Identification and Ecotoxicity of Pharmaceuticals in the Surface Waters of the Upper Mississippi and Lower Illinois Rivers

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Overview

- The need to evaluate pharmaceuticals and EDCs in major rivers in the midwest

- Overall scope of the project

- Sampling, extraction, & analysis

- Toxicity testing

- Ending Comments
Pharmaceuticals in the Water

- Estimated 30-90% excreted as active ingredient\(^1\)
- Discharged into surface waters at similar rates as pesticides\(^2\)
- Studies have found that they are prevalent in waters\(^3,4\)


\(^4\) B. Halford. 2008. Pharmaceuticals have been finding their way into our environment for a long time, but just what are they doing there? *C&E News*. 86 (8), 13-17.
Pharmaceuticals in the Water

- Some have even found seasonal relationship\(^5\)

- Widespread and in the midwest.....

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Total Number of Retail Prescription Drugs Filled at Pharmacies, 2006
(http://www.statehealthfacts.org)

In Units of 100,000

- Alaska
- District of Columbia
- Wyoming
- North Dakota
- Indiana
- Virginia
- New Jersey
- Missouri
- Tennessee
- Georgia
- Michigan
- North Carolina
- Ohio
- Illinois
- Pennsylvania
- New York
- Florida
- Texas
- California

* Denotes states with no data available.
Overall scope of the project

- Sampling, extraction, and analysis of selected pharmaceuticals, veterinary antibiotics and endocrine disrupting compounds (EDCs) from surface waters
  - Along the Mississippi River (MR) & the Illinois River (IR)
  - In tributaries to both rivers

- Use results to determine chemicals to use in aquatic toxicity tests
  - Individually
  - In mixtures at concentrations found in surface waters
Sampling

- Identification of sampling sites
- Weekly or biweekly sampling
- Periodic hourly sampling (24-48hr)
  - ISCO automated samplers

Use the results to go into the toxicity assays
Sampling Locations

- Alongside river
  - Above and below convergence

- Tributaries
  - Downstream feedlots

- Water Treatment facilities
  - Pre- and post
### Some of the Compounds of Interest (~33%)

<table>
<thead>
<tr>
<th>Compound</th>
<th>Compound</th>
<th>Compound</th>
</tr>
</thead>
<tbody>
<tr>
<td>17-a-ethynylestradiol</td>
<td>Estrone</td>
<td>Lipitor</td>
</tr>
<tr>
<td>5,5-diphenylhydantoin</td>
<td>Fluoxetine HCl</td>
<td>Naproxen</td>
</tr>
<tr>
<td>Acetaminophen</td>
<td>Gemfibrosil</td>
<td>Nexium</td>
</tr>
<tr>
<td>Amoxicillin trihydrate</td>
<td>Genistin</td>
<td>Ranitidine HCl</td>
</tr>
<tr>
<td>b-estradiol</td>
<td>Ibuprofen</td>
<td>Sertraline HCl</td>
</tr>
<tr>
<td>Ciprofloxacin</td>
<td>Irgasan</td>
<td>Singulair</td>
</tr>
<tr>
<td>Coumestrol</td>
<td>Levofloxacin</td>
<td>Sulfamethoxazole</td>
</tr>
<tr>
<td>Diltiazem HCl</td>
<td>Lexapro</td>
<td>Synthroid</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Trimethoprim</td>
</tr>
</tbody>
</table>
Extraction

- 1 L samples
- Solid phase extractions
  - In development
    - 1L H₂O samples (varying pH depending on analyte)
    - Oasis HLB (hydrophilic lipophilic balance) (50-500mg)
      - N-vinylpyrrolidone and divinylbenzene
    - Pretreatment W/5mL each of MTBE, MeOH, H₂O, then sample, dry, and elute w/90:10 MeOH:MTBE
- Analytes & Matrix interferences

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Analysis

- **LC-MS/MS**
  - Varian 310 (triple quadrupole)
  - ESI ionization source (pos & neg)
  - Current detection limits range between 500 ng/L down to 50 ng/L (ppt)
## LC-MS/MS Parameters

(for some analytes)

<table>
<thead>
<tr>
<th>Pharmaceuticals</th>
<th>Precursor Ion</th>
<th>Daughter Ion</th>
<th>ESI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acetaminophen</td>
<td>150.9</td>
<td>109.9</td>
<td>+</td>
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<tr>
<td>Gemfibrozil</td>
<td>396.1</td>
<td>317.9</td>
<td>-</td>
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<tr>
<td>Ibuprofen</td>
<td>293.7</td>
<td>249.6</td>
<td>-</td>
</tr>
<tr>
<td>Naproxen</td>
<td>415.2</td>
<td>178.1</td>
<td>-</td>
</tr>
<tr>
<td>Sulfamethoxazole</td>
<td>310.3</td>
<td>148.1</td>
<td>+</td>
</tr>
<tr>
<td>Fluoxetine</td>
<td>249.1</td>
<td>120.7</td>
<td>+</td>
</tr>
<tr>
<td>Diclofenac Sodium</td>
<td>205.0</td>
<td>160.7</td>
<td>-</td>
</tr>
<tr>
<td>Amoxicillin Trihydrate</td>
<td>172.9</td>
<td>157.6</td>
<td>-</td>
</tr>
<tr>
<td>Trimethoprim</td>
<td>250.1</td>
<td>156.0</td>
<td>+</td>
</tr>
<tr>
<td>Diltiazem HCl</td>
<td>291.1</td>
<td>230.1</td>
<td>+</td>
</tr>
</tbody>
</table>
Toxicity Testing

- Individual compounds
- Combinations
- Seasonality⁵ (if concentrations vary)

Toxicity Testing

- **Organism:** *Daphnia magna*
  - Static renewal
    - 96hr toxicity tests
  - Pulsed exposures
    - Based on 24-48 hour sampling

- **Endpoints**
  - Mortality
  - Reproduction
  - Molting
Ending Notes

- Very early in the process

- There are a lot of ecotoxicological questions that need to be answered
  - Bioavailability to organisms
  - Aquatic toxicity (lethality)
  - Sublethal effects

- Down the road.....biomonitoring
Comments/Questions