The Connecticut Agricultural Experiment Station

Hemp Phytoremediation and Degradation of PFAS: A Trial at the Former Loring Air Force Base"

Sara Nason, Chelli Stanley, Richard Silliboy, Maggie Blumenthal, Weilan Zhang, Yanna Liang, Sara Thomas, **Nubia Zuverza-Mena**, Jason White, Christy Haynes, Vasilis Vasiliou, Shan Huang, Peter Jaffe, Bryan Berger

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Durham, NC
NIH TAC

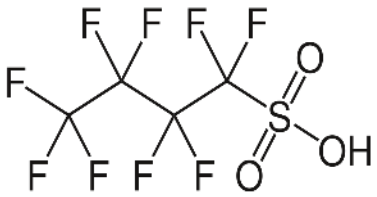
NIH Tribal Advisory Committee (TAC) Meeting

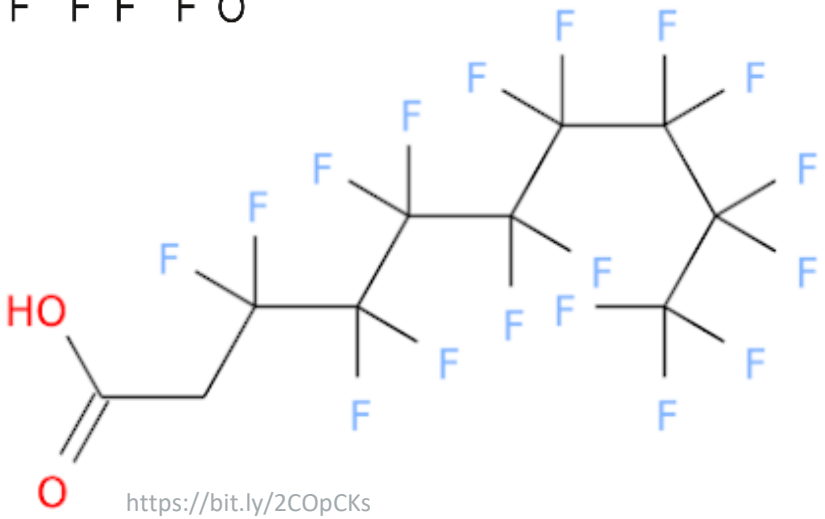
Per and polyfluoroalkyl substances (PFAS)

- PFAS are a class of contaminants
- There are more than 15,000 PFAS compounds
- Can repel oil and water, heat resistant
  - Applications such as
    - Non-stick pans
    - Raincoats
    - Firefighter foams
- They are very difficult to degrade,
  - they have strong C-F bonds
- PFAS are AKA forever chemicals
- No practical methods to remove from the environment ☹

PFAS

- Ultra short chain 2-3 carbons
- Short chain 4-7 carbons
- Long chain more than 8





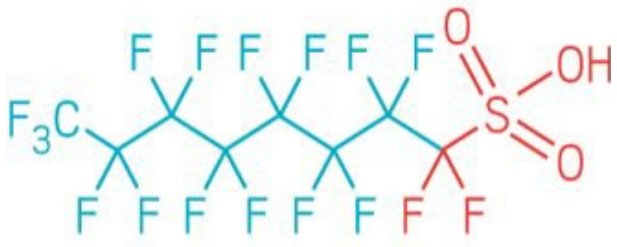
https://bit.ly/2COpCKs

Perfluoroalkyl acid substance (PFAS)	Acronym
Perfluorohexanoic acid	PFHxA
Perfluoroheptanoic acid	PFHpA
Perfluorooctanoic acid	PFOA
Perfluorononanoic acid	PFNA
Perfluorodecanoic acid	PFDA
Perfluoroundecanoic acid	PFUnA
Perfluorododecanoic acid	PFDoA
Perfluorotridecanoic acid	PFTrDA
Perfluorotetradecanoic acid	PFTA
Perfluorobutanesulfonic acid	PFBS
Perfluorohexanesulfonic acid	PFHxS
Perfluorooctanesulfonic acid	PFOS
N-methyl perfluorooctane-sulfonamidoacetic acid	NMeFOSAA
N-ethyl perfluorooctane-sulfonamidoacetic acid	NEtFOSAA

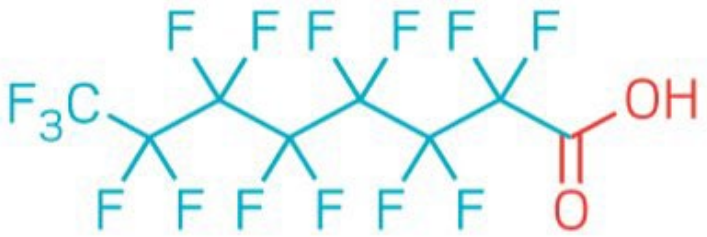
PFAS

- In use since the 1940s, recently aware of hazard
- Highly toxic at very low concentrations
- Related to high cholesterol
- Linked to hormonal imbalances

- Two of the most prevalent:



Perfluorooctane sulfonic acid, PFOS,



Perfluorooctanoic acid, PFOA,

Hemp Phytoremediation

- Upland Grassroots – a community group dedicated to cleaning land using fiber hemp

- Hemp (*Cannabis sativa*) characteristics:
  - Fast growing
  - High water uptake
  - Big

- Goal: Phytoextraction

Site Information: Burn House

- Upland Grassroots – CAES. Possible project?
- The burn house and parking lot located at the former Loring Airforce Base in northern Maine were used for firefighting drills and testing
- The land now belongs to the Aroostook Band of the Mi'kmaq Nation
- The US Airforce investigated PFAS at the site in 2015-2017. No remediation completed.

Baker, 2018

Site Details

Ditch that receives parking lot drainage

Burn house

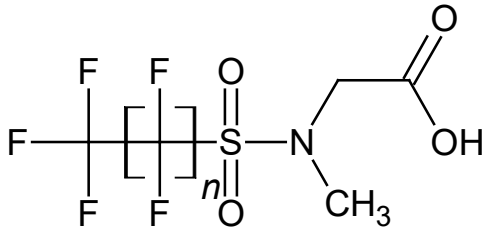
Bowl shaped parking lot with a drain in the northeast corner

Google Earth, 2022

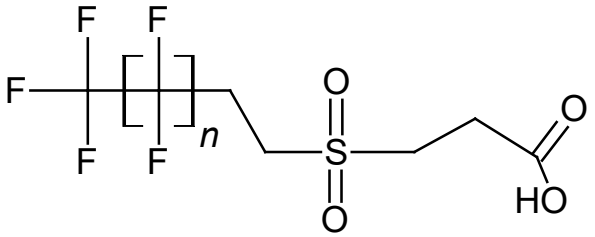
Previous results from soil analysis at Loring AFB

- PFOS identified as main PFAS contaminant (up to 150 ppb)
  - Other sulfonic and carboxylic acids also detected
- Non-targeted analysis found 15 different classes of PFAS
  - Indicate both PFOS and fluorotelomer based foams were used
- PFAS concentrations are highest in the drainage ditch, lowest at the south end of the site

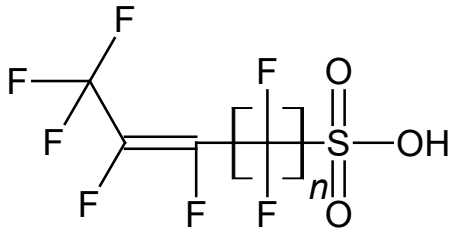
Sulfonamide-N-methyl alkyl acid



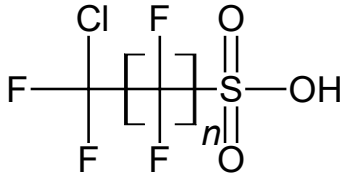
Fluorotelomer sulfone



Unsaturated Sulfonic Acid



Chlorinated Sulfonic Acid



Nason et al., 2021

Site Details

Ditch that receives parking lot drainage

Hemp Growth Plot 5: High PFAS area

Hemp Growth Plots 1-4: Lower PFAS area

Burn house

Bowl shaped parking lot with a drain in the northeast corner

Google Earth, 2022

Field Team

- 4 hemp varieties were tested in each growth plot
- Sown May 30, 2022
- Harvested August 22, 2022

Plant growth results

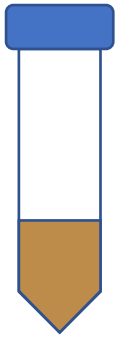
ChinMa variety hemp grew the best

Other hemp varieties struggled to grow and produced minimal biomass

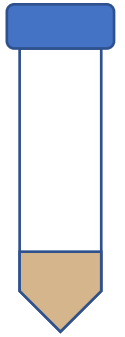
PFAS measurement: Extraction + LC-HRMS

Soil air dried and weighed into tubes

Shaken 5 minutes in 400 mM ammonium acetate in methanol

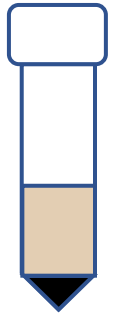


Centrifuged, supernatant decanted



Repeat 3x, combine all supernatant

Extracts dried down, reconstituted, cleaned-up with GCB



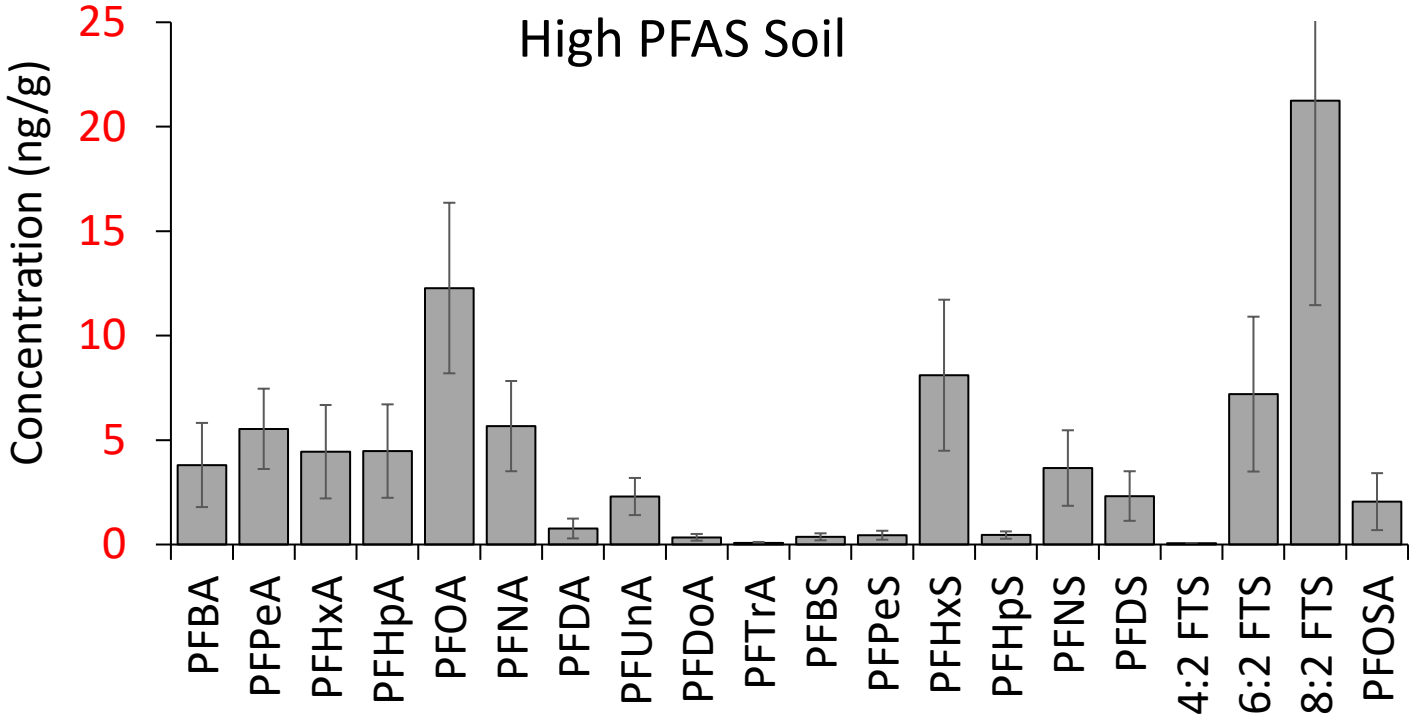
High speed centrifuge

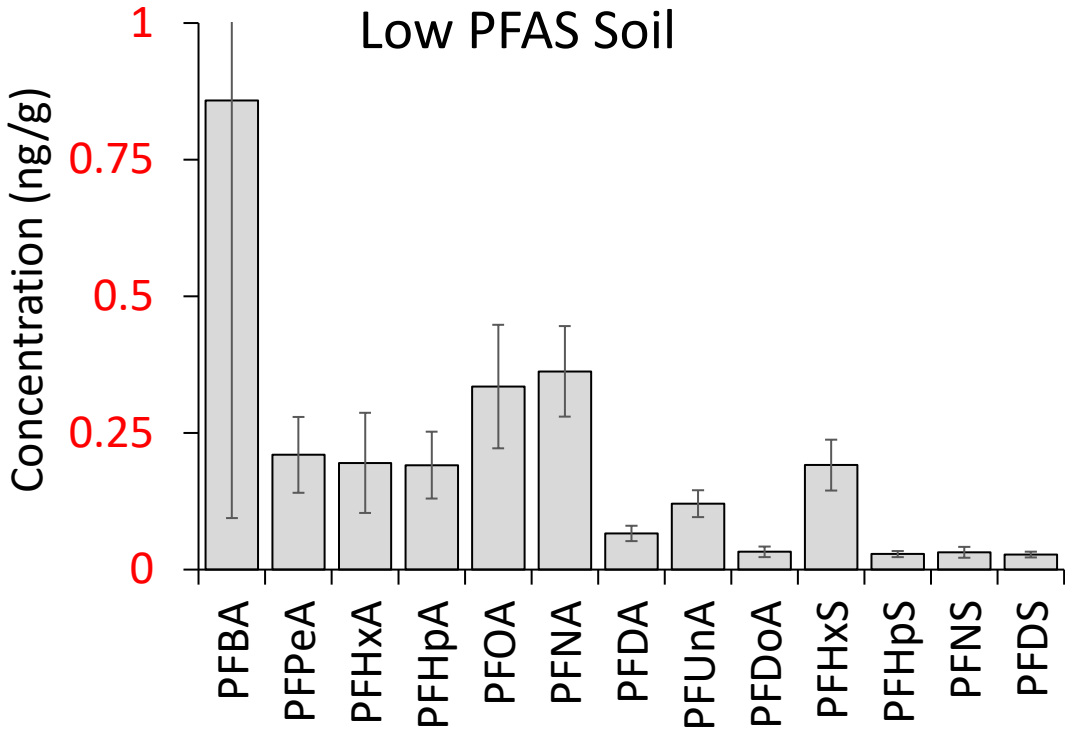
HPLC coupled with a Q-Exactive Orbitrap MS

Nason et al., 2021

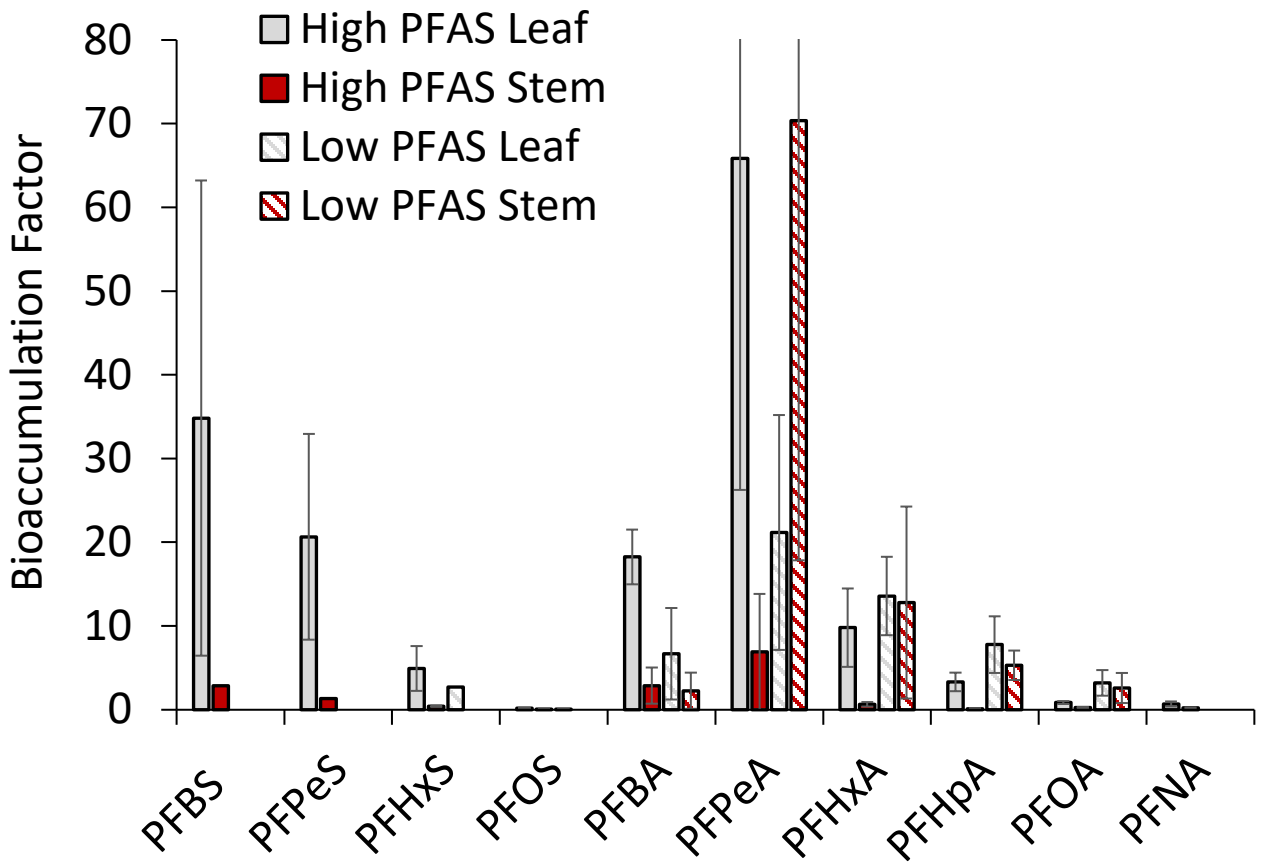
PFAS in soil

- PFOS is the primary PFAS present: 107 ng/g in high soil and 7.5 ng/g in low soil
- Soil concentrations compared before and after hemp growth for ChinMa growth plots – No significant differences found

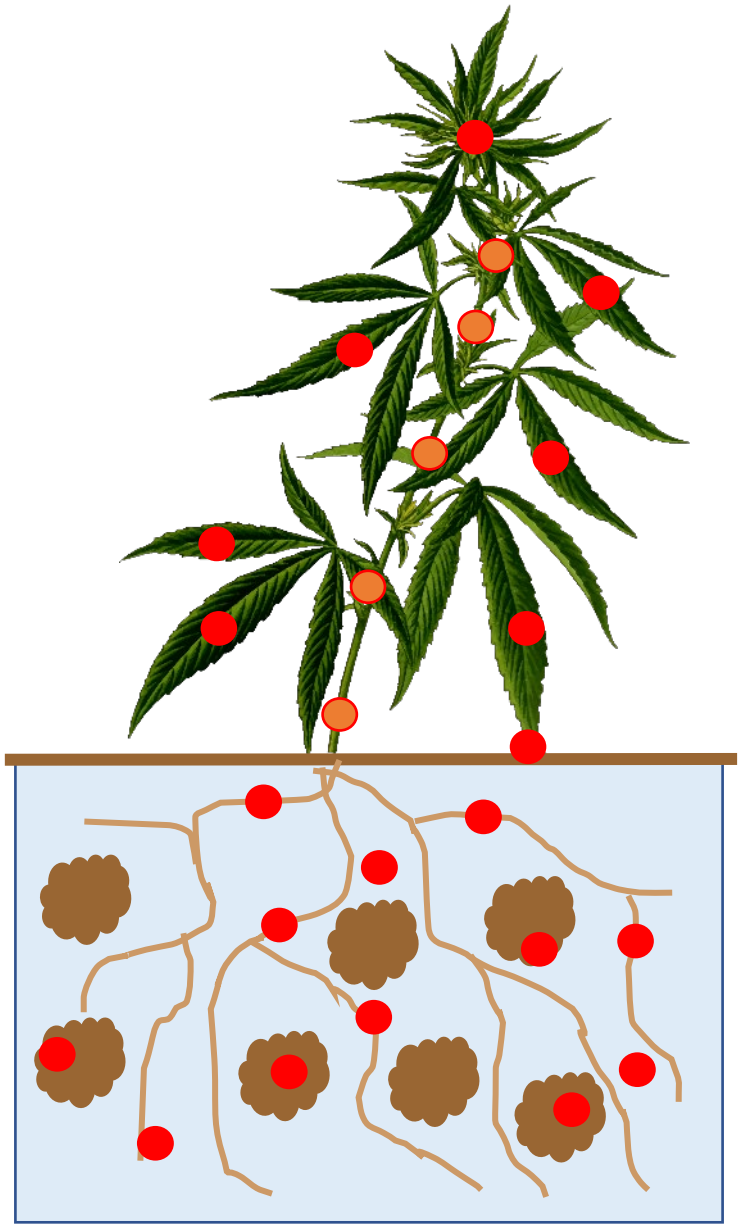




PFAS bioaccumulation in hemp

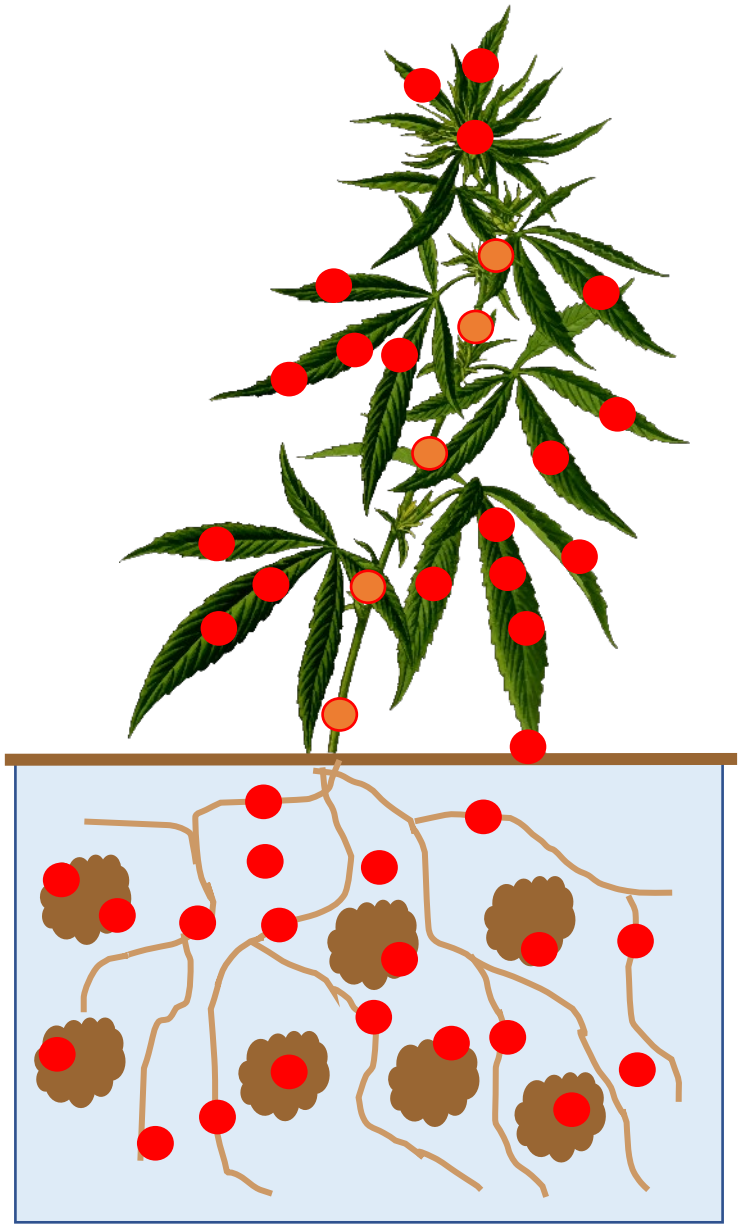


- **Some of the contaminant was removed**
- Bioaccumulation - dependent on PFAS chain length
  - Less with longer chain compounds
- Different bioaccumulation patterns in low and high PFAS soil
  - HIGH PFAS plot - Leaf concentrations higher than stems
  - LOW PFAS PLOT - Leaf and stem concentrations similar
  - Stem bioaccumulation greater in low PFAS soil than high PFAS soil



PFAS saturation in hemp stems?

- Contaminants move with transpired water from roots -> stems -> leaves
  - Sorption occurs along the way
  - Contaminant is left behind in leaves when water is transpired
- Hypothesis: a higher fraction of PFAS reach the leaves when a greater amount of PFAS are taken up
- Hemp stems contain the useful fibers for making cloth, rope, bricks, etc.



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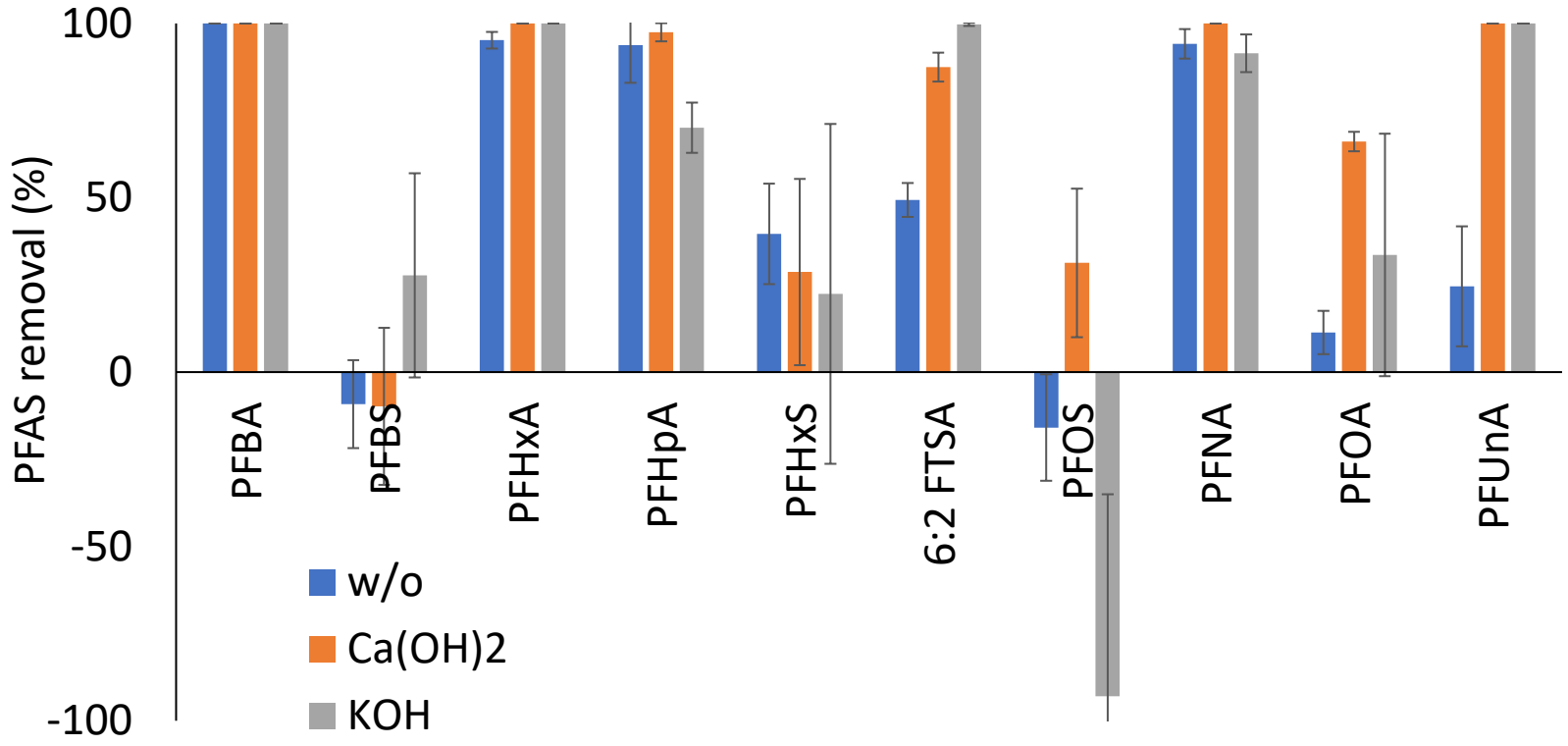
What happens to the PFAS-filled hemp?

- Transfer to a landfill
- Use fibers to make consumer products
  - Fate of PFAS in products is unknown
- Put hemp into a PFAS degradation process
  - Hydrothermal liquefaction (HTL)
    - Produces biofuel
  - Microbial degradation

Hydrothermal liquefaction (HLF)

- Dried hemp tissue is heated in a high-pressure system
  - $Ca(OH)_2$ or KOH may be added to enhance degradation
- Biofuel is produced

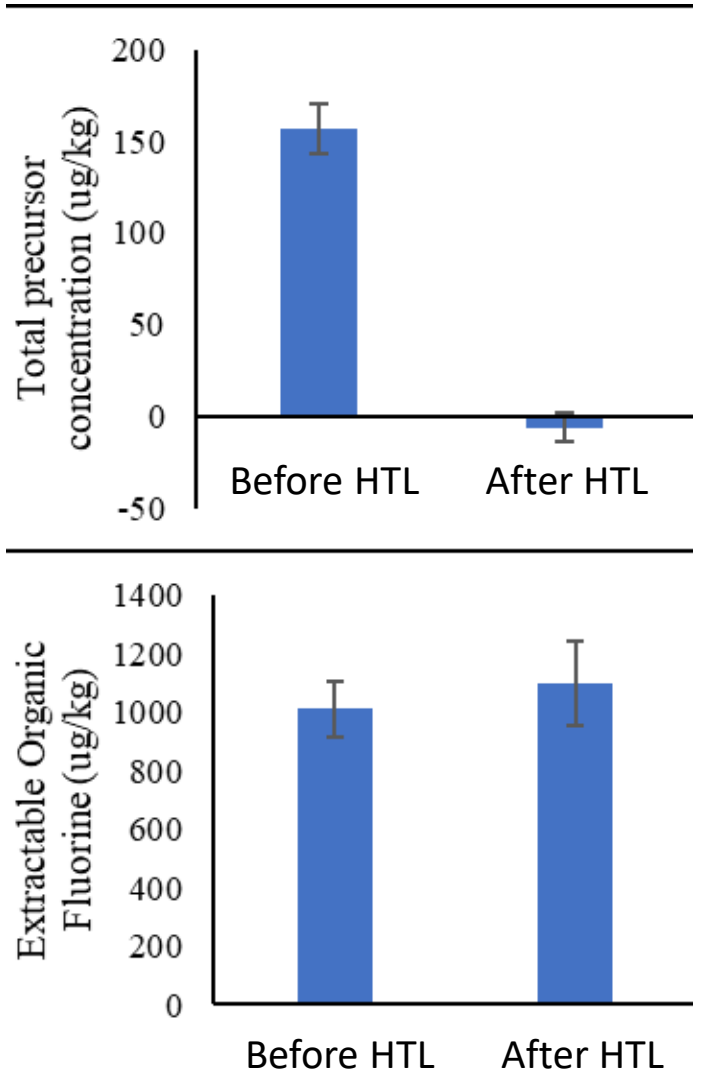
- Carboxylic acids are removed very well
- Some sulfonic acids are partially removed
- Are some PFAS produced as transformation products of other compounds?



Additional PFAS Measures

- Total Oxidizable Precursor assay indicates nearly complete removal of precursor PFAS after HLF

- Extractable Organic Fluorine measurements decrease when bases are added during HLF



Next steps

- Investigate microbial degradation of PFAS in hemp
  - Collaboration with Jaffé lab at Princeton University
- Non-targeted analysis of PFAS in plants and soil
  - What other PFAS are taken up by plants?
  - Are new PFAS formed in the plants?
  - Are there precursors present that explain some of the HLF results?
- NIH NIEHS R01ES032712

Investigate soil amendments to increase plant uptake of long chain PFAS.

“Understanding and enhancing PFAS phytoremediation mechanisms using novel nanomaterials”

- Just awarded from EPA program EPA-G2023-STAR-J1 Research for Understanding PFAS Uptake and Bioaccumulation in Plants and Animals in Agricultural, Rural, and Tribal Communities

“Novel, bio-enabled strategies to prevent per- and polyfluoralkyl substances accumulation in crops and food webs”

Summary and Conclusions

- There are currently no viable methods to remove PFAS from the soil.
- Phytoremediation is not a comprehensive solution for PFAS remediation, but has potential.
- Community impacts are significant and essential to in solving environmental problems.
- This project promoted community engagement which was key to get the attention from the government and industry into problems that require funding.
- Small improvements are meaningful. Even though we did not observe a significant removal of PFAS from soil, we were able to detect PFAS in plants, this PFAS uptake can make a difference over time.

References

- Nason, S. L.; Stanley, C.J.; PeterPaul C.E.; Blumenthal, M.F.; Zuverza-Mena, N.; Silliboy, R.J. (2021). A community based PFAS phytoremediation project at the former Loring Airforce Base. *iScience* 24, 102777 *https://www.cell.com/iscience/pdf/S2589-0042(21)00745-8.pdf*
- Nason, S. L., Koelmel, J., Zuverza-Mena, N., Stanley, C., Tamez, C., Bowden, J. A., Godri Pollitt, K. J. (2021). Software Comparison for Nontargeted Analysis of PFAS in AFFF-Contaminated Soil. *Journal of the American Society for Mass Spectrometry*, *32*, 840-846. DOI: 10.1021/jasms.0c00261
- Nason, S. L.; Thomas, S.; Stanley, C.; Silliboy, R., Blumenthal, M.; Zhang, W.; Liang, Y.; Jones, J.P.; Zuverza-Mena, N.; White, J.C.; Haynes, C.L.; Vasiliou, V.; Timko, M.P.; Berger, BW. (2024). A comprehensive trial on PFAS remediation: hemp phytoextraction and PFAS degradation in harvested plants. *Environ. Sci.: Adv*., 3, 304. DOI: 10.1039/d3va00340j

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