

# Freshwater Bivalves as an Alternative to Fish Populations in Canadian Environmental Effects Monitoring

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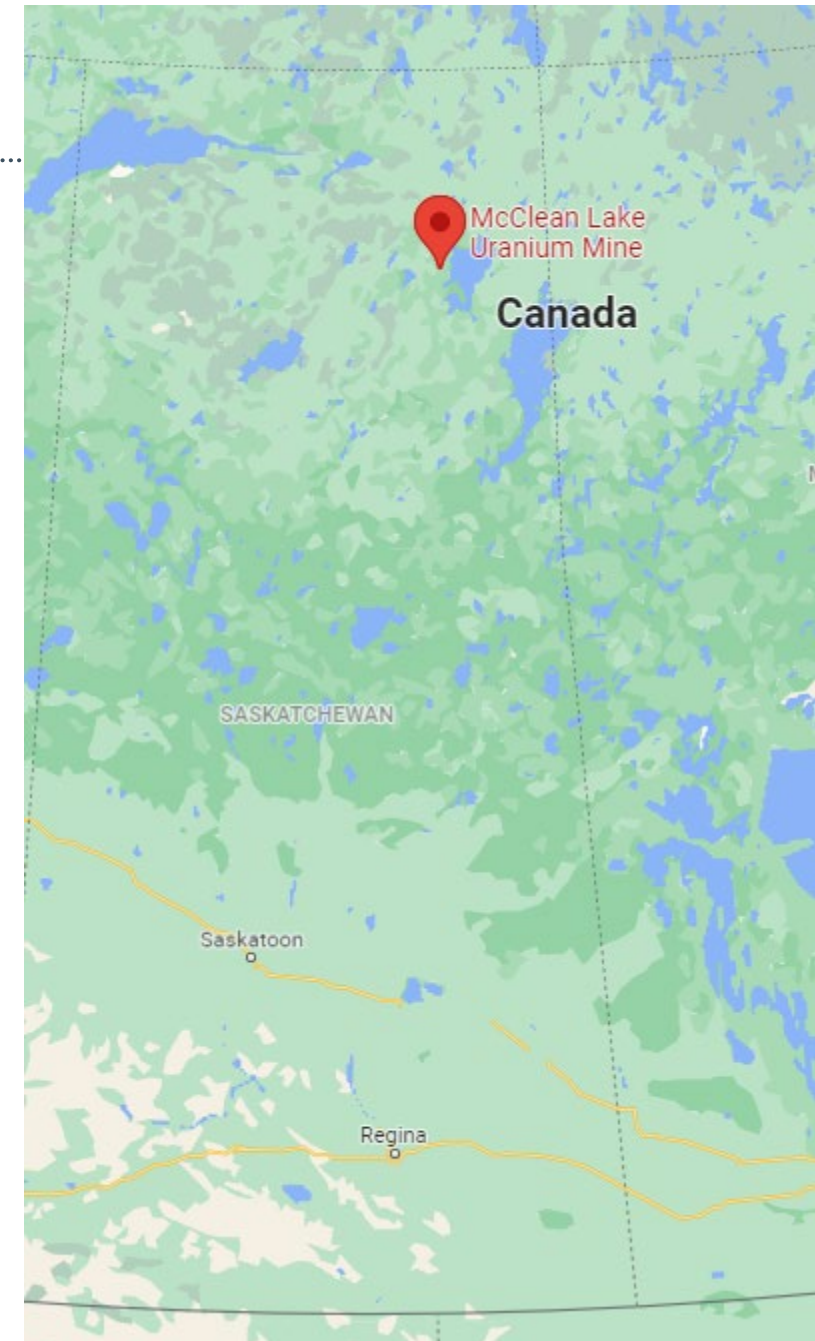
# INTRODUCTION

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- Metal and Diamond Mining Effluent Regulations (MDMER) under the *Fisheries Act*
  - Requires mines to conduct EEM studies
    - **Effluent** and **water quality** monitoring
    - **Biological** monitoring
  - Imposes effluents limits (pH, cyanide, metals, TSS)
  - Prohibits the discharge of effluent that is acutely lethal to fish

# McClean Lake

- Orano Canada's McClean Lake Operation
  - Uranium mining and milling facility since 1996
  - Located in Northern Saskatchewan
  - Orano has completed six biological EEM studies pursuant to the company's requirements under the MDMER
  - The latest study was completed in 2019 and submitted to ECCC in 2020



# ENVIRONMENTAL EFFECTS MONITORING

- EEM Biological Monitoring Studies
  - Fish population to assess effects on fish health;
  - Benthic invertebrate community to assess fish habitat or food;
  - Fish tissue to assess the human usability of the fisheries resources.
- Fish Population Survey Effect Indicators
  - MDMER effect indicators are **survival** (age), **growth** (size at age), **reproduction** (relative gonad size) and **condition** (condition, relative liver size).
  - Standard lethal fish survey is recommended
    - Non-lethal fish survey or Alternative Methods survey may be considered

# ALTERNATIVE METHODS

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- Site-Specific Reasons for Alternative Methods
  - Hazardous conditions
  - Unsuitable habitat for sampling
  - Presence of confounding factors
- Alternative Methods
  - Mesocosm (artificial stream) survey
  - Wild Molluscs
  - Caged bivalve survey
- McClean Lake
  - Prior EEM phases, fish community survey utilizing burbot and slimy sculpin
  - Asymmetrical harvest of fish from Exposure by external party (2017)
  - Wild bivalve survey in 2019

# WILD BIVALVE SURVEY

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- Study Design
  - Control-Impact design: reference vs. mine-effluent exposed areas on Collins Creek
  - Lethal survey of Pisidiidae clams as alternative to fish
  - Pisidiidae
    - Small freshwater clams
    - Widely distributed
    - Lifespans 3 years, maturity at year 1
    - Reproductive effort can be quantified
  - Suitable for the McClean site as they are abundant and collected with ease



# FIELD COLLECTIONS

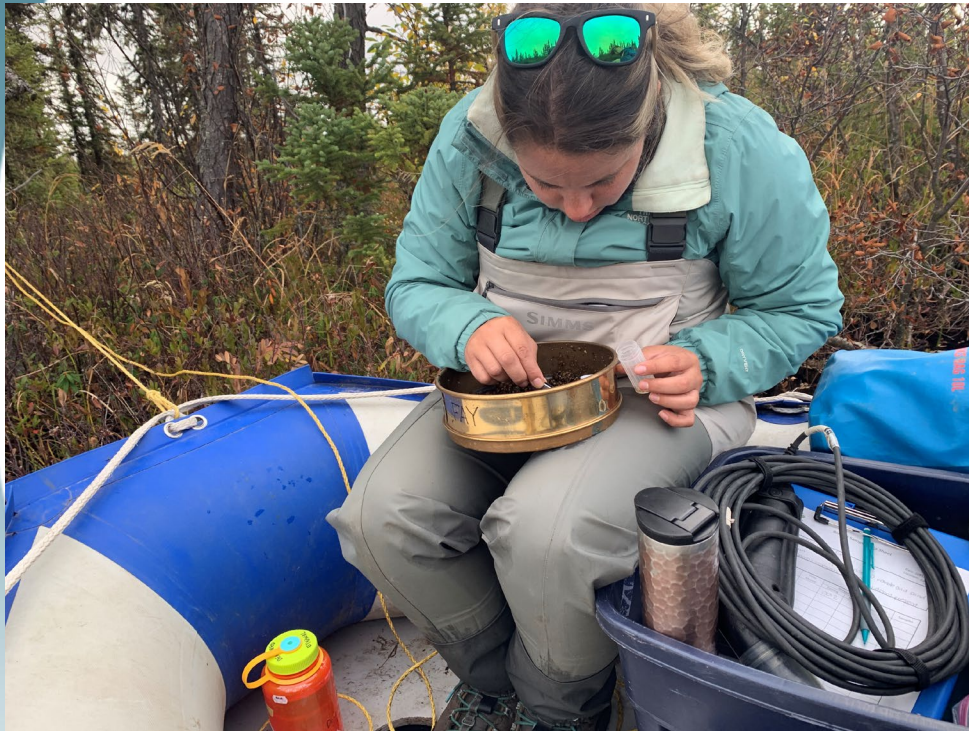
- Erosional habitats (riffles)
- Kick and sweep of materials under large substrate





# FIELD COLLECTIONS

- Initial processing in the field to collect a min of 400 adult *Pisidium* and 400 adult *Sphaerium*





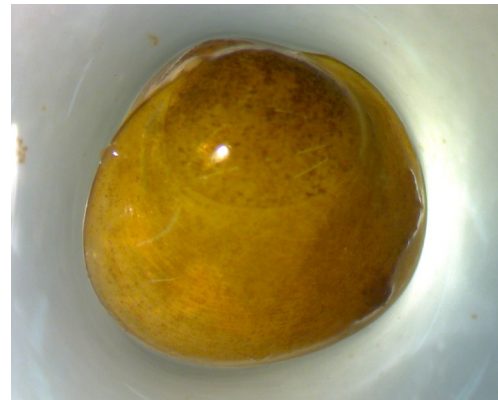
# LABORATORY PROCESSING

- Identification to species (4 species identified)
- Sentinel species selection
  - *Sphaerium nitidum*
  - *Pisidium casertanum*
- Sample sizes
  - 100 adults (per Section 9.3.4.1, EC 2012)

*Pisidium sp.*



*Sphaerium nitidum*

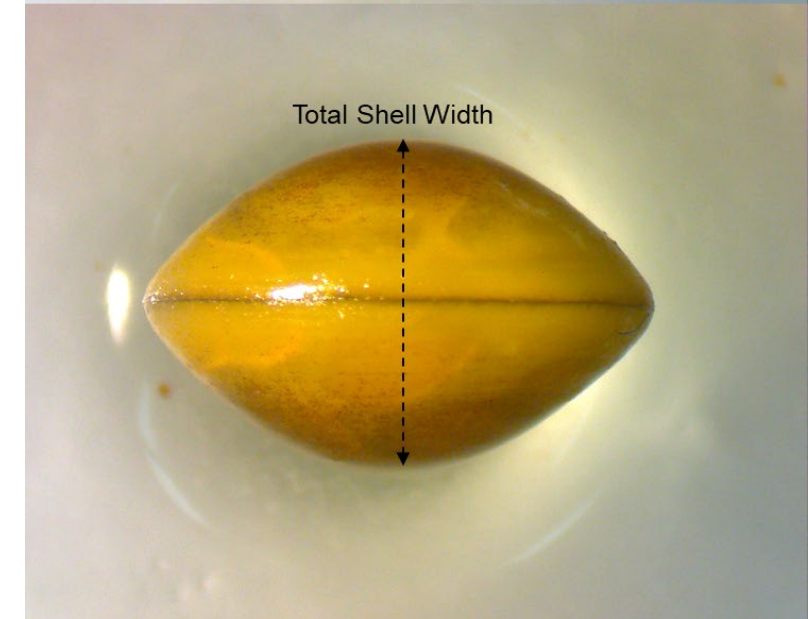
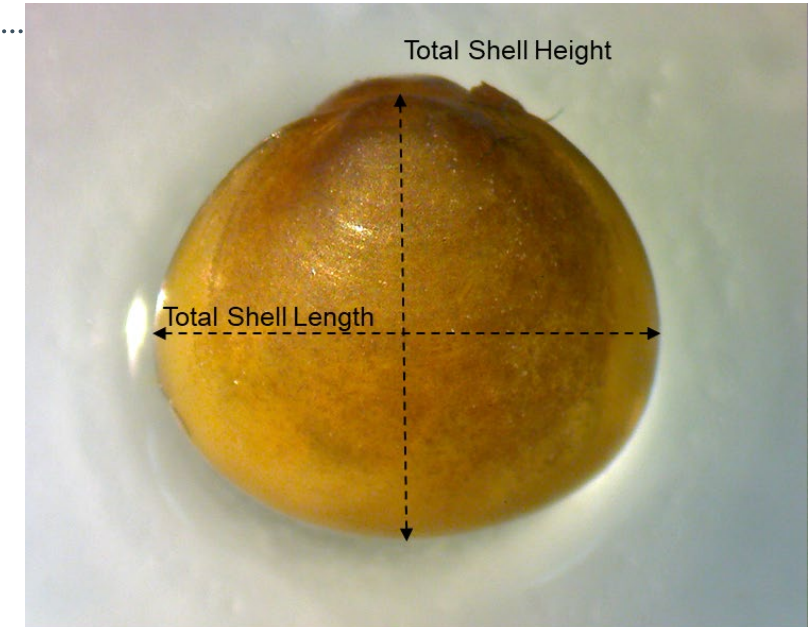


*Musculium partumeium*



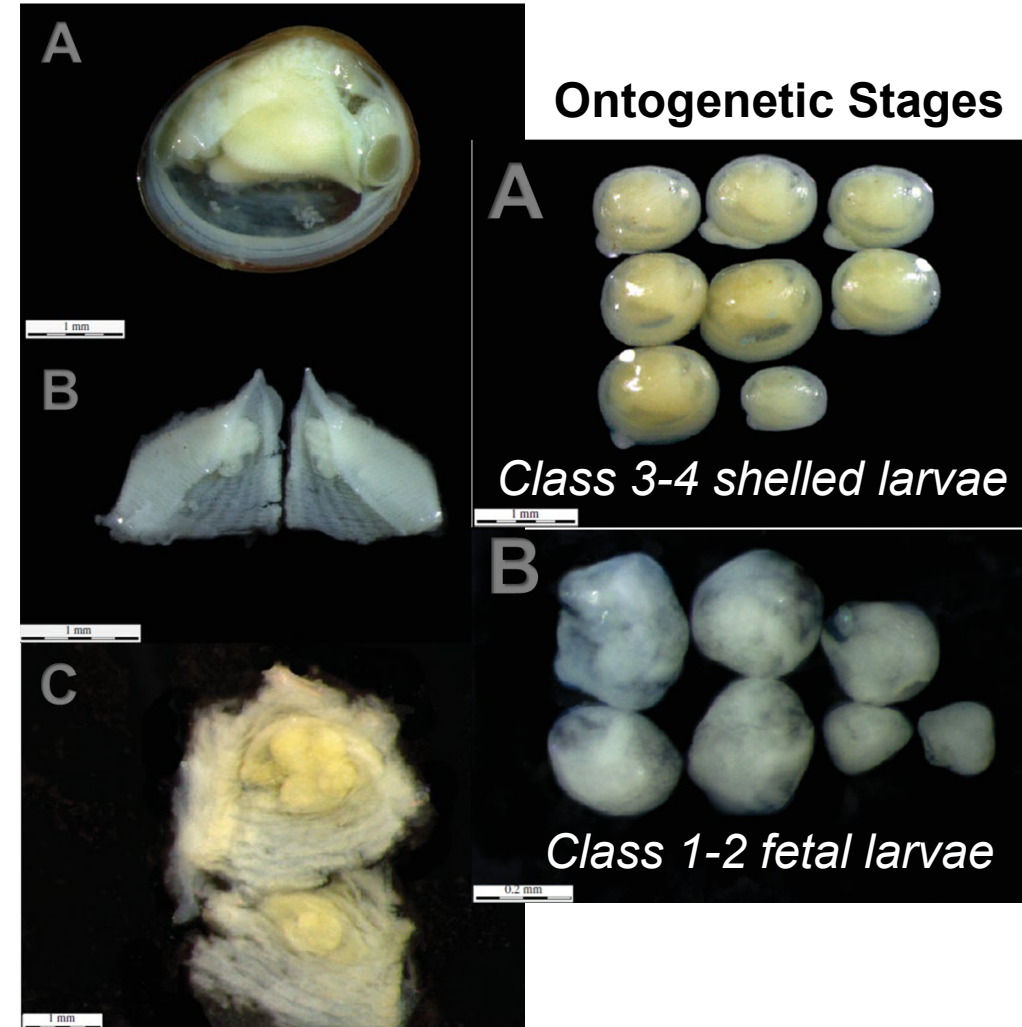
# LENGTH MEASUREMENTS

- Bivalve processing involved measuring:
  - Total Shell Length
  - Total Shell Height
  - Total Shell Width
  - Shell Volume ( $L \times H \times W$ )
- Ocular micrometer fitted to stereomicroscope
  - Converted to mm
- QAQC
  - 10% measurements repeated
  - Average % difference 2.4%



# DISSECTIONS

- Dissected N = 50 for estimate of # larvae of various ontogenetic stages
- Marsupial sacs dissection, embryos removed and measured
- Morphological types of ontogenetic stages described for *Sphaerium*
  - Class 1-2 larvae
  - Class 3-4 larvae





# WEIGHT MEASUREMENTS

- Weighed N = 50 with microbalance (resolution 1  $\mu\text{g}$ )
  - Whole Animal Wet Weight (WAWW)
  - Whole Animal Dry Weight (WADW)
  - Dry Shell Weight (DSW)
  - Ash Free Dry Weight (AFDW)
- DSW and AFDW obtained by burning in a muffle furnace at 550 °C for 2 hours until all the organic material was removed and only ashed shells remained.



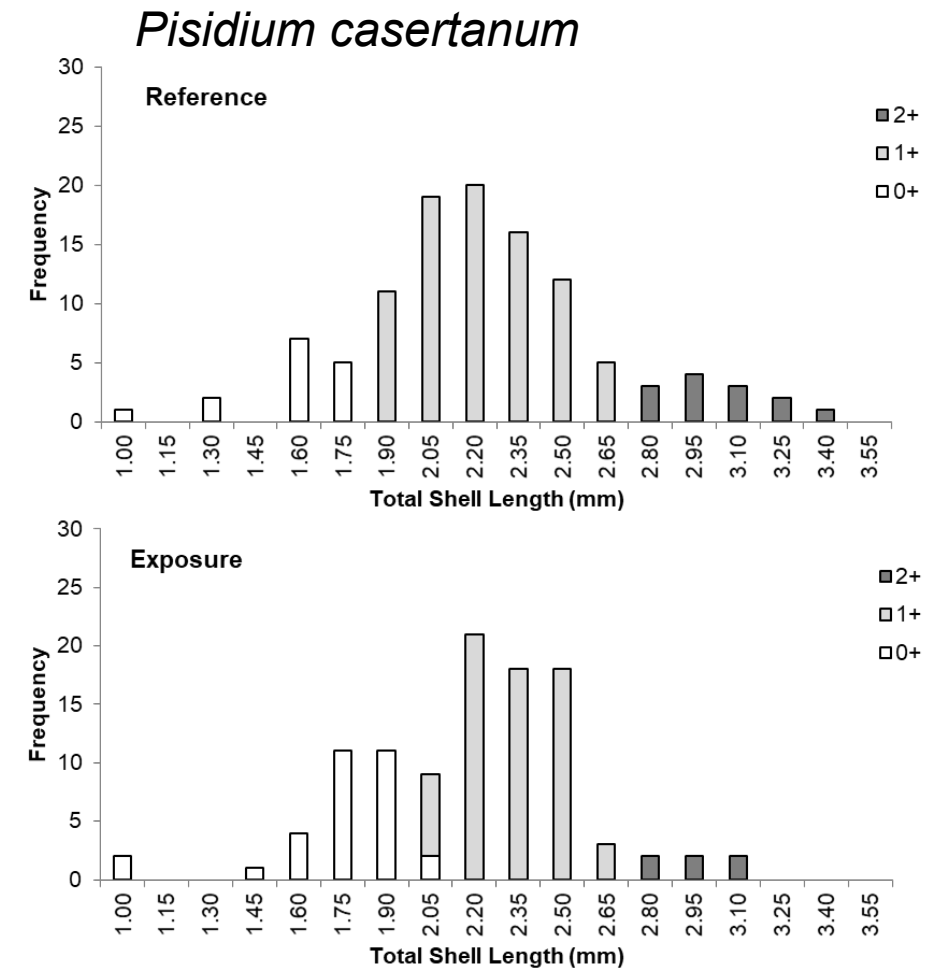
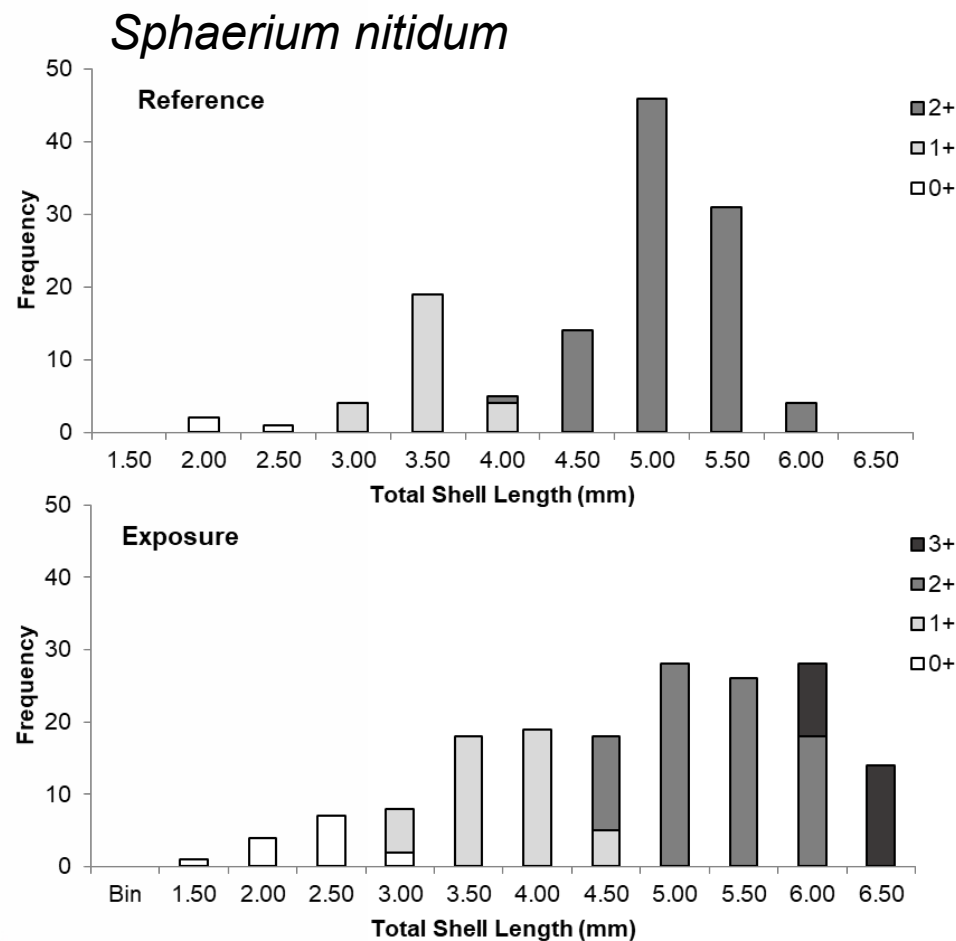
# DATA ANALYSES

Fish survey effect indicators and endpoints for various study designs and the appropriate statistical analyses

Effect Indicator		Standard Lethal Fish Survey	Non-Lethal Fish Survey	Wild Bivalve
Survival	Core	Age (ANOVA)	Length-frequency analysis (2 sample Kolmogorov-Smirnov test)	Shell length-frequency analysis (2 sample Kolmogorov-Smirnov test)
	Supporting	Age and length-frequency distributions (2 sample Kolmogorov-Smirnov test)	Age frequency distributions (2 sample Kolmogorov-Smirnov test)	Percent of 1+ and 2+ individuals (Confidence limits on percentages)
Energy Use (Growth)	Core	Size (body weight) at age (ANCOVA)	Size (length and weight) of YOY (age 0+) fish at end of growth period (ANOVA)	WADW, AFDW and shell Length at age (ANCOVA)
	Supporting	Size (length) at age (ANCOVA)	Size (length and weight) at age (ANCOVA)	
Energy Use (Reproduction)	Core	Gonad size at body weight (ANCOVA)	Relative abundance of YOY (% composition of YOY) (Confidence Limits on proportions)	Brood size (ANOVA)
	Supporting	Gonad size at length (ANCOVA)		Brood size (class 3-4) (ANOVA)
Energy Storage (Condition)	Core	Body weight relative to length (ANCOVA)	Body weight relative to length (ANCOVA)	WADW relative to length (ANCOVA)
		Liver size at body weight (ANCOVA)		AFDW relative to length (ANCOVA)

# AGE ASSIGNMENTS

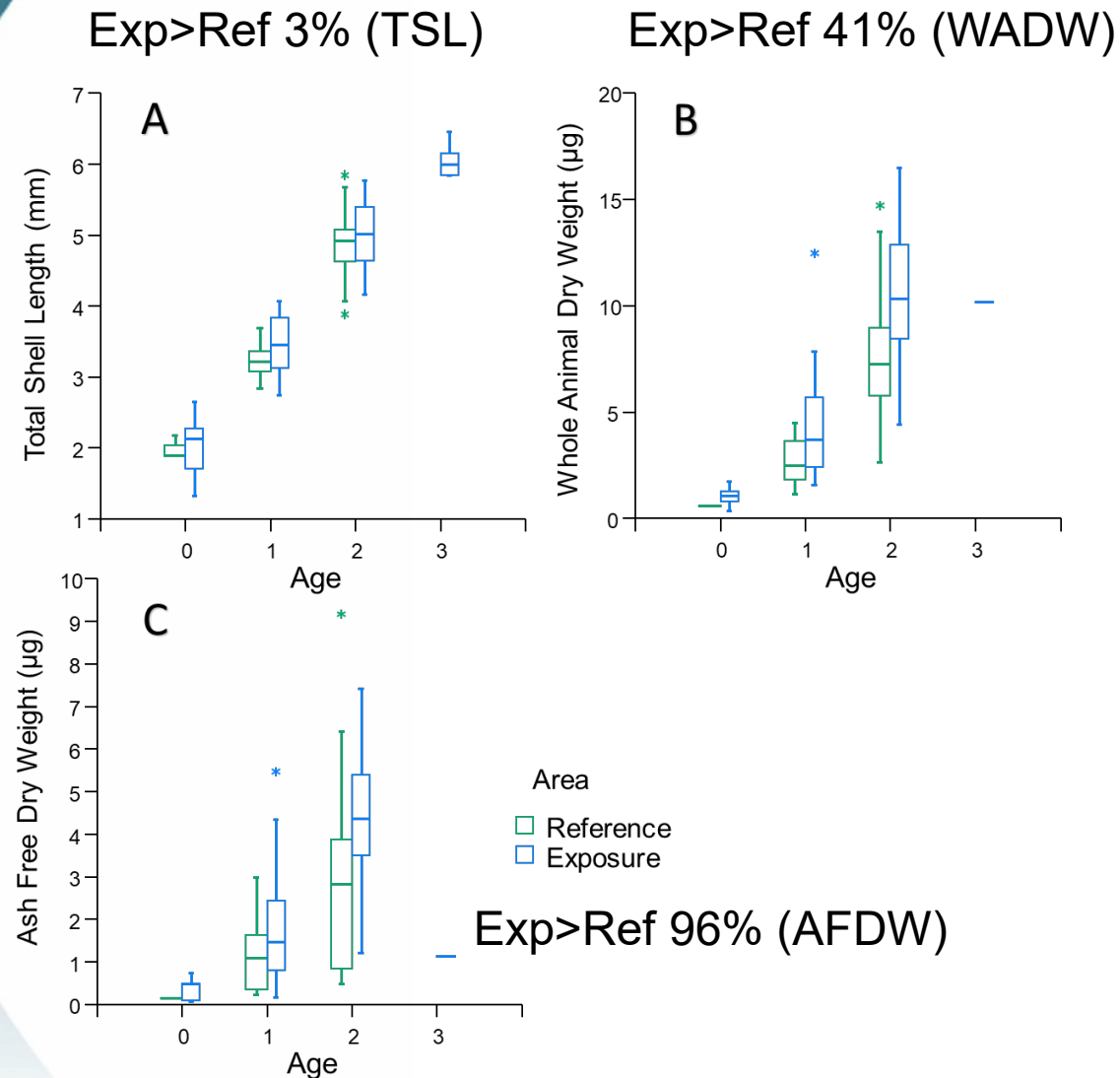
- Bivalve age classes were assigned to each individual from length-frequency distributions



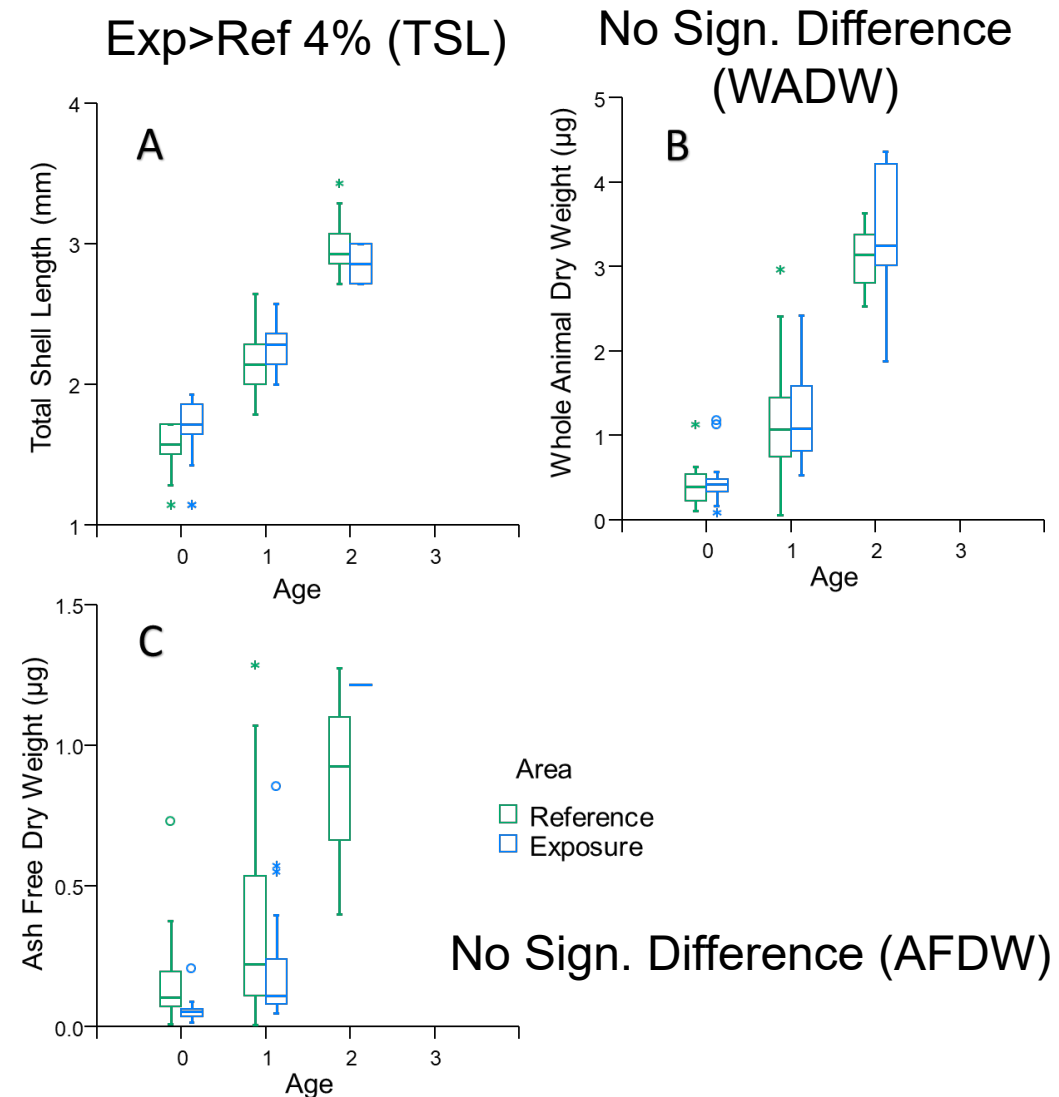


# RESULTS - GROWTH

## *Sphaerium nitidum*

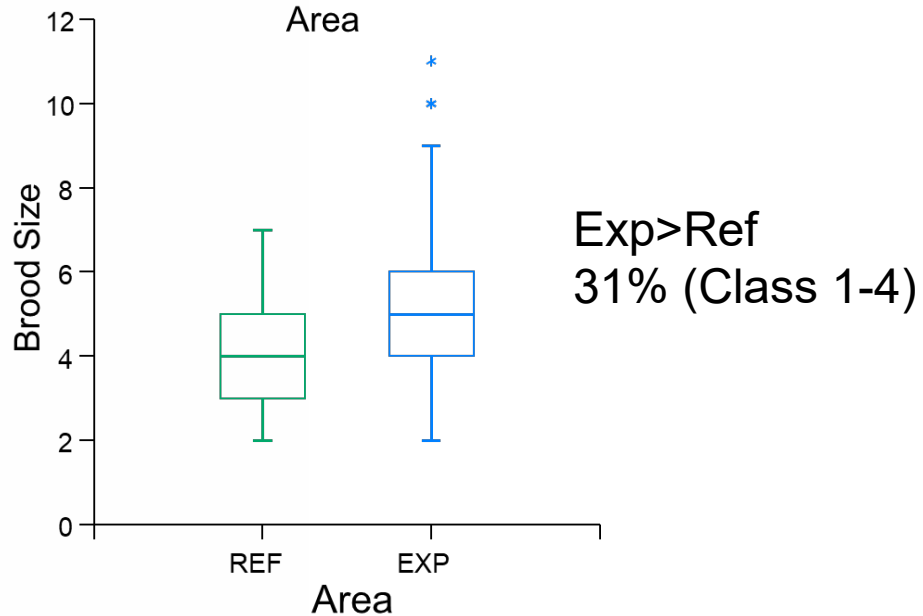
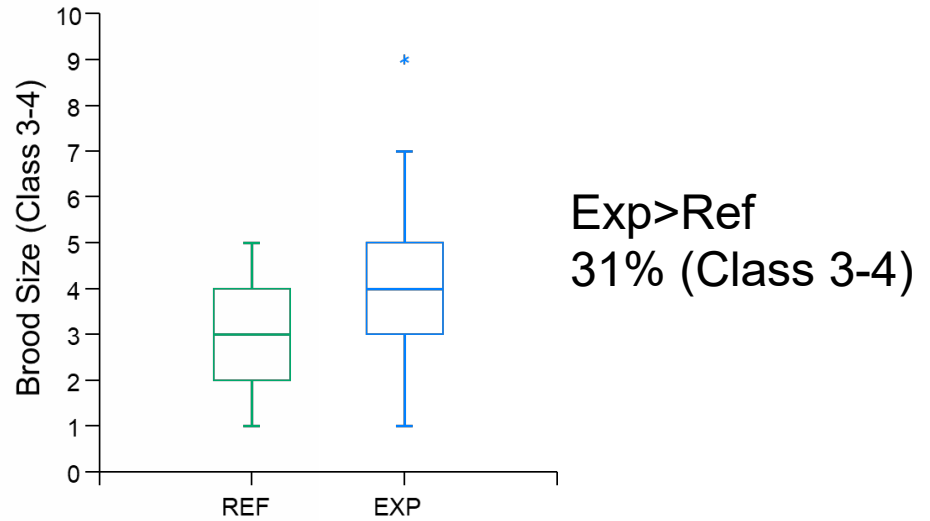


## *Pisidium casertanum*

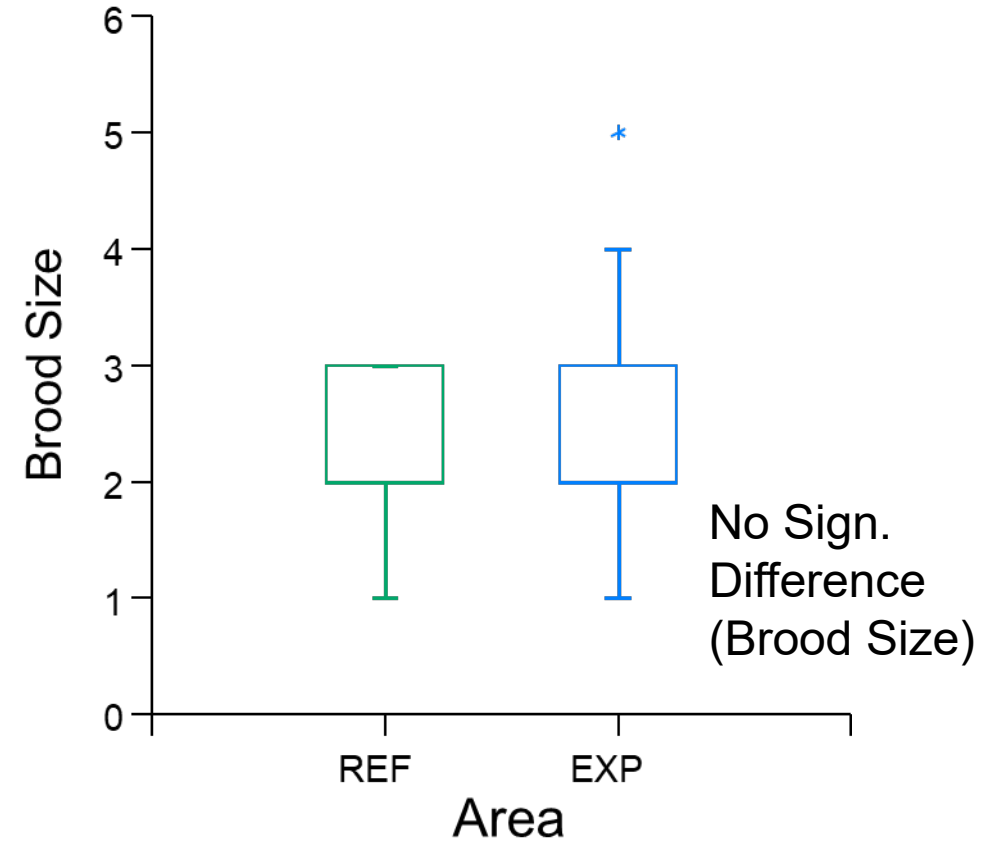


# RESULTS - REPRODUCTION

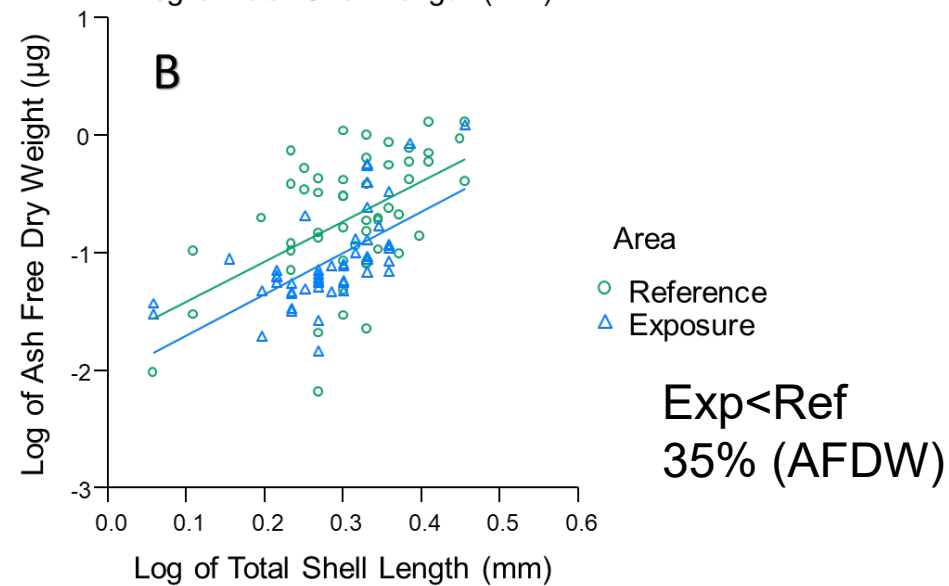
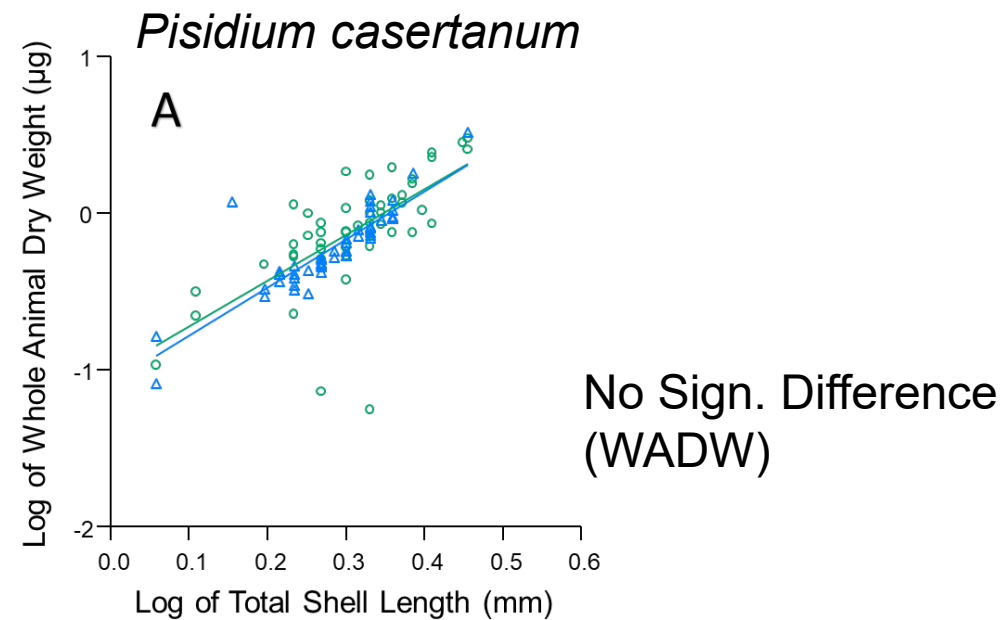
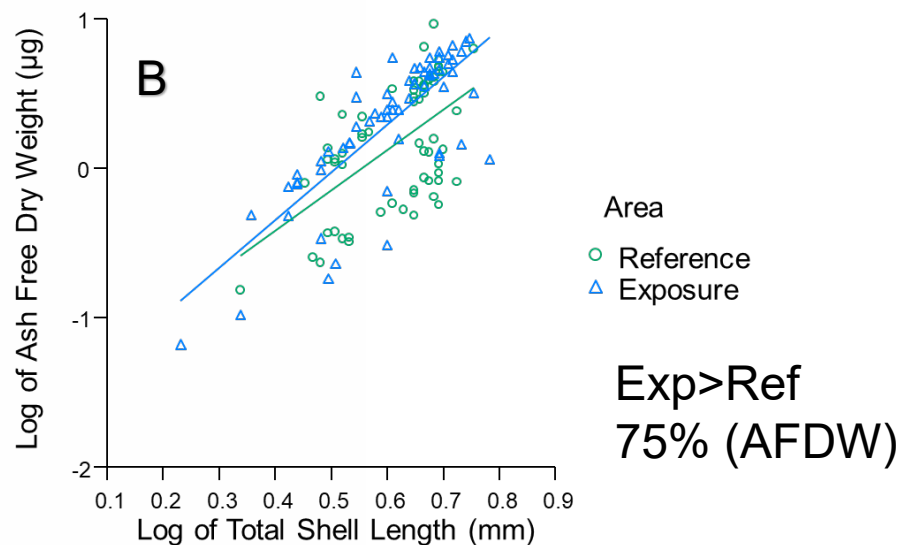
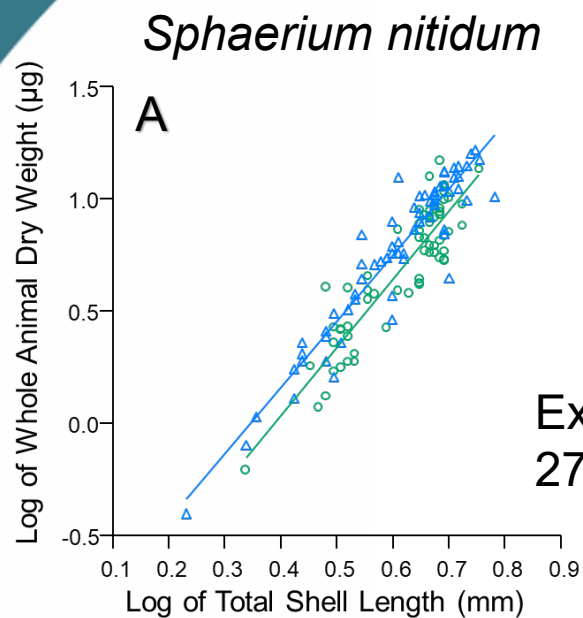
## *Sphaerium nitidum*



## *Pisidium casertanum*



# RESULTS - CONDITION





# CONCLUSIONS

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- Phase 6 bivalve survey produced significant differences in survival, growth and energy storage
- Variations in effect variables for bivalves in 2019 were large ( $> \text{CES}$ ), but generally consistent with variations in effect variables for fish in the previous cycle.
- Feasible alternative to fish population survey
- Adequately provided required data and meeting EEM program objectives
- Cost effective, required 1 day sampling per area and collection of data in the lab was similar to effort for fish dissections



Thank You.  
Questions?