



Toxicity of Weathered Petroleum Hydrocarbons Fraction 3 in Field Subarctic Soils to Soil Invertebrates

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I want to acknowledge

**I work, live, and play on the traditional
gathering place, travelling route and
home for many Indigenous Peoples.**

Progressive
Aboriginal
RELATIONS

COMMITTED

Canadian Council for
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Project Background



- Taiga Plains ecozone of Northwest Territories
- Bioremediation of petroleum hydrocarbon (PHC) contaminated soils using on-site biocells
 - ~5 years treatment
- PHC F2 degradation met guidelines

PHC Fraction 2 (F2) = >C10 to C16

PHC Fraction 3 (F3) = >C16 to C34



Project Challenges

- Rate of PHC F3 degradation declines with time
 - Still exceeding generic Canadian Council of Ministers of the Environment (CCME) guidelines
- Treatability range 2500 to 5000 mg/kg PHC F3
 - Industrial Guidelines
 - 2500/1700 mg/kg (coarse/fine)
 - Residential/Parkland Guidelines
 - 1300/300 mg/kg (coarse/fine)
- Limited by ecological (eco) soil contact exposure pathway
- Generic guidelines concerns
 - Agronomic species
 - Minimal weathering





Project Objective

- Developing site-specific remediation objectives for PHC F3 for the eco-contact pathway that are protective of native plants and invertebrates found at the Site.

Project History – Soil Fauna and Rooting Depth Field Survey



**Majority of
invertebrate
population <20 cm**

**Rooting Depth
is <1 m**



Project History – Plant Toxicity – Remtech 2020



**No plant toxicity
observed to several site-
specific species at PHC F3
concentrations between
1600 and 5950 mg/kg**



Ecotoxicology
<https://doi.org/10.1007/s10646-022-02585-9>

RESEARCH



Assessment of the toxicity of weathered petroleum hydrocarbon impacted soils to native plants from a site in the Canadian Subarctic

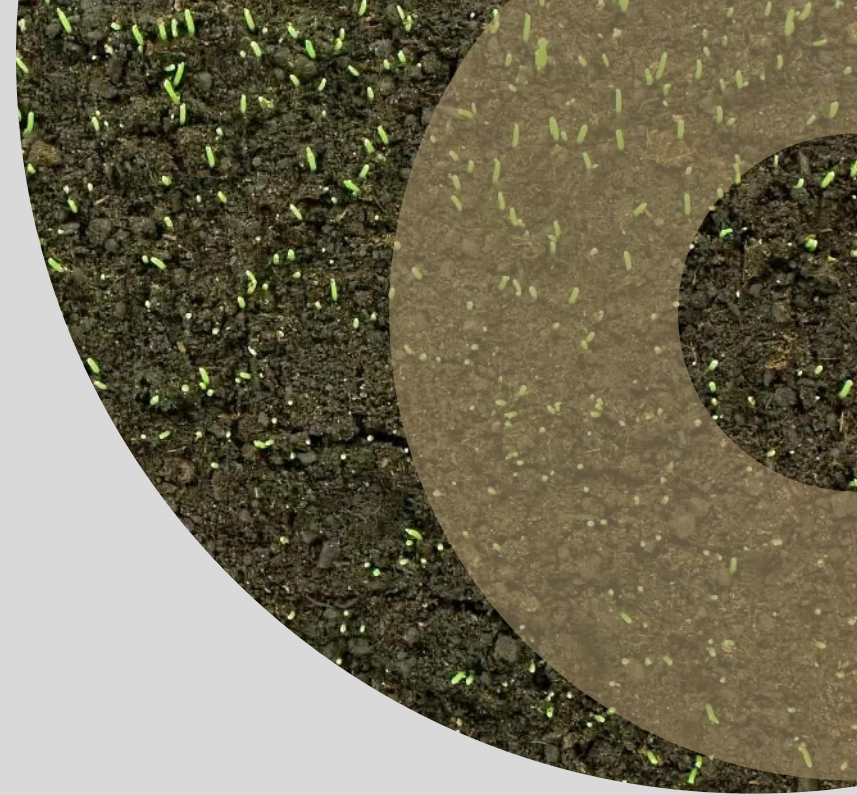
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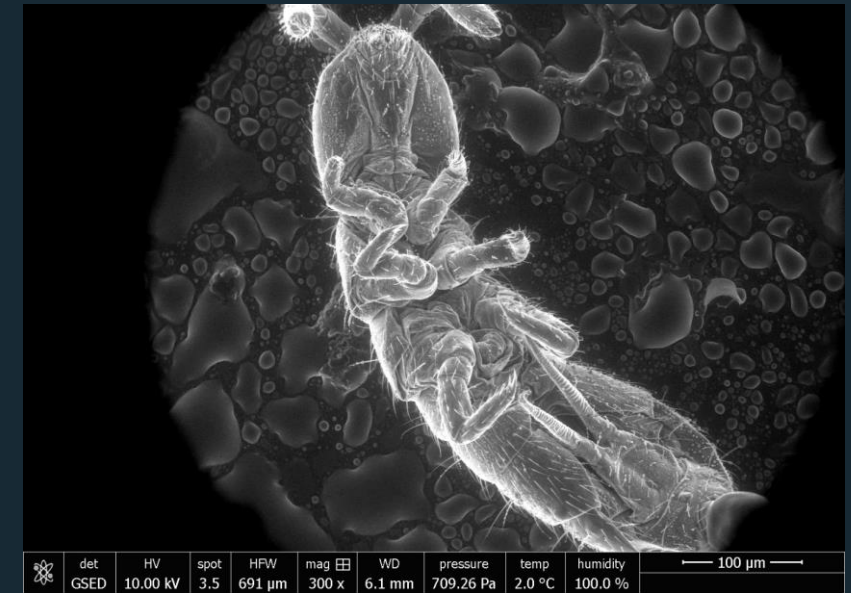
Abstract

Soil Invertebrate Toxicity to Weathered PHC F3



Soil Invertebrate Community Survey

- Identified 2 dominant groups of soil invertebrates inhabiting the site
 - Acari - Class containing ticks and mites (71 – 93%)
 - Entognatha (specifically Collembola) - Order containing springtails (5 – 25%)
- Other less dominant groups include (2 – 4%):
 - beetles, flies, thrips, spiders, centipedes and roundworms



Ventral view of a *Folsomia bisetosa*



Dorsal view of an Oribatid mite

Acknowledgement to taxonomist Jeff Batigeli

Soil Invertebrate Species

- 3 Collembola and 1 Oribatid mite species selected for the study
 - *Folsomia candida*
 - Model springtail species that has been well studied
 - Reproduces rapidly via parthenogenesis
 - *Proisotoma minuta*
 - Another commonly studied springtail species
 - Widely distributed in Canada and other parts of the world
 - *Folsomia bisetosa*
 - Springtail species endemic to the subarctic ecosystem
 - Not well studied
 - *Oppia nitens*
 - Most abundant arthropod across cold ecosystems in Canada
 - Opportunistic feeders that occupy several trophic levels

Folsomia sp.



P. minuta individual



Acknowledgement to Juliska Princz, ECCC

Test Soils

- Toxicity tests were conducted with 3 pairs of background (BG) and biotreated contaminated soils
 - Paired based on texture and proximity to each other
- MEBG- BAT3
- BIBG - BIT5
- BIBG - 018X

Top of Riffle Sampler



Side of Riffle Sampler



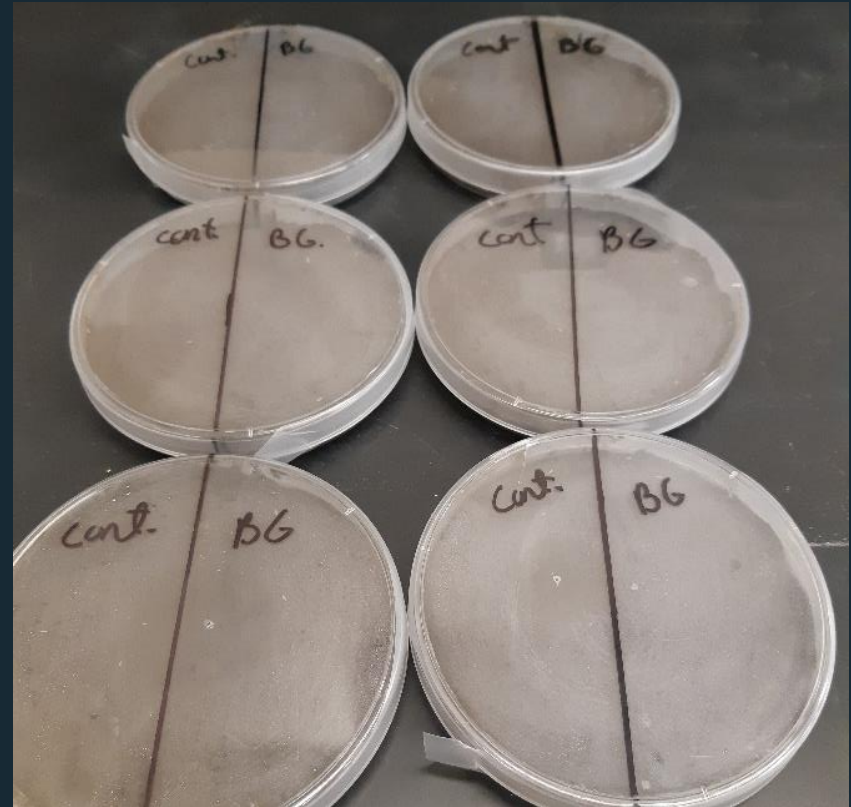
Summary of site soils

Treatment Group	Soil ID	Grain Type	PHC F2	PHC F3	PHC F4	TPH	CCME PHC F3 Guidelines (2008)
Background	MEBG	Fine	<10	86	60	149	1300
	BIBG	Fine	17	33	<10	56	1300
Contaminated	BIT5	Fine	33	5880	3570	9470	1300
	BAT3	Fine	45	4940	2560	7550	1300
	018X	Coarse	11	1140	1070	2220	300

Test Types

Avoidance test

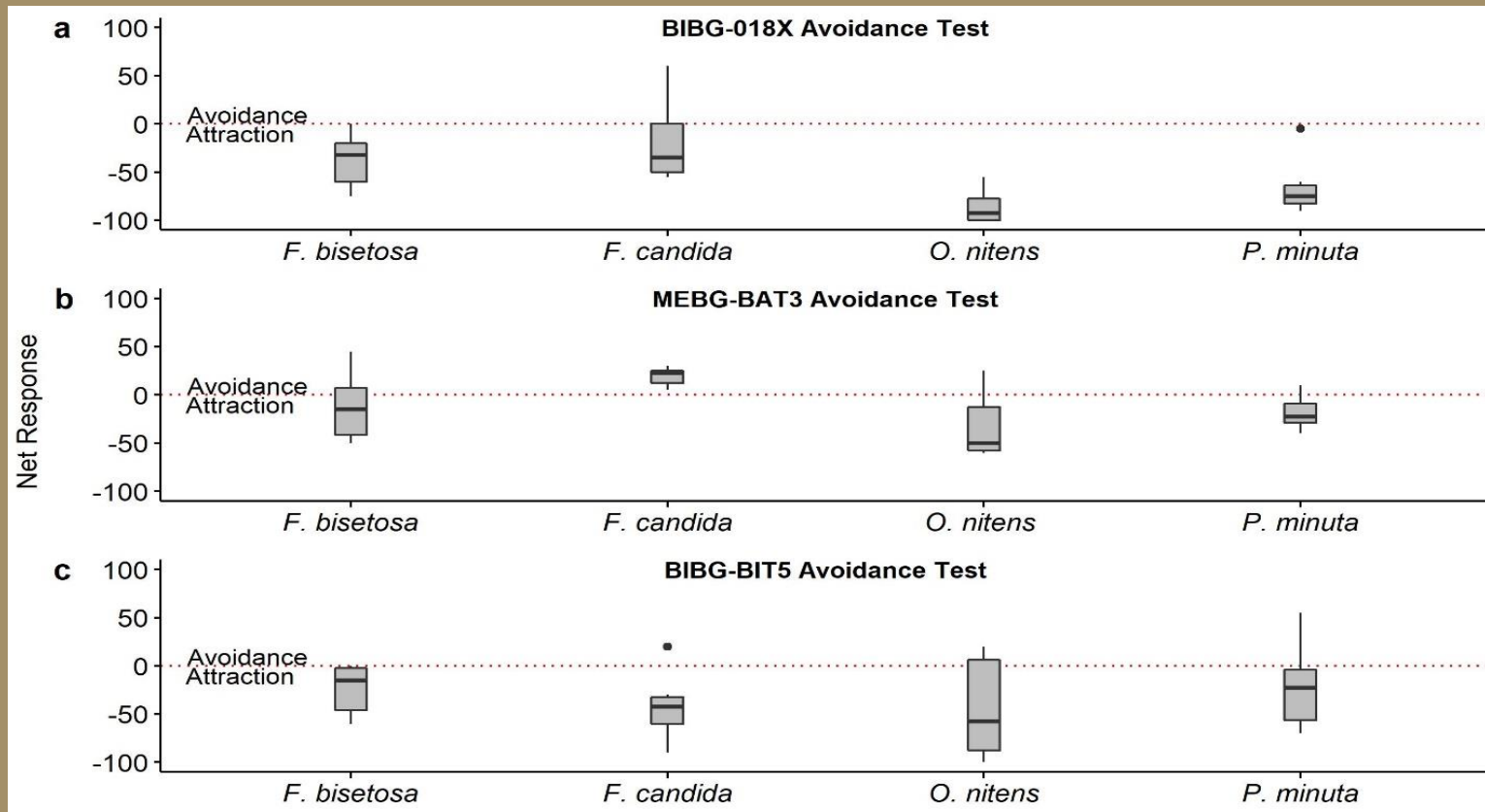
- Behavioural toxicity
- Habitat preference
- Performed with all 4 soil invertebrates in replicates of 6
- 20 individual organisms



Treatment Group	Soil ID	PHC F3	CCME F3 Guidelines (2008)
Background	MEBG	86	1300
	BIBG	33	1300
Contaminated	BIT5	5880	1300
	BAT3	4940	1300
	018X	1140	300

Avoidance Results

- 48 hour avoidance tests
- Generally, 4 soil invertebrate species were found to be attracted to the PHC-contaminated soils



Test Types

Mortality and reproduction

- Single-concentration test
- 21 or 28-day exposure test at $20 \pm 2^{\circ}\text{C}$ (16h light: 8h dark)
- Conducting using *P. minuta* and *F. candida* in replicates of 6
- 10 age-synchronized juveniles

Treatment Group	Soil ID	PHC F3	CCME F3 Guidelines (2008)
Background	MEBG	86	1300
	BIBG	33	1300
Contaminated	BIT5	5880	1300
	BAT3	4940	1300
	018X	1140	300

Mortality Results

- Single-concentration tests
- No significant adverse effect on survival of *F. candida* or *P. minuta*

Species	Grain Type	Soil ID	Adult Survival
<i>F. candida</i>	Fine	MEBG	8.8
		BAT3	8.7
		BIBG	8.8
		BIT5	9.2
	Coarse	BIBG	9.4
		018X	8.8
<i>P. minuta</i>	Fine	MEBG	9.3
		BAT3	9.2
		BIBG	9
		BIT5	8.2
	Coarse	BIBG	8.5
		018X	8.5

ATC60 Growth Chamber



Reproduction Results

- Single-concentration tests
- No significant adverse effect on reproduction of *F. candida* or *P. minuta*

Species	Grain Type	Soil ID	Number of Progeny
<i>F. candida</i>	Fine	MEBG	203
		BAT3	205
		BIBG	194
		BIT5	194
	Coarse	BIBG	145
		018X	158
<i>P. minuta</i>	Fine	MEBG	127
		BAT3	189
		BIBG	138
		BIT5	115
	Coarse	BIBG	181
		018X	237



Assessment of Collembola Sensitivities

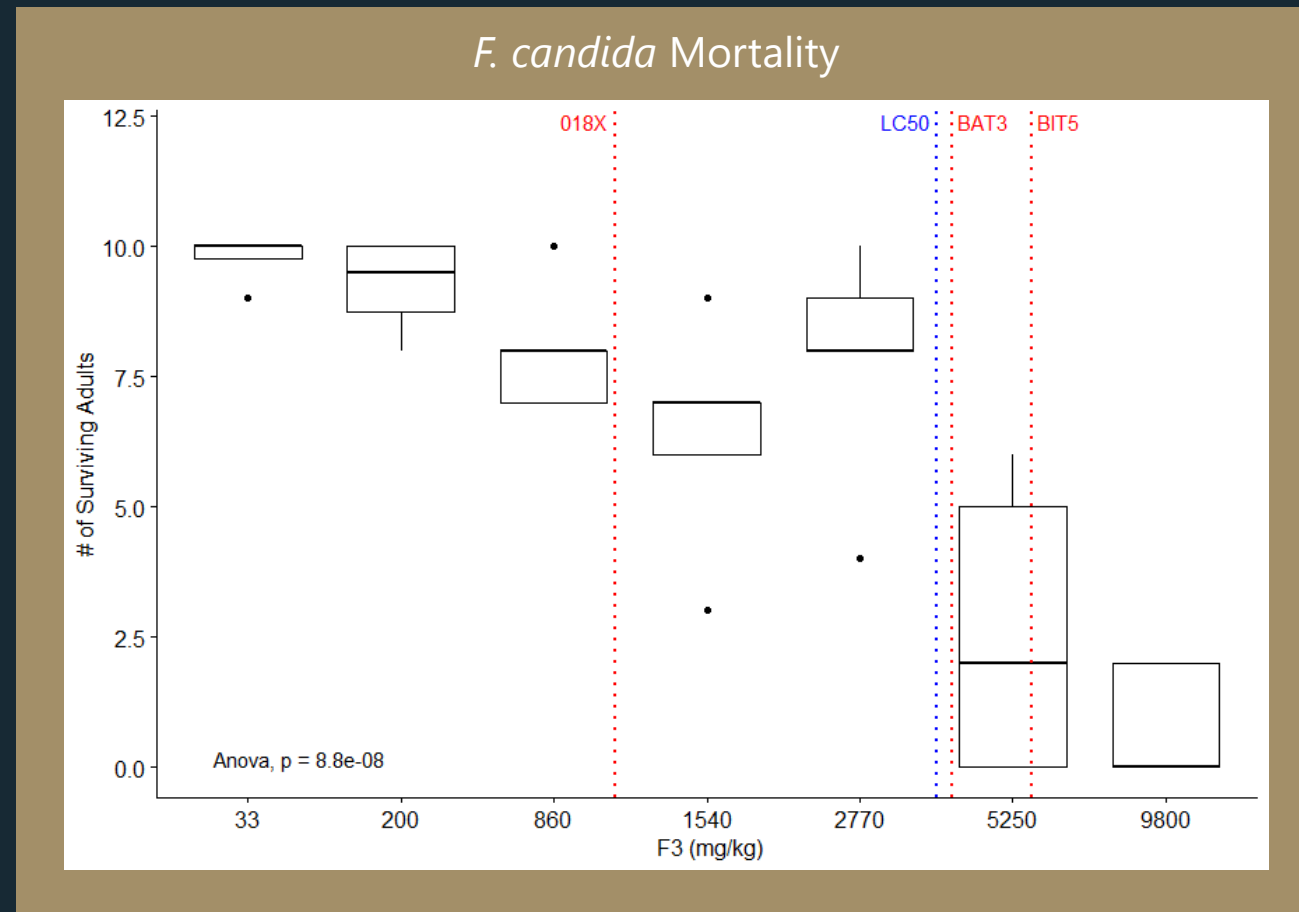
- Reference toxicant tests using boric acid were performed to:
 - i) verify sensitivity of the colonies used
 - ii) assess precision and accuracy of the single-concentration tests

Species	Boric Acid Concentration (mg/kg)	Mortality (%)	LC50 (mg F3/kg)	Previously calculated LC50 (mg/kg)
<i>P. minuta</i>	0	0.0	801 ± 130	507 to 774 (EC 2014)
	250	7.0		
	500	7.3		
	1000	63		
	15000	90		
	20000	100		
<i>F. candida</i>	0	6	1280 ± 82	800 to 1483 (EC 2014)
	250	3		
	500	10		
	1000	30		
	15000	70		
	20000	100		

Toxicity of Fresh PHC-contaminated Soil

Performed toxicity tests using *F. candida* exposed to soils freshly spiked with crude oil

- LC50 = 4017 mg /kg PHC F3 (mortality)

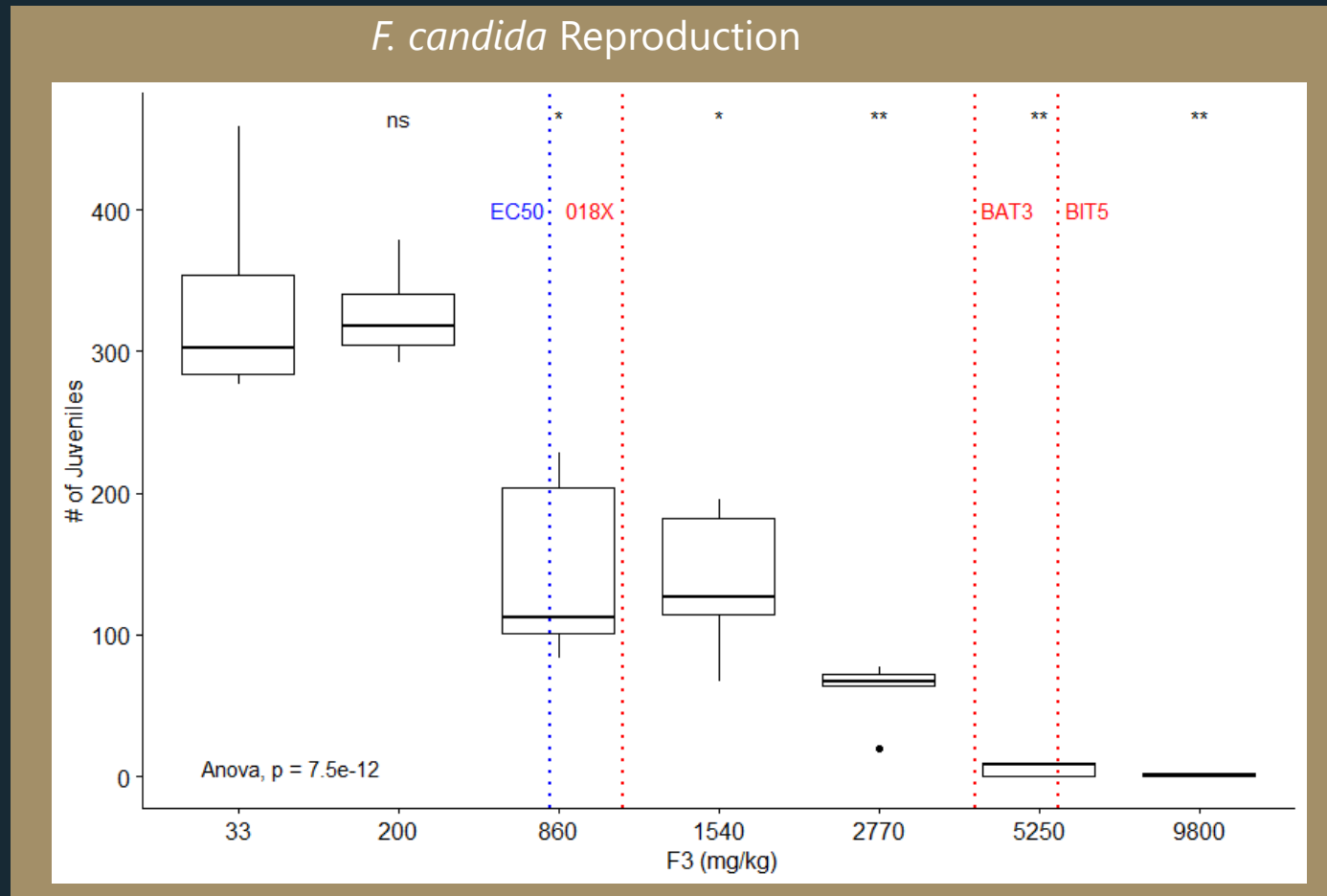


- Red lines = weathered PHC impacted soils, no observed effects concentrations
- Blue line = LC50 for fresh/unweathered PHC impacts in soils

Toxicity of Fresh PHC-contaminated Soil

Performed toxicity tests using *F. candida* exposed to soils freshly spiked with crude oil

- EC50 = 840 mg/kg PHC F3 (reproduction)



- Red lines = weathered PHC impacted soils, no observed effects concentrations
- Blue line = LC50 for fresh/unweathered PHC impacts in soils

Significance

- Current concentrations of weathered PHC F3 impacts in soil at the Site are having no significant adverse effects on native plant and soil invertebrate species
 - Likely due to 2 factors:
 - i) Loss and/or conversion of lower molecular weight compounds with higher toxic potential
 - ii) Reduced bioavailability of residual PHCs due to adsorption to soil particles





Future Work

- Link transcriptome of *F. candida* to effects observed at the individual level
- Link soil invertebrate toxicity to bioavailability of freshly spiked soils using solid phase microextraction (SPME)



Summary and Next Steps

- **Current generic guidelines overly conservative for weathered contamination present at the Site**
 - Risk-based approaches
- Site-specific PHC F3 guideline
- Publish findings and share results with scientific community

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Taxonomist Jeff Battigelli (NAIT)

Thank you!

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