Iron Toxicity to Aquatic Life

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\textsuperscript{1}\textsuperscript{2}\textsuperscript{3}


A Re-calculation of the Chronic Iron Criterion Value


Not talking about the 0.3 mg/l drinking water standard
Iron (Fe) is prevalent and unique

History of the current national iron (Fe) standard

How Criterion Values for aquatic life are derived

A re-calculation of the Chronic Iron Criterion Value

Not talking about the 0.3 mg/l drinking water standard

Literature Search
Single Species Experiments
Mesocosms Experiments

Fe x Cu x Zn experiments

Overall Observations
Questions / Discussion
>23,000 abandoned mines in Colorado per USEPA

Map: USGS Mineral Resources Data System (MRDS)
Numerous mine photos
Ferrous $\text{Fe}^{++}$ Fe (II)

Ferric $\text{Fe}^{+++}$ Fe (III)
Iron Hydroxide $\text{Fe(OH)}_3$ Yellow
Iron Oxides $\text{Fe}_2\text{O}_3$ Red/Rusty
Floc (controversial term)
Precipitates
An estimated 20,000-50,000 mines in the western United States produce acid mine drainage (AMD) which seriously affects 5,000-10,000 miles of streams (USDA 1993) and has been described as the greatest water quality problem in the Rocky Mountain region (Mineral Policy Center 1997).

Image:

**Bold Statement:** "In arid western states ... 75-80% of terrestrial vertebrates..."


Chaney, E., W. Elmore, and W. S. Platts. 1990. Livestock grazing on western riparian areas. USEPA

Ferrous Fe++ Fe (II)
Ferric Fe+++ Fe (III)
Iron Hydroxide Fe(OH)₃
Iron Oxides Fe₂O₃ Red/Rusty Floc (controversial term)
Precipitates
Deposition of Ferric Fe

More Toxic
Low Dissolved Oxygen
Low pH

Less Toxic
Fe^{+++}, Ferric
Direct Effects

- Aqueous toxicant passes gill membrane and a chemical mode of toxicity occurs

Indirect Effects

- Turbidity reduces Primary Productivity
- Deposition reduces habitat and induces drift
- Leaf litter decay rates are reduced
- Prey species is lost due to direct toxicity

based on field observations

- 1976 RED BOOK
- CDPHE John Woodling
- 1000µg/L = 1mg/L
- Total Recoverable
- Not experimentally derived
based on laboratory toxicity tests

Disclaimer:
It is not a “Standard” until the state or nation adopts it formally.

What I am showing is a ‘Calculated Criterion Value’ or ‘Calculated Final Chronic Value’
based on laboratory toxicity tests

96 Hour Survival

PPB

0.0
0.2
0.4
0.6
0.8
1.0

50
100
150
200

LC$_{50}$ = 110ppb

based on laboratory toxicity tests
based on laboratory toxicity tests

EC\(_{20}\) = For Chronic

LC\(_{50}\) = 110 ppb

based on laboratory toxicity tests

LC\(_{50}\) = 110 ppb
Based on laboratory toxicity tests

**Acute Criteria/Standard (96h)**
- Genus 1 = 10ppb
- Genus 2 = 20ppb
- Genus 3 = 70ppb
- Genus 4 = 100ppb
- Genus 5 = 110ppb
- Genus 6 = 210ppb
- Genus 7 = 217ppb
- Genus 8 = 621ppb
- Genus 9 = 638ppb
- Genus 10 = 780ppb
- Genus 11 = 990ppb

95% of Organisms

**Chronic Criteria/Standard**
- Genus 1 = 01ppb
- Genus 2 = 05ppb
- Genus 3 = 08ppb
- Genus 4 = 10ppb
- Genus 5 = 20ppb
- Genus 6 = 21ppb
- Genus 7 = 217ppb
- Genus 8 = 280ppb

95% of Organisms

\[ \text{EC}_{20} = \text{Effect Concentrations that affects 20\% of organisms} \]

**Estimate the 95th percentile with line fitting**
How Many Taxa?

- ~11,000 invertebrate taxa
- ~1,200 fish
- 500-1200 algae
- Meiofauna
- Fungus
- Bacteria
- Many interactions

Minimum 8 Taxa:
- At least 1 Salmonid
- At least 1 non-Salmonid fish
- At least 1 pelagic crustacean
- At least 1 benthic crustacean
- ...one from a 3rd family in Chordata
- ...one from a phylum other than Chordata and Arthropoda
- At least one insect
- ...one from a phylum not represented

Minimum 8 Taxa:
- Literature search:
  - EPA’s ECOTOX Database
  - Web of Science
  - Ebsco
  - N.I.H’s PubMed
  - Kay.... Our librarian
  - Google Scholar
  - Google.com .fr .at .au .be .ca...
Minimum 8 Taxa:

8

“Acceptable” Literature
-EPA & ASTM
-Highly oxygenated
-Neutral pH (6.5-9)
-Ferric only
-≥ 25d (≥7 for Daphnia)
-Genera in Colorado

Minimum 8 Taxa:

CPW experiments:
Highly oxygenated
Neutral pH (6.5-8.5)
> 30d
Genera in Colorado
Neutralized Fe(III)Cl₂-6 H₂O with Sodium Hydroxide
Vigorously aerated Fe stock

Red= CPW experiments
### 8 Minimum Taxa

#### At least 1 Salmonid

<table>
<thead>
<tr>
<th>Scientific name</th>
<th>Common name</th>
<th>Chronic Value µg/l</th>
<th>SMCV µg/l</th>
<th>GMCV µg/l</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Salvelinus fontinalis</em></td>
<td>brook trout</td>
<td>10500</td>
<td>10365</td>
<td>10365</td>
<td>Smith and Sykora 1976</td>
</tr>
<tr>
<td><em>Salvelinus fontinalis</em></td>
<td>brook trout</td>
<td>10231</td>
<td></td>
<td></td>
<td>Sykora et al. 1972</td>
</tr>
<tr>
<td><em>Salmo trutta</em></td>
<td>brown trout</td>
<td>5146</td>
<td>5146</td>
<td>5146</td>
<td>Co. Parks &amp; Wildlife</td>
</tr>
<tr>
<td><em>Oncorhynchus kisutch</em></td>
<td>coho salmon</td>
<td>1889</td>
<td>1889</td>
<td>2510</td>
<td>Smith and Sykora 1976</td>
</tr>
<tr>
<td><em>Oncorhynchus mykiss</em></td>
<td>rainbow trout (egg mort)</td>
<td>1483</td>
<td>3335</td>
<td></td>
<td>Goettl and Davies 1977</td>
</tr>
<tr>
<td><em>Oncorhynchus mykiss</em></td>
<td>rainbow trout</td>
<td>7500</td>
<td></td>
<td></td>
<td>Steffens et al. 1993</td>
</tr>
<tr>
<td><em>Prosopium williamsoni</em></td>
<td>mt. whitefish (growth)</td>
<td>935</td>
<td>935</td>
<td>935</td>
<td>Co. Parks &amp; Wildlife</td>
</tr>
</tbody>
</table>

Existing Fe Standard: 1000µg/L = 1mg/L

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### At least 1 non-Salmonid fish

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<thead>
<tr>
<th>Scientific name</th>
<th>Common name</th>
<th>Chronic Value µg/l</th>
<th>SMCV µg/l</th>
<th>GMCV µg/l</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Pimephales promelas</em></td>
<td>fathead minnow</td>
<td>569</td>
<td>1067</td>
<td>1067</td>
<td>Birge et al. 1985</td>
</tr>
<tr>
<td><em>Pimephales promelas</em></td>
<td>fathead minnow</td>
<td>2000</td>
<td></td>
<td></td>
<td>Smith et al. 1973</td>
</tr>
</tbody>
</table>

Existing Fe Standard: 1000µg/L = 1mg/L
### 8 Minimum Taxa

#### At least 1 pelagic crustacean

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<tr>
<th>Scientific name</th>
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<th>Chronic Value µg/l</th>
<th>SMCV µg/l</th>
<th>GMCV µg/l</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Daphnia carinata</em></td>
<td>cladoceran</td>
<td>2419</td>
<td>2419</td>
<td>1130</td>
<td>Van Dam et al. 1998</td>
</tr>
<tr>
<td><em>Daphnia longispina</em></td>
<td>cladoceran</td>
<td>1690</td>
<td>1690</td>
<td></td>
<td>Randall et al. 1999</td>
</tr>
<tr>
<td><em>Daphnia magna</em></td>
<td>cladoceran</td>
<td>4380</td>
<td>890</td>
<td></td>
<td>Biesinger and Christensen 1972</td>
</tr>
<tr>
<td><em>Daphnia pulex</em></td>
<td>cladoceran</td>
<td>958</td>
<td>958</td>
<td></td>
<td>Dave 1984</td>
</tr>
</tbody>
</table>

Existing Fe Standard: 1000µg/L = 1mg/L

### 8 Minimum Taxa

#### At least 1 benthic crustacean

<table>
<thead>
<tr>
<th>Scientific name</th>
<th>Common name</th>
<th>Chronic Value µg/l</th>
<th>SMCV µg/l</th>
<th>GMCV µg/l</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Orconectes limosus</em></td>
<td>crayfish</td>
<td>22000</td>
<td>22000</td>
<td>22000</td>
<td>Boutet and Chaisemartin 1973</td>
</tr>
</tbody>
</table>

Existing Fe Standard: 1000µg/L = 1mg/L
### 8 Minimum Taxa
**One from a 3rd family in Chordata**

<table>
<thead>
<tr>
<th>Scientific name</th>
<th>Common name</th>
<th>Chronic Value µg/l</th>
<th>SMCV µg/l</th>
<th>GMCV µg/l</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bufo boreas</td>
<td>boreal toad</td>
<td>2798</td>
<td>2798</td>
<td>2798</td>
<td>Co. Parks &amp; Wildlife</td>
</tr>
</tbody>
</table>

**Existing Fe Standard:**
1000µg/L = 1mg/L

### 8 Minimum Taxa
**One from a phylum other than Chordata and Arthropoda**

<table>
<thead>
<tr>
<th>Scientific name</th>
<th>Common name</th>
<th>Chronic Value µg/l</th>
<th>SMCV µg/l</th>
<th>GMCV µg/l</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Dugesia dorotocephala</em></td>
<td>planarian</td>
<td>40134</td>
<td>40134</td>
<td>40134</td>
<td>Co. Parks &amp; Wildlife</td>
</tr>
</tbody>
</table>
Minimum Taxa

At least one insect

<table>
<thead>
<tr>
<th>Scientific Name</th>
<th>Common name</th>
<th>Chronic Value µg/l</th>
<th>SMCV µg/l</th>
<th>GMCV µg/l</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Hexagenia</em></td>
<td>mayfly</td>
<td>7863</td>
<td>7863</td>
<td>7863</td>
<td>Co. Parks &amp; Wildlife</td>
</tr>
</tbody>
</table>

Existing Fe Standard: 1000µg/L = 1mg/L

Minimum Taxa

one from a phylum not represented

<table>
<thead>
<tr>
<th>Scientific name</th>
<th>Common name</th>
<th>Chronic Value µg/l</th>
<th>SMCV µg/l</th>
<th>GMCV µg/l</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Lumbriculus variegatus</em></td>
<td>worm</td>
<td>880</td>
<td>880</td>
<td>880</td>
<td>Co. Parks &amp; Wildlife</td>
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</tbody>
</table>

Existing Fe Standard: 1000µg/L = 1mg/L
<table>
<thead>
<tr>
<th>Rank</th>
<th>Scientific name</th>
<th>Common name</th>
<th>Chronic Value (µg/L)</th>
<th>SMR V (µg/L)</th>
<th>GMCV (µg/L)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>12</td>
<td>Dugesia dorotocephala</td>
<td>Planarian</td>
<td>40134</td>
<td>40134</td>
<td>40134</td>
<td>This study</td>
</tr>
<tr>
<td>11</td>
<td>Dromiector limosus</td>
<td>Crayfish</td>
<td>22000</td>
<td>22000</td>
<td>22000</td>
<td>Monnet and Chaisemartin 1973</td>
</tr>
<tr>
<td>10</td>
<td>Chironomus riparius</td>
<td>Midge</td>
<td>19811</td>
<td>19811</td>
<td>19811</td>
<td>Radford 1997</td>
</tr>
<tr>
<td>9</td>
<td>Salvelinus fontinalis</td>
<td>Brook trout</td>
<td>9237</td>
<td>9237</td>
<td>9237</td>
<td>Sykora et al. 1975</td>
</tr>
<tr>
<td>8</td>
<td>Hexagenia limbata</td>
<td>Mysis</td>
<td>7863</td>
<td>7863</td>
<td>7863</td>
<td>This study</td>
</tr>
<tr>
<td>7</td>
<td>Salmo trutta</td>
<td>Brown trout</td>
<td>5146</td>
<td>5146</td>
<td>5146</td>
<td>This study</td>
</tr>
<tr>
<td>6</td>
<td>Oncorhyncus kisutch</td>
<td>Coho salmon</td>
<td>4830</td>
<td>4009</td>
<td>5056</td>
<td>Smith and Sykora 1976</td>
</tr>
<tr>
<td></td>
<td>Oncorhyncus kisutch</td>
<td>Coho salmon</td>
<td>3300</td>
<td>3300</td>
<td>3300</td>
<td>Brenter and Cooper 1978</td>
</tr>
<tr>
<td>5</td>
<td>Bufo boreas</td>
<td>Boreal toad (tadpole)</td>
<td>3145</td>
<td>3145</td>
<td>3145</td>
<td>This study</td>
</tr>
<tr>
<td>4</td>
<td>Daphnia magna</td>
<td>Cladoceran</td>
<td>4380</td>
<td>4380</td>
<td>2048</td>
<td>Biesinger and Christiansen 1972</td>
</tr>
<tr>
<td></td>
<td>Daphnia pulex</td>
<td>Cladoceran</td>
<td>958</td>
<td>958</td>
<td>958</td>
<td>Birge et al. 1985</td>
</tr>
<tr>
<td>3</td>
<td>Prosopium williamsoni</td>
<td>Mountain whitefish</td>
<td>1318</td>
<td>1318</td>
<td>1318</td>
<td>This study</td>
</tr>
<tr>
<td>2</td>
<td>Lumbriculus variegatus</td>
<td>Worm</td>
<td>870</td>
<td>870</td>
<td>870</td>
<td>This study</td>
</tr>
<tr>
<td>1</td>
<td>Pimephalas promelas</td>
<td>Fathead minnow</td>
<td>910</td>
<td>688</td>
<td>688</td>
<td>Birge et al. 1985</td>
</tr>
<tr>
<td></td>
<td>Pimephalas promelas</td>
<td>Fathead minnow</td>
<td>520</td>
<td>520</td>
<td>520</td>
<td>Smith et al. 1973</td>
</tr>
</tbody>
</table>

Existing Fe Standard: 1000µg/L = 1mg/L
<table>
<thead>
<tr>
<th>RANK</th>
<th>GENUS</th>
<th>GMCV</th>
<th>Ln(GMCV)</th>
<th>Ln(GMCV)^2</th>
<th>P=R/N+1</th>
<th>P^0.5</th>
</tr>
</thead>
<tbody>
<tr>
<td>4</td>
<td>Daphnia</td>
<td>2048</td>
<td>7.6246</td>
<td>58.1348</td>
<td>0.3077</td>
<td>0.5547</td>
</tr>
<tr>
<td>3</td>
<td>Prosopium</td>
<td>1318</td>
<td>7.1839</td>
<td>51.6080</td>
<td>0.2308</td>
<td>0.4804</td>
</tr>
<tr>
<td>2</td>
<td>Lumbriculus</td>
<td>870</td>
<td>6.7685</td>
<td>45.8125</td>
<td>0.1538</td>
<td>0.3922</td>
</tr>
<tr>
<td>1</td>
<td>Pimephales</td>
<td>688</td>
<td>6.5338</td>
<td>42.6904</td>
<td>0.0769</td>
<td>0.2774</td>
</tr>
</tbody>
</table>

| SUM  | 28.1108 | 198.2457 | 0.7692 | 1.7047 |
| SUM SQUARED | 790.2155 | 39301.3616 | 0.5917 | 2.9059 |

\[ S^2 = \frac{\text{SUM}(\ln(GMCV))^2 - (\text{SUM}(\ln(GMCV))^2)}{\text{SUM}(P) - \text{SUM}(P^{0.5})^2/4} \]

\[ L = \frac{\text{SUM}(\ln(GMCV)) - S(\text{SUM}(P^{0.5}))}{4} \]

\[ A = S(\sqrt{0.5}) + L \]

\[ FCV = e^A \]

N= Number of Genera = 26
R= Rank

Is this protective?

499 µg/l Fe

< 50% of national standard
We used the least toxic form of Fe

Some tests were static renewal...
Using mostly pelagic taxa

Should have used benthic organisms

Deposition of Fe floc
### Ranking of Test Materials

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<td>40134</td>
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<tr>
<td>11</td>
<td>Orconectes limosus</td>
<td>Crayfish</td>
<td>20000</td>
<td>20000</td>
<td>20000</td>
<td>Boutet and Chaisemartin 1973</td>
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<td>10</td>
<td>Chironomus riparius</td>
<td>Midge</td>
<td>19811</td>
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<td>688</td>
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</tbody>
</table>

**EPA & ASTM say:**

**DO NOT FEED THE ANIMALS**


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**Growth**

Longer Duration Sublethal Endpoints

Early Life Stages

Reproduction

**Existing Fe Standard:** 1000µg/L = 1mg/L
Here Comes Pete’s #1 Favorite Graphic

Ecotoxicological Levels of Biological Organization

Ecosystem responses
- Productivity, decomposition, nutrient cycling, food web structure

Community responses
- Direct: loss of sensitive species, reduced species richness
- Indirect: competition, predation

Population responses
- Abundance, sex ratios, age structure, recruitment, genetic structure

Individual responses
- Mortality, growth, reproduction, behavior

Biochemical, physiological responses
- Respiration, metabolism, Metallothionein, MFO, AChE, DNA damage


Increasing ecological relevance and spatiotemporal scale

Increasing mechanistic understanding and specificity
Favorite Graphic Number 2: Trade-Off of Control and Realism in Ecotoxicological Experiments

- **Biomonitoring**
  - Can not show causation

- **Environmental Manipulation Natural Experiments**
  - Illegal

- **Mesocosm Experiments**
  - Can be done legally

- **Field Experiments**
  - Illegal

- **Laboratory Toxicity Experiments**
Biomonitoring

Environmental Manipulation
Natural Experiments

Mesocosm Experiments

Field Experiments

Laboratory Toxicity Experiments

1976 - Presence of Fish in one basin ~1000 µg/l Fe

1976 - Presence of Fish in one basin ~1000 µg/l Fe

1976 - Presence of Fish in one basin ~1000 µg/l Fe
1976 - Presence of fish in one basin ~1000 µg/l Fe
2007 Quantile regression across many sites in Ohio.
210 µg/l to 1,740 µg/l

Biomonitoring

Environmental Manipulation Natural Experiments

Mesocosm Experiments

Field Experiments

Laboratory Toxicity Experiments

499 µg/l
Mesocosm Experiments

• What is it?.... I will give an example

• Mandated in Europe

• Will be “acceptable” data or required by EPA

• Typically find organism are more sensitive

• Multiple species interacting, Diet, Function, etc.

• Increased environmental realism
Deposition of Fe floc
Use random number generator to select trays

8 or 9 of these in a reference stream

Mesocosm Experiments

- Multiple species
- Species interactions
- Small instars
- More environmentally realistic
Based on Densities reported here:

African Elephant

Stolen From: http://patell.org/

Hyrax

Zebra

Arctopsyche grandis

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Tanytarsini

Baetis
Community Mesocosm Experiment

Only **10** days

**SUB-chronic**

100% **Ferric** Fe

Existing Fe Standard: 1000µg/L = 1mg/L
Mesocosm Video

After 4 days:
0.4 mg/L  1.0 mg/L  2.5 mg/L  6.2 mg/L  15.6 mg/L

Deposition of Fe per day in Stream Mesocosms

\[ y = 1.59x + 4.395 \]

\[ R^2 = 0.97 \]
Ecosystem: Community Metabolism/Production

Community: Structure & Diversity

Population: Abundance

Individual: Drift Behavior

Biochemical

Increasing ecological relevance and spatiotemporal scale

Increasing mechanistic understanding and specificity

Mesocosm Community Metabolism

Approximate Depth of Stream Mesocosms

Rubber Stoppers, Note: located at corners to purge air prior to assessment

Food Grade Vinyl Tubing

2 L Food Containers

Barbed Hose Fittings

Peristaltic Pump

Vinyl Tubes

Sealed in Epoxy

Note: end tube can flush to epoxy to purge air. Epoxy can be substituted for silicone or a rubber stopper

Clear Acrylic Tube

Dissolved Oxygen Probe
Mesocosm Community Metabolism

<table>
<thead>
<tr>
<th>Total Fe (mg/L)</th>
<th>mgO₂/h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>1.2</td>
</tr>
<tr>
<td>0.4</td>
<td>1.4</td>
</tr>
<tr>
<td>1</td>
<td>1.1</td>
</tr>
<tr>
<td>2.5</td>
<td>0.8</td>
</tr>
<tr>
<td>6.5</td>
<td>0.6</td>
</tr>
<tr>
<td>15</td>
<td>0.8</td>
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</tbody>
</table>

Existing Fe Standard: 1000µg/L = 1mg/L

Ecosystem: Community Metabolism/Production

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Increasing ecological relevance and spatiotemporal scale

Increasing mechanistic understanding and specificity
Mesocosm Community Response to Fe Floc
(Treatment = mg/L total Fe)
Only 3 replicates – Error bars are Standard Deviation NOT standard Error
Mesocosm Community Response to Fe Floc

Existing Fe Standard: 1000µg/L = 1mg/L

<table>
<thead>
<tr>
<th>Genus (or tribe/subfamily)</th>
<th>EC$_{20}$ (µg/L)</th>
<th>After only 10 days...</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rhithrogena sp.</td>
<td>&gt; 14073</td>
<td>3 taxa unprotected</td>
</tr>
<tr>
<td>Ephemerella sp.</td>
<td>&gt; 14073</td>
<td>by our 499 µg/L</td>
</tr>
<tr>
<td>Sweltsa sp.</td>
<td>&gt; 14073</td>
<td></td>
</tr>
<tr>
<td>Brachycentrus sp.</td>
<td>7558</td>
<td></td>
</tr>
<tr>
<td>Baetis sp.</td>
<td>4870</td>
<td></td>
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<tr>
<td>Capnia sp.</td>
<td>3697</td>
<td></td>
</tr>
<tr>
<td>Cinygmula sp.</td>
<td>1882</td>
<td></td>
</tr>
<tr>
<td>Taenionema sp.</td>
<td>1626</td>
<td></td>
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<tr>
<td>Heterlimnius sp.</td>
<td>1282</td>
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<tr>
<td>Prostoia sp.</td>
<td>1176</td>
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<tr>
<td>Orthocladiinae</td>
<td>776</td>
<td></td>
</tr>
<tr>
<td>Micrasema sp.</td>
<td>356</td>
<td></td>
</tr>
<tr>
<td>Epeorus sp.</td>
<td>335</td>
<td></td>
</tr>
<tr>
<td>Tanytarsini</td>
<td>234</td>
<td></td>
</tr>
</tbody>
</table>

EC$_{20}$ calculated with TRAP, a toxicology specific program made by the US EPA
Biomonitoring

Environmental Manipulation
Natural Experiment

? µg/l

Mesocosm Experiments

10 day

Field Experiments

499 µg/l

Laboratory Toxicity Experiments

30 day

Environmental Realism

Control & Repeatability

- Dugesia dorotocephala
- Orconectes limosus
- Chromadora riparius
- Rhithrogena sp.
- Sialis sp.
- Ephemereleia sp.
- Salvelinus fontinalis
- Hexagenia limbata
- Brochyncentrus sp.
- Salmo trutta
- Baetis sp.
- Capnia sp.
- Oncorhynchus kisutch and O. mykiss
- Buflo boneas
- Daphnia magna and D. pulex
- Oxygastra sp.
- Proastra sp.
- Laiophleba variegatus
- Orthocodia
- Pimephales promelas
- Microcentrus
- Epeorus sp.
- Tanypus sp.

Genus Mean Chronic Value (µg/l Total Iron)

Percentile

100

80

60

40

20

0

100

1000

10000

Final Chronic Value, including Mesocosm

Single Species Final Chronic Value

Current USEPA Chronic Criterion

5th Percentile
<table>
<thead>
<tr>
<th>RANK</th>
<th>GENUS</th>
<th>GMCV</th>
<th>Ln(GMCV)</th>
<th>Ln(GMCV)**2</th>
<th>P=R/N+1</th>
<th>P**0.5</th>
</tr>
</thead>
<tbody>
<tr>
<td>4</td>
<td>Pimephales</td>
<td>688</td>
<td>6.5338</td>
<td>42.6904</td>
<td>0.1481</td>
<td>0.3849</td>
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<tr>
<td>3</td>
<td>Microsema sp.</td>
<td>356.29</td>
<td>5.8757</td>
<td>34.5244</td>
<td>0.1111</td>
<td>0.3333</td>
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<tr>
<td>2</td>
<td>Epeorus sp.</td>
<td>334.5</td>
<td>5.8126</td>
<td>33.7867</td>
<td>0.0741</td>
<td>0.2722</td>
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<tr>
<td>1</td>
<td>Tanytarsini (Tribe)</td>
<td>233.65</td>
<td>5.4538</td>
<td>29.7442</td>
<td>0.0370</td>
<td>0.1925</td>
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SUM

<table>
<thead>
<tr>
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</thead>
<tbody>
<tr>
<td>23.6760</td>
</tr>
<tr>
<td>660.5527</td>
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</table>

SUM SQUARED

<table>
<thead>
<tr>
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</thead>
<tbody>
<tr>
<td>140.7457</td>
</tr>
<tr>
<td>19809.3584</td>
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</tbody>
</table>

S2 = SUM(LnGMCV)/2 - (SUM(LnGMCV)/2)^2/4 - SUM(P)/SUM(P**0.5)/4

L=(SUM(LnGMCV)-S*SUM(P**0.5))/4

A=S*SQRT(0.5) + L

FCV=EXP(A)

N= Number of Genera = 26
R= Rank

251 µg/l Fe
25% of national standard

Environmental Realism

Control & Repeatability

1976 - Presence of Fish in one basin ~1000 µg/l Fe
2007 Quantile regression across many sites in Ohio. 210µg/l to 1,740 µg/l

Mesocosm Experiments

499 µg/l

Laboratory Toxicity Experiments
Is this protective?

• Only 10 days duration. Not 30 days. Not a life cycle

If someone wants to sponsor a 30 day trial please contact Prof. William H Clements at Colorado State University.

2011 - Fe x Zn+Cu Periphyton and Invertebrate Communities

<table>
<thead>
<tr>
<th></th>
<th>No Cu &amp; Zn</th>
<th>LM: 10 µg/L Cu</th>
<th>HM: 50 µg/L Cu</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Fe x3</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>600 µg/l Fe x3</td>
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<td></td>
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</tr>
</tbody>
</table>

10 day exposure
Ecosystem
Community
Population
Individual

Increasing ecological relevance and spatiotemporal scale

Biochemical: Enzymes
Cu + Zn + Fe accumulation, Protein

Periphyton Accumulation of Cu & Zn

ANOVA
Fe: p<0.001
M: p<0.001

ANOVA
Fe: p<0.001
M: p<0.001
Fe*M: p<0.01
Ecosystem

Community: Structure & Diversity of Diatoms

Population: Diatom Density

Individual

Biochemical

Increasing ecological relevance and spatiotemporal scale

Increasing mechanistic understanding and specificity
Overall Observations:

- Pelagic vs. Benthic
- Benthic vs. Benthic
- Small vs. Large
- Motility
- Algivorous
Overall Observations:

- Flow Through vs. Static Renewal
- Mesocosm vs. Lab.
- Naturally Colonized Substrate
- Longer Duration
• Evidence that 1,000 µg/l is under-protective  
• Using single species experiments we derived a Final Chronic Value of 499 µg/l total Fe.

• Evidence that 499 µg/l is under-protective  
• Using mesocosm experiments we derived a Final Chronic Value of 251 µg/l total Fe.

• Seems to match field based studies