Mycoremediation of Naphthenic Acid Fraction Compounds Sourced from Oil Sands Process Affected Water

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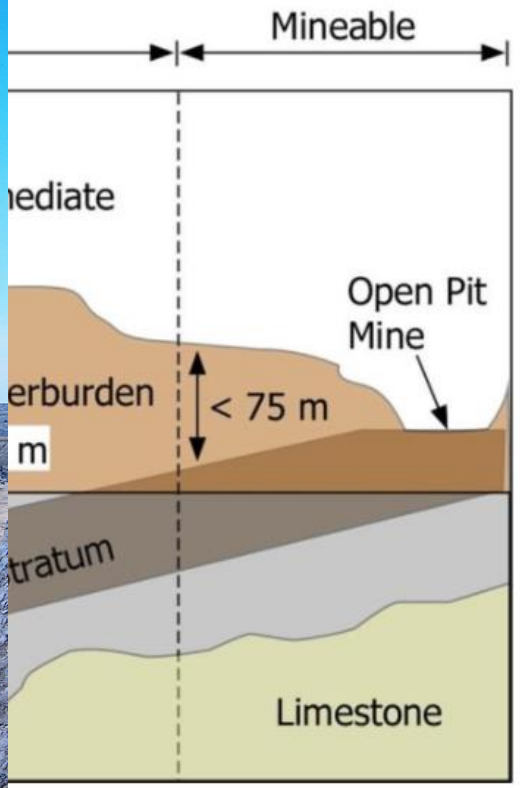
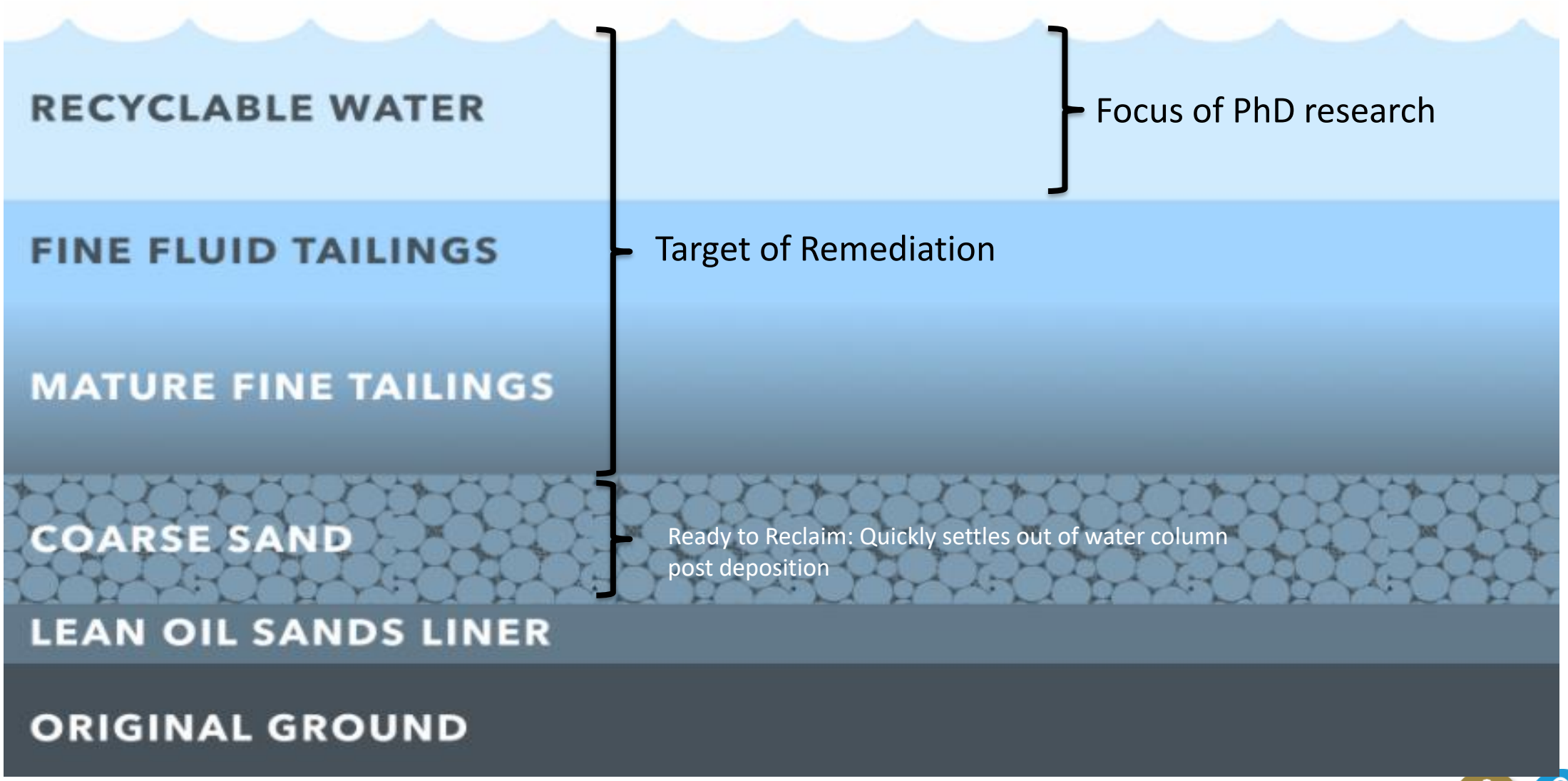
Oil Sands Background



https://awrl.ca/overview/

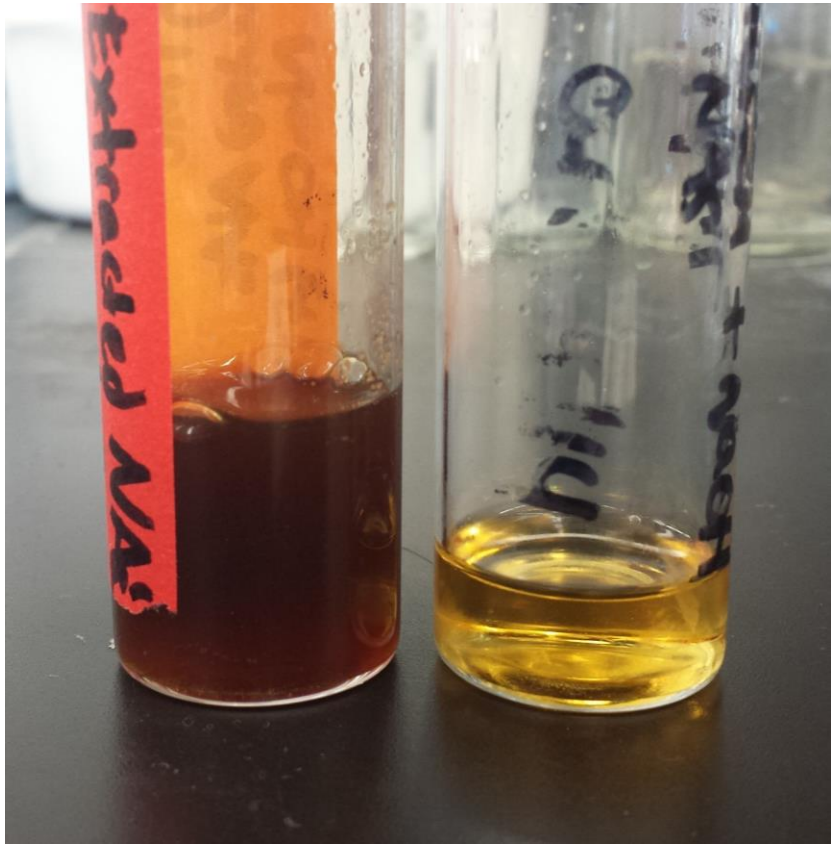
Oil Sands Tailings Ponds



https://www.oilsandsmagazine.com/technical/mining/tailings-ponds

Oil Sands Process Affected Water

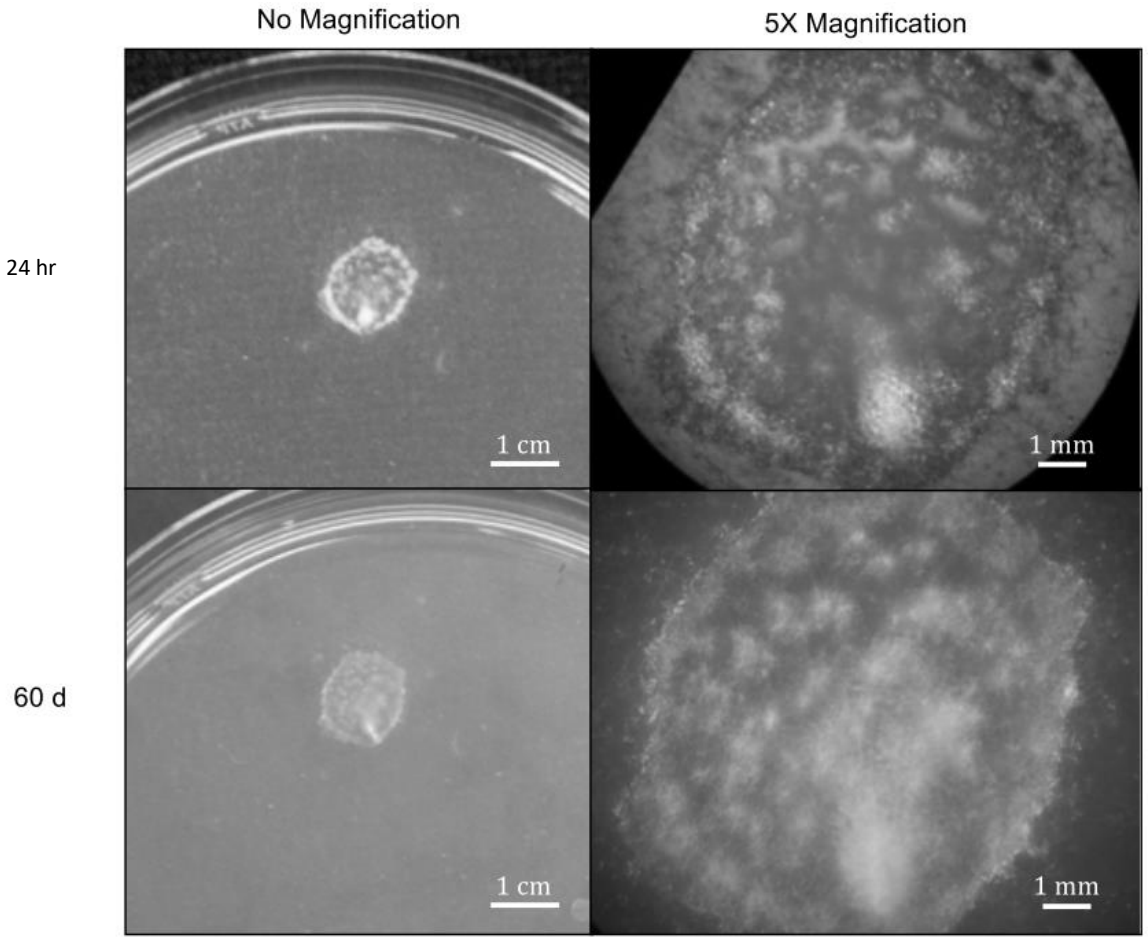
- Classical definition of NAs general structural formula of $C_nH_{2n+z}O_2$
- Currently termed naphthenic acid fraction compounds (NAFCs)
- Concentration in OSPW varies (<1 mg/L to 120 mg/L)
- Main contributor to toxicity
- O_2^- group and larger more complex compounds with ≥17 carbons attributed to toxicity



OSPW Sourced NAFCs

Dzidic, I., Somerville, A.C., Raia, J.C., Hart, H.V., 1988. Determination of Naphthenic Acids in California Crudes and Refinery Wastewaters by Fluoride-Ion Chemical Ionization Mass-Spectrometry. Analytical Chemistry 60, 1318-1323.
Hughes, S.A., Mahaffey, A., Shore, B., Baker, J., Kilgour, B., Brown, C., Peru, K.M., Headley, J.V., Bailey, H.C., 2017. Using ultrahigh-resolution mass spectrometry and toxicity identification techniques to characterize the toxicity of oil sands process-affected water: The case for classical naphthenic acids. Environmental Toxicology and Chemistry 36, 3148-3157.

Trichoderma harzianum



- pH growth range 2-9
- Inhibitory salinity ≥60g/L
- Inhibitory NA concentration 2400mg/L

Isolate grows on a single drop of pure NAFCs

Miles, S.M., Hofstetter, S., Edwards, T., Dlusskaya, E., Cologgi, D.L., Gänzle, M., Ulrich, A.C., 2019. Tolerance and cytotoxicity of naphthenic acids on microorganisms isolated from oil sands process-affected water. Science of The Total Environment 695, 133749.

Project Objective. NAFC/NA degradation characterization

Research Questions:

- Can an OSPW sourced fungus *Trichoderma Harzianum* degrade NAFCs and decrease toxicity?
- Can an OSPW sourced fungus *Trichoderma Harzianum* degrade model NA compounds with varying structures?

T. harzianum Degradation Study

Merichem Microcosm experiment

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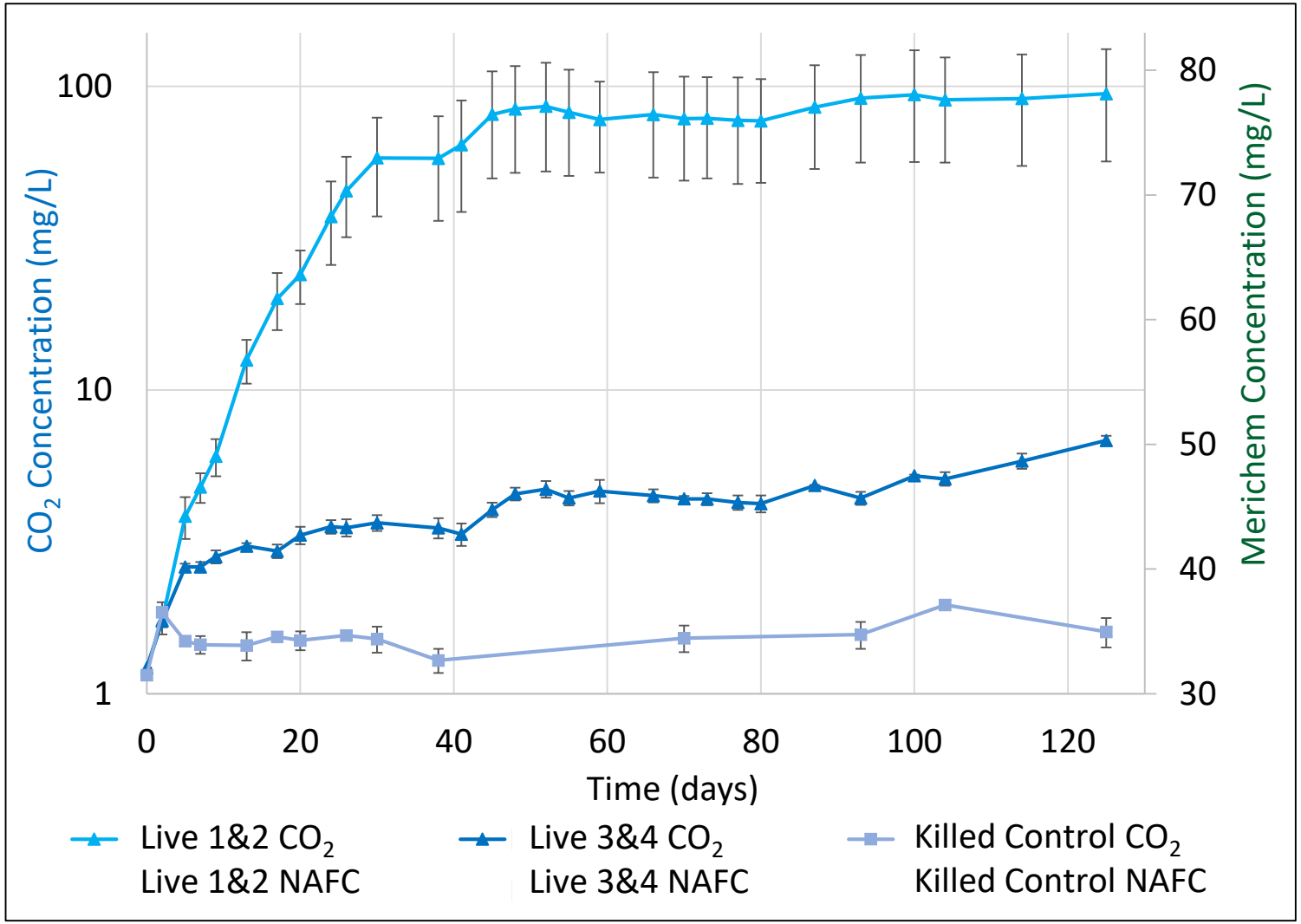
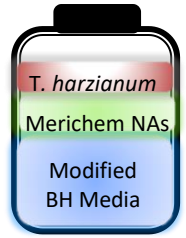


Live *T. harzianum* treatment



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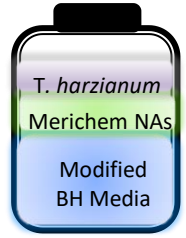

T. harzianum Degradation Study

Merichem Microcosm experiment

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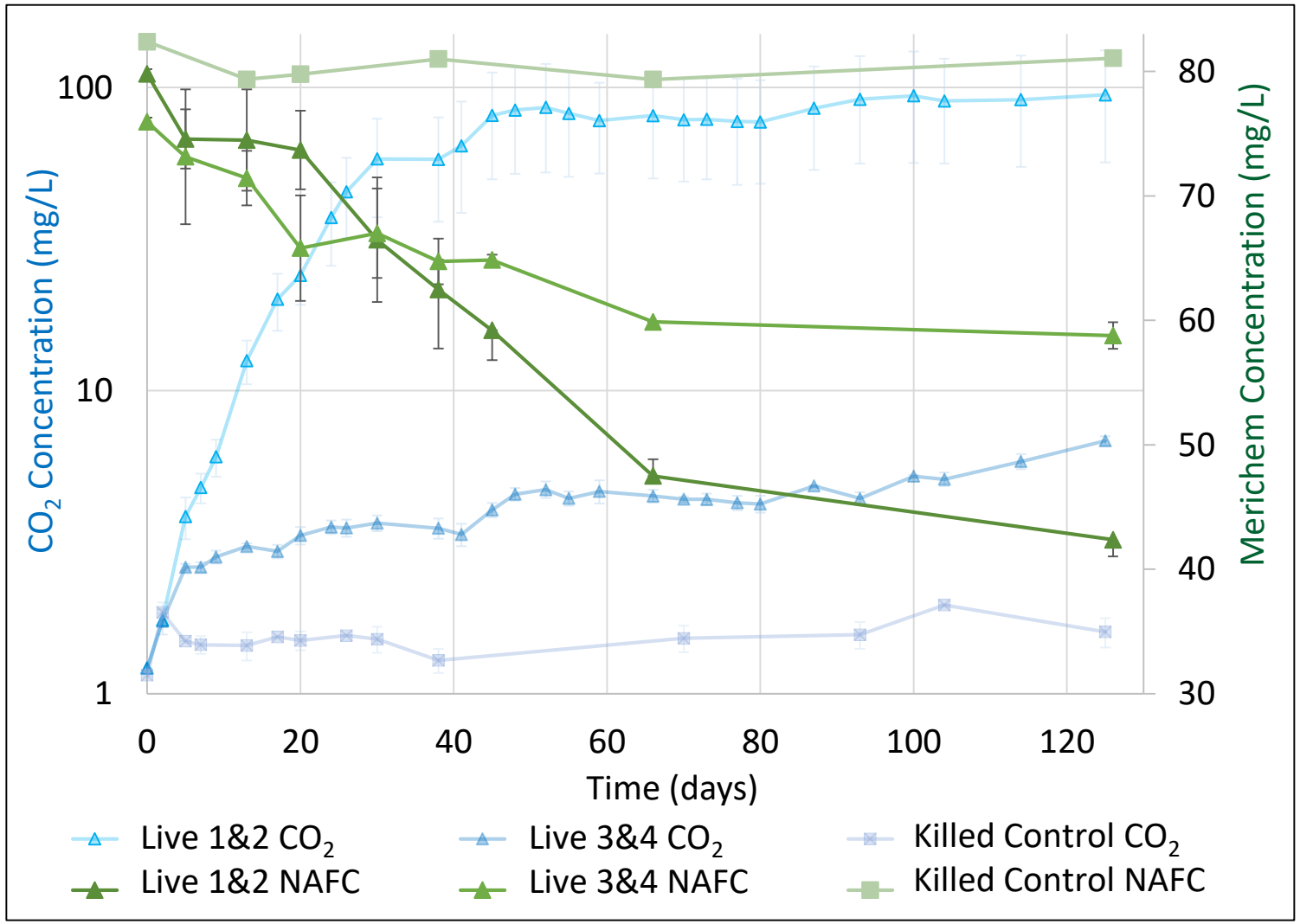

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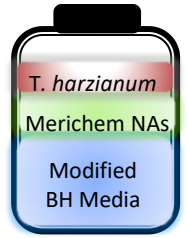

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T. harzianum Degradation Study

Merichem Microcosm experiment

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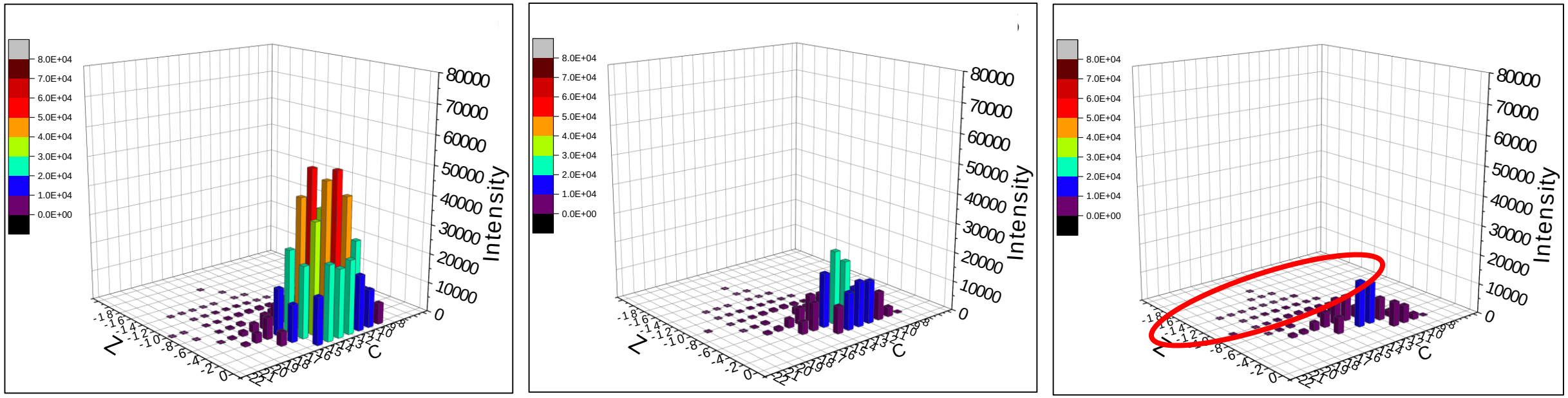
Day 0 → **Day 105** → **Day 183**



Total O_2^- decreased by 88% in 183 days; however, 80% of removal was completed before 105 days

Decrease by day 105	Decrease by day 183
Z= 0 100%	Z= 0 100%
Z= -2 68%	Z= -2 83%
Z= -4 51%	Z= -4 67%

Over 183 days decrease of 77% compounds containing ≤16 carbons;
and 85% reduction in compounds containing ≥17 or more carbons

T. harzianum Degradation Study

OSPW Sourced NAFCs Microcosm experiment

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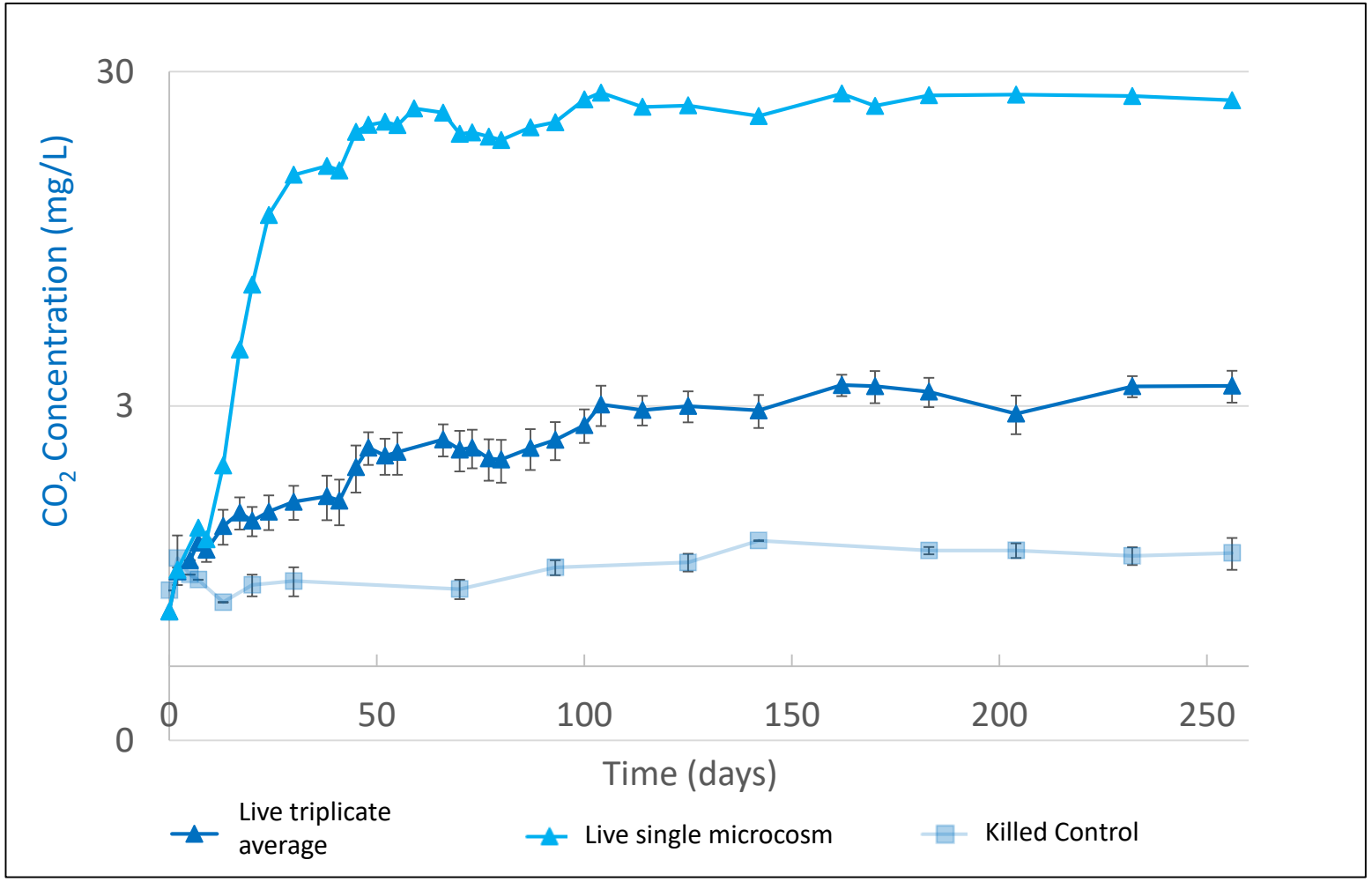
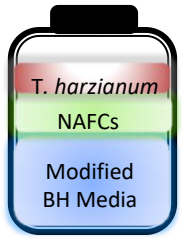


Live *T. harzianum* treatment



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T. harzianum Degradation Study

OSPW Sourced NAFCs Microcosm experiment

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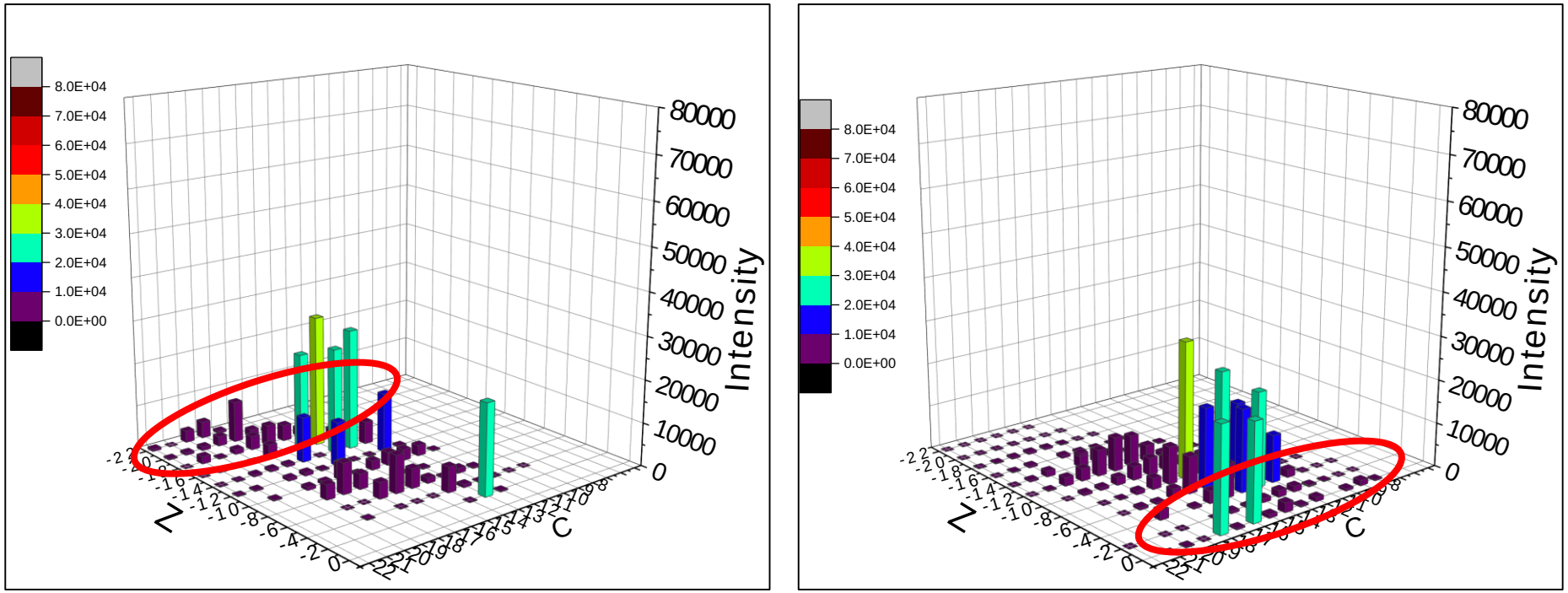
Day 0 → **Day 183**



In 183 days

- 30% increase in total intensities
- increase in Z=0 series
- 13% decrease in compounds containing ≥17 carbons

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Microtox™ toxicity bioassay

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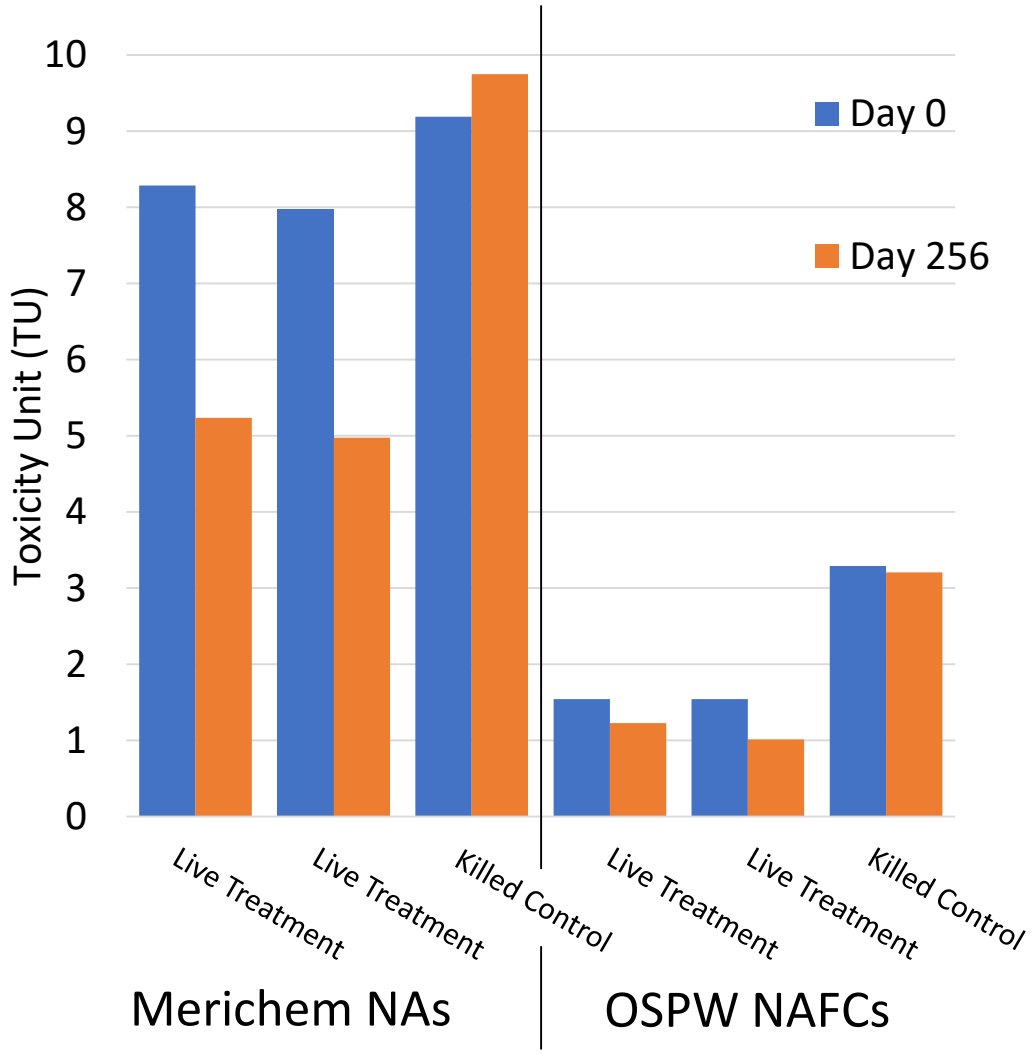

T. harzianum Degradation Study

Model Compound Selection

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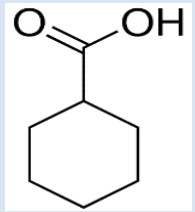
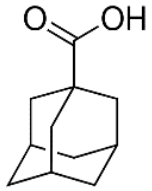
Model NAFC	Formula	Rationale	Reference
Cyclohexane Carboxylic Acid	 $C_7H_{12}O_2$	Single ring compound	(Demeter et al., 2014;Herman et al., 1993)
Adamantane-1-Carboxylic Acid	 $C_{11}H_{16}O_2$	Tricyclic compound	(Rowland et al., 2011)

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Cyclohexane Carboxylic Acid (CHCA) Microcosm experiment

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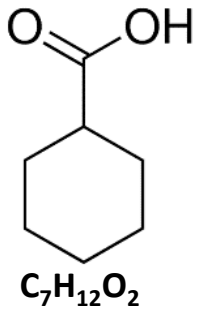
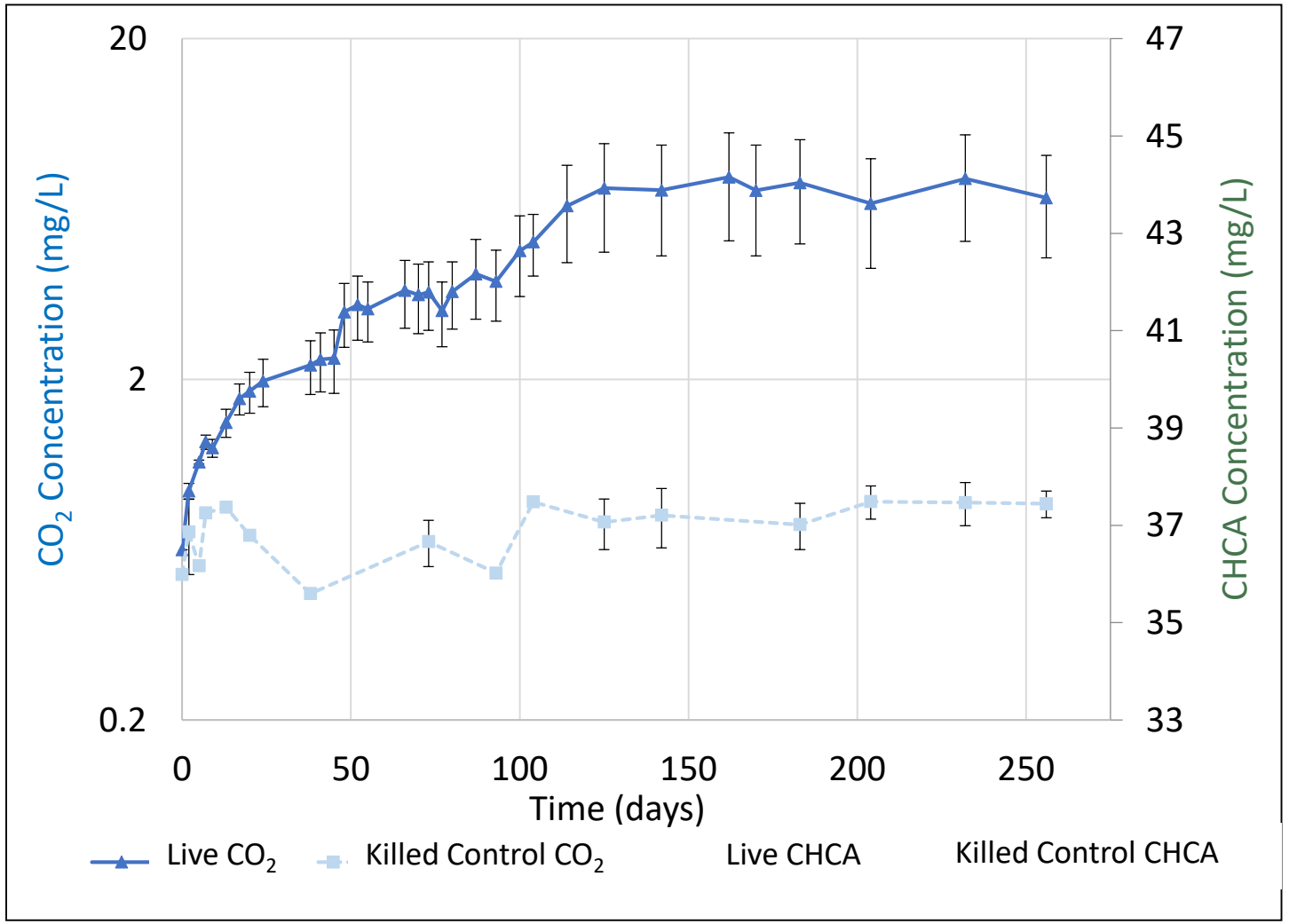
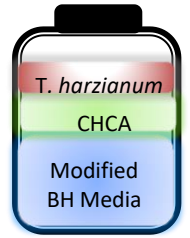


$C_7H_{12}O_2$



Live *T. harzianum* treatment



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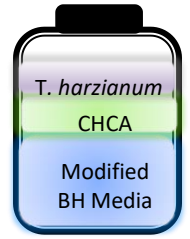

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Cyclohexane Carboxylic Acid (CHCA) Microcosm experiment

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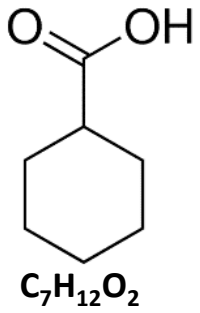

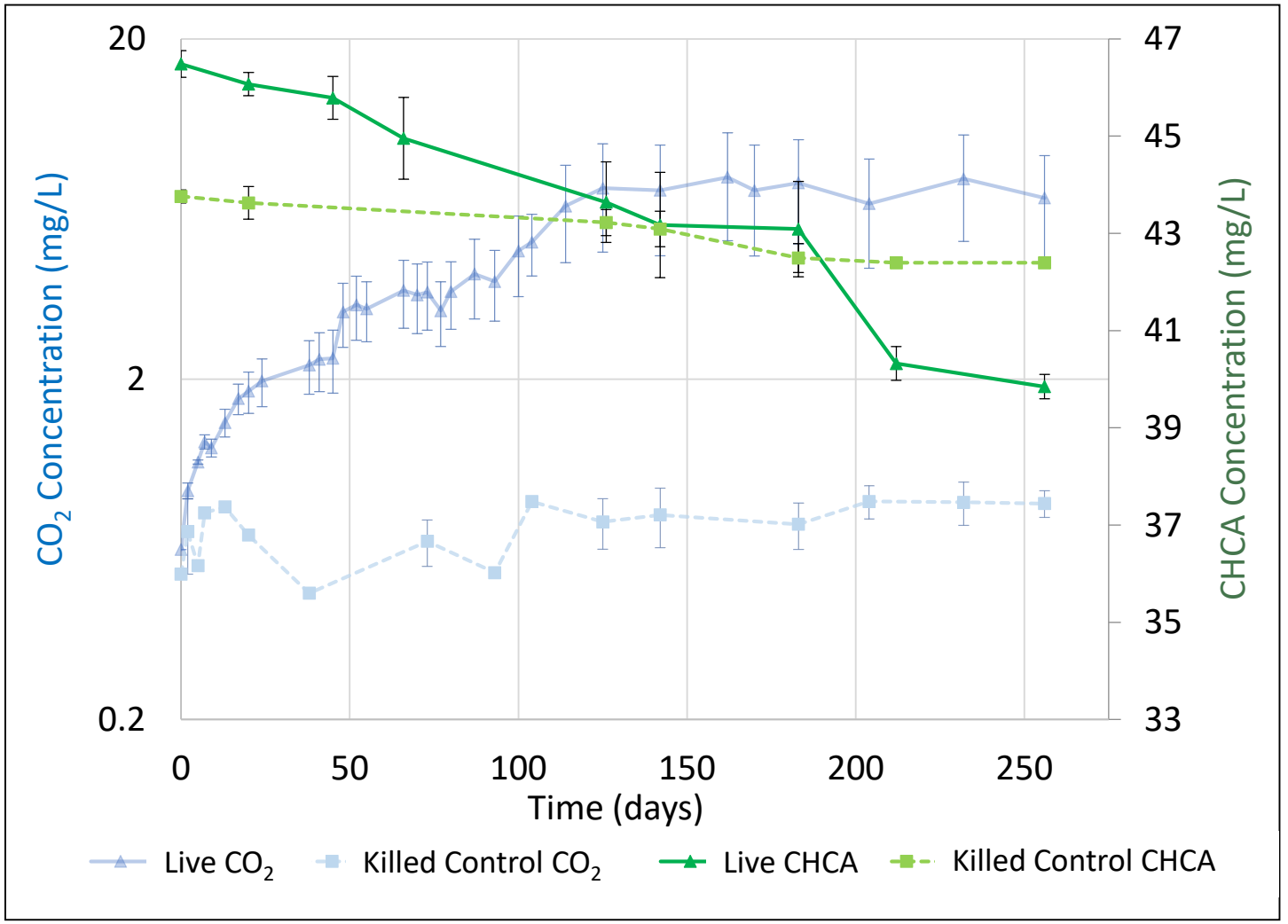

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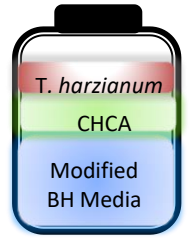

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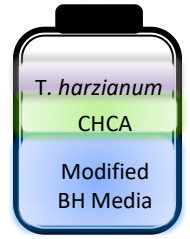

T. harzianum Degradation Study

Adamantane-1-Carboxylic Acid (ADA) Microcosm experiment

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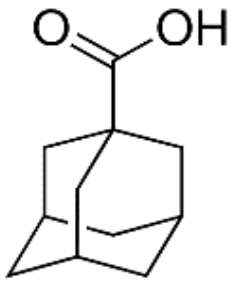
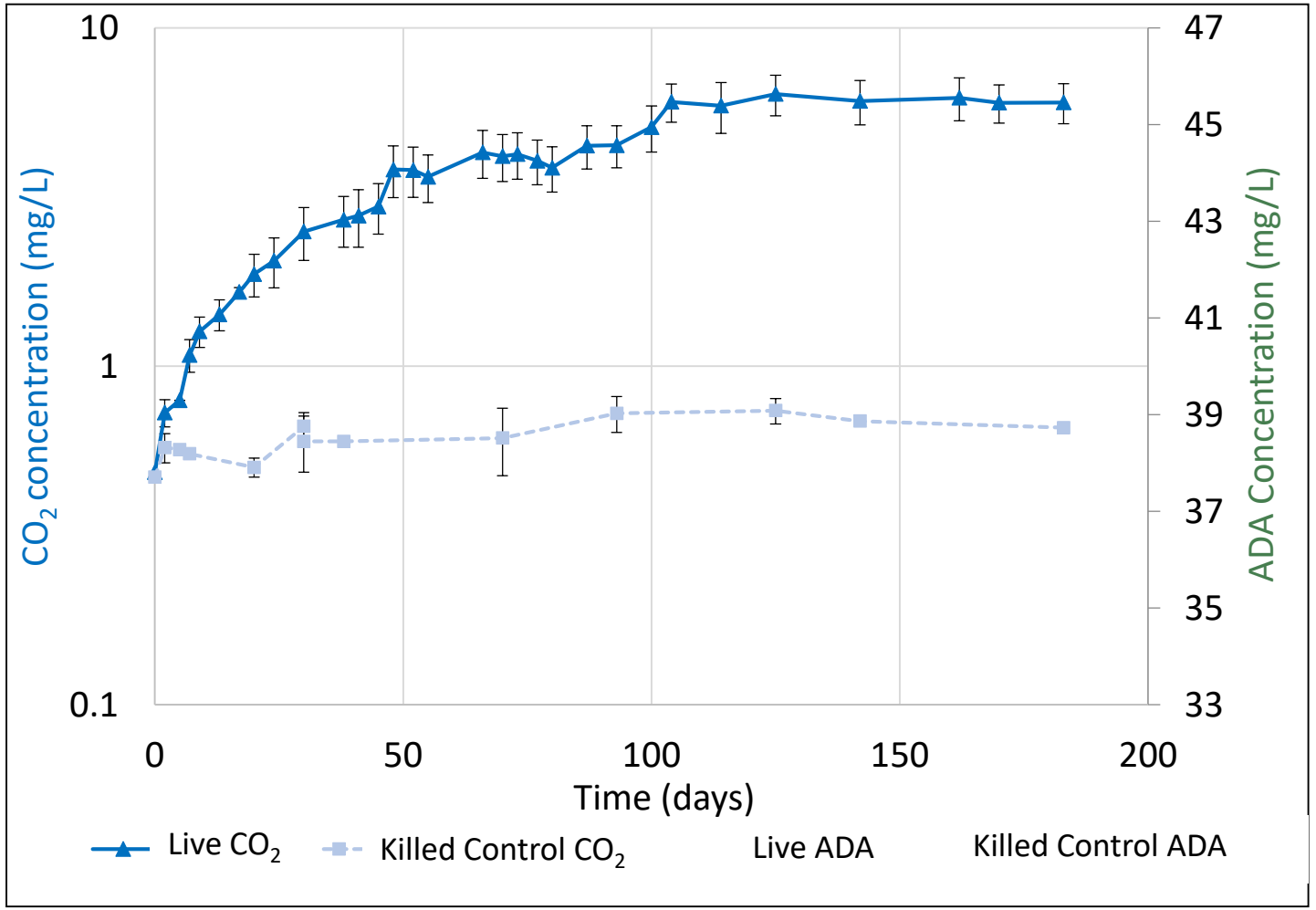


$C_{11}H_{16}O_2$



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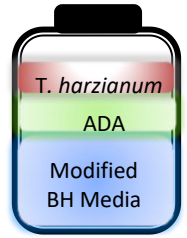

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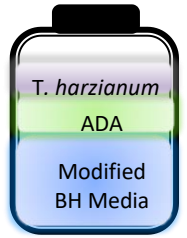

T. harzianum Degradation Study

Adamantane-1-Carboxylic Acid (ADA) Microcosm experiment

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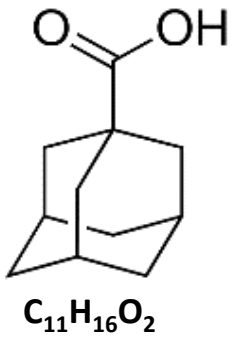
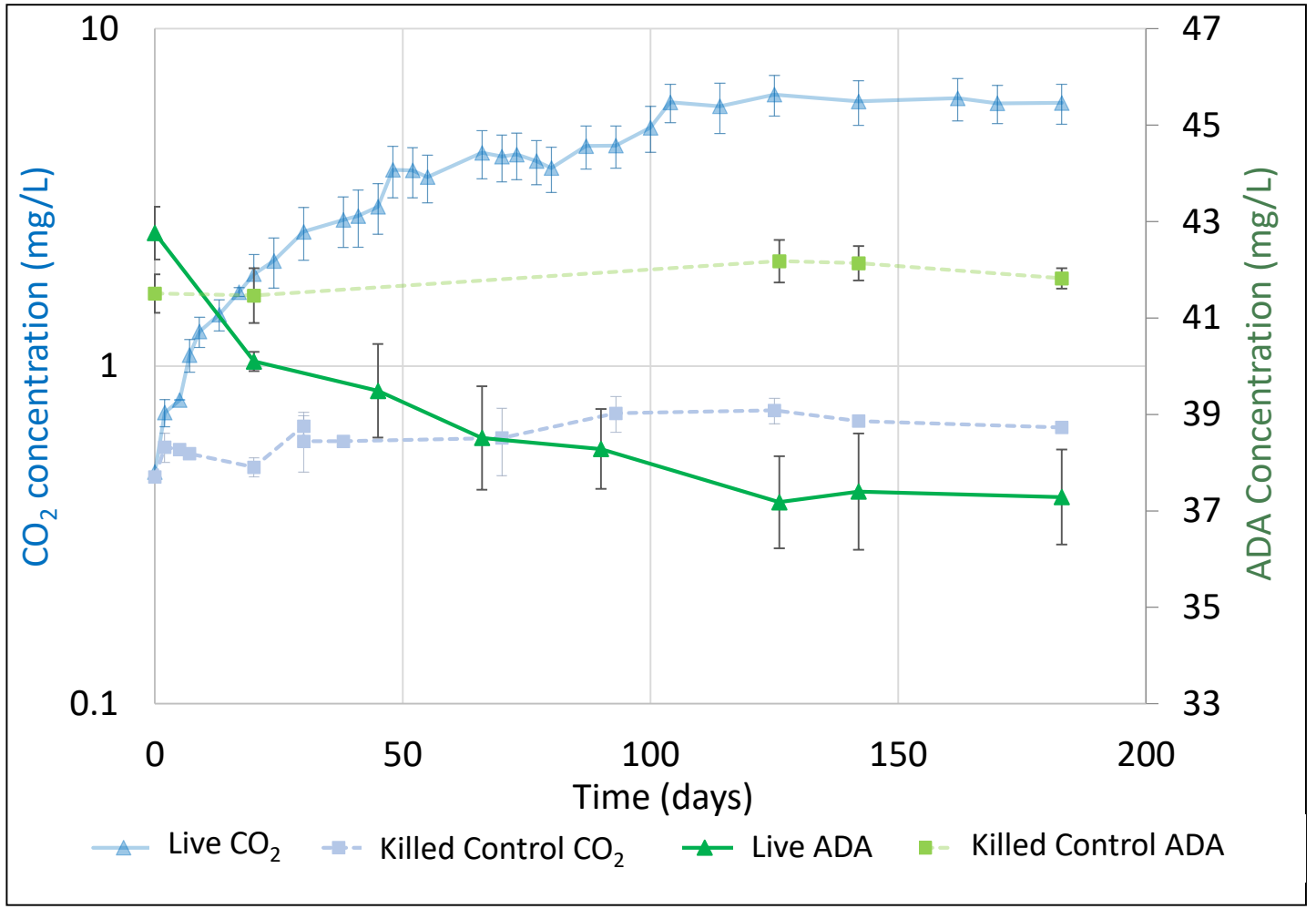


$C_{11}H_{16}O_2$



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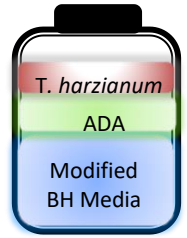

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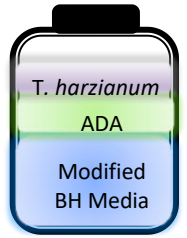

Summary

Research Questions: *Can an OSPW sourced fungus Trichoderma Harzianum degrade NAFCs and decrease toxicity?*

- 23-47% reduction in Merichem NAs in 126d 85% reduction in compounds containing ≥17 carbons
- 59% and 52% decrease in toxicity for Merichem NAs, and OSPW NAFCS respectively

Research Question: *Can an OSPW sourced fungus Trichoderma Harzianum degrade model NAFC compounds with varying structures?*

- 14% of CHCA in 256d was degraded
- 13% of ADA in 183d was degraded

Although complete degradation was not observed, any degradation is important as a fungal isolate from OSPW has never demonstrated the ability to degrade NAFCs

Acknowledgements

Special thanks to:

Dr Ania Ulrich

Ulrich Lab Group

Dr. Evelyn Asiedu

Amy-Lynne Balaberda

Demeter, M. A., J. Lemire, I. George, G. Yue, H. Ceri & R. J. Turner (2014) Harnessing oil sands microbial communities for use in ex situ naphthenic acid bioremediation. *Chemosphere,* 97**,** 78-85.

Dzidic, I., A. C. Somerville, J. C. Raia & H. V. Hart (1988) Determination of Naphthenic Acids in California Crudes and Refinery Wastewaters by Fluoride-Ion Chemical Ionization Mass-Spectrometry. *Analytical Chemistry,* 60**,** 1318-1323.

Herman, D. C., P. M. Fedorak & J. W. Costerton (1993) Biodegradation of Cycloalkane Carboxylic-Acids in Oil Sand Tailings. *Canadian Journal of Microbiology,* 39**,** 576-580.

Hughes, S. A., A. Mahaffey, B. Shore, J. Baker, B. Kilgour, C. Brown, K. M. Peru, J. V. Headley & H. C. Bailey (2017) Using ultrahigh-resolution mass spectrometry and toxicity identification techniques to characterize the toxicity of oil sands process-affected water: The case for classical naphthenic acids. *Environmental Toxicology and Chemistry,* 36**,** 3148-3157.

Liu, Q., B. Montoya & B. Martinez. 2018. Accelerated Dewatering of Mature Fine Tailings Through Microbial Induced Calcium Carbonate Precipitation In *International Oil Sands Tailings Conference*.

Liu, Q. W. & B. M. Montoya. 2020. Experimental Study of Consolidation Behavior of Mature Fine Tailings Treated with Microbial Induced Calcium Carbonate Precipitation. In *Biogeotechnics,* eds. E. Kavazanjian, J. P. Hambleton, R. Makhnenko & A. S. Budge, 196-204.

Miles, S. M., E. Asiedu, A.-l. Balaberda & A. C. Ulrich (2020) Oil sands process affected water sourced Trichoderma harzianum demonstrates capacity for mycoremediation of naphthenic acid fraction compounds. *Chemosphere,* 258**,** 127281.

Miles, S. M., S. Hofstetter, T. Edwards, E. Dlusskaya, D. L. Cologgi, M. Gänzle & A. C. Ulrich (2019) Tolerance and cytotoxicity of naphthenic acids on microorganisms isolated from oil sands process-affected water. *Science of The Total Environment,* 695**,** 133749.

Rowland, S. J., A. G. Scarlett, D. Jones, C. E. West & R. A. Frank (2011) Diamonds in the Rough: Identification of Individual Naphthenic Acids in Oil Sands Process Water. *Environmental Science & Technology,* 45**,** 3154-3159.

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