

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

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30th Annual Society of
Environmental Toxicology and
Chemistry Meeting

21 November 2009

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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



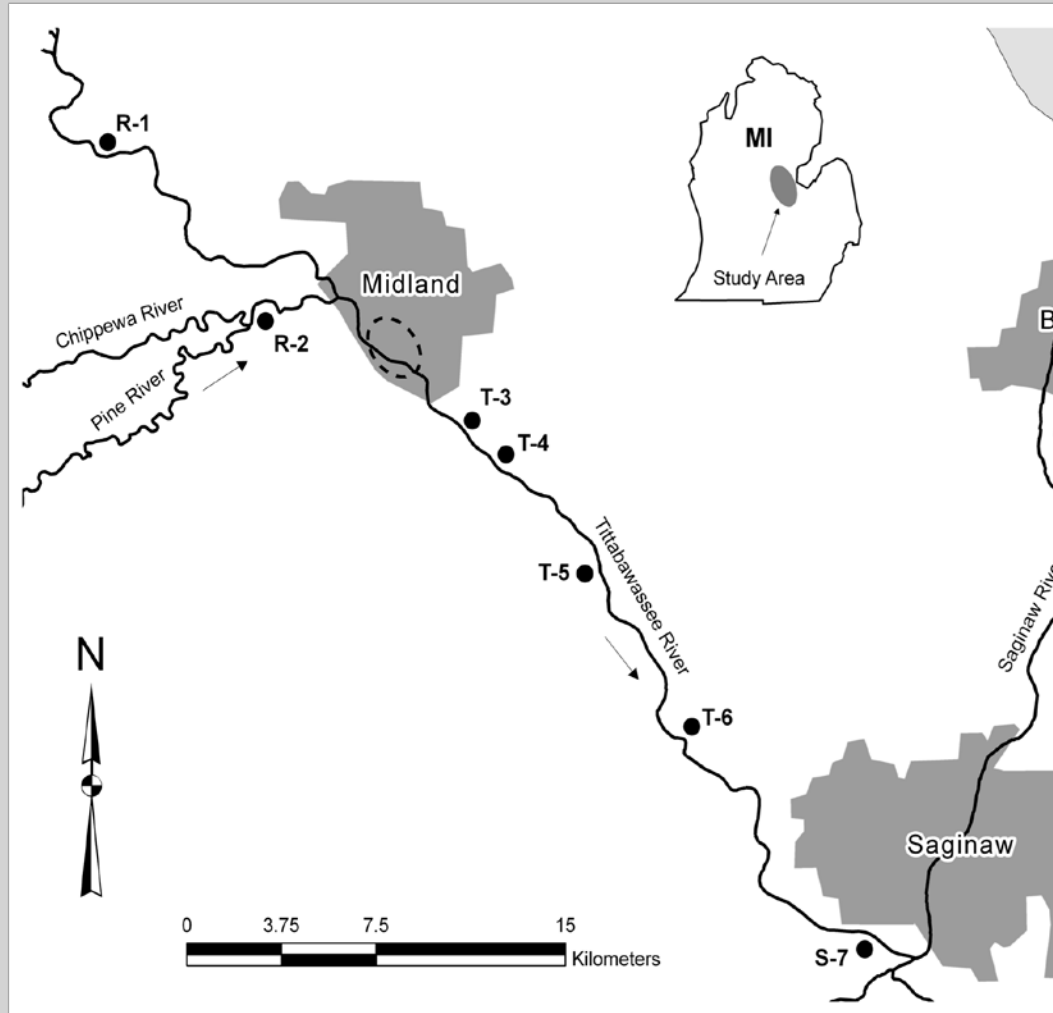
- Tittabawassee River Ecological Risk Assessment (ERA), Midland, MI, USA
 - Multiple line of evidence approach
 - Michigan State University's Aquatic Toxicology Laboratory
- Receptor species include: great horned owl (*Bubo virginianus*), great blue heron (*Ardea herodias*), kingfisher (*Ceryle alcyon*), tree swallow (*Tachycineta bicolor*), Eastern bluebird (*Sialis sialis*), house wren (*Troglodytes aedon*), American robin (*Turdus migratorias*) and American mink (*Mustela vison*)

Mammalian Exposure



- The greatest dioxin-like exposure expressed as mammalian TEQs is from PeCDF and TCDF
 - Mink - mean liver sum TEQs = 400 ± 74 ng/kg ww
 - $TEQ_{PeCDF} = 56\%$ Total
 - $TEQ_{TCDF} = 7\%$ 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

Tittabawassee River – Site map



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



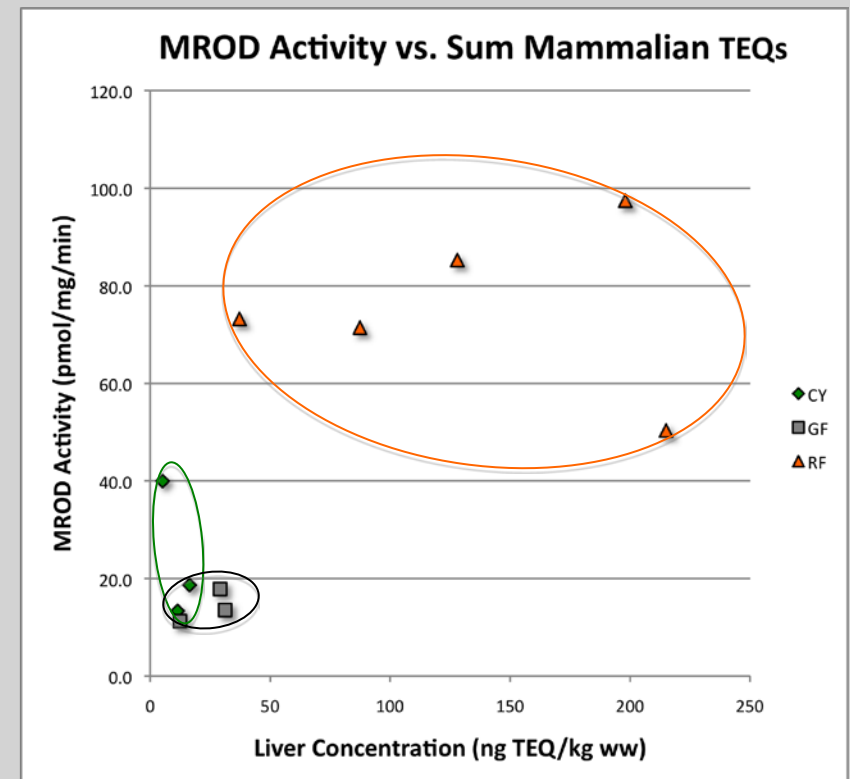
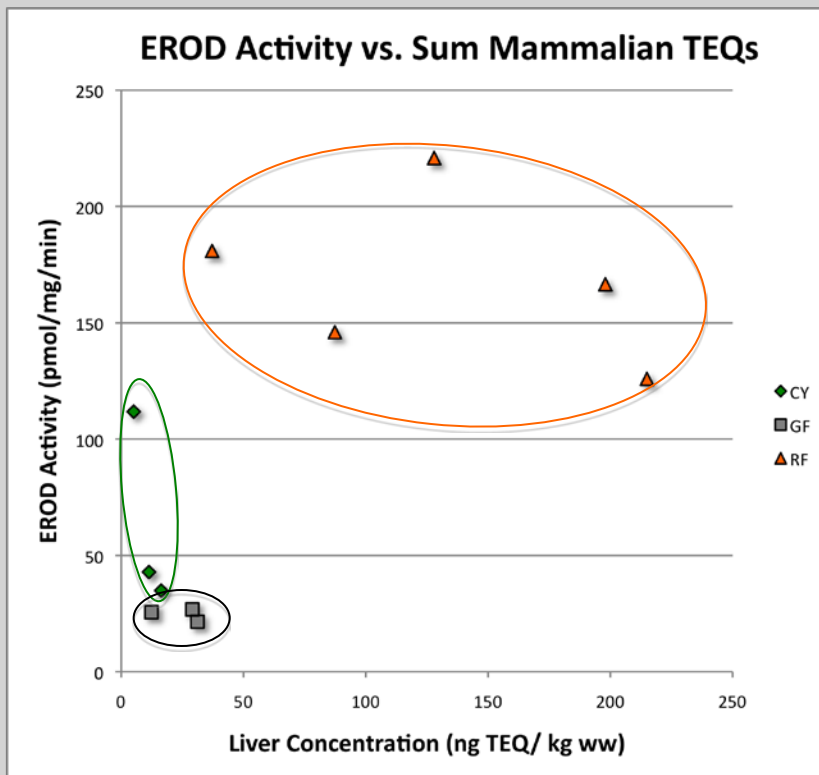
- Board certified pathologist examined lungs, heart, liver, lymph nodes, kidneys, bladder and spleen
- P450 activity and protein concentration were measured using a Fluoroscan Ascent microplate fluorometer
- Liver tissue concentrations determined by EPA extraction method 8290 and High Resolution Mass 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

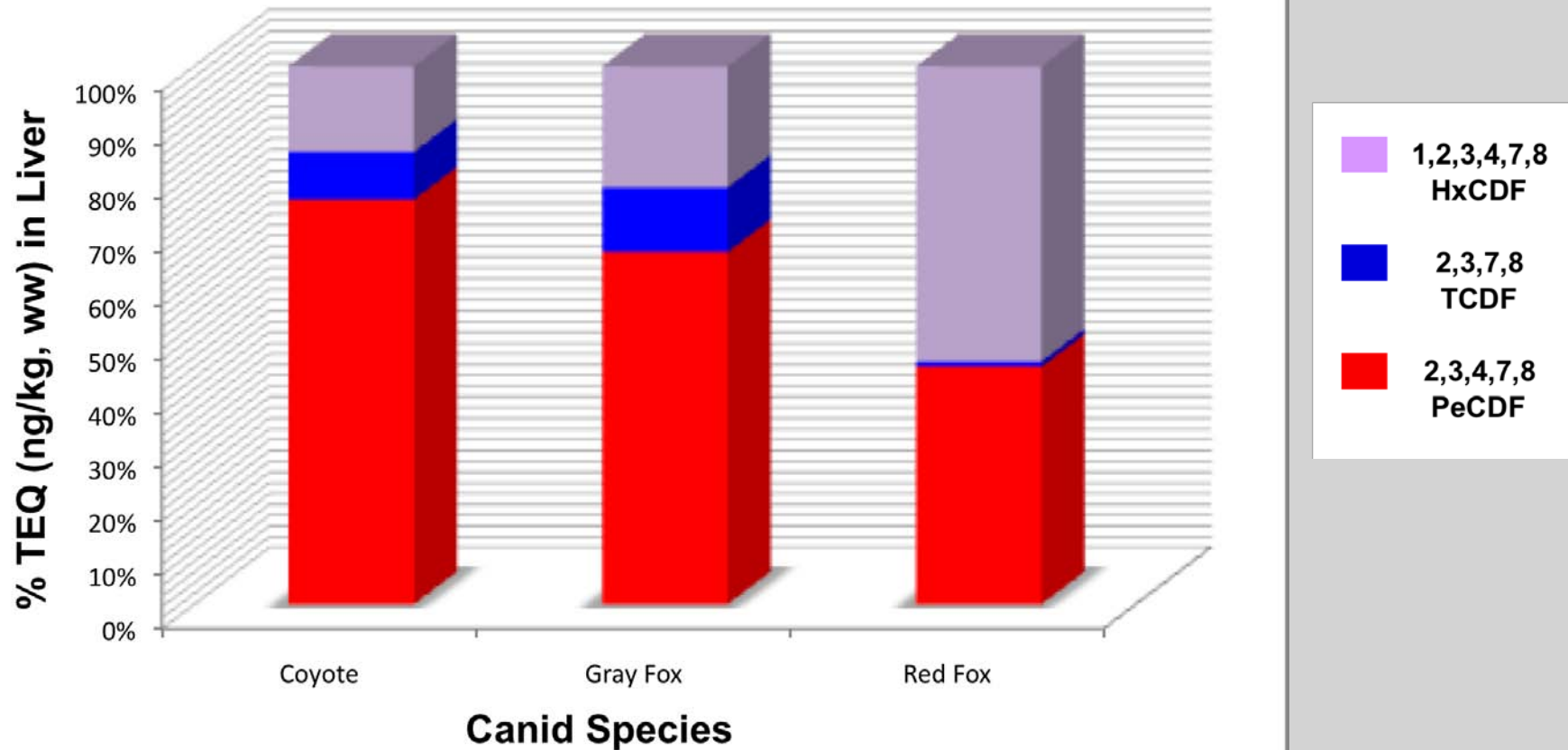
Hepatic TEQs and Enzyme Activity

- Enzyme Activity and Canid Hepatic TEQs



Results – % PCDF TEQs in Liver

PCDFs in Tittabawassee River floodplain Canids



Dietary Percentages (literature-based)



	Coyote	Red Fox
Deer	40	None
Small mammal	20	24
Shrew ¹	2	1.5
Rabbit	20	40
Bird	15	14
Invertebrate	2	4
Plant	1	7
Soil ²	2.8	2.8

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

²soil 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



	Coyote	Red Fox
Dietary concentration ¹ (ng TEQ/kg ww food)	28 (87)	27 (149)
Food ingestion rate ² (kg/kg bw/d)	0.023	0.10
Average daily dose ³ (ng TEQ/kg bw/d)	0.83 (2.6)	3.6 (17)

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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