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science for a changing world

Great Lakes RESTORATION

Development of tools to control filter-feeding aquatic invasive species including Asian carps and dreissenid mussels.

M.P. Gaikowski, J.J. Amberg, T.D. Hubert and J.A. Luoma
USGS Upper Midwest Environmental Sciences Center

U.S. Department of the Interior
U.S. Geological Survey

U.S. Geological Survey

Federal source for science about the Earth, its natural and living resources, natural hazards, and the environment.

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U.S. Geological Survey

Six focus areas: Climate and Land Use Change, Core Science Systems, Ecosystems, Energy, Minerals and Environmental Health, Natural Hazards and Water.

Provides Scientific support to the Department of Interior agencies that manage natural resources, such as the U.S. Fish & Wildlife Service, National Park Service, and Bureau of Land Management.

18 biology-focused research Centers across the Nation.

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USGS-UMESC is an integrated research facility

Mission – UMESC provides the scientific information needed by managers, decision makers, and the public to protect, enhance and restore the ecosystems in the Upper Mississippi River Basin, the Midwest and worldwide.

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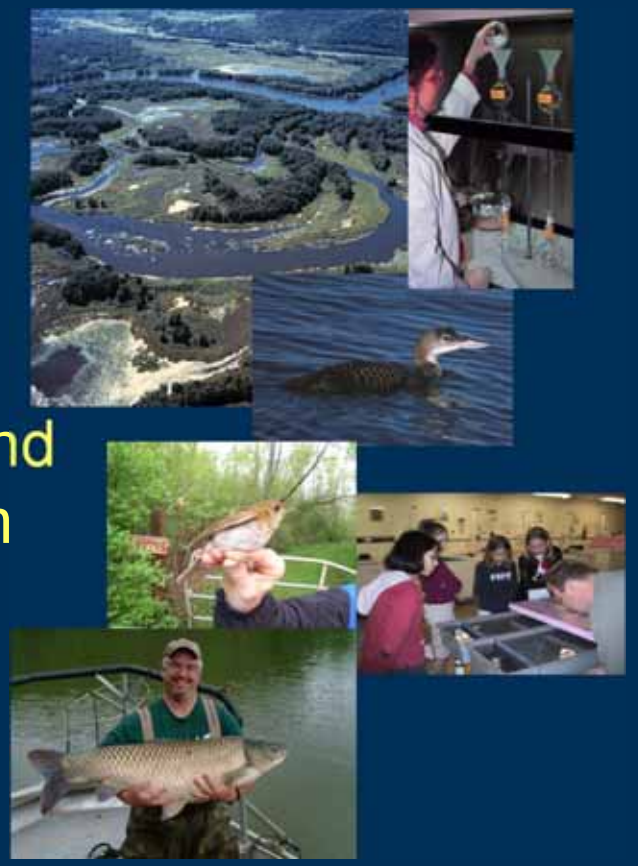
UMESC Research and Monitoring Themes

1. Large River Ecosystems
2. Geospatial Sciences and Decision Support System Development
3. Invasive Species Control
4. Fisheries Management Chemical and Drug Development and Registration
5. Endangered Species
6. Contaminant Effects on Wildlife



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USGS Invasive Species Research

- Prevention of AIS introduction
- Early detection / rapid assessment of AIS
- Monitoring & forecasting of AIS distribution
- Effects of AIS on native species
- Control / management of AIS
- Information dissemination

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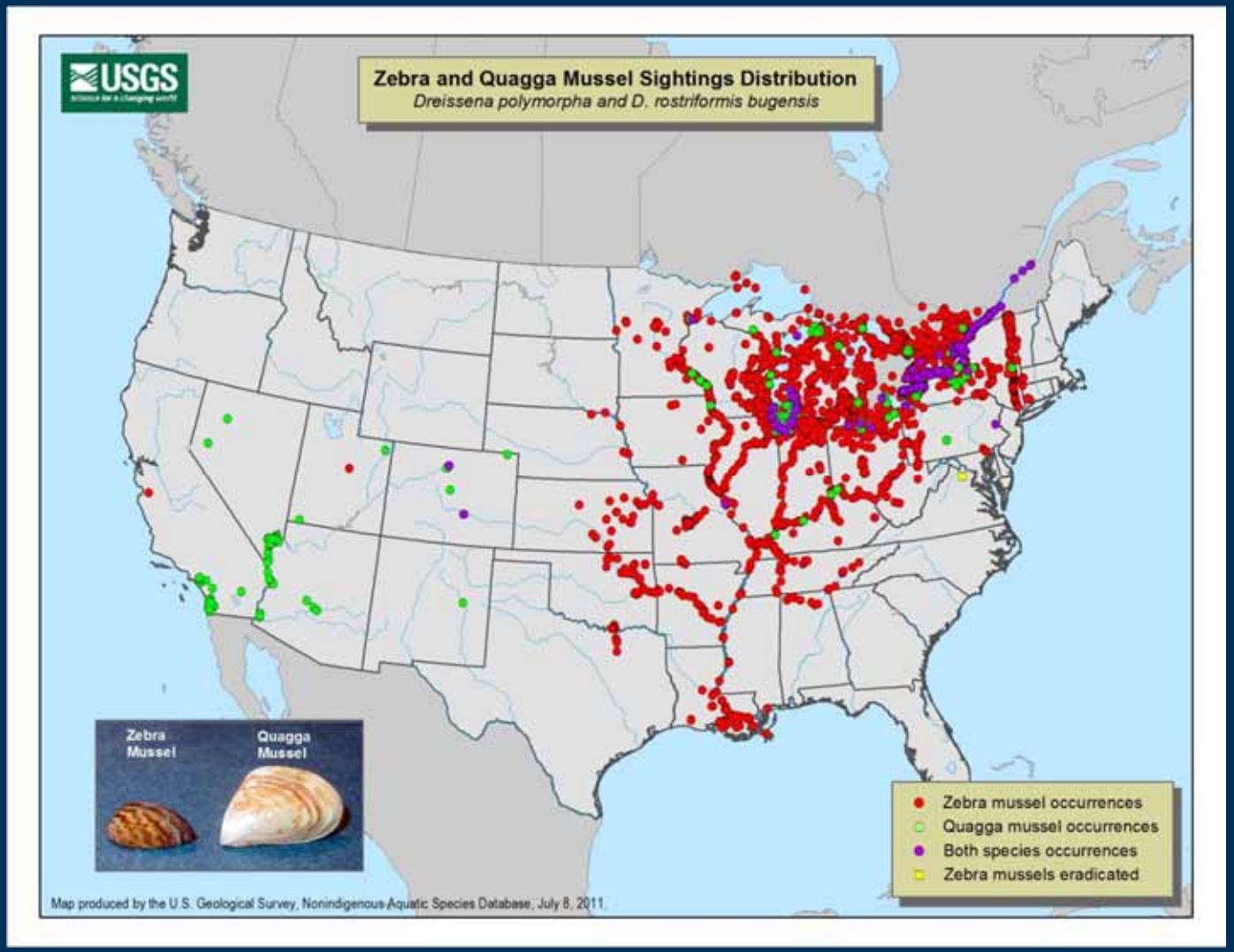
Dreissenid mussel distribution



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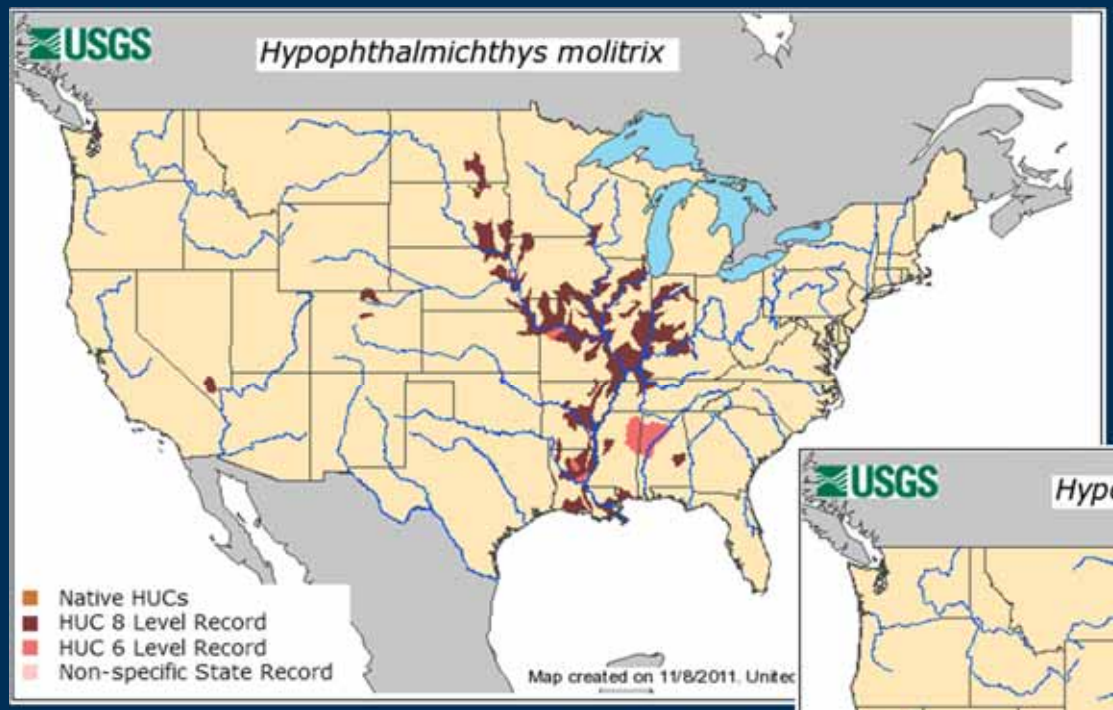
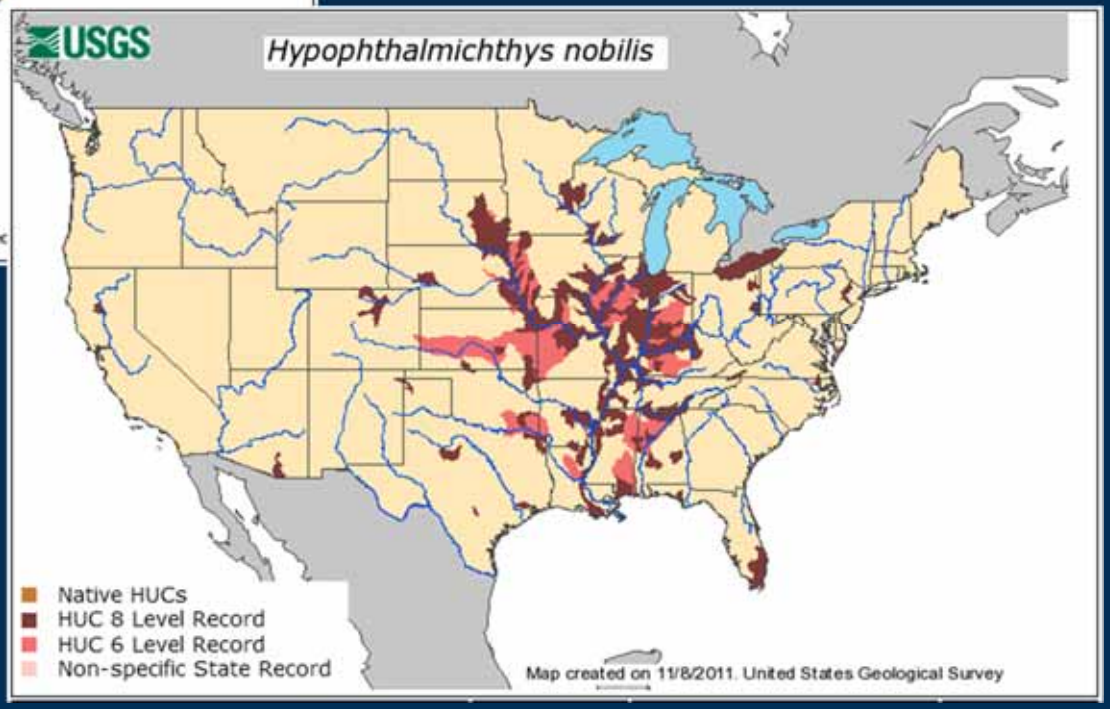
Asian carp distribution



Silver carp



Bighead carp

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Asian carp distribution

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AIS control challenges

Four current biocides

- antimycin
- rotenone
- 3-trifluoromethyl-4-nitrophenol (TFM)
- niclosamide

Minimal specificity

- TFM & niclosamide for sea lamprey
- Non-target effects

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Success for AIS control tools

1. Selective
   - Limited effect on native species
2. Scaleable
   - Mussel beds to lake trout spawning beds
   - Backwaters to large rivers
3. Economics
   - Application $ << Resource value $

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AIS research goals

- **Develop microparticle formulations for selective delivery of control agents to filter-feeding aquatic invasive species**
- Identify and evaluate potential selective biocides for bighead and silver carp and dreissenid mussels
- Evaluate physical methods to limit Asian carp and dreissenid mussel populations.
- Evaluate current molecular surveillance techniques and develop next-generation molecular surveillance techniques
- **Evaluate the potential of ZEQUANOX® to control dreissenid mussels in open water.**

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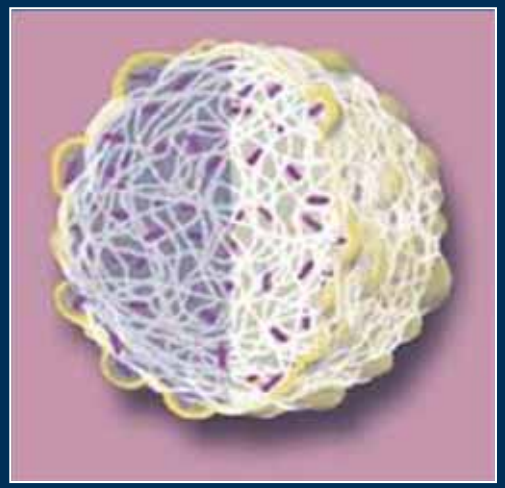
AIS research goal

Develop microparticle formulations for selective delivery of control agents to filter-feeding aquatic invasive species



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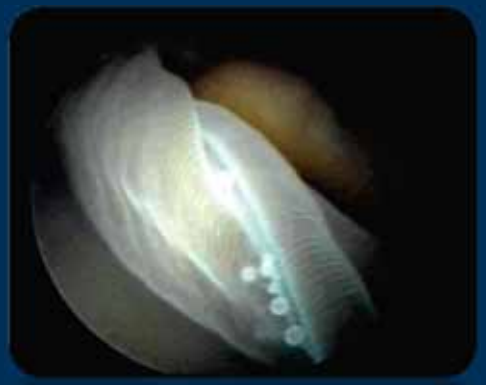
Control agent-laden microparticles

1. ‘Hold’ the agent
2. Appropriately sized
3. Readily consumed
4. Targeted release
5. Scaled production

Black sandshell Zebra mussel



Biobullets® on zebra mussel gill

Biobullet®-laden psuedofeces expelled from native mussel

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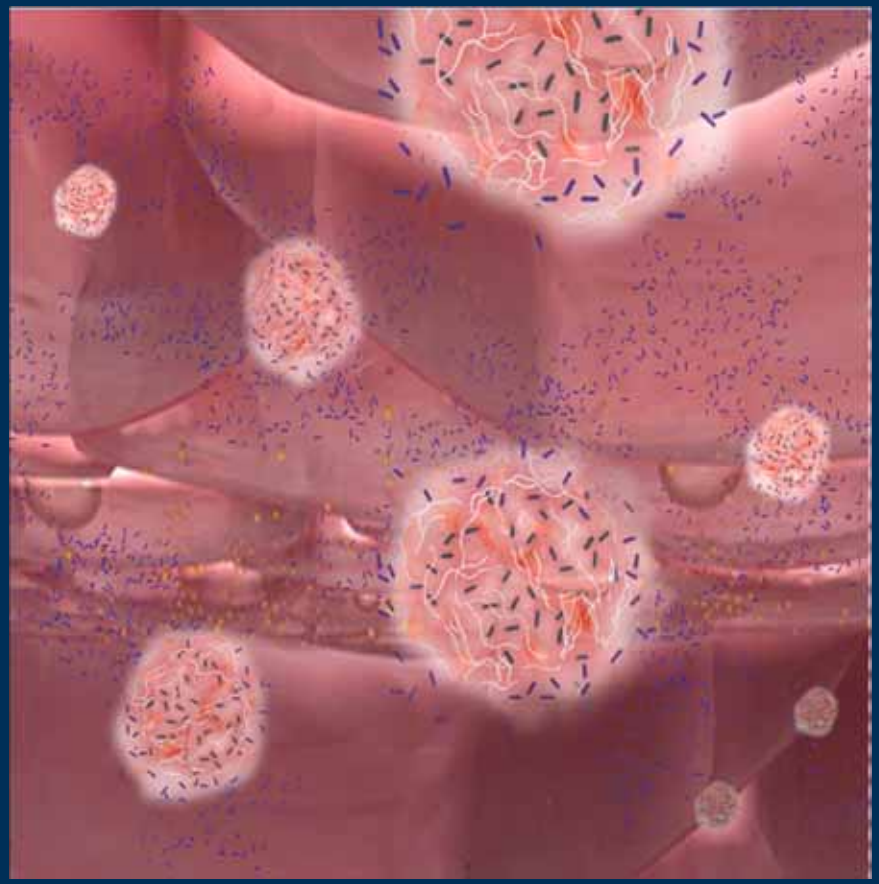
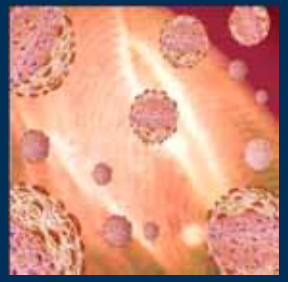
Potential application to dreissenid mussels





Enzymatic release of control agent
in targeted species

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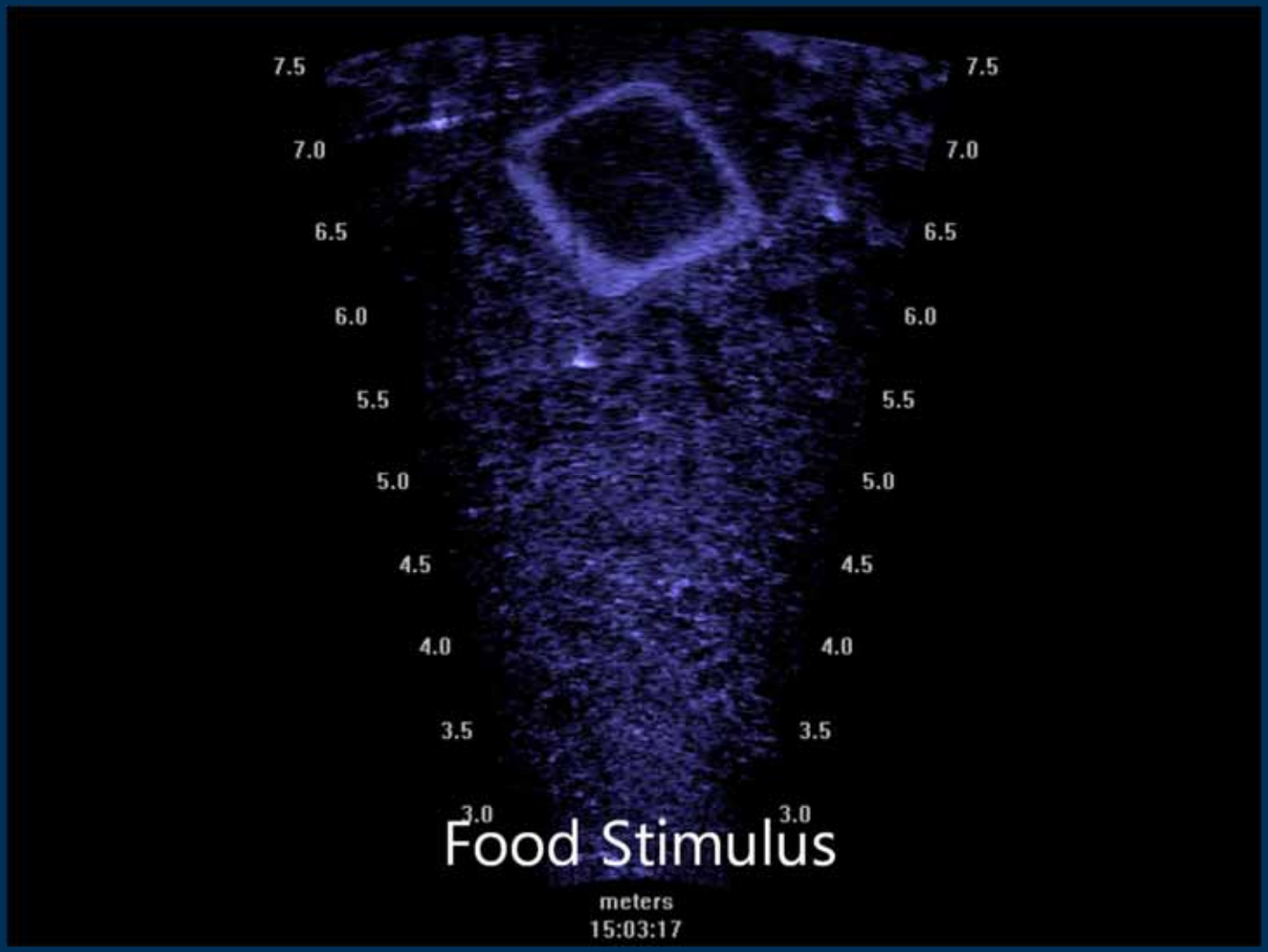
Potential application to Asian carp



Video courtesy
Dr. Ed Little,
USGS-CERC

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Developing microparticles

1. Feeding characteristics
   - Filtration rate
   - Size selectivity
2. Digestive processes
   - pH / Digestive enzymes
   - Temporal changes
3. Control agent
   - Selection
   - Loading
4. Registration

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Developing species-specific microparticles for Asian carps or dreissenid mussels

Current research:

- Compare digestive enzyme activity in AIS versus native fish
- Evaluate particle retention by AIS
- Determine lethal control agent levels for AIS vs native fish or mussels

Future research:

- Evaluate microparticles to control Asian carp or dreissenid mussels and non-target effects on native fish
  - Lab, field, and environmental fate
- Register microparticles with EPA

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Digestive physiology

- **Mussels**
  - **Mississippi River (Winona, MN)**
    1. **Zebra mussel *Dreissena polymorpha***
    2. **Threeridge *Amblema plicata***
    3. **Pocketbook *Lampsilis cardium***
- **Fish**
  - **Illinois R. (IL), Wabash R. (IN), Jim R. (SD)**
    1. **Bighead carp *Hypophthalmichthys nobilis***
    2. **Silver carp *H. molitrix***
    3. **Gizzard shad *Dorosoma cepedianum***
    4. **Bigmouth buffalo *Ictiobus cyprinellus***
- **2010-2012 (bi-monthly depending on flow)**
- **Digestive system enzymes assayed**
- **Expanding to larval aquatic insects**

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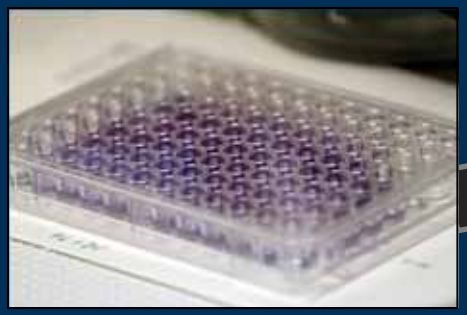
Enzyme assays – AIS vs native species



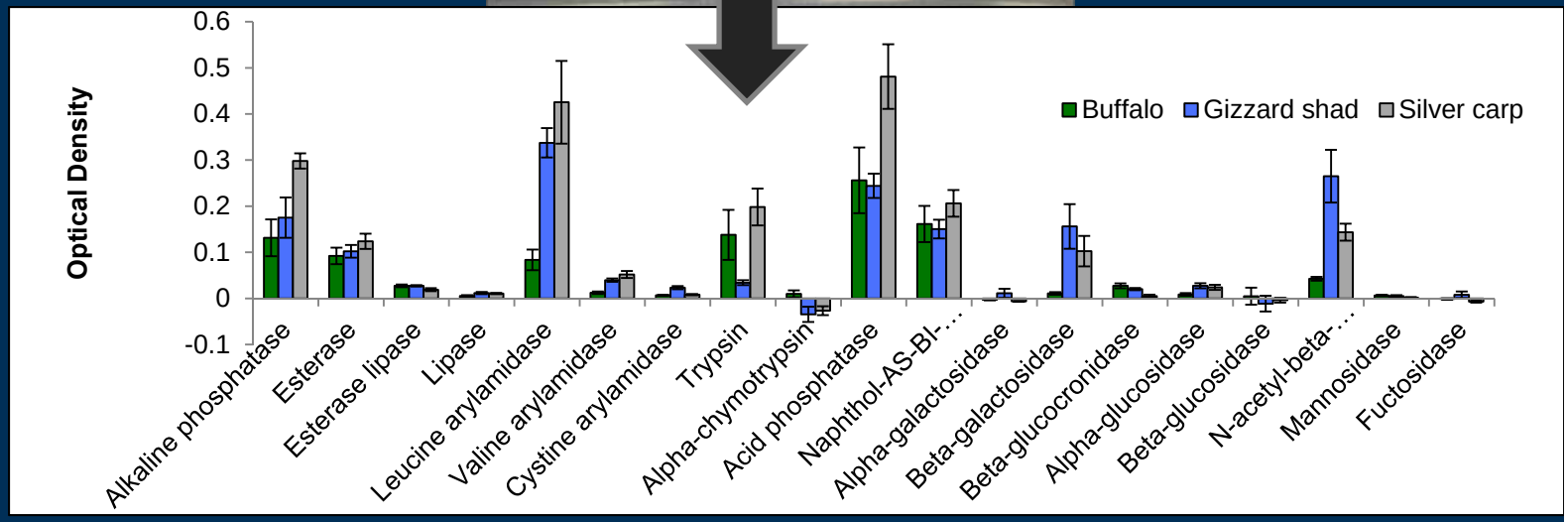
19 digestive enzymes
Normalized to equal amounts of protein
Zebra mussels vs. two native mussel species
Silver carp vs. two native planktivorous fish

Gizzard shad

Bigmouth buffalo

Silver carp

Easy to visualize
>intensity = >enzyme



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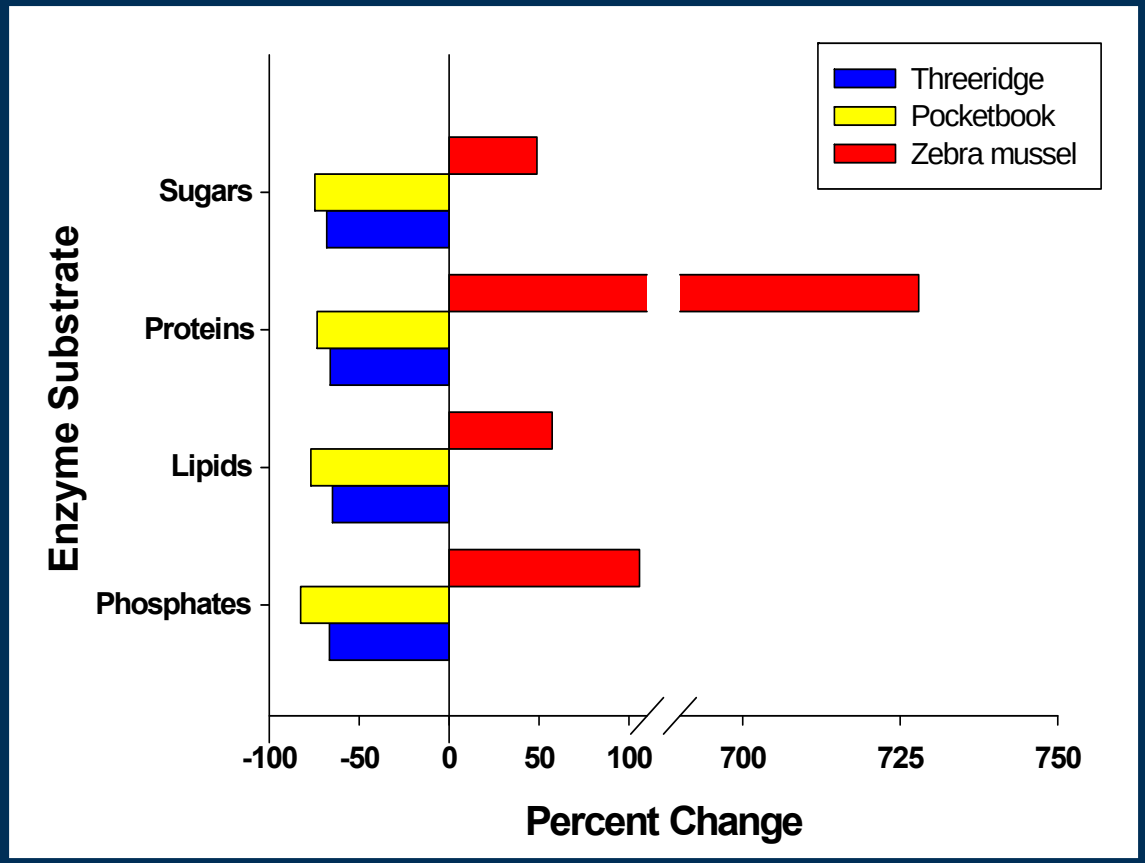
Change in mussel enzyme activity



- Samples collected August & September (2010)
- 7°C change between collections
- Expanded to more seasons, locations and aquatic insects

Bars = 10 individuals

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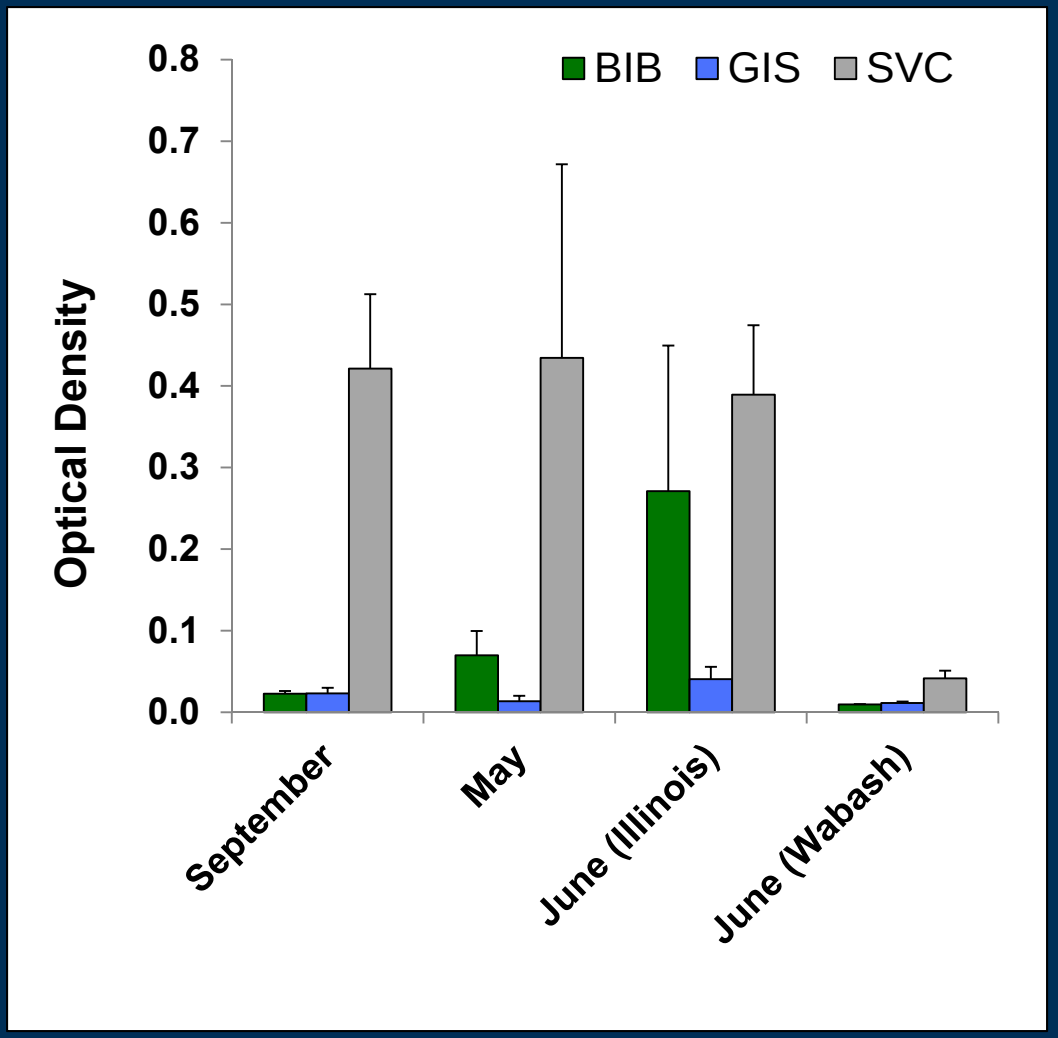
Silver carp vs native planktivores

β-galactosidase

Converts lactose to glucose and galactose

Common source: Plants

*Commonly used in molecular biology to screen bacteria



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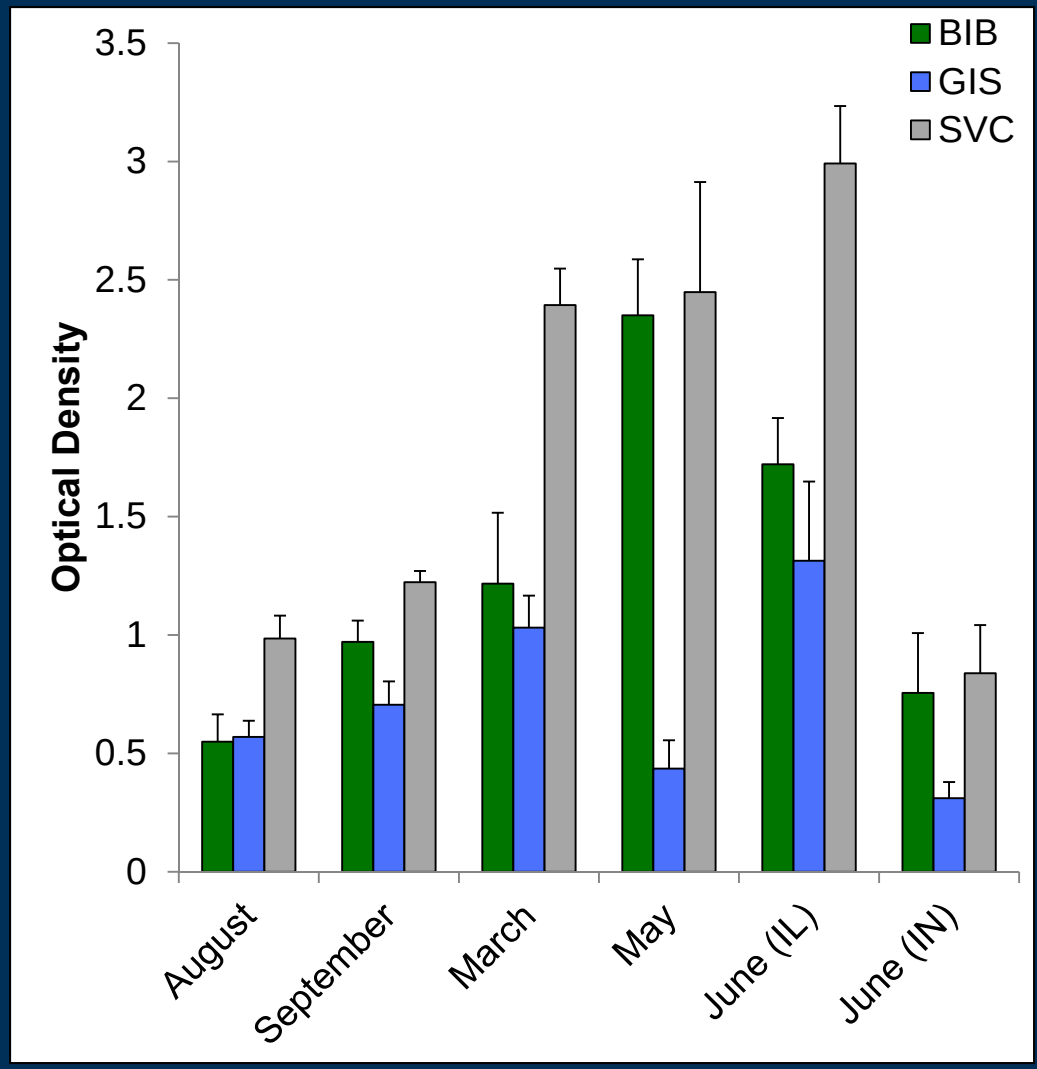
Silver carp vs native planktivores

Phosphatases
Higher in SVC in August, September and March

Always higher in SVC compared to GIS



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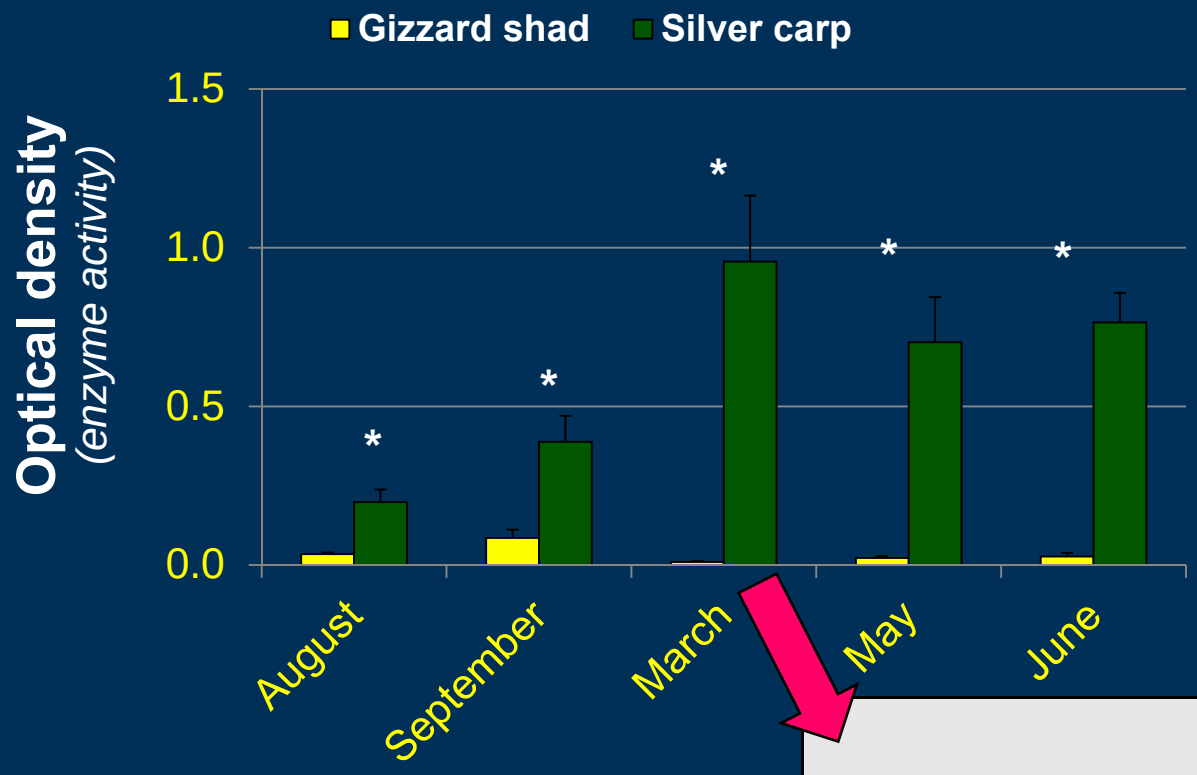
Silver carp vs native planktivores

Trypsin



Species	% with food in gut (March)
Gizzard shad	0
Bigmouth buffalo	0
Silver carp	**100**

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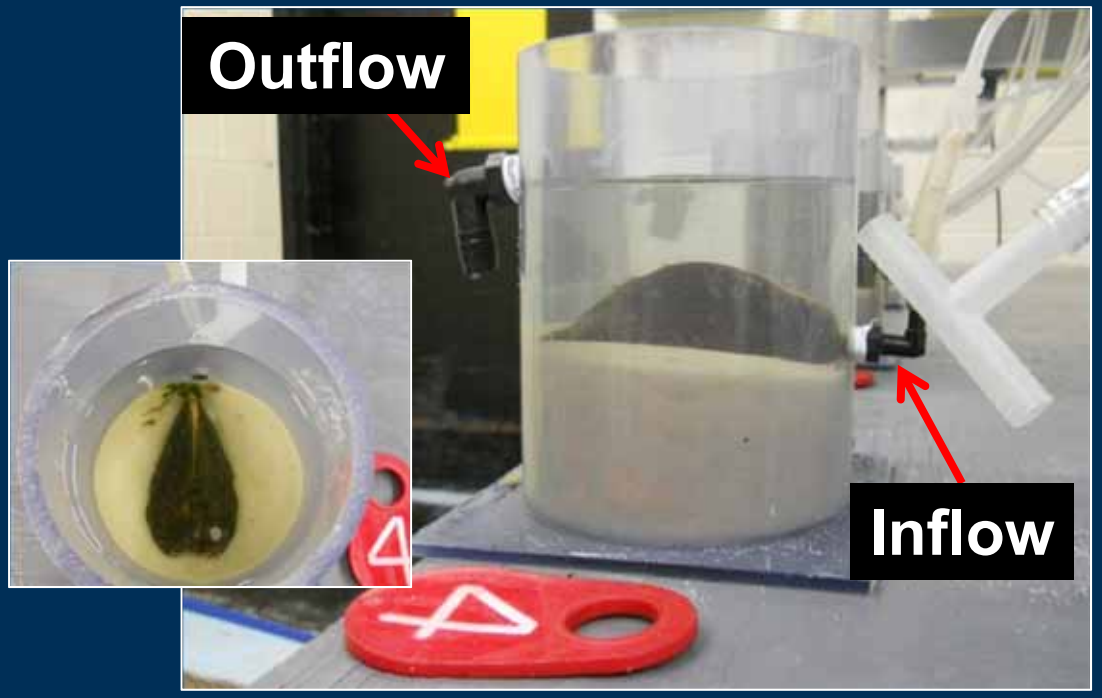
Mussel filtration

Fatmucket[1]
(*Lampsilis siliquoidea*)

Threeridge[1]
(*Amblema plicata*)



Flow through system (similar to Filgueira et al., 2006)

Zebra mussel[2]
(*Dreissena polymorpha*)

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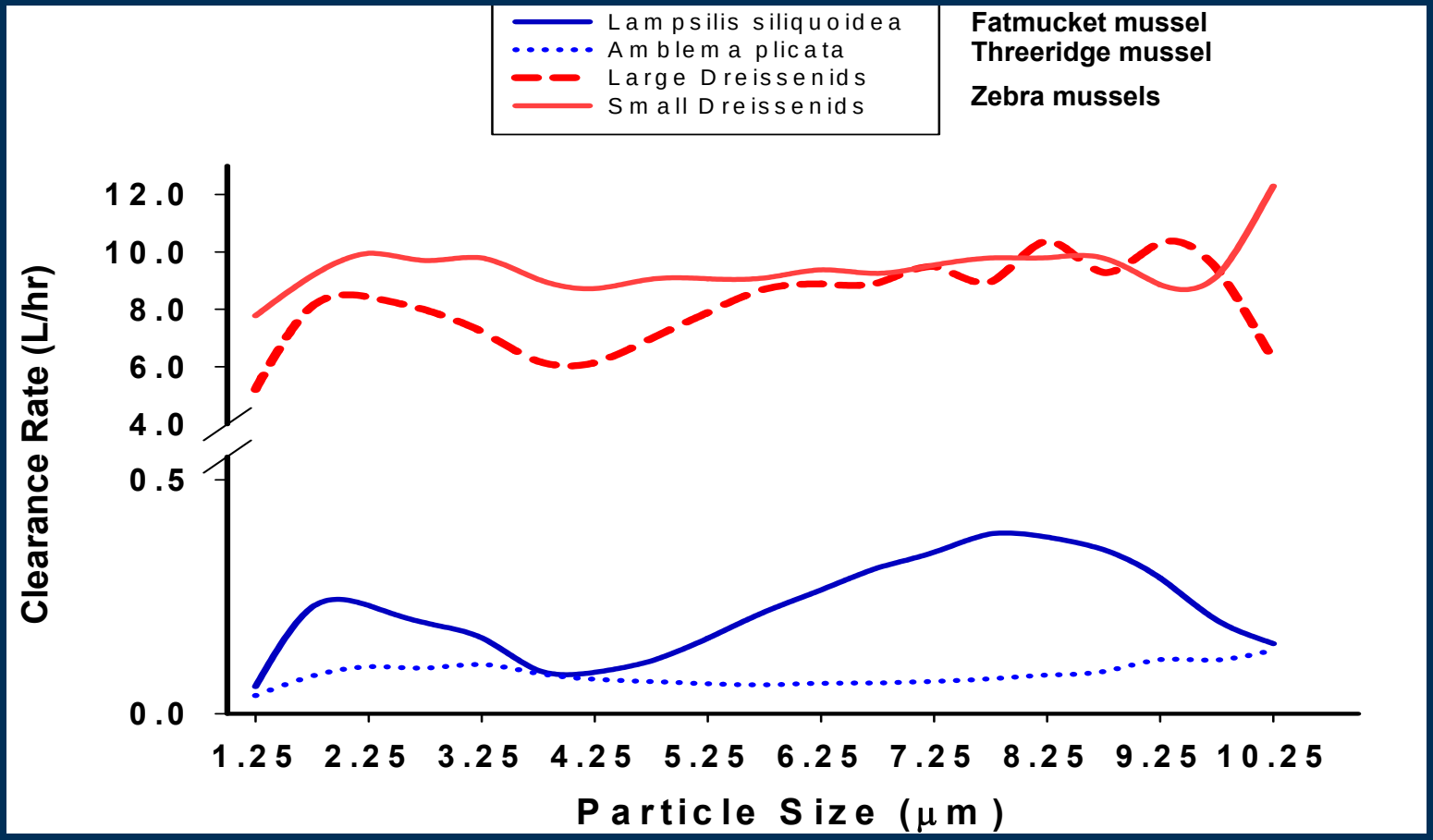
[1]Photo source: IL NHS
[2]Photo source: USGS

Mussel filtration



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Points represent the mean of 3 independent trials

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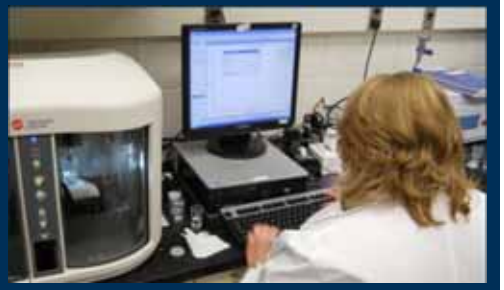
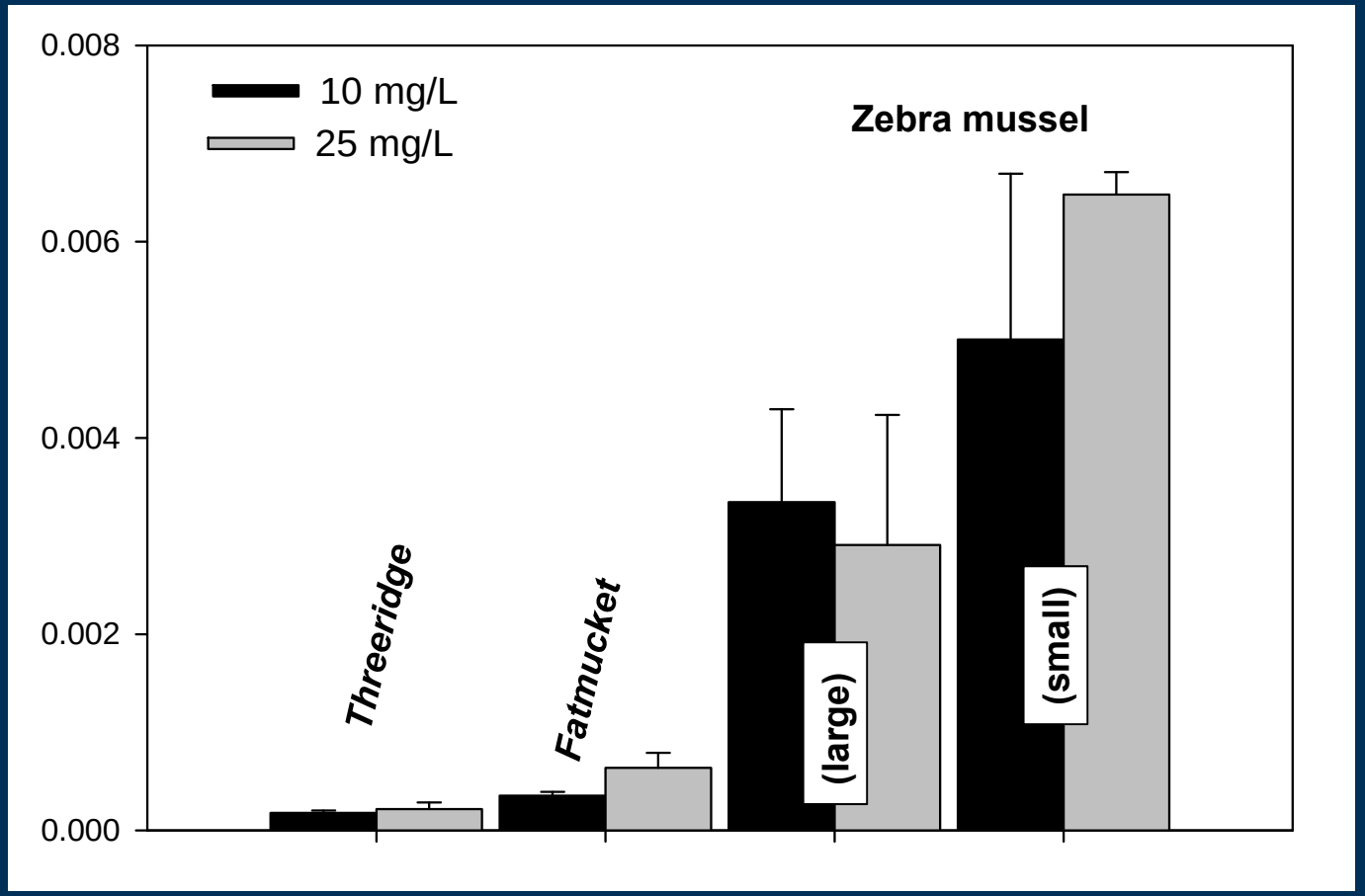
Mussel filtration





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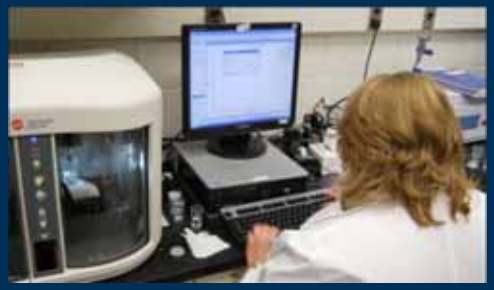
Mussel filtration



Specific gravity of algae cells ~ 1

- Removal
  - ~0.003 mL/h/g dry wt
  - = 3.0 mg/h/g dry wt
- Dose = mass x load x available
  - 3.0 mg/h/g x 5% x 75% = ~0.1 mg/g/h

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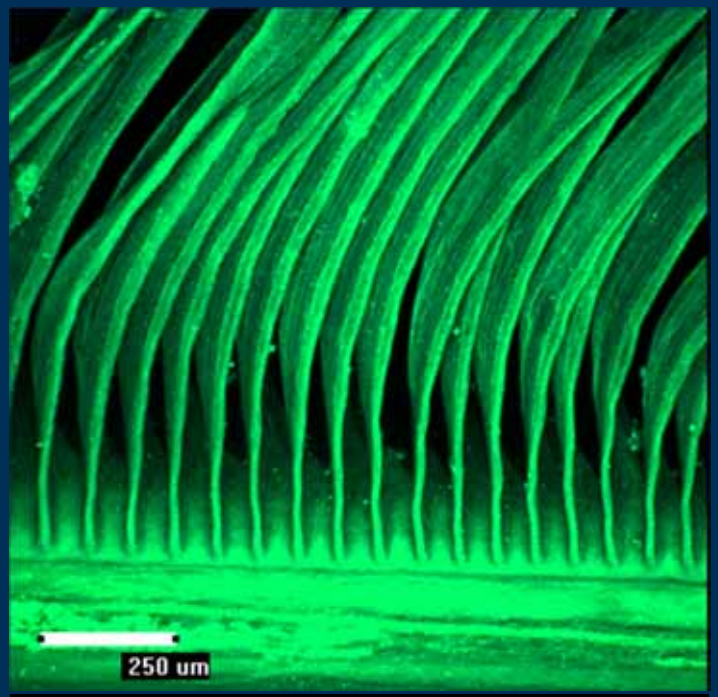
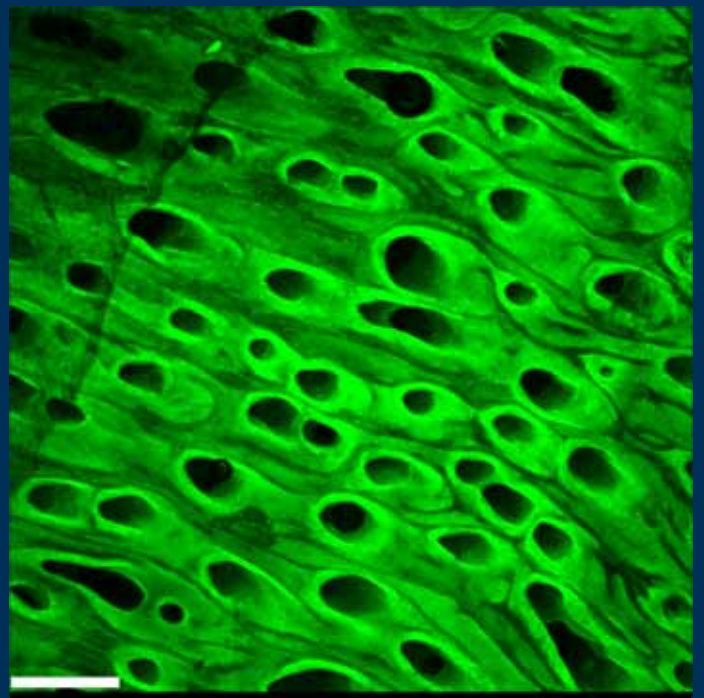
Asian carp filtration



Gizzard shad gill



SVC gill

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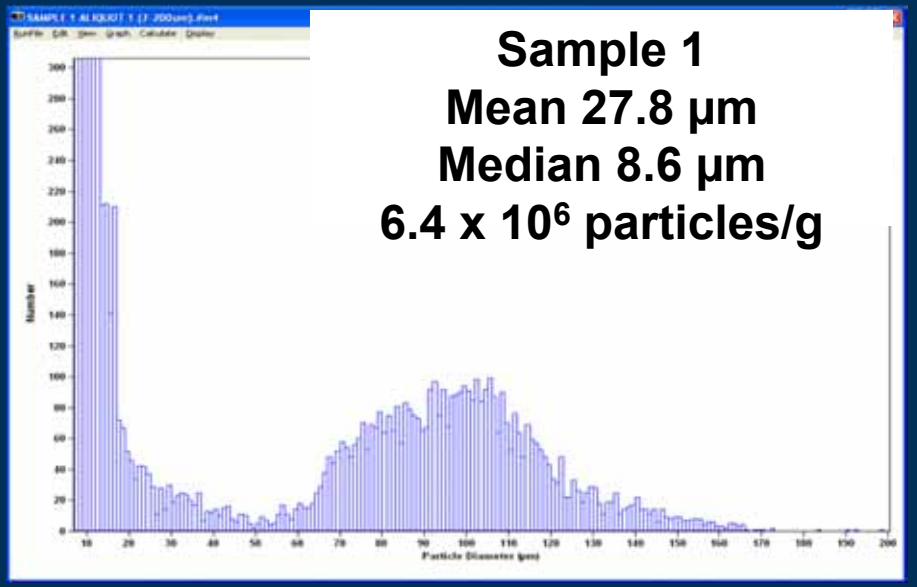
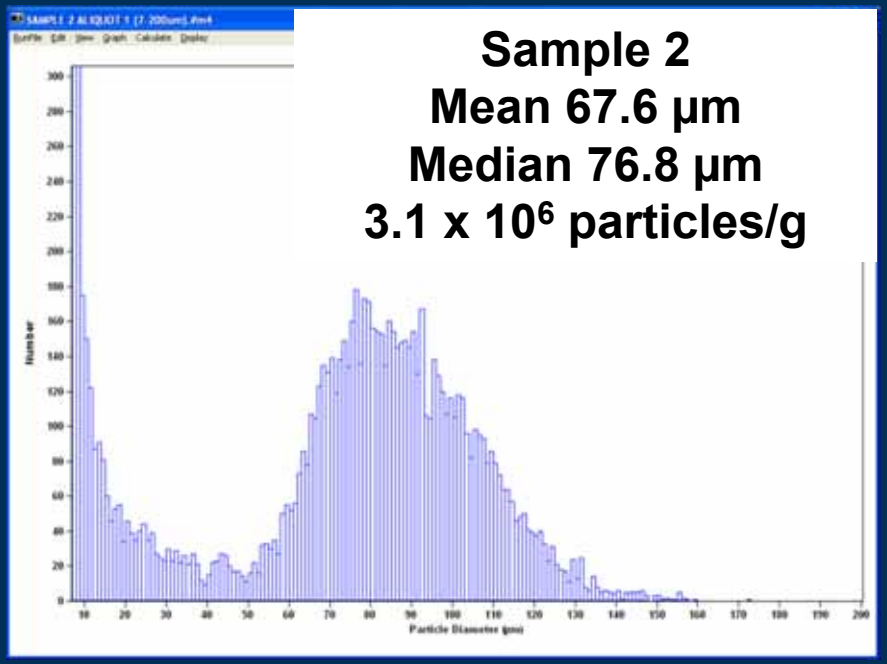
Asian carp filtration



Sample 1
Mean 27.8 µm
Median 8.6 µm
6.4 x 10^6 particles/g

- 5 g/90 L water
- Sample 1 = 3.6 x 10^5 particles/L (~355 particles/mL)
- Sample 2 = 1.7 x 10^5 particles/L (~172 particles/mL)
- SVC, BHC, Hybrid carp
  - 35-40 g



Sample 2
Mean 67.6 µm
Median 76.8 µm
3.1 x 10^6 particles/g

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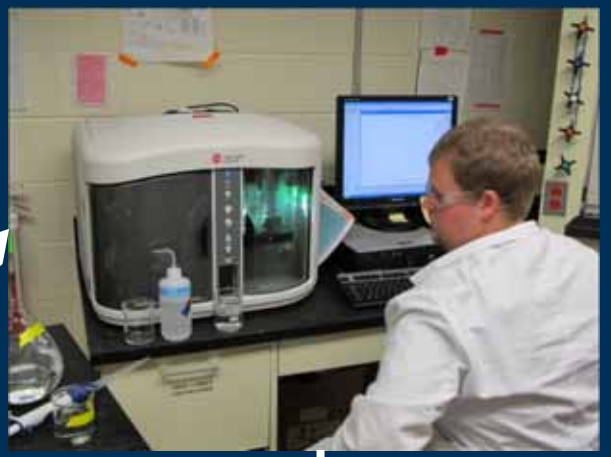
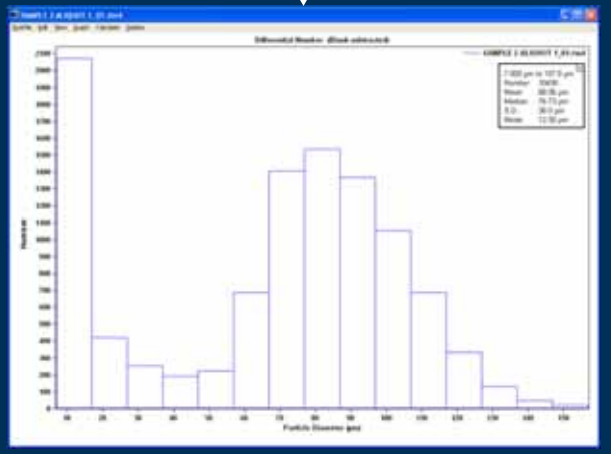
Asian carp filtration





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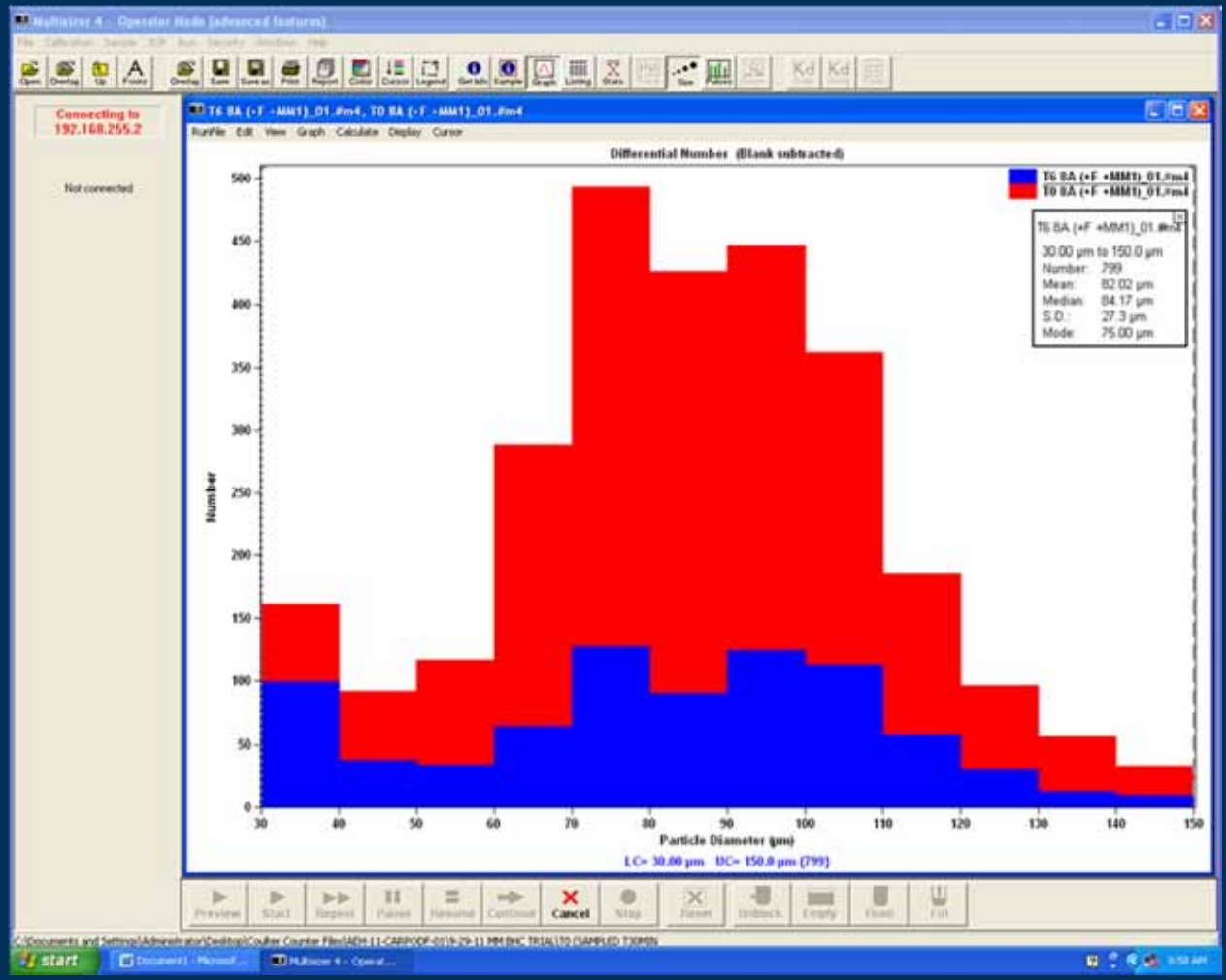
Asian carp filtration

BHC – Sample 1

Red bars = 0-h

Blue bars = 6-h



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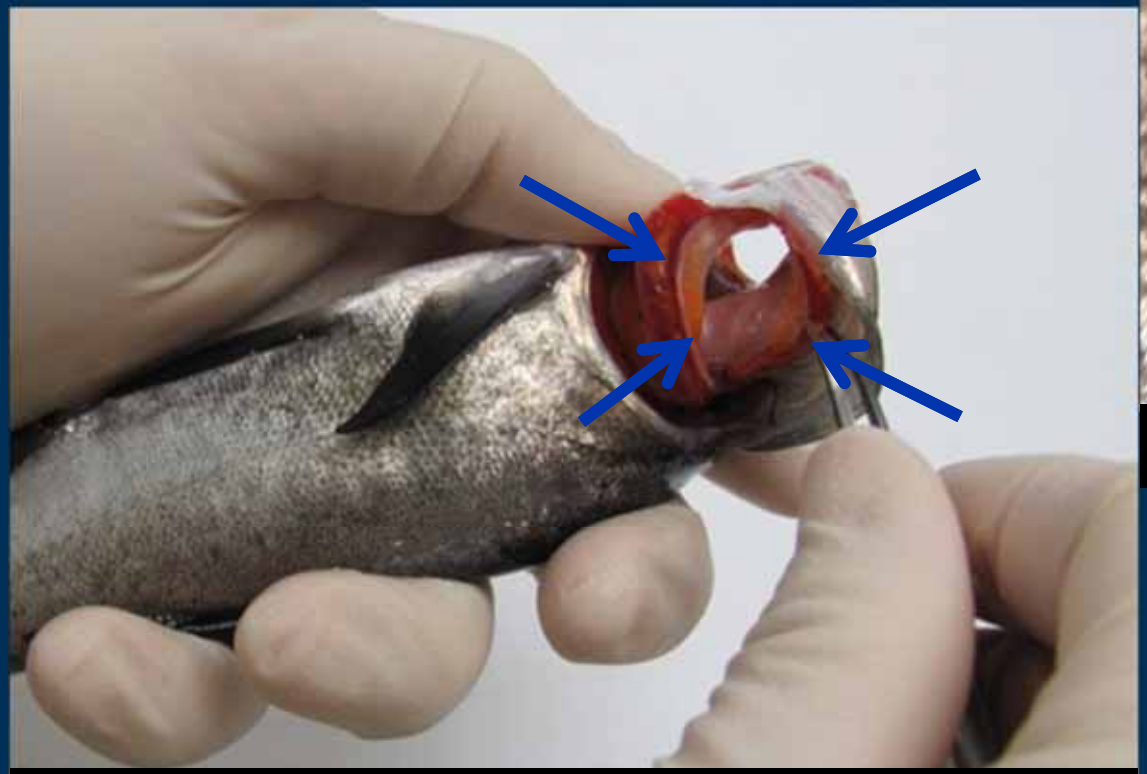
Asian carp filtration

Removal of microparticles



BHC gill

SVC gill

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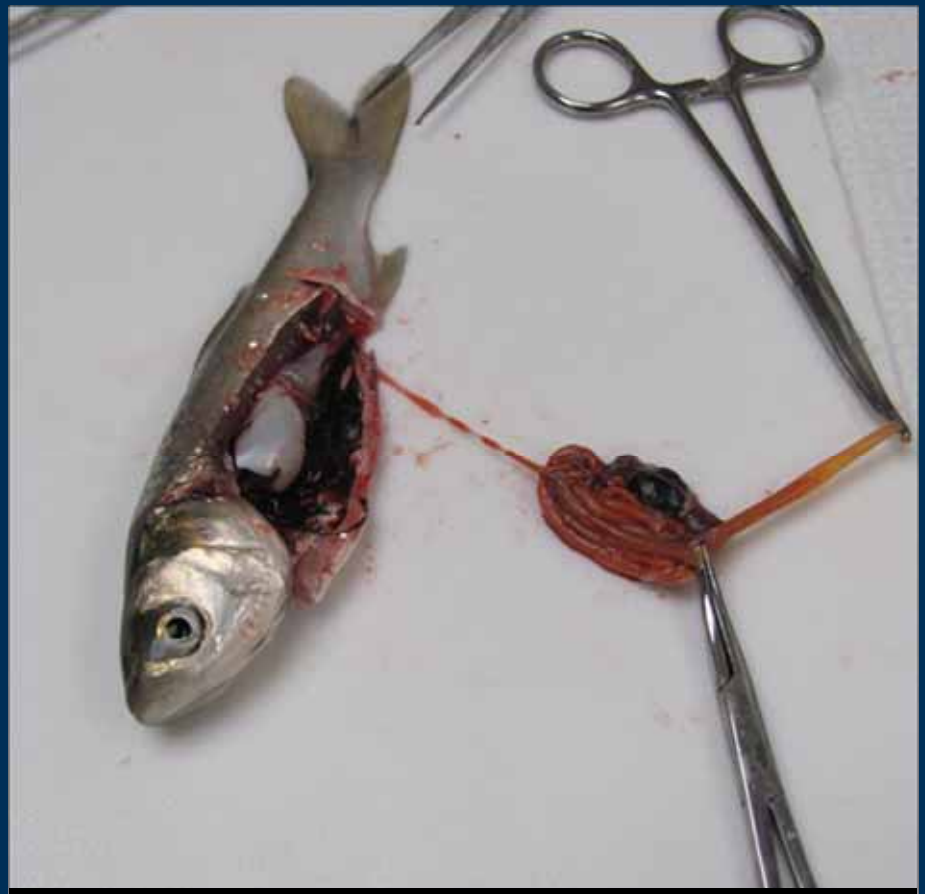
Asian carp filtration

Removal of microparticles

particles retained in the GIT of a BHC



particles retained in the GIT of a SVC

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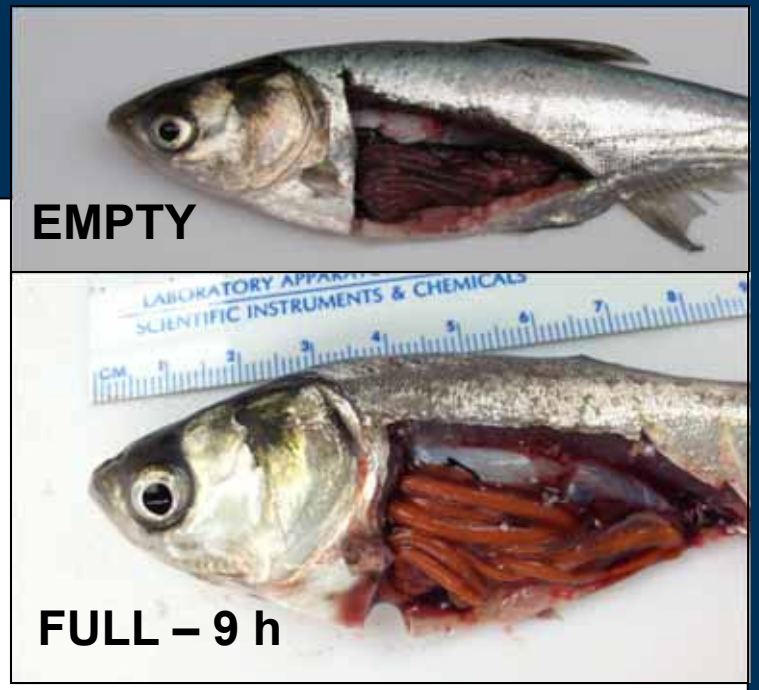
Asian carp filtration

Removal of suspended particles (PROVISIONAL DATA)



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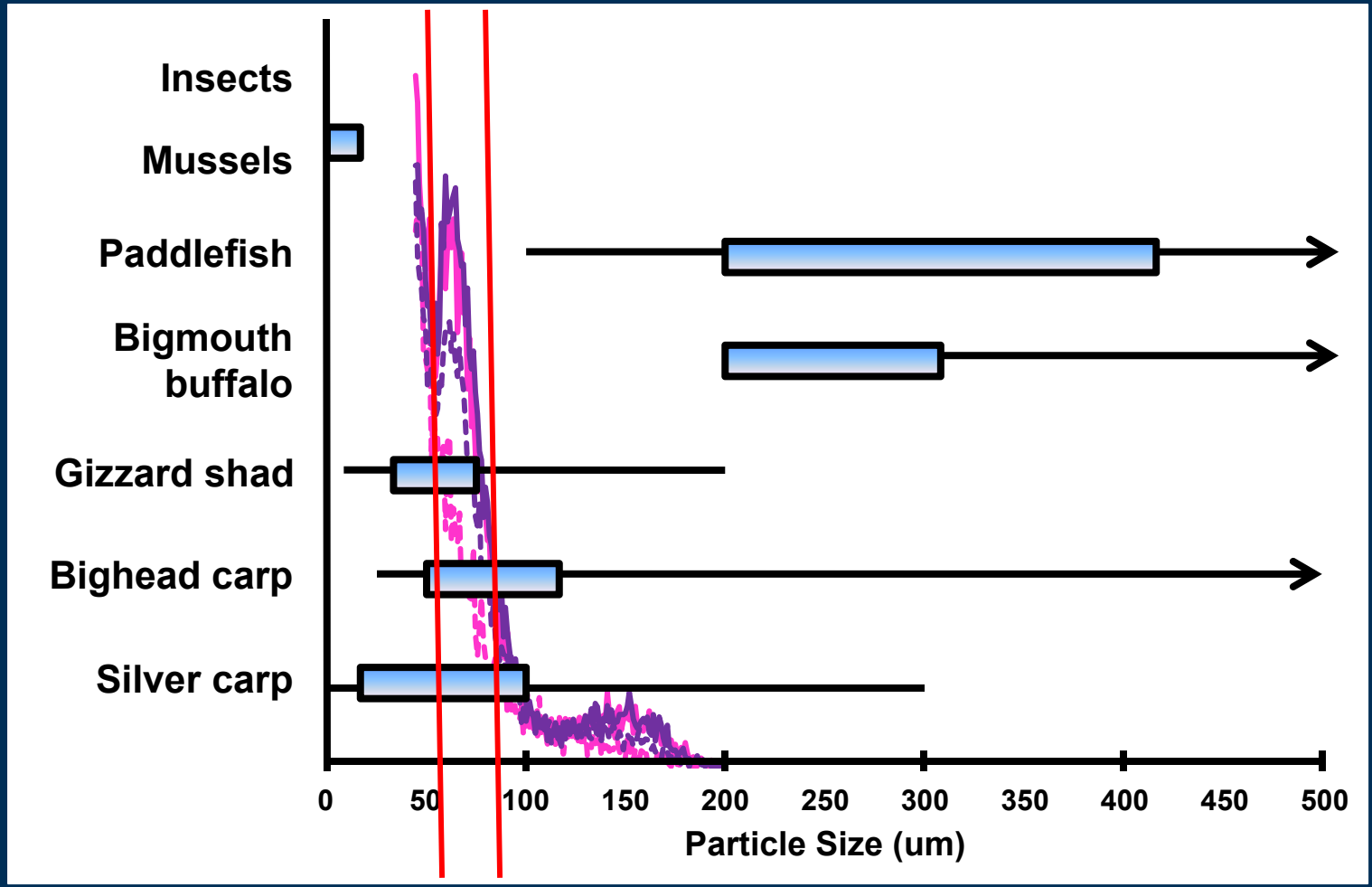
Each point represents the mean of 4 independent experimental units

Planktivore particle overlap



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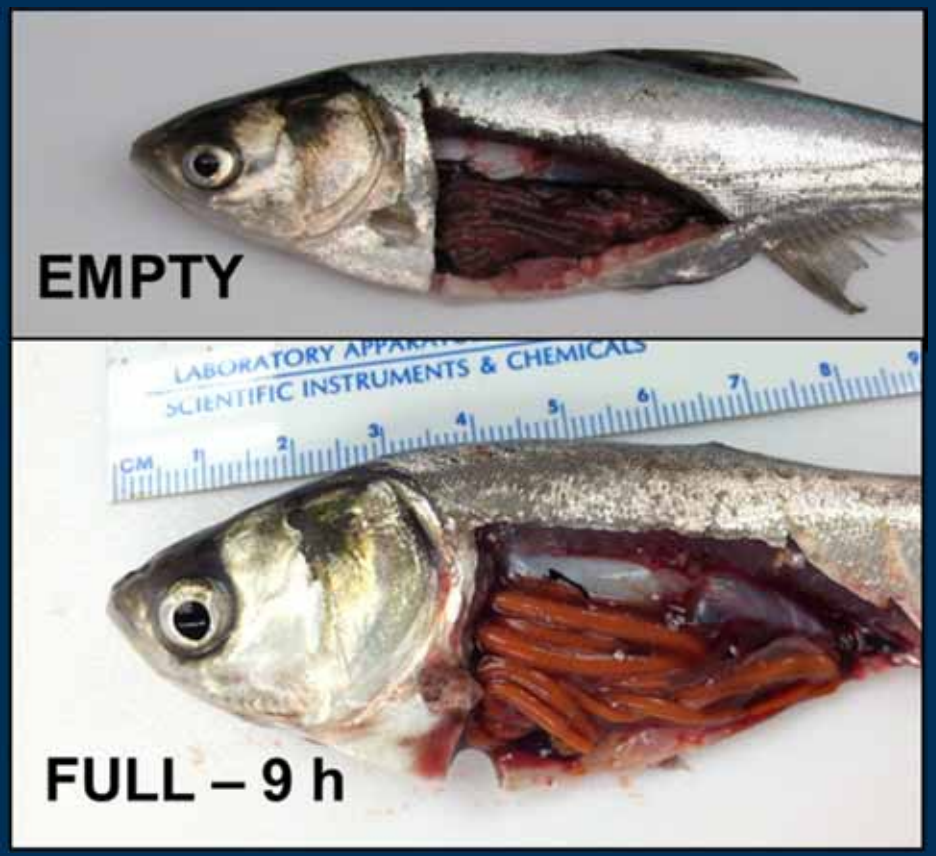
Asian carp filtration

Microparticle retention

- Extract GI tract
- Rank (quartiles) foregut and mid-hindgut fullness
- Extract GI tract contents
  - Foregut – most tests
  - Mid/hindgut limited
  - Entire GIT – most tests



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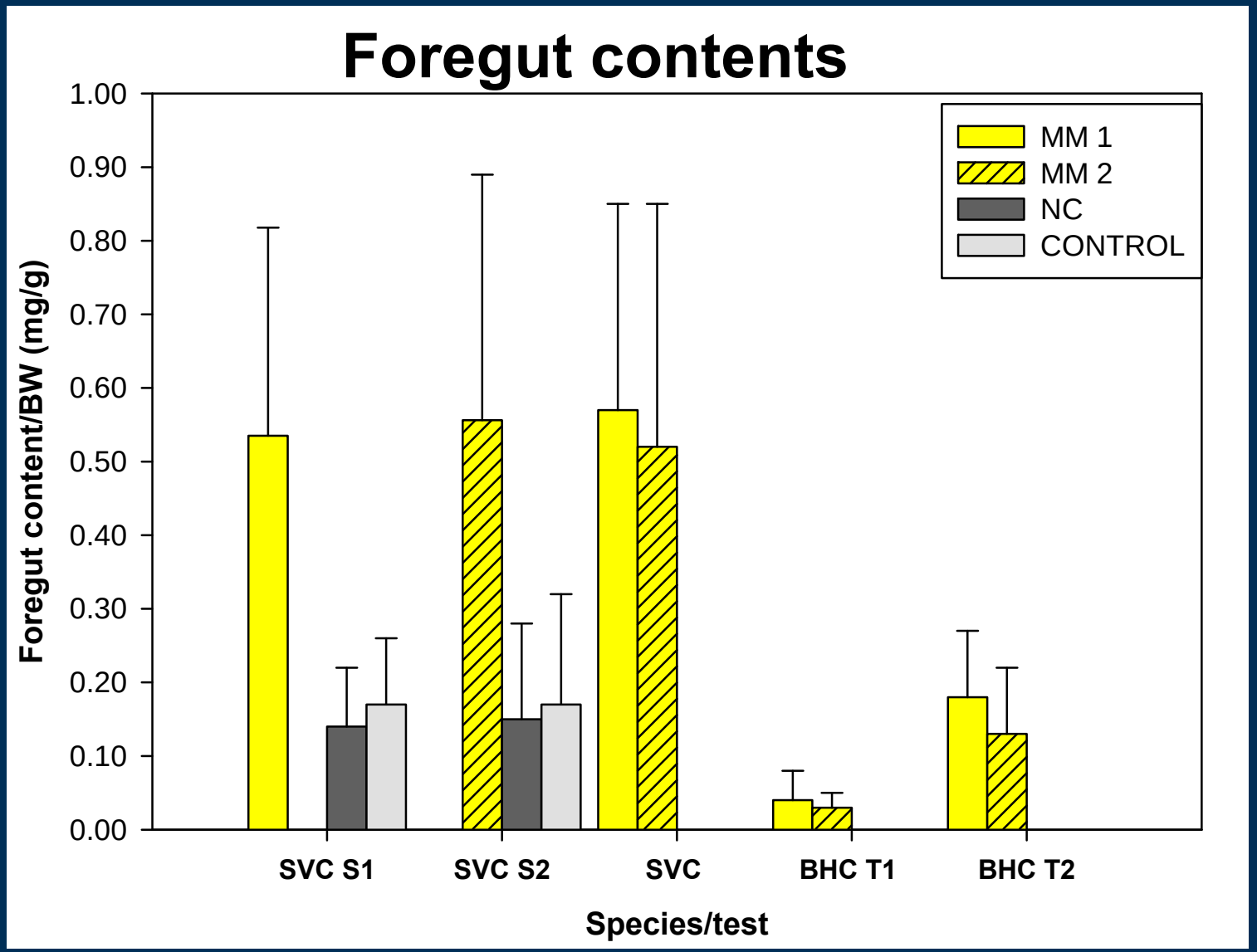
Asian carp microparticle retention



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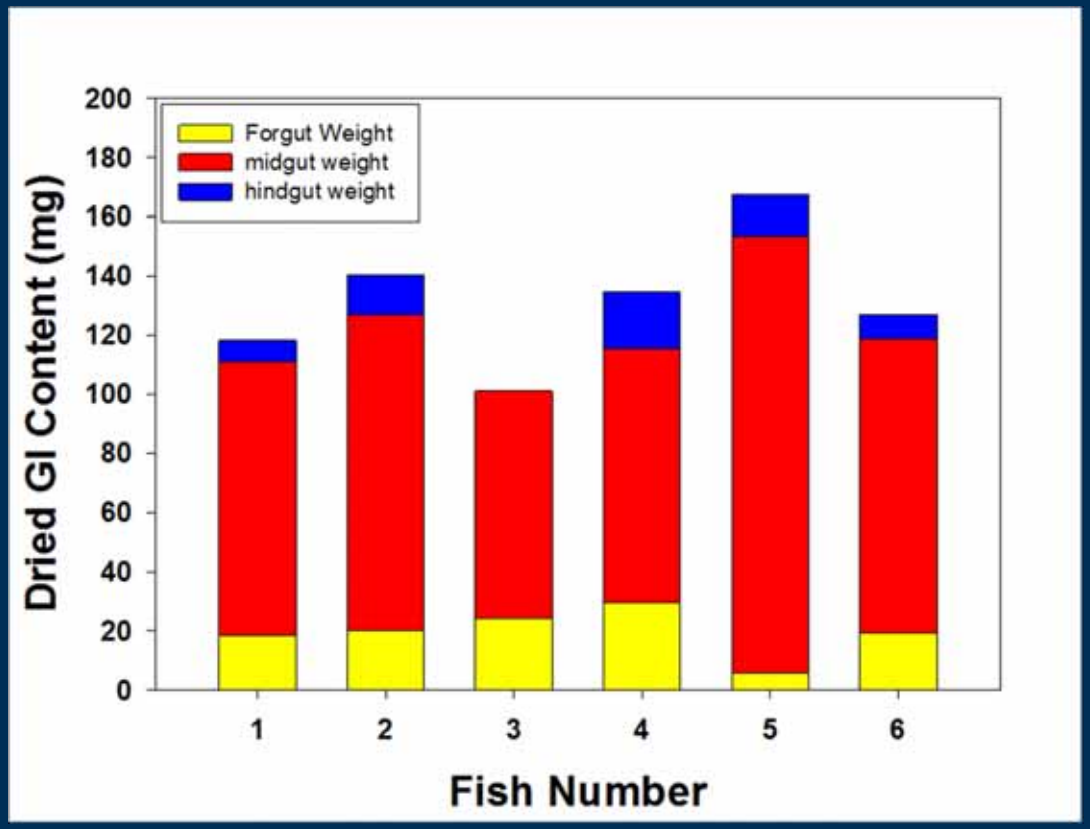
Asian carp GIT particle distribution



6 SVC @ level 4 after exposure to Sample 2

GIT contents (mg)

- Foregut
  - 19.7 ± 7.1 mg
  - 6.1 x 10^4 particles
- GIT
  - 131.4 ± 20.5 mg
  - 4.1 × 10^5 particles

Particle distribution

- Foregut ~ 16%
- Mid/hindgut ~ 84%

Particle mass/BW

- Foregut ~ 0.6 mg/kg BW
- GIT ~ 4500 mg/kg BW

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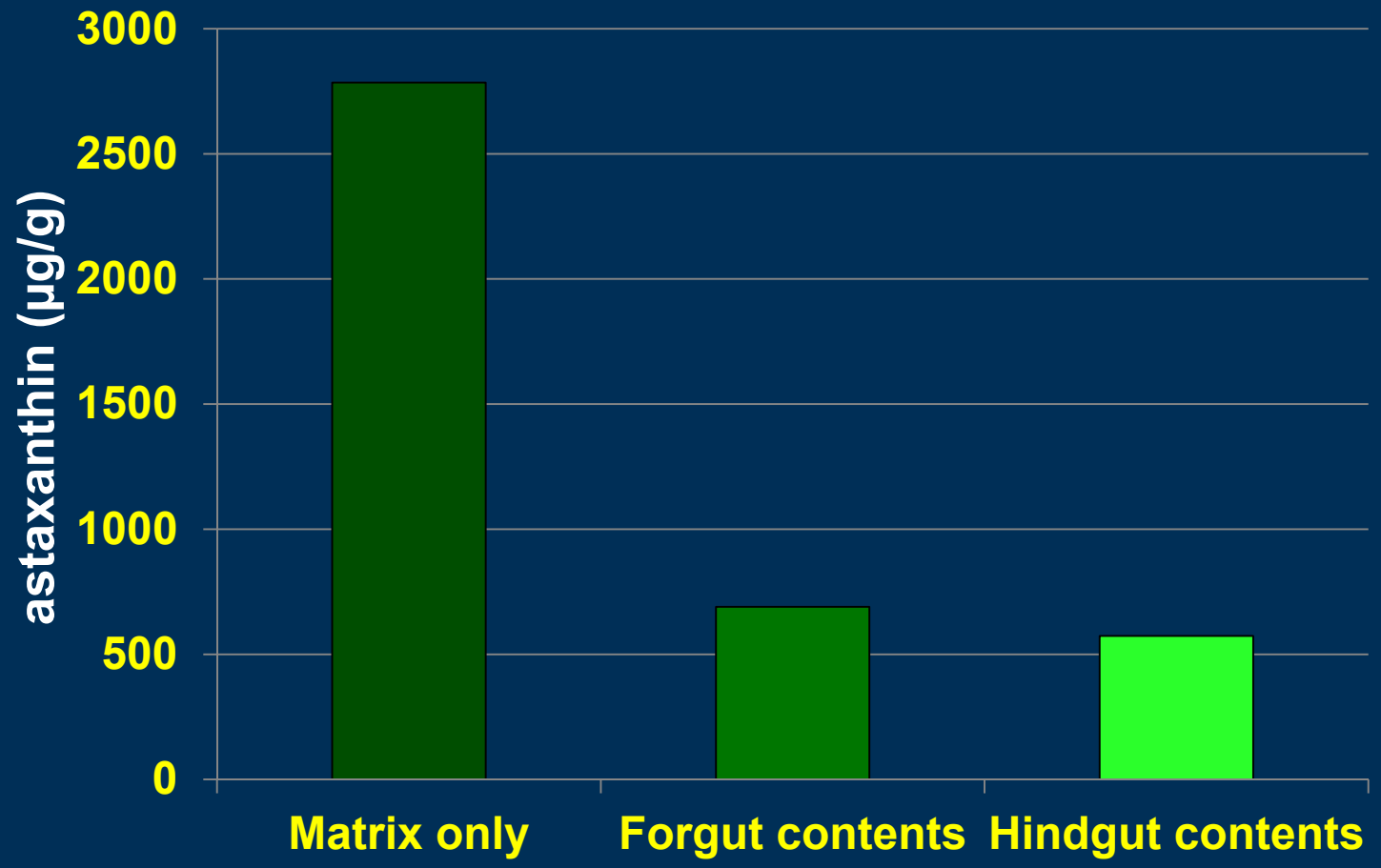
Asian carp microparticles – agent release



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Asian carp potential dose delivery

- Dose (D) = M × L × A
  - M = Mass consumed
  - L = % loading (5% w/w)
  - A = % available (75%)

SVC

Foregut: 600 mg/kg × 5% × 75% = ~ 23 mg/kg dose

Entire GIT: 4500 mg/kg × 5% × 75% = ~ 170 mg/kg dose

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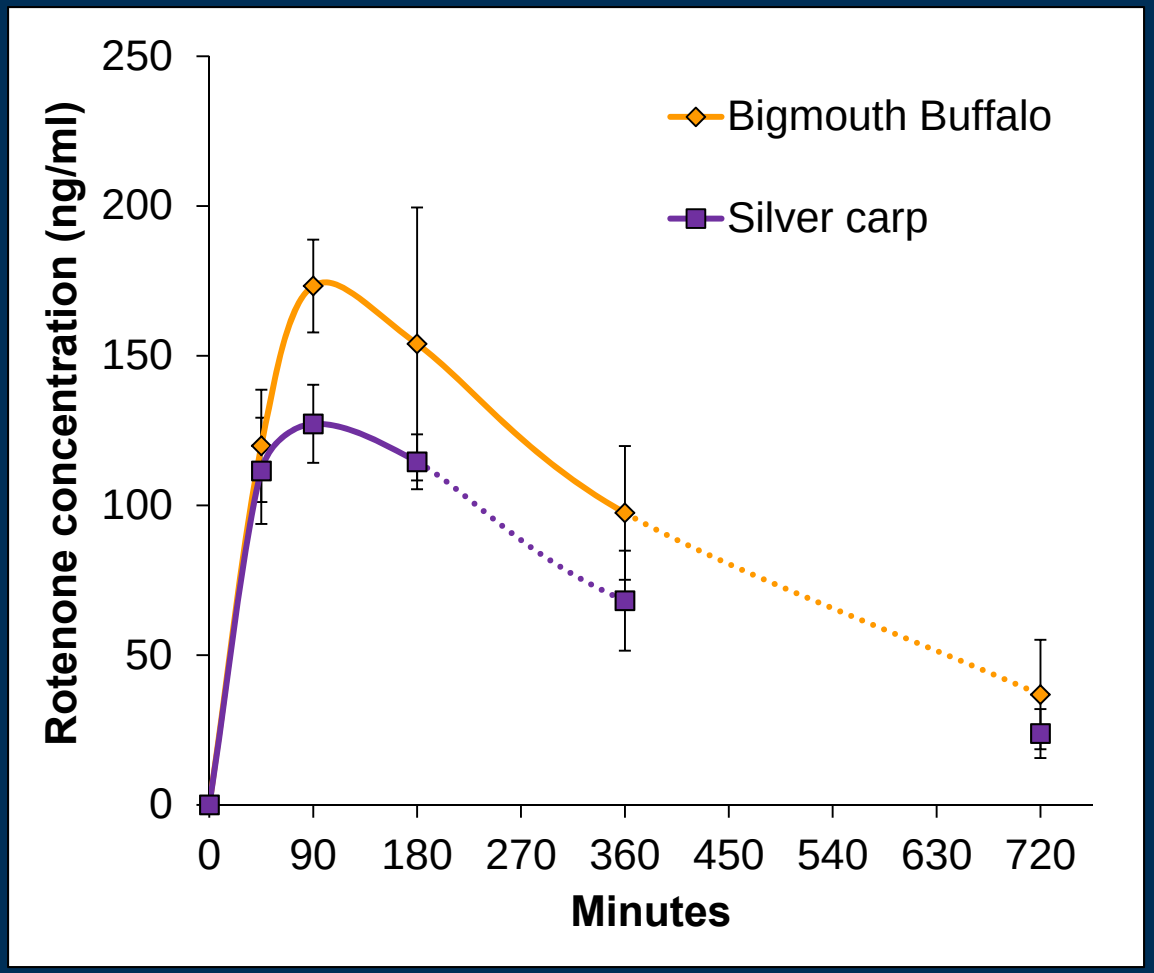
Asian carp response to rotenone

- Active absorption
- Similar uptake and excretion
- Silver and bighead carp use different molecular processes to respond to rotenone exposure



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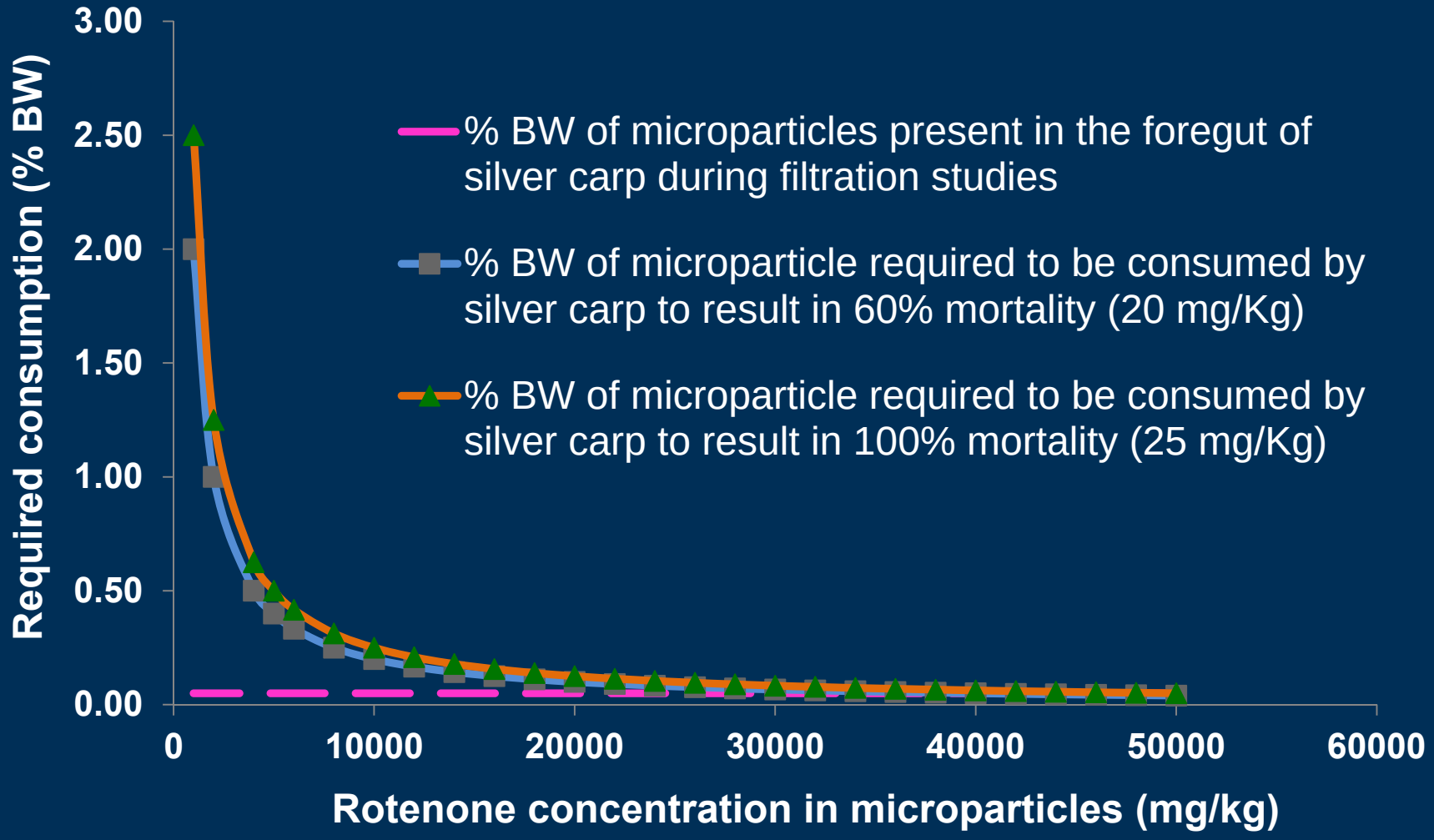
Toxicity of rotenone microparticles



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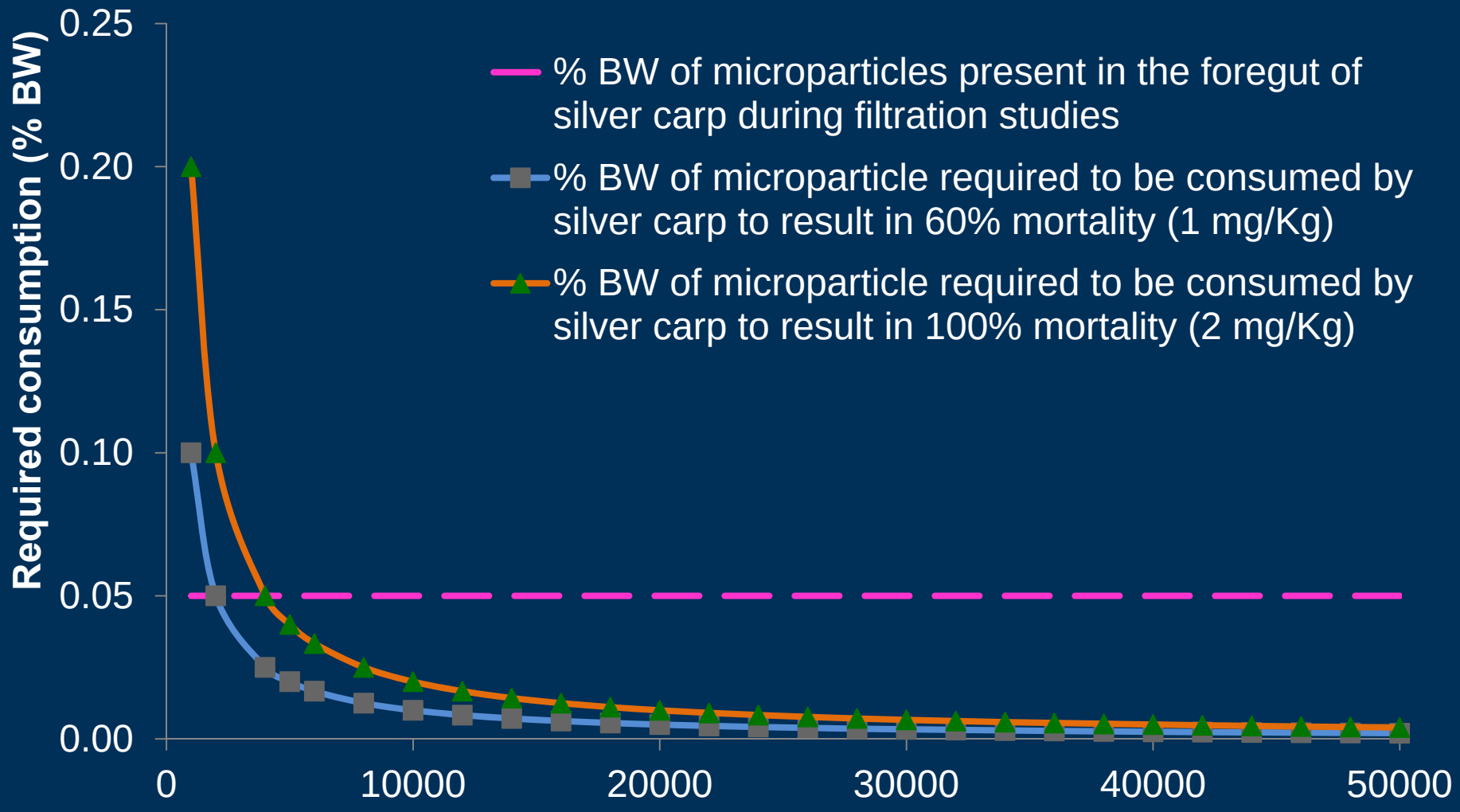
Toxicity of antimycin microparticles – RBT



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Microparticles characteristics

1. **Particle degradation**

   Measure particle size in water (20 °C) over time

   - microscopically
   - Coulter Counter

2. **Particle Settling**

   Measure particle settling in water (20 °C)

T_0

T_6

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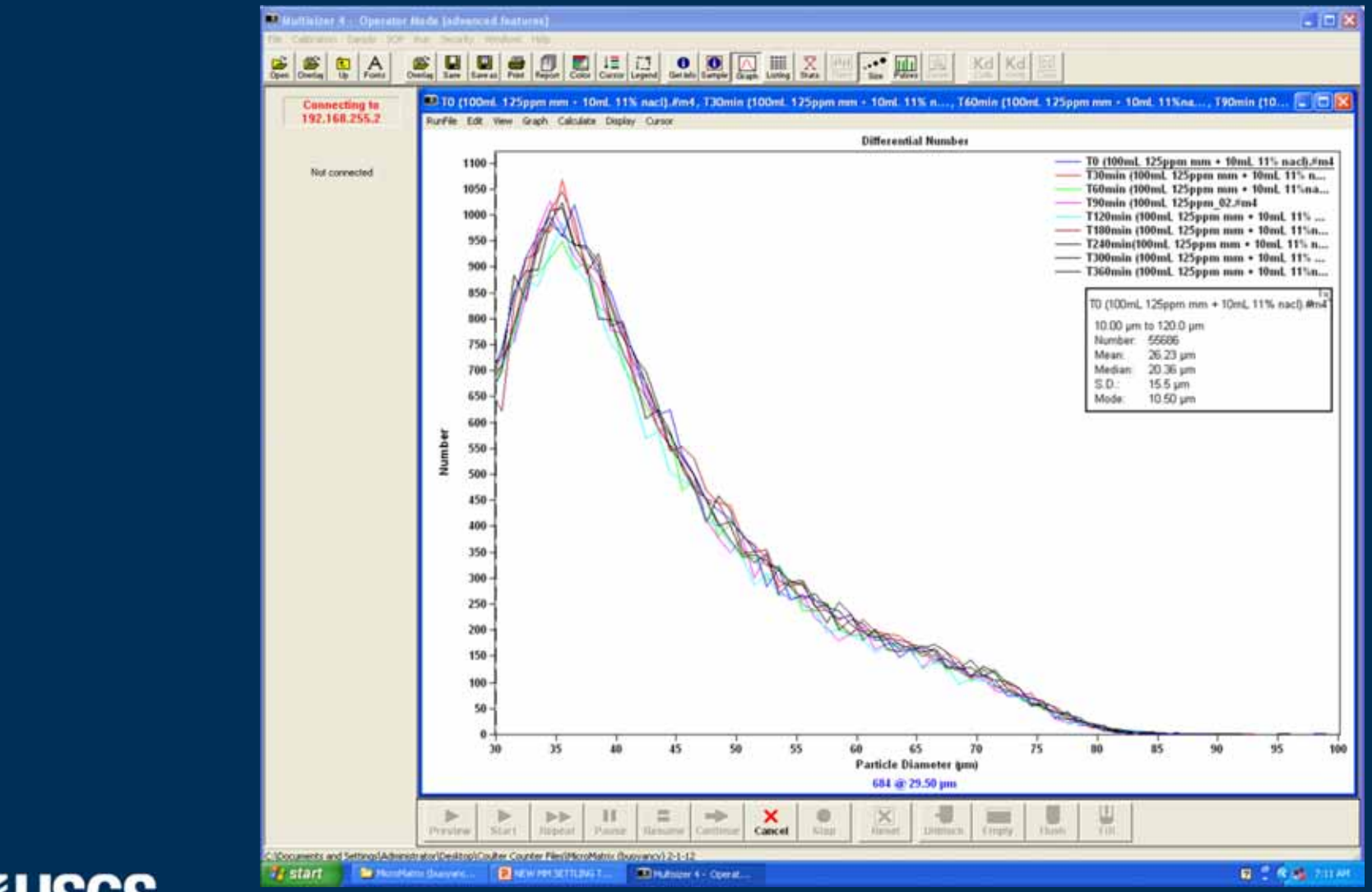
Microparticles characteristics



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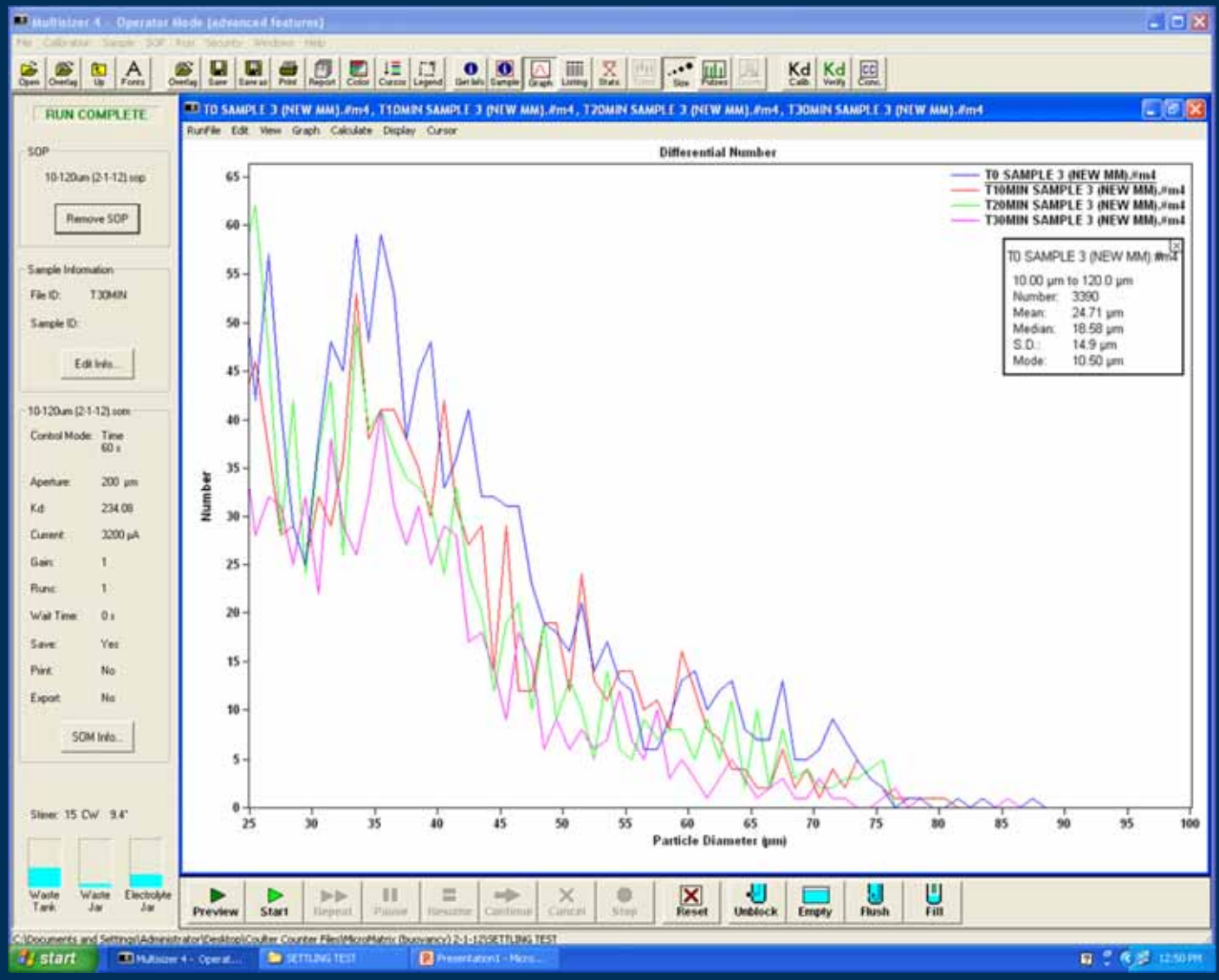
Microparticle settling



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Summary – Zebra mussel

- Filtration rate >> natives
  - Incorporate low levels of a toxicant?
- Minimal change in zebra mussels digestive enzymes with decreasing water temperature
  - Potential seasonal application?
  - Increased phosphatases and proteases - potential release target?
- Incorporate control agent into microparticles
  - Initiate/complete lab exposure trials

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Summary – Asian carp

- Asian carp retain preliminary microparticles
  - Determine filtration and gut evacuation rates
- Activity of certain digestive enzymes are higher in Asian carp vs. native planktivores
  - Trypsin, phosphatases – potential particle release
  - Active feeding earlier than natives
- Incorporate control agent into microparticles
  - Initiate/complete lab exposures
  - Confirm dose

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Registration of control agent in a microparticle

U.S. Environmental Protection Agency

- Existing control agents
  - likely considered a new “formulation”
  - shorter registration path
    (product chemistry, user safety,
    environmental fate, ecological effects)
- New control agents
  - full registration

USGS

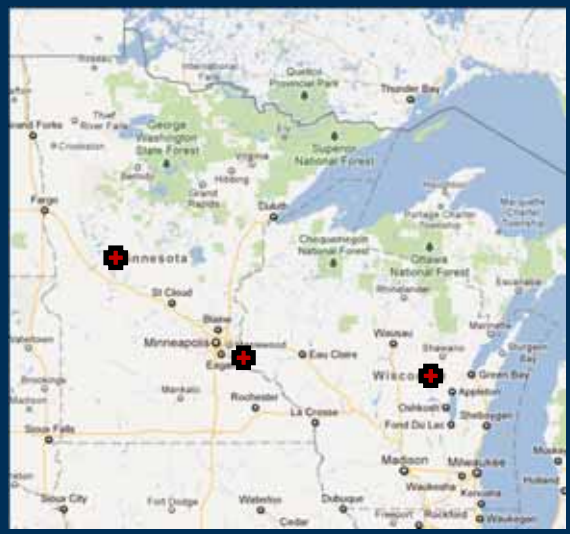
Use of *Pseudomonas fluorescens* (*Pf*-CL145A; ZEQUANOX®) to control dreissenid mussels

Current research:

- Assess the safety of *Pf*-CL145A to three life stages (glochidia, juvenile, subadult) of 7 native mussel species
- Assess the safety of *Pf*-CL145A to 10 native fish species

Future research:

- Assess efficacy and potential non-target effects of *Pf*-CL145A used to control zebra mussels in open water around native mussel beds and propagation cages
- Assess efficacy of *Pf*-CL145A to control zebra mussel veligers in fish transport water

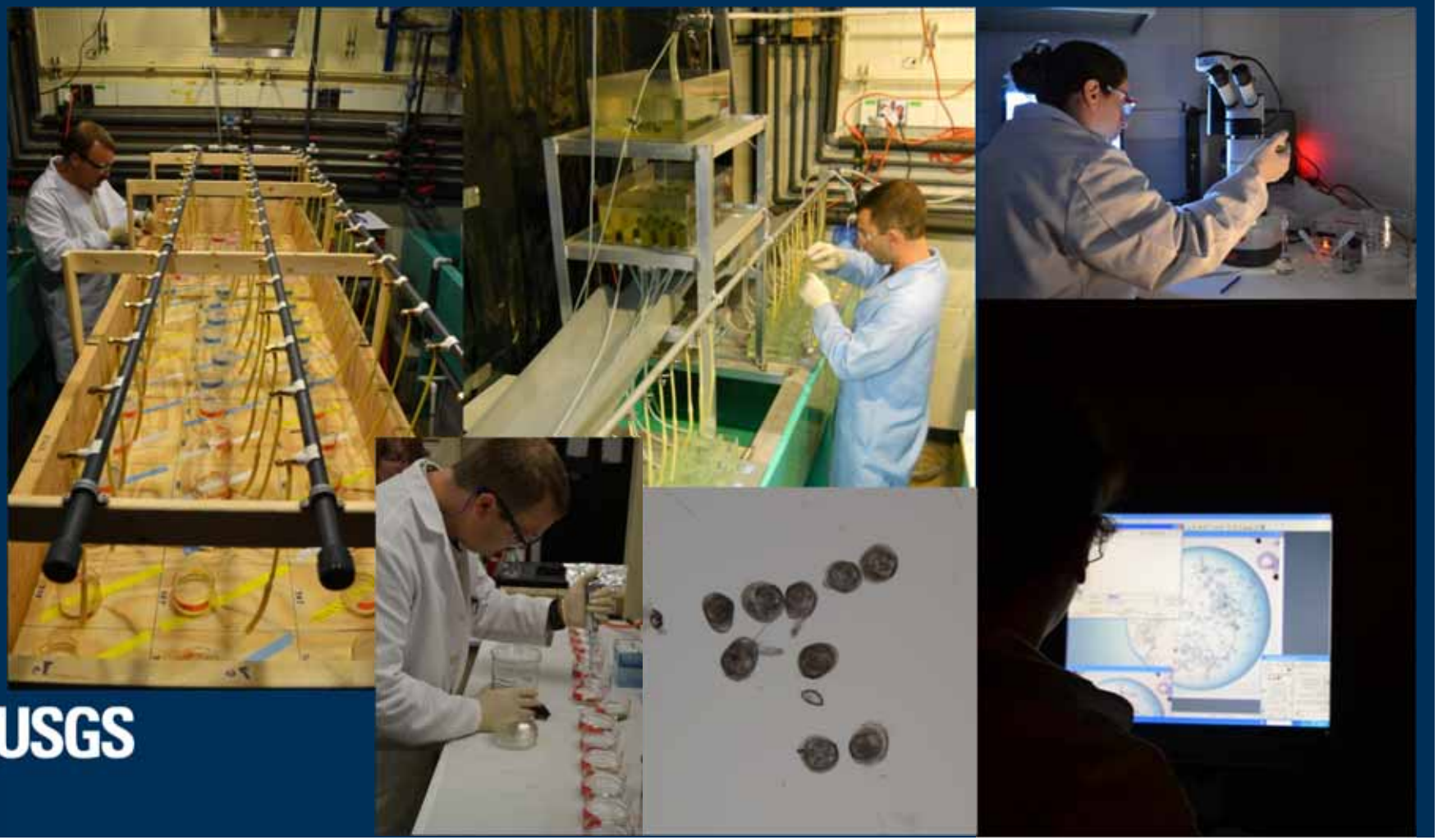


USGS

http://cida.usgs.gov/glri/projects/invasive_species/zm_control.html

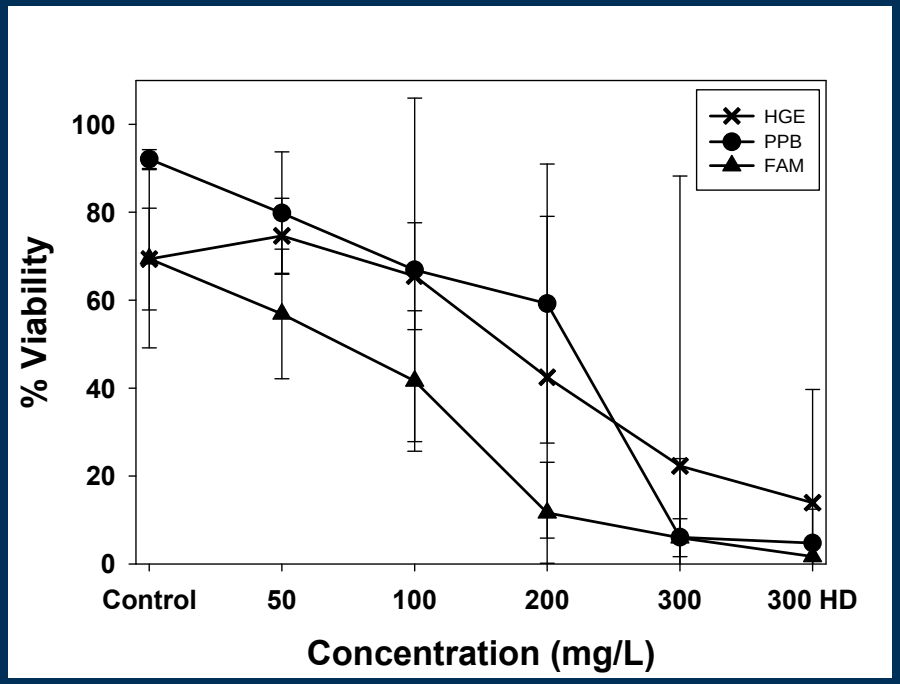
UMESC laboratory trials with *Pf*-CL145A

- Research according to Good Laboratory Practice regulations with controlled environmental conditions
- Designed to support EPA registration for open water uses



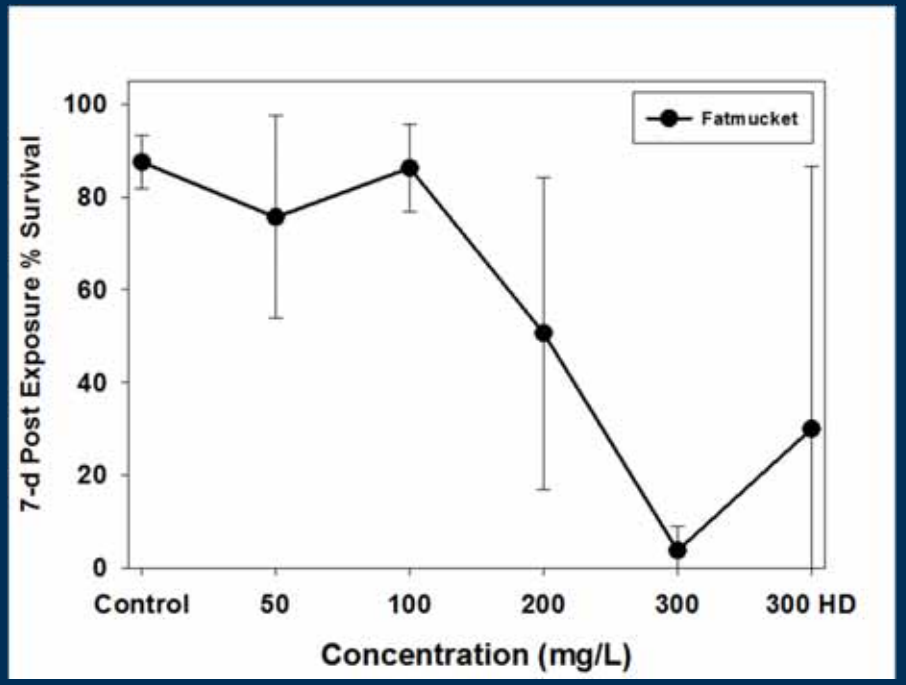
USGS

UMESC laboratory trials with *Pf*-CL145A



Glochidia viability

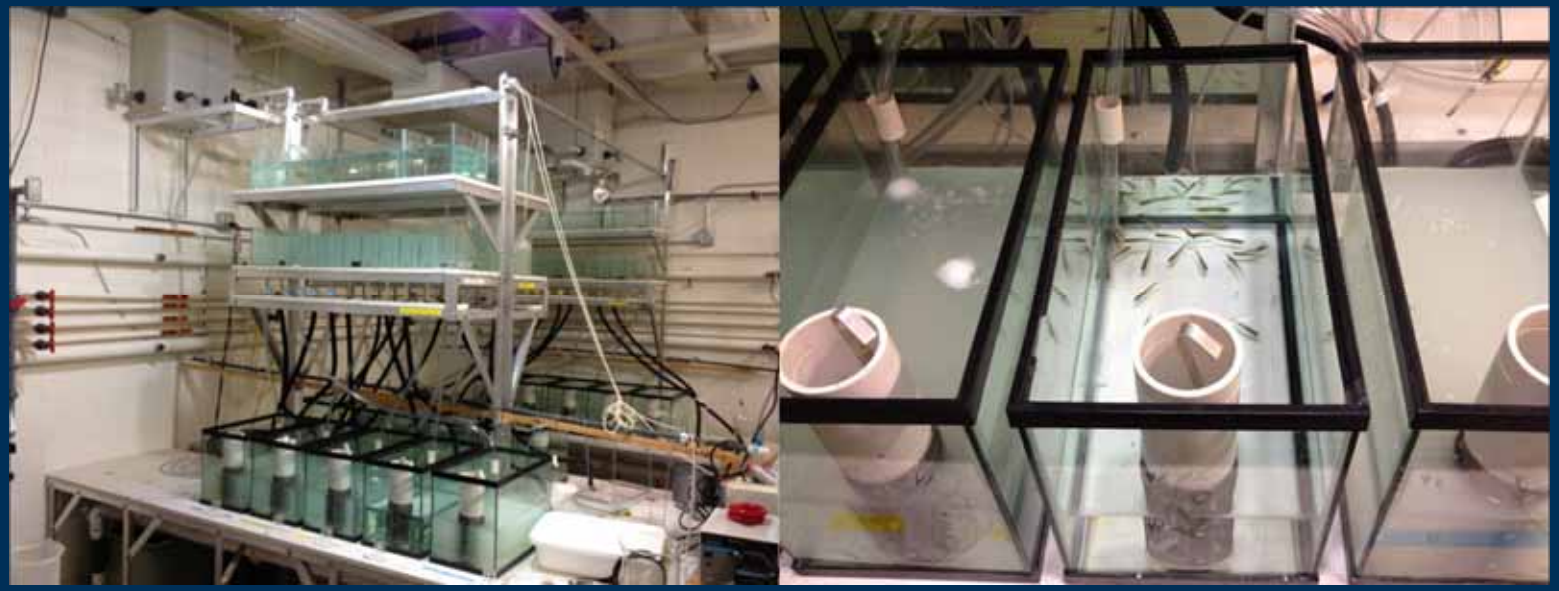
Juvenile viability



USGS

USGS PROVISIONAL DATA

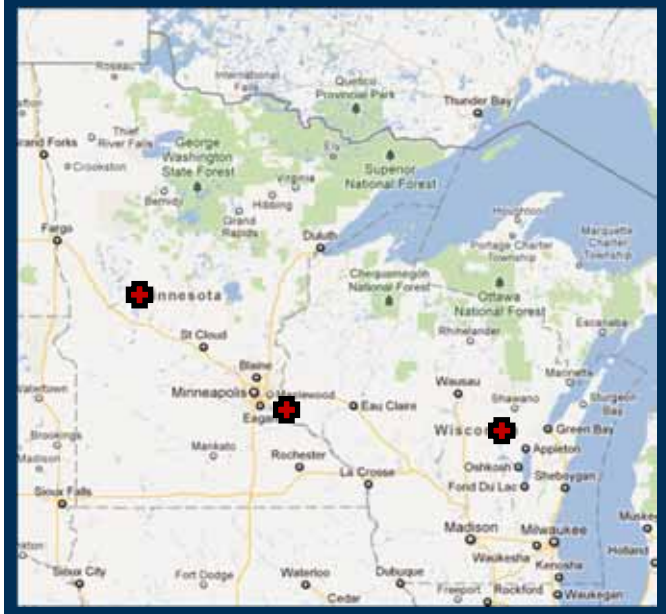
UMESC laboratory trials with *Pf*-CL145A



Evaluate effects of *Pf*-CL145A on fish
- in progress

USGS

Controlled field trials with *Pf*-CL145A



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  - $ Great Lakes Restoration Initiative
  - $ US Geological Survey
  - $ US Fish and Wildlife Service – Science Support Project

- More information
  - http://www.umesc.usgs.gov/
  - http://cida.usgs.gov/glri/projects/invasive_species/zm_control.html

NEW YORK State Museum

Marrone Bio Innovations

U.S. FISH & WILDLIFE SERVICE

Minnesota DEPARTMENT OF NATURAL RESOURCES

WISCONSIN DEPT. OF NATURAL RESOURCES

USGS

Great Lakes RESTORATION

Questions?

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