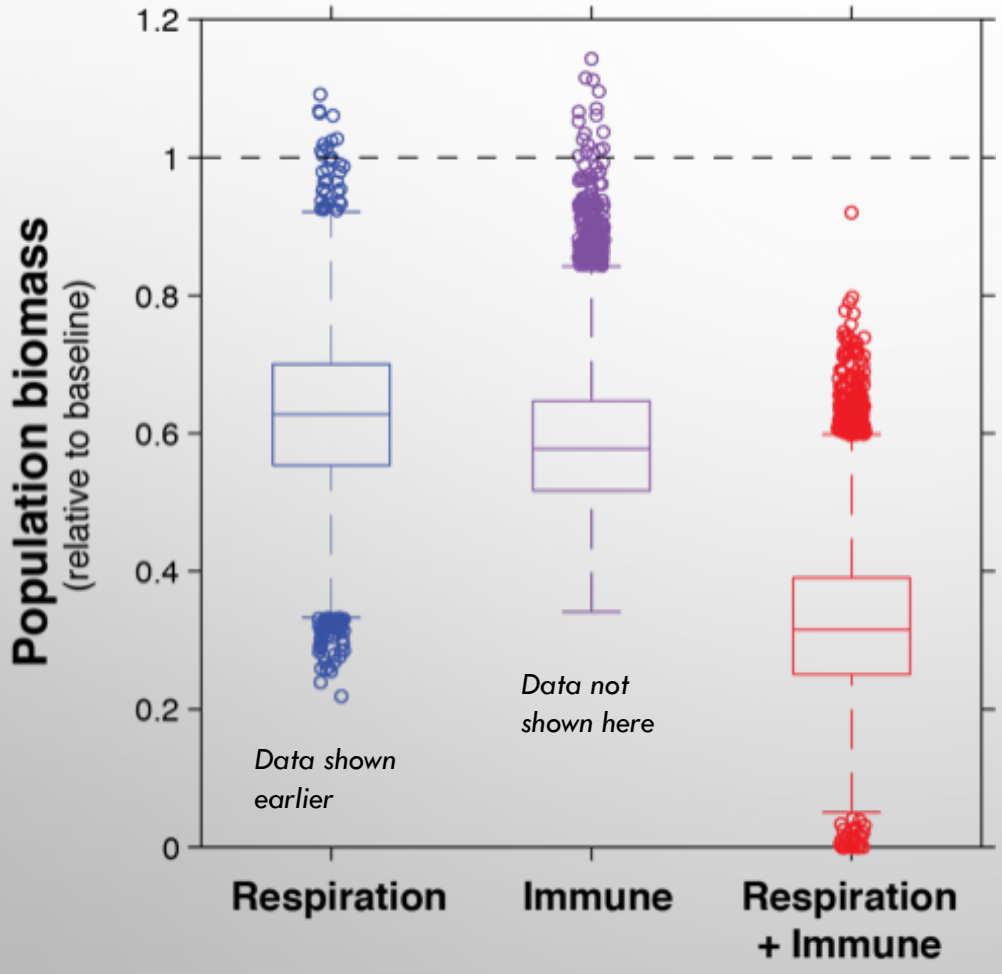
The challenge in applying AOPs to microplastics (and eventually nanoplastics) is that an AOP is defined as a series of key events that begins with a molecular initiating event. Microplastics are a diverse group of contaminants that don't have an easily defined MIE and instead appear to induce a more general oxidative stress response.



Ankley et al. 2010, Ankley and Edwards 2018, Yong et al. 2020

Extrapolating to population level effects …





- Age-structured black sea bass population model
- **High-exposure scenario:** acute effects (from mid-concentrations) seen in each generation
- Decreased immune response →increased mortality
- Increased respiration → decreased growth rate

Easter et al. 2020, White et al. 2017, Brander and White in prep.

Current work…

Fundamental Research

Exposure Characterization

Effects Analysis

Tools and Methods Development

GOAL: To provide foundational information about what physicochemical properties (e.g., composition, micro, nano sizes, shape) and environmental components (e.g., salinity, primary and secondary producer characteristics, lipid content, transformations) are determinative of risk.

Modeling

Community Engagement

Government Agencies & Regulators

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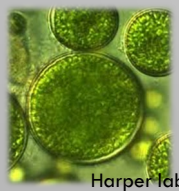
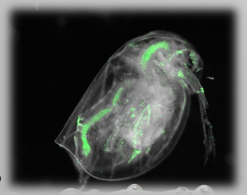
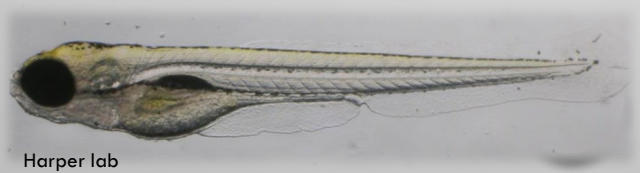


Harper lab





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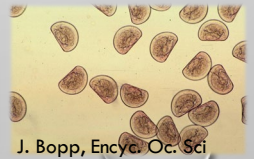
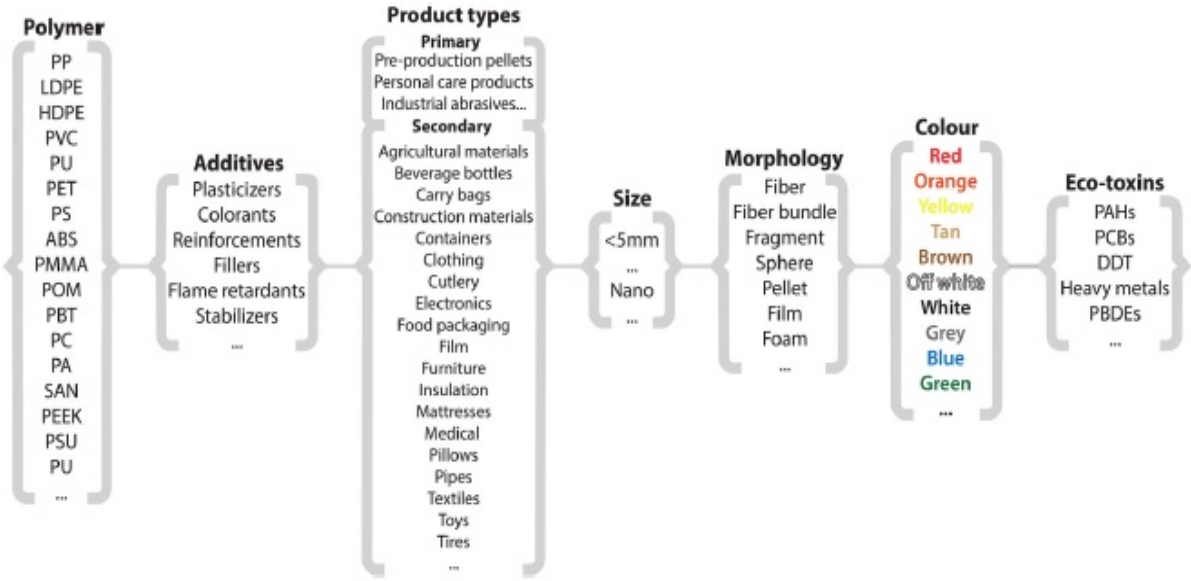


J. Bopp, Encyc. Oc. Sci

Focus on shape, size and abiotic factors such as salinity…



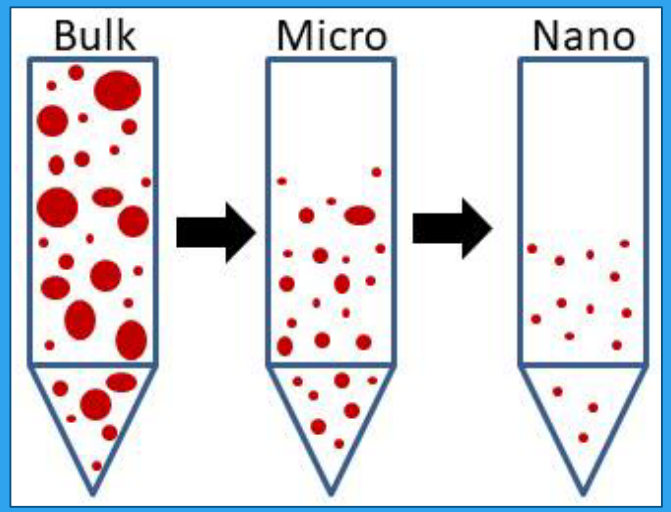
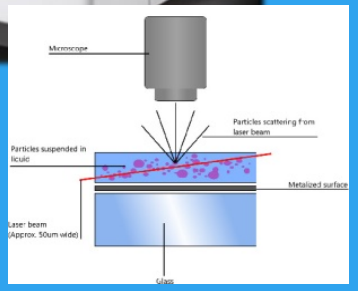


Image from B. Harper, OSU



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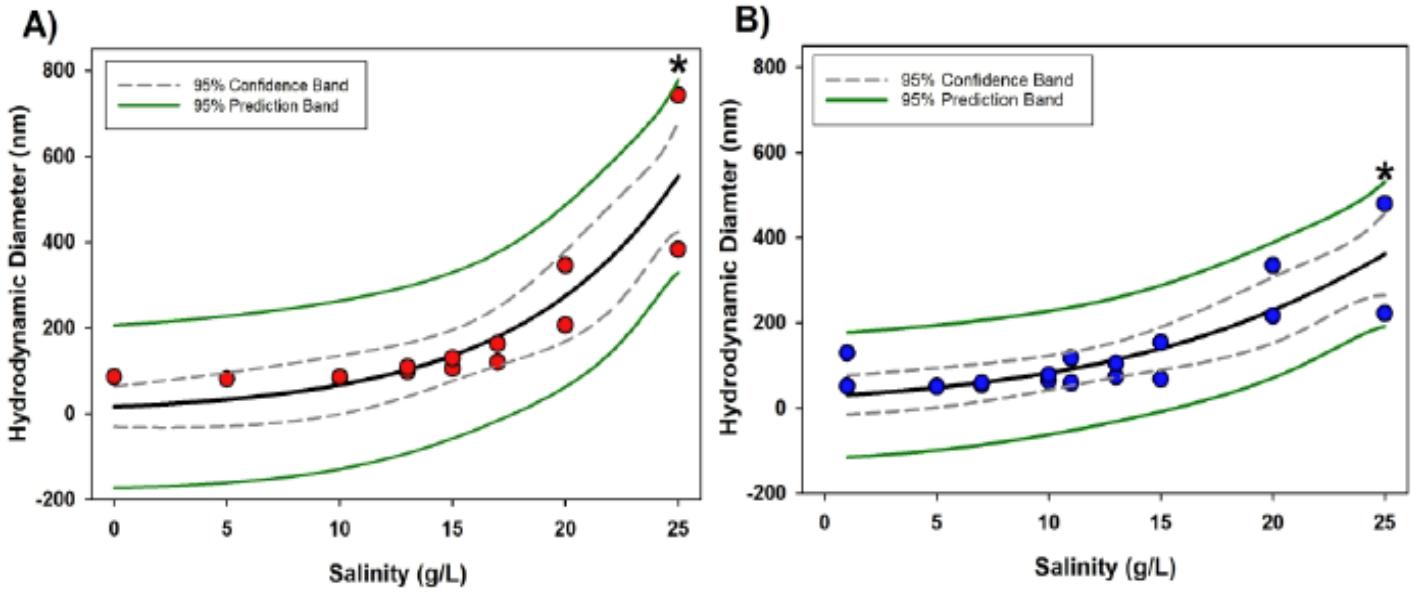
Nanoplastic agglomeration increases with salinity …



Hydrodynamic diameter comparison of 50 nm Visiblex™ blue and red color dyed PS nanospheres in 0-25 g/L salinity gradient. Grey dashed and solid green lines indicate 95% confidence and prediction bands, respectively for regression fit shown as solid black line. Asterisk (*) represents a significant change in hydrodynamic diameter.

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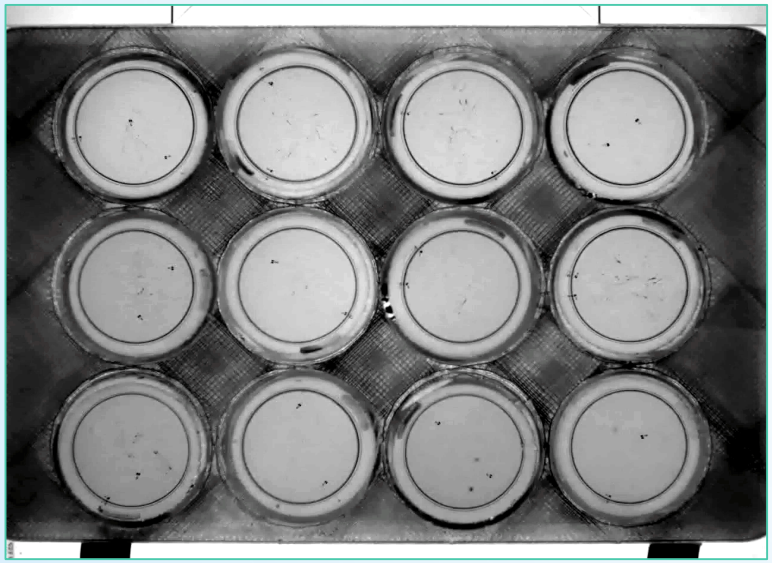
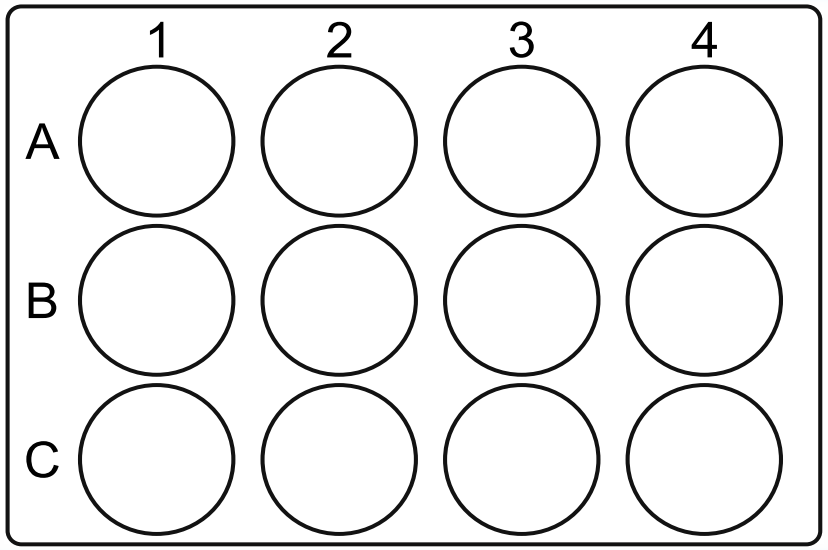
Boenisch et al. in revision, *ET&C*

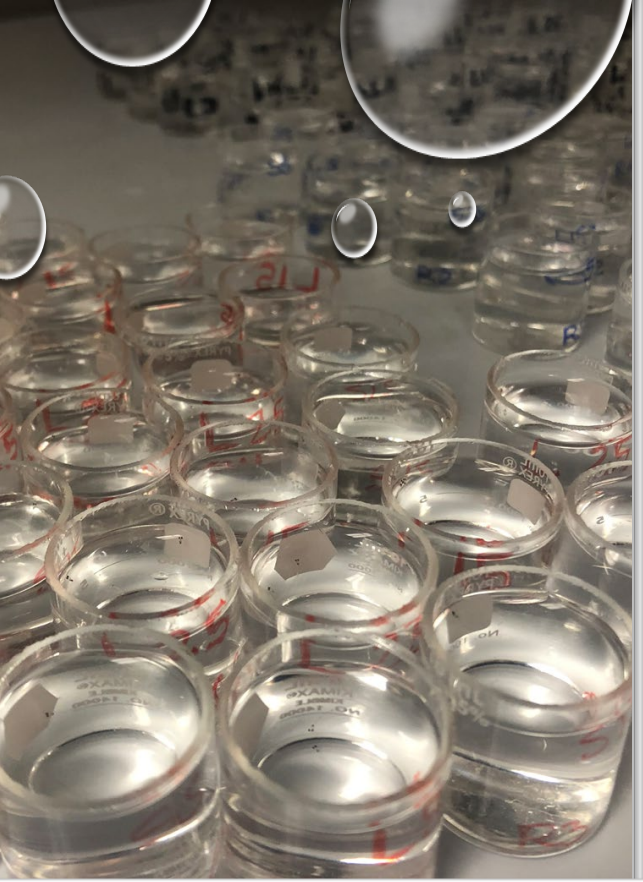
So many plastics, so little time…

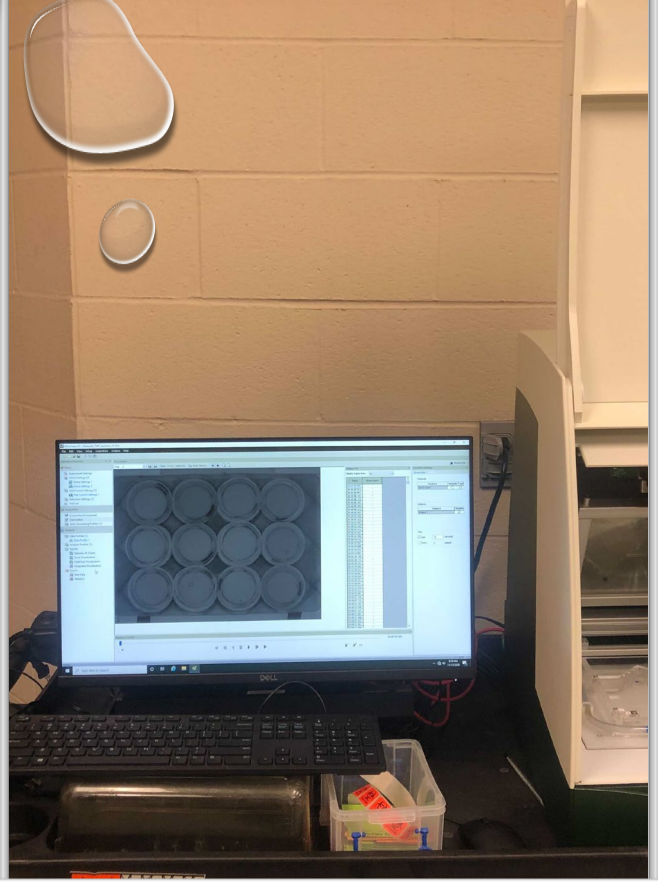


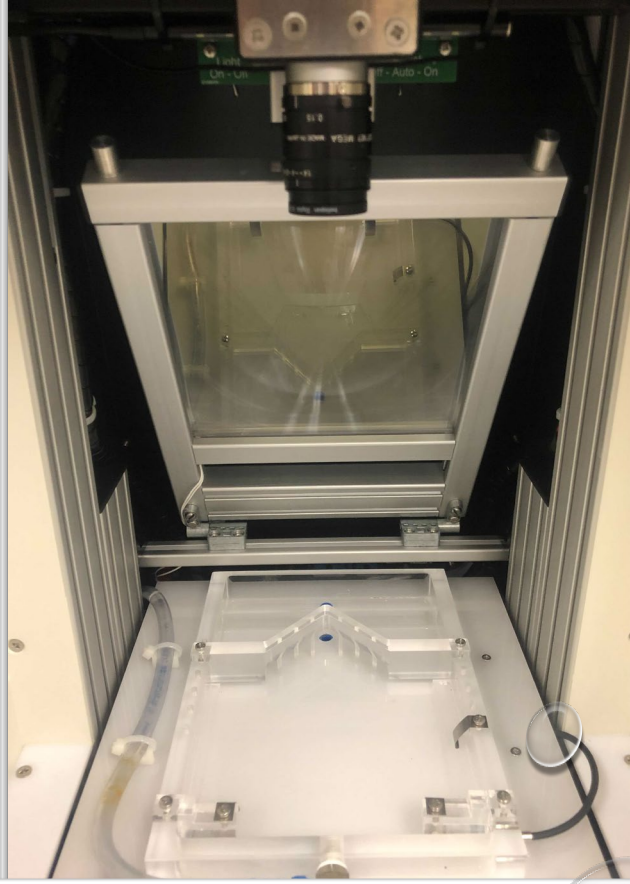


High throughput testing using adapted fish early life stage toxicity tests across plastic types, sizes (micro and nano), and concentrations … our lab group focuses on the estuarine model species *Menidia* and invertebrate *Americamysis bahia* (mysid shrimp).

https://www.thetimes.co.uk/article/fish-larvae-eat-plastic-washed-into-the-sea-2whsn63qk







*Danio*vision Observation Chamber

(aka *Menidia*vision)

Microplastic exposure can cause alterations in swimming.

Daniovision, widely used for behavior tracking, carries out pattern analysis on video images of observed animals.

Mundy et al. 2020, Frank et al. 2019, Pannetier et al. 2019, Choi et al. 2020

Inland silverside

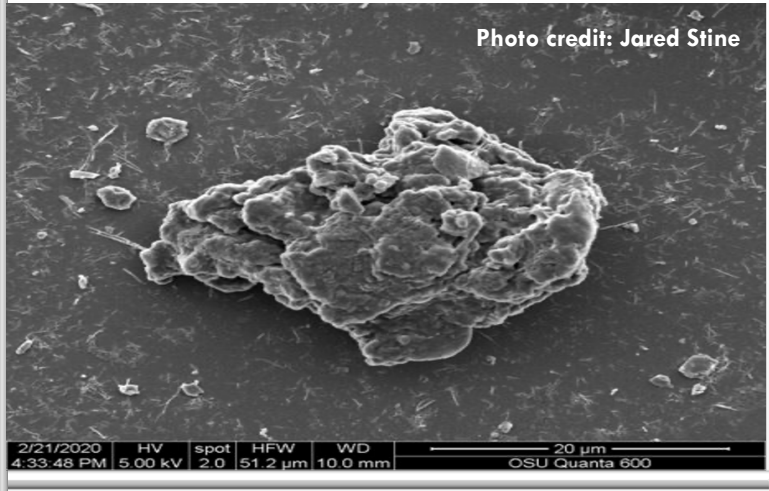
Tire wear particle exposures

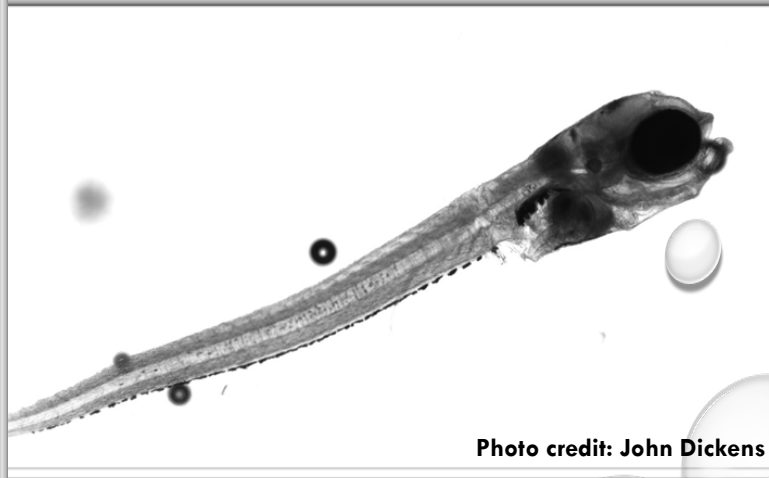
TWP now known to be a sizeable fraction of microplastic pollution in urban estuaries (e.g. Charleston SC, San Francisco, CA USA), and is detected in euryhaline fish species (e.g. menhaden).

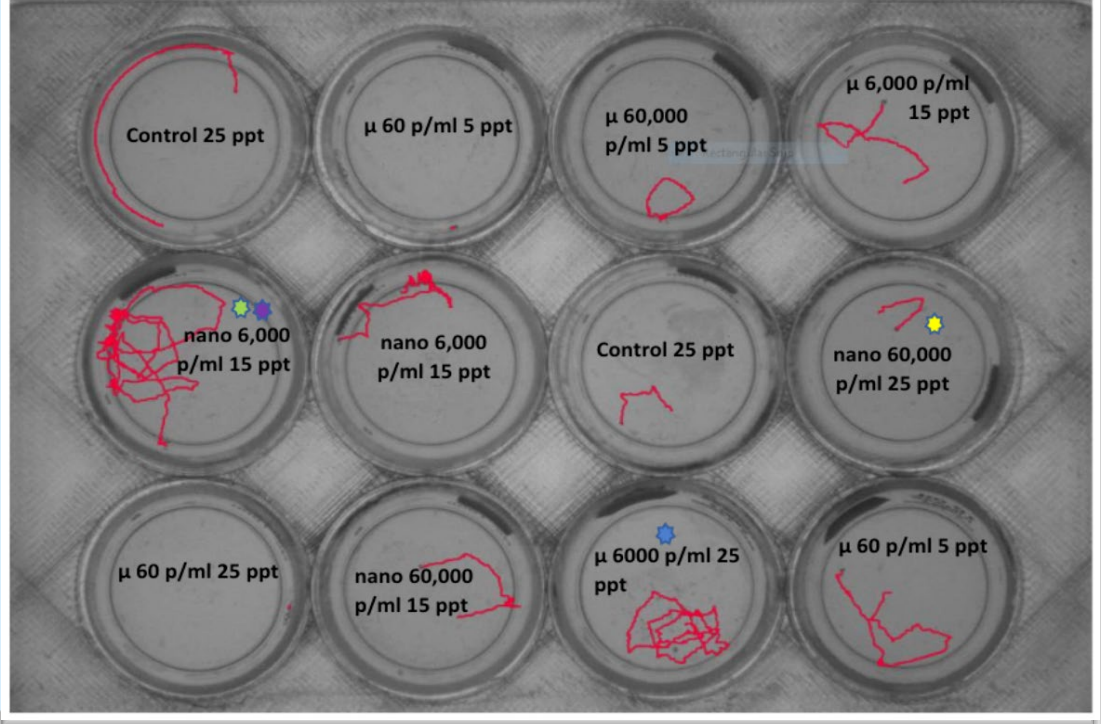
Exposed *Menidia* to micro (1-20 µm) and nano (<1 µm) fractions (produced onsite) across 3 salinities to 60, $6x10^3$, and $6x10^5$ particles per mL.

Embryos hatched out into wells containing plastics, exposed for 96 hours post hatch with daily renewal.

Parker et al. 2020, Leads & Weinstein 2019, Gray et al. 2018, SFEI 2019



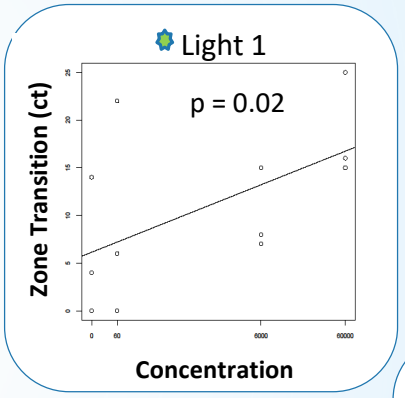


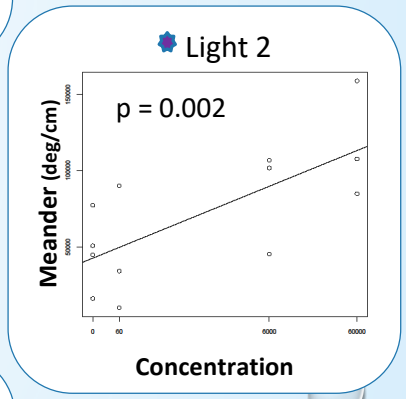


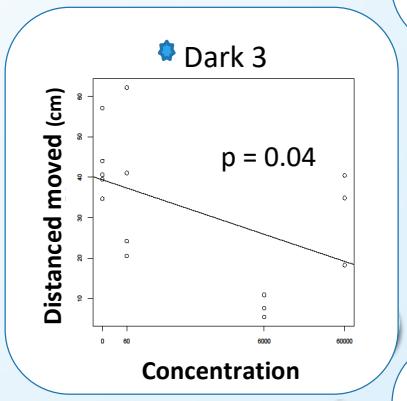
Results

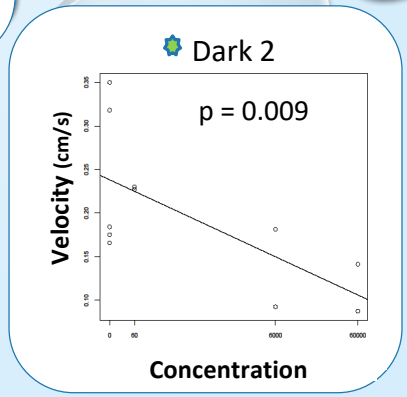
Generally silversides exposed to both nano and micro fraction exhibit an increase in erratic swimming behavior with increasing concentration

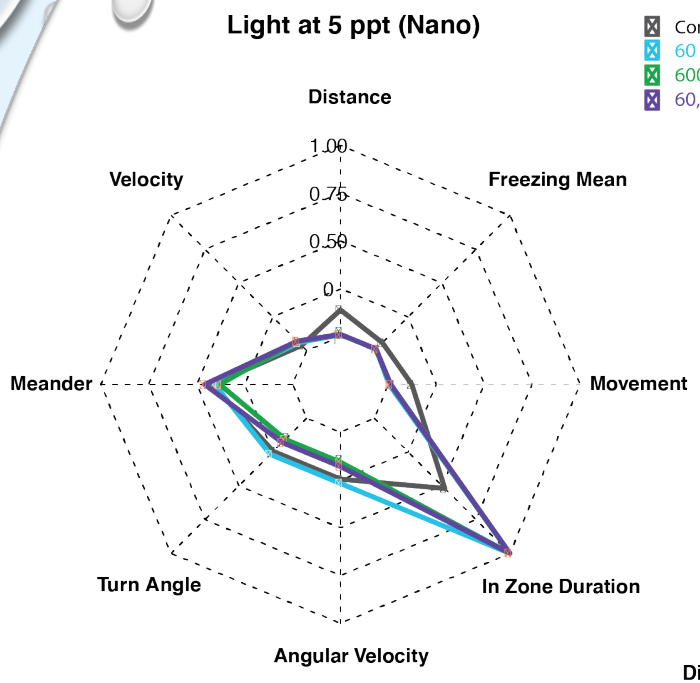
This has been seen in other fish species such as medaka and sheepshead minnows

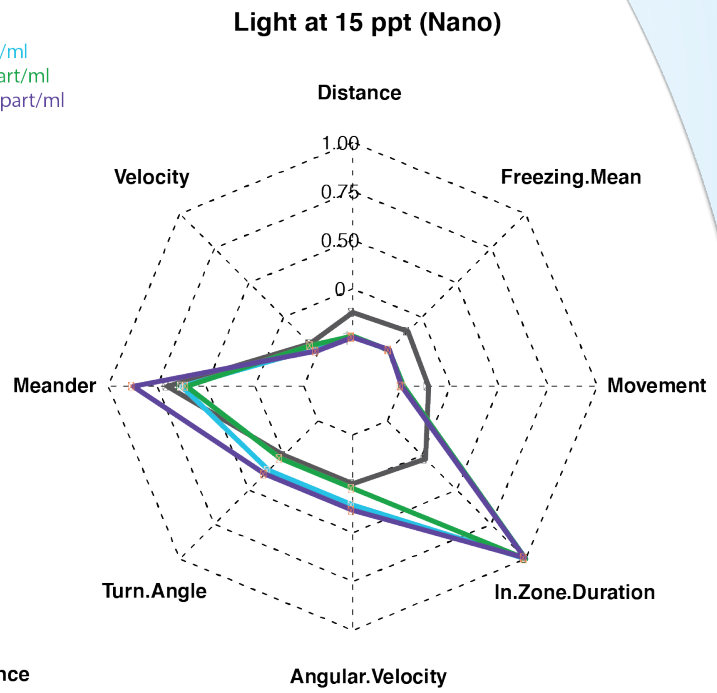






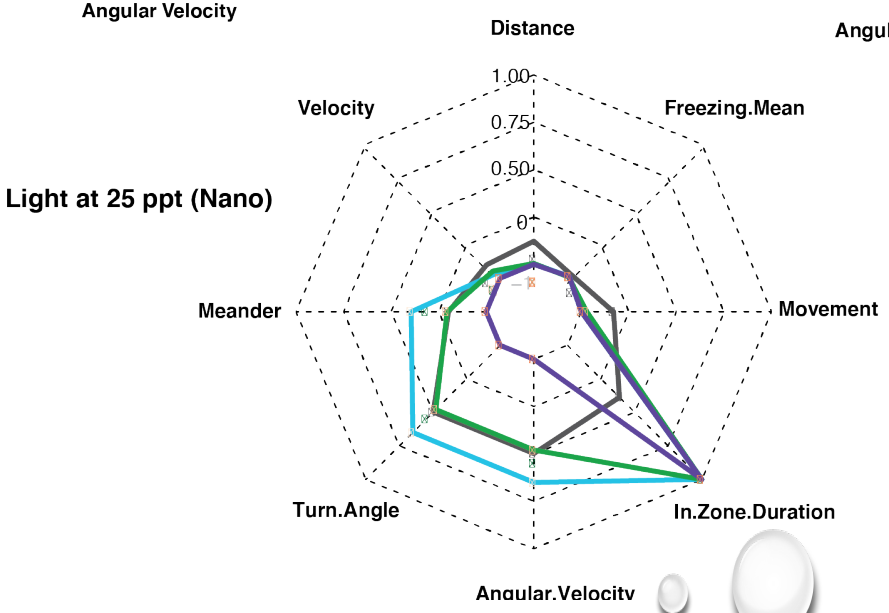






Results

Salinity may have subtle impacts on behavioral responses



Summary & Future directions

Silverside cleared with CUBIC™

Photo credits: John Dickens

TWP + salinity may impact behavior, salinity, analyses ongoing

Estuarine results as well as freshwater and marine to be incorporated into Risk Assessment Framework

High throughput tests with additional polymers in Menidia and mysid shrimp.

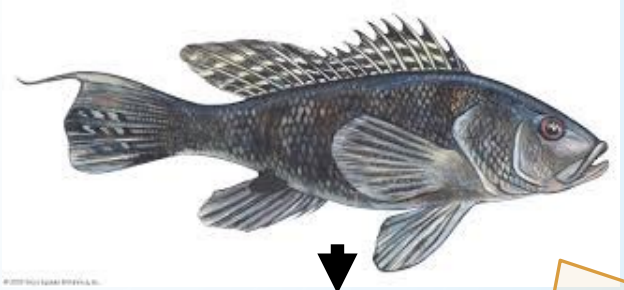
Pacific Northwest Consortium on Plastics

Prioritize for chronic repro & F1 impact testing

RNA seq across polymer types, other endpoints

Next steps: RNAseq Analysis

1. Raw RNA seq reads will be aligned to a reference transcriptome
2. Alignment reads will be assigned to genes
3. Quantify differential gene expression
4. Finally, functional analysis is performed to reveal molecular mechanisms of toxicity



mRNA

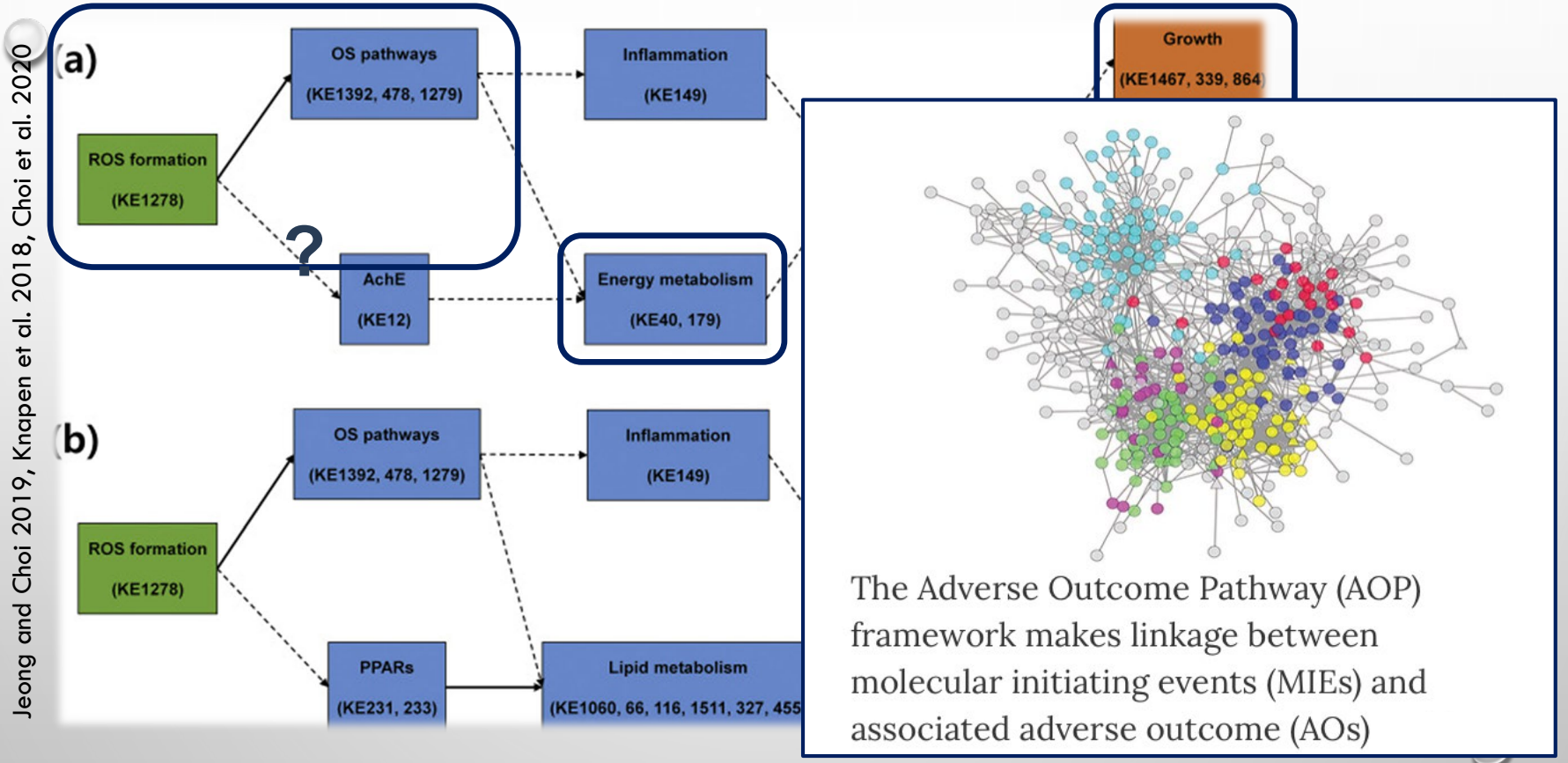
cDNA

Sequence

Align/Quantify DEG's! Functional analysis

Reference transcriptome

On the whole looking across studies …



We see evidence from our work and that of others that fits with this putative AOP, there may be some overlap between polymer types, shapes, and sizes but an eventual AOP will be a network with many nodes.

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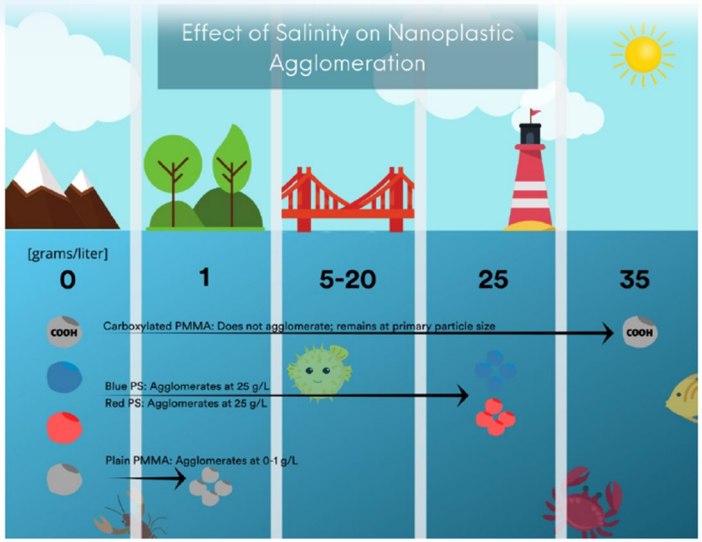
More at www.pnwmicroplastics.org



Nanoplastic agglomeration behavior across experimental salinity gradients varies depending on plastic type and surface chemistry.

PLASTICAST

PLASTICAST is a media project within the PNW Consortium on Plastics. Hosted by John Dickens, M.S. student at Oregon State University, PLASTICAST is an episodic series featuring scientists, legislators, and all interested stakeholders on the latest in plastic pollution research and policy.

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PNW Consortium at SETAC SciCon2:
https://t.co/DFw4Bd3mda?amp=1

www.pnwmicroplastics.org

Next teleconference 11/25 3pm PST