Canidae Exposure to Polychlorinated Dibenzofurans (PCDFs) of the Tittabawassee River Floodplain, Midland, MI

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Introduction

- Tittabawassee River, Midland, MI

- Presence of polychlorinated dibenzo-\(p\)-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs) in river sediment and floodplain soils

- Particularly, 2,3,7,8-tetrachlorodibenzofuran (TCDF) and 2,3,4,7,8-pentachlorodibenzofuran (PeCDF) which comprise as much as 90% of the total 2,3,7,8-tetrachlorodibenzo-\(p\)-dioxin (TCDD) equivalents (TEQs)
Tittabawassee River Project

- Tittabawassee River Ecological Risk Assessment (ERA), Midland, MI, USA
  - Multiple line of evidence approach
  - Michigan State University’s Aquatic Toxicology Laboratory

- Receptor species include: great horned owl (*Bubo virginianus*), great blue heron (*Ardea herodias*), kingfisher (*Ceryle alcyon*), tree swallow (*Tachycineta bicolor*), Eastern bluebird (*Sialis sialis*), house wren (*Troglodytes aedon*), American robin (*Turdus migratorias*) and American mink (*Mustela vison*)
The greatest dioxin-like exposure expressed as mammalian TEQs is from PeCDF and TCDF
- Mink - mean liver sum TEQs = 400 ± 74 ng/kg ww
  - $\text{TEQ}_{\text{PeCDF}} = 56 \% \text{ Total}$
  - $\text{TEQ}_{\text{TCDF}} = 7 \% \text{ Total}$ (Zwiernik et al. 2008)

While much attention has focused on bird species and mink, no work has been done to evaluate the exposure of large long-lived mammals associated with terrestrial food chains in the floodplain
Objectives

• Determine:
  – Large terrestrial mammalian predators present within the Tittabawassee River floodplain
  – Histological abnormalities
  – Examine a biomarker of exposure
  – Hepatic TEQ concentrations
  – Average Daily Dose (ADD)
  – The species most exposed to PCDFs based on enzyme activity and hepatic mammalian TEQs
Resident Floodplain Predators

• 3 genera represented within the Canidae family

• Coyote (Canis latrans), red fox (Vulpes vulpes) and gray fox (Urocyon cinereoargenteus)
Canidae Home Ranges

- Coyote
  - 10 - 40 km²

- Red Fox
  - 1 - 5 km²

- Gray Fox
  - 0.13 - 3.1 km²
Field Methods - Time

- Canids (11) were trapped and collected during the state regulated trapping season immediately downstream of Midland, Michigan in the fall of 2006
  - 3 - coyote (11/3 - 11/10)
  - 5 - red fox (10/27 - 11/10)
  - 3 - gray fox (10/19 - 11/10)
Field Methods - Trapping
Field Methods – Field Sampling

- Field lab sampling
  - Sub-sample of liver collected immediately upon euthanization and placed in liquid nitrogen for subsequent measurement of cytochrome P450 enzymatic activity; ethoxyresorufin O-deethylase (EROD) and methoxyresorufin O-deethylase (MROD) as a biomarker of exposure

  - Hide removed and carcass frozen at – 20°C until full necropsy
Laboratory Necropsy Methods

- Animals necropsied at the Michigan State University (MSU) University Research Containment Facility (URCF)

- Examined by a board certified pathologist from MSU’s Diagnostic Center for Population and Animal Health (DCPAH)

- Morphological measurements, gross observation and tissue collection
Analysis Methods

- Board certified pathologist examined lungs, heart, liver, lymph nodes, kidneys, bladder and spleen
- P450 activity and protein concentration were measured using a Fluoroscan Ascent microplate fluorometer
- Liver tissue concentrations determined by EPA extraction method 8290 and High Resolution Mass Spectrometry (HR-MS)
- Gastrointestinal contents dried, weighed and sorted
Results – Histology and Activity

• Histology
  – No gross or histological abnormalities

• Liver EROD and MROD:
  – Red fox ranged from 71.4 to 221 pmol/mg/min
  – Coyote ranged from 13.4 to 112 pmol/mg/min
  – Gray fox ranged from 11.3 to 17.8 pmol/mg/min
Hepatic TEQs and Enzyme Activity

- Enzyme Activity and Canid Hepatic TEQs
Results – % PCDF TEQs in Liver
Dietary Percentages (literature-based)

<table>
<thead>
<tr>
<th></th>
<th>Coyote</th>
<th>Red Fox</th>
</tr>
</thead>
<tbody>
<tr>
<td>Deer</td>
<td>40</td>
<td>None</td>
</tr>
<tr>
<td>Small mammal</td>
<td>20</td>
<td>24</td>
</tr>
<tr>
<td>Shrew(^1)</td>
<td>2</td>
<td>1.5</td>
</tr>
<tr>
<td>Rabbit</td>
<td>20</td>
<td>40</td>
</tr>
<tr>
<td>Bird</td>
<td>15</td>
<td>14</td>
</tr>
<tr>
<td>Invertebrate</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>Plant</td>
<td>1</td>
<td>7</td>
</tr>
<tr>
<td>Soil(^2)</td>
<td>2.8</td>
<td>2.8</td>
</tr>
</tbody>
</table>

\(^1\) proportion of shrew is estimated based on relative abundance of shrews to other small mammals in Michigan

\(^2\) soil ingestion is percent of dry weight of food ingestion; reported by Beyer et al 2004 for red fox
## Dietary Concentration and Average Daily Dose Estimate

<table>
<thead>
<tr>
<th></th>
<th>Coyote</th>
<th>Red Fox</th>
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</thead>
<tbody>
<tr>
<td>Dietary concentration¹</td>
<td>28 (87)</td>
<td>27 (149)</td>
</tr>
<tr>
<td>(ng TEQ/kg ww food)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Food ingestion rate²</td>
<td>0.023</td>
<td>0.10</td>
</tr>
<tr>
<td>(kg/kg bw/d)</td>
<td></td>
<td></td>
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<tr>
<td>Average daily dose³</td>
<td>0.83 (2.6)</td>
<td>3.6 (17)</td>
</tr>
<tr>
<td>(ng TEQ/kg bw/d)</td>
<td></td>
<td></td>
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</tbody>
</table>

All concentration data displayed as median with 95th centile in parentheses

¹based on site-specific data from MSU ERA studies on Tittabawassee River

²red fox ingestion is based on Sargeant 1978; coyote is from Laundre and Hernandez 2003
Conclusions

• 3 genera of canids (coyote, red fox and gray fox) are present in the Tittabawassee River floodplain

• These canids are exposed to detectable levels of PCDFs including PeCDF, TCDF and 1,2,3,4,7,8 hexachlorodibenzofuran (HxCDF) and can therefore be considered a *receptor* species of the Tittabawassee River
Conclusions

• EROD/MROD activity confirms exposure of these dioxin-like compounds

• Preliminary findings indicate that:
  – no morphological or histological abnormalities in 3 species/11 individuals of large mammalian terrestrial predators exposed to hepatic tissue levels of PCDFs ranging from 5.14 to 214 ng sum mammalian TEQ/kg, ww
Conclusions

• Preliminary findings also indicate that of the canids known to be on-site the red fox appears to be the most highly exposed to dioxin-like compounds
  
  – Tissue and dietary exposure assessments agree
Further Study and Future

• Age of individuals by cementum annuli count to look at variability in exposure

• Bioaccumulation Factors (BAFs)

• Jaw histology – biomarker potential
Other presentations...

- Multiple lines of evidence risk assessment for belted kingfisher exposed to PCDD/DF in the Tittabawassee River floodplain, Midland, MI USA
- A site-specific, multiple lines of evidence risk assessment of great horned owl (Bubo virginianus) exposure to PCDD/DFs in the Tittabawassee River floodplain in Midland, MI USA
- Assessing the exposure and condition of American robins along the Tittabawassee River using a multiple lines of evidence approach
- Adult and nestling band returns and long-term survival monitoring of three passerine species inhabiting the Tittabawassee River basin
- Concentrations of PCDDs and PCDFs in eggs and diets of wood ducks and hooded mergansers
- The Effects of TCDD, PeCDF and TCDF on Development of Maxillary and Mandibular Squamous Epithelial Proliferation in Mink
Thank you!

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