Factors related to growth inhibition in juvenile mussels exposed to ambient stream conditions

Wendell R. Haag  
US Forest Service

Jacob Culp  
KY Division of Water

Monte McGregor  
KY Dept Fish and Wildlife Resources

Steve Price  
University of Kentucky

Lesley Sneed  
Kentucky State University
Background: 2015 study

- High survival
- Very low growth in defaunated streams
Background: 2015 study

Nutrients, TOC, temperature

Pesticides, NO₃

\[ \log y = 0.032(x) - 1.578 \]
\[ R^2 = 0.434, P = 0.0055 \]

\[ \log y = -0.095(x) - 1.722 \]
\[ R^2 = 0.577, P < 0.0001 \]
2016 study goals

• Repeated 2015 study at subset of 10 sites from 2015

• Measured diatom and bacterial abundance and assemblage composition

• Measured C and N stable isotopes in mussels, periphyton, and seston

• Examined physiological status with metabolomics
Study design

- Exposed juvenile mussels in silos to ambient conditions in 10 streams; five low-growth and five ‘normal’ growth streams

- Pocketbook mussel, *Lampsilis cardium*: 4 months old, mean length = 5.0 mm, mass = 0.026 g.

- 3 silos/site; 25 mussels/silo

- 91-day exposure period (average; June-September)
## Study streams

<table>
<thead>
<tr>
<th>Low-growth sites</th>
<th>‘Normal-growth’ sites</th>
</tr>
</thead>
<tbody>
<tr>
<td>Drakes Creek</td>
<td>Beech Fork</td>
</tr>
<tr>
<td>Horse Lick Creek*</td>
<td>South Fork Licking River</td>
</tr>
<tr>
<td>Little River</td>
<td>South Fork Kentucky River</td>
</tr>
<tr>
<td>Nolin River*</td>
<td>Rockcastle River</td>
</tr>
<tr>
<td>Red River</td>
<td>Russell Creek</td>
</tr>
</tbody>
</table>
Methods – mussel food resources

• Sampled diatom assemblages at each site:
  • Replicate, quantitative rock scrapings and sediment samples.
  • One sample/site, approx. mid-way through study.
  • Diatoms identified and counted (EnviroScience, Inc)

• Sampled bacterial assemblages
  • Collected one composite sediment sample/site
  • Microbes identified to major groupings with fatty acid methyl ester profiles (FAME).
Methods – isotopes

- Sampled periphyton and seston at each site
  - One sample/site, approx. mid-way through study
  - Periphyton: same methods as for diatoms
  - Seston: 9-500 ml samples filtered through 50 µm filter, composited, and 50 ml subsample pulled across 0.7 µm filter
  - Samples frozen

- Initial sample of mussels frozen prior to deployment
- Subsample of mussels frozen mid-way through study (July)
- Final sample frozen at end

All samples analyzed at University of Georgia
Methods – metabolomics

- Final sample flash-frozen at end of study
- Samples pooled from Little and Red rivers
- Samples analyzed by Metabolon, Inc., Durham, NC
- Measured 417 named biochemicals
Results

Silos: retrieval = 87%, range = 33-100%
median survival = 74%
Results

The chart shows the mean increase in length (mm) for different creeks and streams. The Y-axis represents the mean increase in length, ranging from -2 to 12 mm. The X-axis lists various creeks and streams, including Horse Lick Creek, Nolin River, Low growth, and Normal growth.
Diatoms

- 227 species present
- Appalachian vs. other streams
- Little relationship to growth
Diatoms
Bacteria

- Seven major groupings identified
- Little pattern with regard to physiology or growth

Non-metric multidimensional scaling
Final stress = 6.059
Stable isotopes
Stable isotopes
Stable isotopes

Mussels

Periphyton

Seston
Metabolomics

- 417 biochemicals detected
- 247 significantly different between low and normal growth groups.
Metabolomics

- Strong separation on PC 2
- PC 2 strongly related to growth
Metabolomics

• Biochemical patterns indicative of following characteristics of low-growth animals:
  • Decreased Krebs Cycle activity
  • Lower amino acid levels and metabolism
  • Increased fatty acid oxidation

= starvation?
Summary

• Not much evidence for major differences in food resources among streams; coarse data

• Stable isotopes: mussels in low growth streams either not obtaining or assimilating available food resources

• Starvation appears to be a consistent trait
Thanks!

- Staff at Center for Mollusk Conservation
- Daniel Boone National Forest
- Ohio River Basin Fish Habitat Partnership
- US Fish and Wildlife Service, Frankfort, KY field office
- Robert Bringolf
- Andrea Fritts
- Carla Atkinson
- Dozens of field helpers