

THE BIG PROBLEM

In Situ Bioremediation Strategies to Destroy PFAS

Keith B. Rapp - PG, RG, CPG Jonna Spanier - PE Brendan McShane - CHMM

> MGWA Conference November 19, 2024



PROBLEM

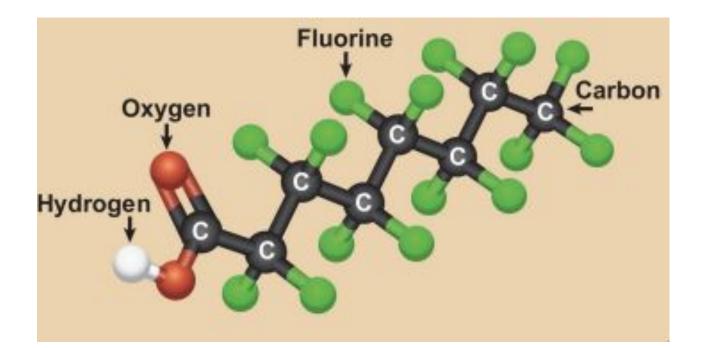
use, extent, exposure routes, persistence, limited remediation alternatives, foreverness

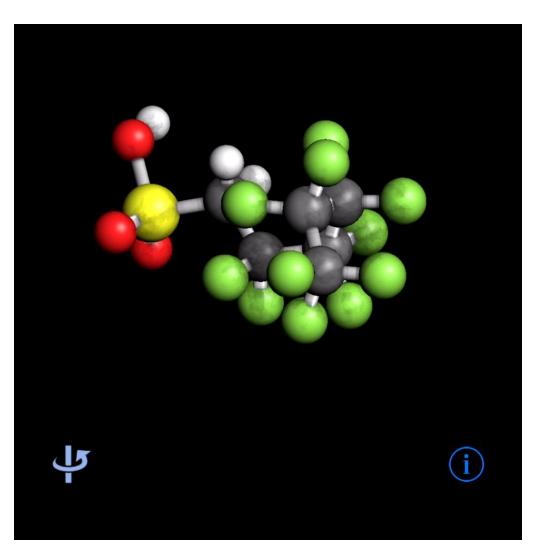
QUESTIONS

bioaccumulation, toxicity, remedial options, waste management, alternatives

PER- AND POLYFLUOROALKYL SUBSTANCES (PFAS)

All PFAS contain a chain of carbon atoms bonded to fluorine atoms



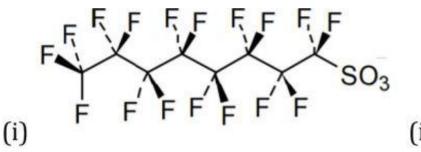


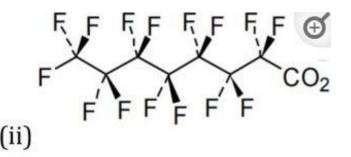
STRUCTURE OF PFAS - (i) PFOS, (ii) PFOA, (iii) PFHxS

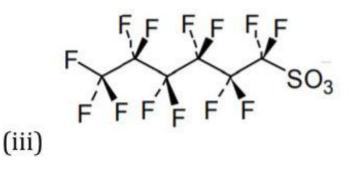
Perfluoroalkyl substances -

short and long carbon chains $(C_{2}-C_{12}^{+})$, fluorine atoms attach to all bonding sites except for the last carbon group

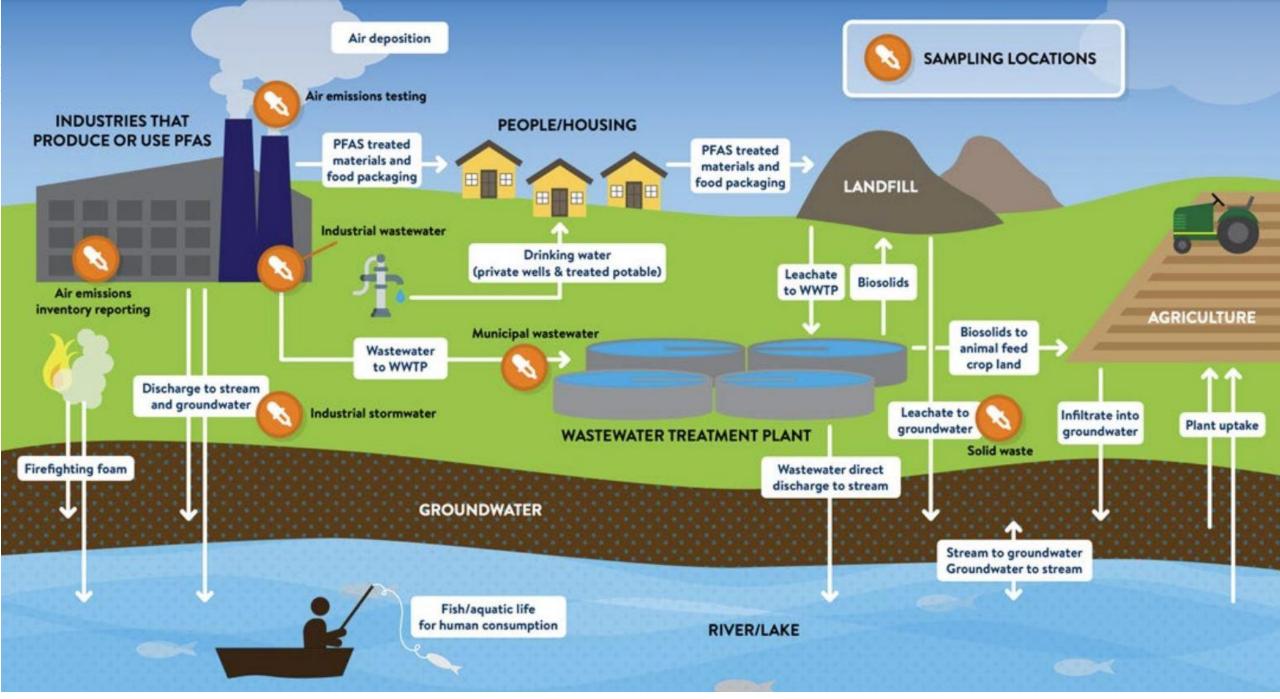
Polyfluoroalkyl substances are not fully fluorinated, and have at least one lapse in the chain not a fluorinated atom







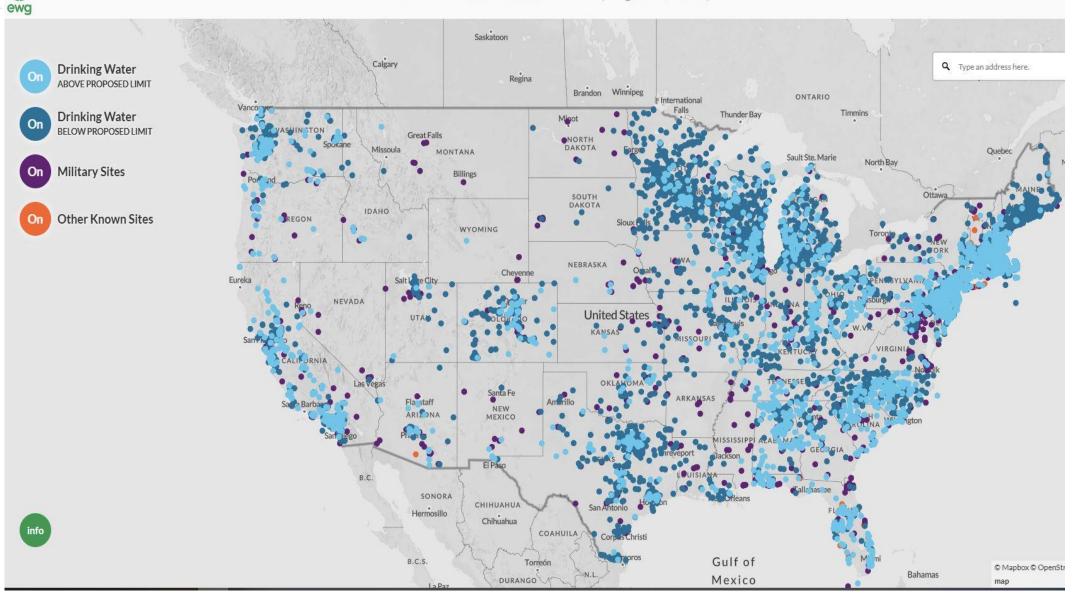
PFAS are a group of nearly 15,000 synthetic chemicals



Source: Minnesota Pollution Control Agency • PFAS Monitoring Plan: Initial findings and next steps

PFAS – It's Everywhere

PFAS contamination in the U.S. (August 9, 2024)



Environmental Working Group, 2024. https://www.ewg.org/interactive-maps/pfas contamination/

HUBRIS

PFAS costs \$50 - \$1,000 per pound, but \$2.7 - \$18 million per pound to remove and destroy from environment

Removing and destroying PFAS from MN water and biosolids \$14 - \$28 billion over 20 years

New "short-chain" PFAS are more difficult and up to 70% more expensive than old "long-chain" PFAS February 2021

Minnesota's PFAS Blueprint

A plan to protect our communities and our environment from per- and polyfluorinated alkyl substances



MI MINNESOTA



TREATMENT TECHNOLOGIES V. REMEDIATION STRATEGIES

<u>**Treatment technologies</u>** exploit a contaminant's chemical and physical properties to immobilize, separate and concentrate, or destroy the contaminant. <u>**Point source solution**</u></u>

<u>Remediation strategies</u> holistically address the entirety of environmental conditions, exposures, environmental media and corrective action options to eliminate mechanisms and conditions in environmental contamination. <u>Field scale solution</u>

TREATMENT TECHNOLOGIES (POINT SOURCE)

Separation technologies

Ion exchange resin Colloidal/granular activated carbon Nanofiltration Reverse osmosis Foam fractionation

Destructive technologies

Electrochemical oxidation Plasma Photocatalysis Sonolysis Supercritical water oxidation

Thermal degradation/incineration

Meegoda et al. A Review of PFAS Destruction Technologies. 2022.



TREATMENT TECHNOLOGIES (SAFF)

- injects air into contaminated water, concentrating PFAS in foam
- 2. foam is removed



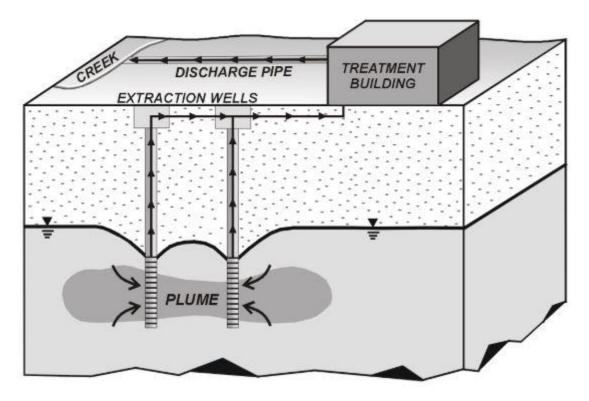


Minnesota is the first state government in U.S. to use this combination of innovative technologies to address "forever chemicals."

MPCA - 2022. https://www.pca.state.mn.us/news-and-stories/mpca-brings-cutting-edge-technology-to-minnesota-to-remove-pfas-from-water

REMEDIATION STRATEGIES

Groundwater pump & treat (P&T)

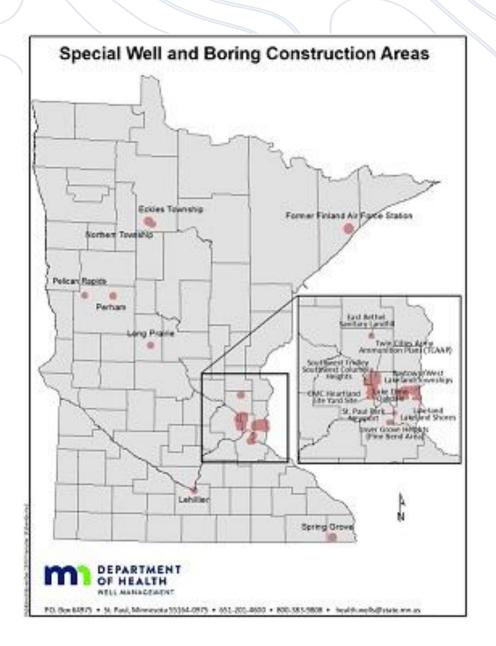


In Situ Sorption (sequestration)



granular AC

powdered AC



CTIONS | p

* StarTribune

A plume of PFAS chemicals under the east metro is moving. The state has a plan to stop it.

Preliminary plans would include a broad and complex system of wells to control the underground flow.

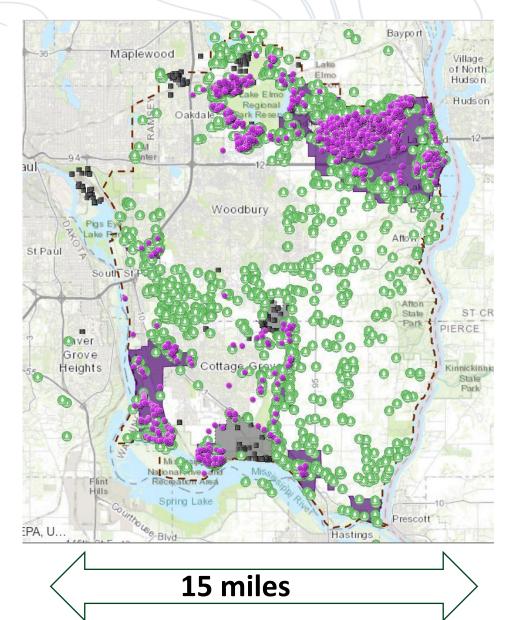
By Chloe Johnson Star Tribune APRIL 19, 2024 - 6:00AM

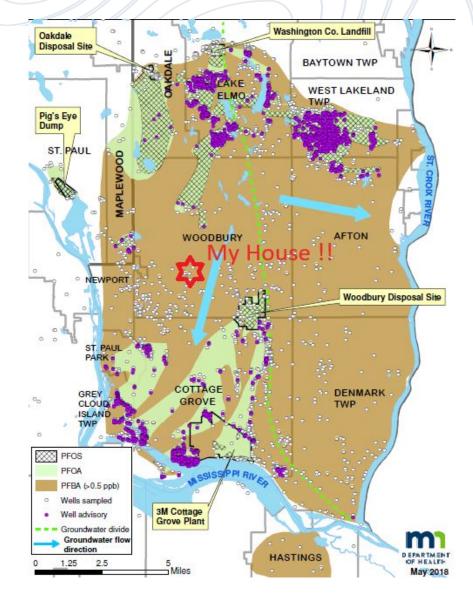


DAVID JOLES, STAR TRIBUNE

3M dumped PFAS sludge in the Washington County landfill for years. It's one of two sites that seeded underground chemical pollution that state officials now say is migrating.

S. WASHINGTON COUNTY, MN

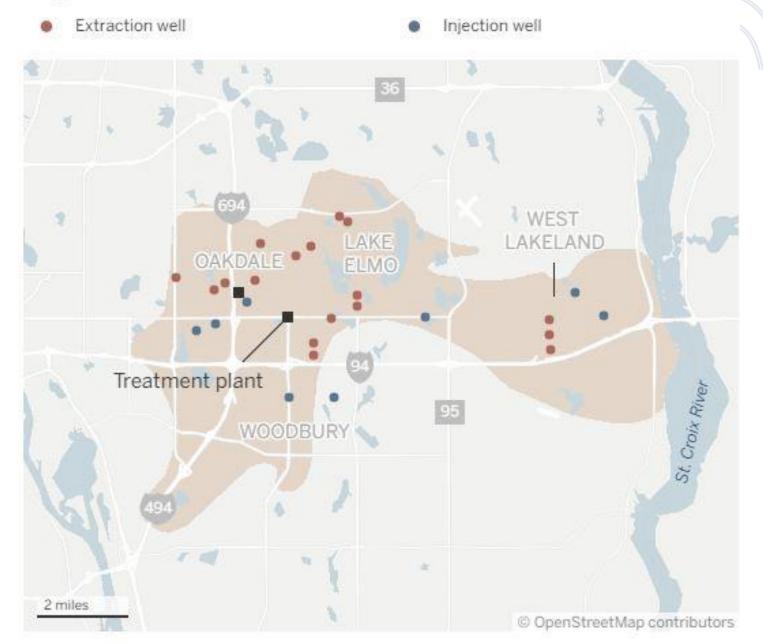




MN MOAT

Minnesota studying "moat" concept – network of extraction wells to pump PFAS-contaminated groundwater, treat it, and re-inject

Proposed infrastructure



Jake Steinberg, Star Tribune • Source: Minnesota Pollution Control Agency, AECOM

ECONOMIC COMPETITION

"Pump & Treat is remediation in perpetuity"



100⁺-million Americans' water impacted with PFAS above federal drinking water standards.

Solving this is THE BIG PROBLEM



Use of naturally occurring or deliberately introduced microorganisms or other life forms to consume or breakdown environmental pollutants

"Environmental cleanup by bioremediation is not considered practical currently. Implementation of [PFAS] bioremediation will require uncovering and understanding the rare microbial successes in degrading these compounds."

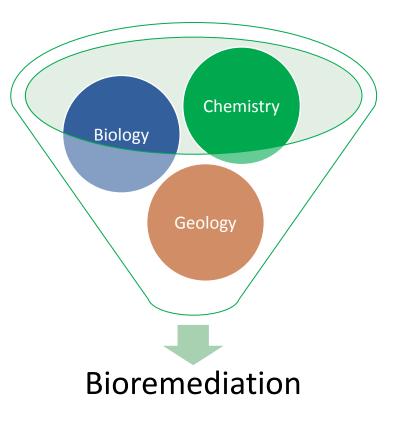
THIS WAY



^{*}L.P. Wackett, et.al. – 2021. Microbial Biotechnology, 15, 773–792

HydroGeoMicroBioChemistry

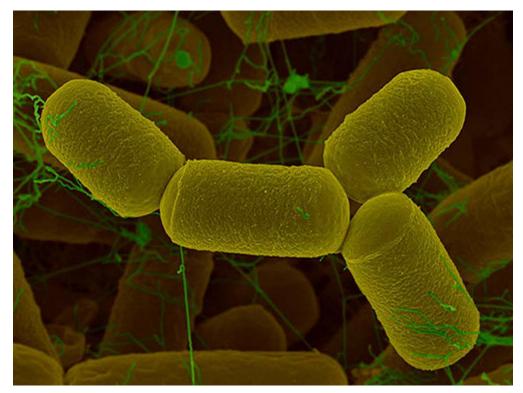
Interaction of chemistry, biology (micro), geology, soil, sediment, rock and groundwater contamination

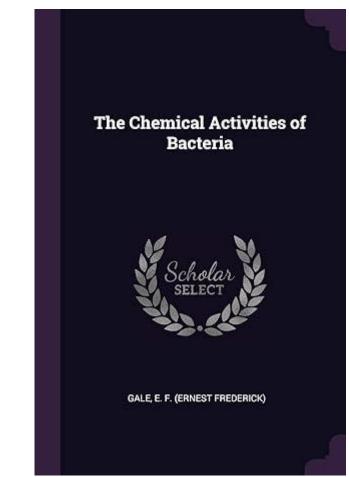




MICROBIAL INFALLIBILITY HYPOTHESIS

".....if there is energy to be gained from a compound [contaminant], a microorganism will figure out how to extract it and create a niche for itself."





E.F. Gale, 1951 - The Chemical Activities of Bacteria

BIOFILM GODFATHER

"Biofilms form in all environments with sufficient nutrients – worldwide"

Costerton (1995)

In nature, >80% of microbes exist within a biofilm

J. William Costerton **The Biofilm Primer** Springer

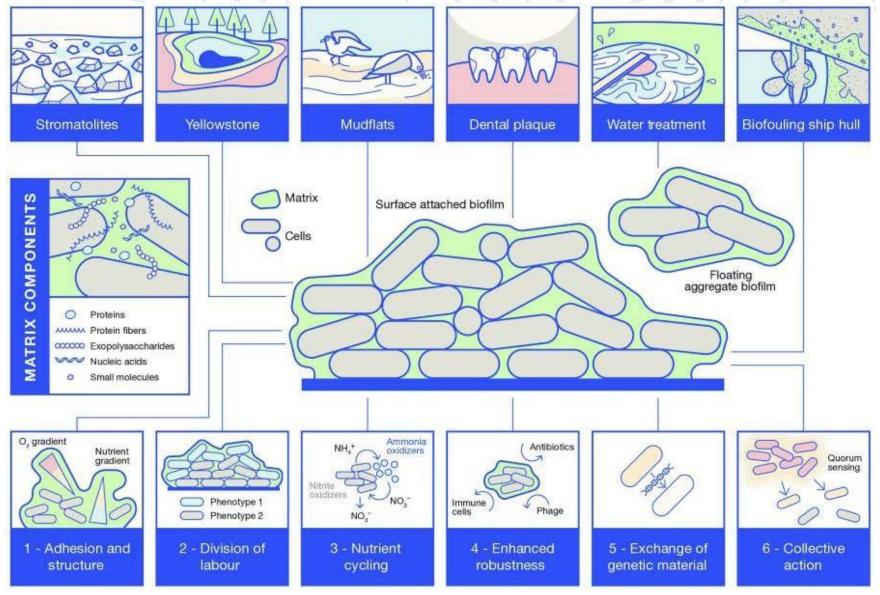
KNOWN PFAS BIODEGRADING MICROBES

Bacteria

Acetobacterium sp.	Desulfovibrio aminophilus	Sporomusa sphaeroides	Pseudomonas sp.
Desulfovibrio aminophilus	Sporomusa sphaeroides	Acidimicrobium sp.	
<i>Burkholderia</i> sp.	Thauera aromatica	Aliivibrio fischeri	Rhodopseudomonas
Desulfococcus	<i>Bacillus</i> spp	Rhodococcus jostii	Dehalobacter sp.
Fungi			
Aspergillus niger (brown rot)	Phanerochaete chrysosporium (white rot)		Phanerochaete chrysosporium
Archaea			
Thaumarchaeota	Euryarchaeota		

BIOFILMS EXECUTE BIOREMEDIATION

BIOFILMS IN THE WORLD



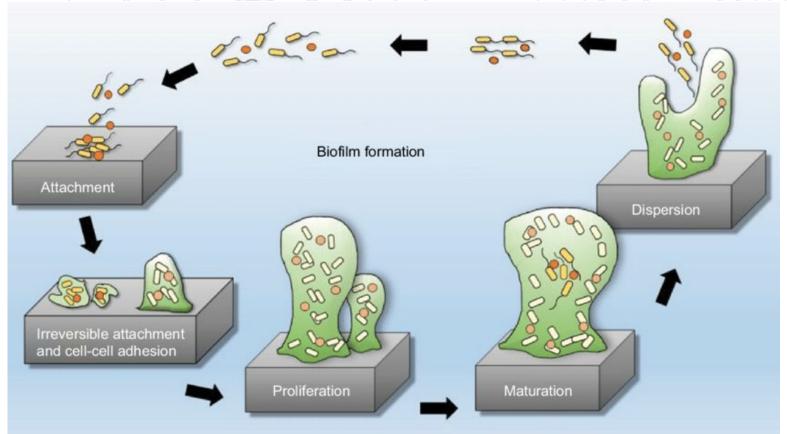
Bamford, NC, et.al., Microbial Primer: An introduction to biofilms – August 1, 2023

BIOFILM DEVELOPMENT

Biofilms form via cell-to-cell communication

Microbes form attachments to <u>surfaces</u>. Proteins signal nearby cells (<u>quorum sensing</u>)

Signaling recruits new cells, horizontal gene transfer, colony aggregates and forms a biofilm. <u>Colony builds</u>



Proteins signal development of exopolysaccharides (EPS), a protective layer, with channels for flow, energy transmission, and horizontal gene transfer. <u>Biofilm matures</u>

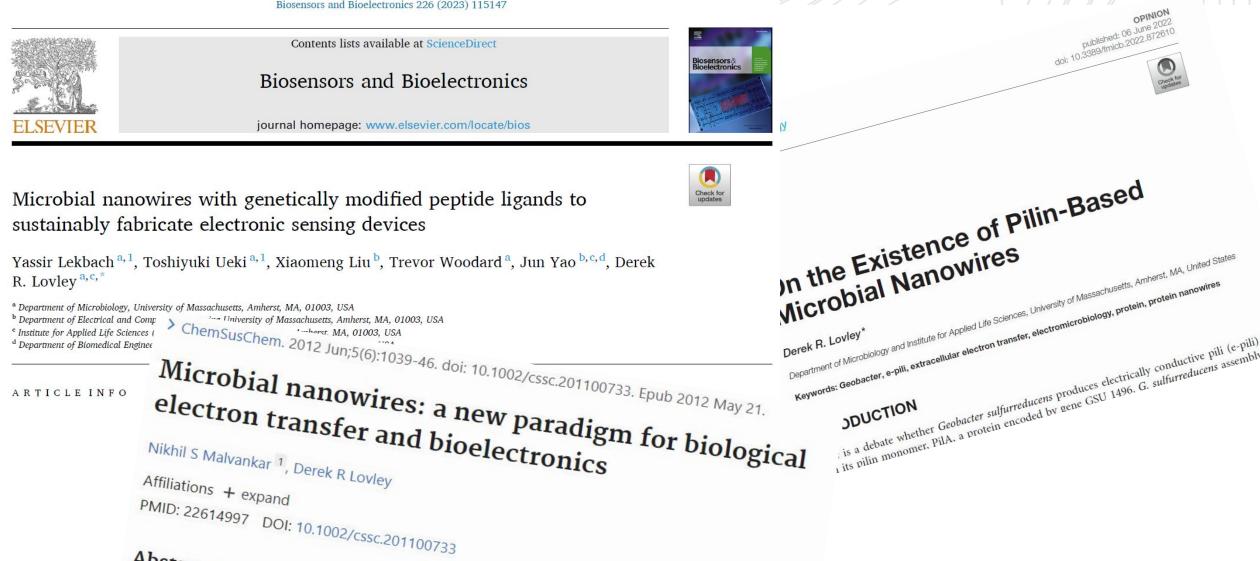
BIOFILM FORMATION

Contaminant destruction occurs at <u>surfaces</u> – pore surfaces, fractures, boundaries.

Planktonic microbes are quickly removed, sessile microbes accomplish remediation



BIOFILM COMMUNICATIONS



Abstract

The discovery that Geoba

Dr. Bonnie Bassler (YouTube - 18:11) https://youtu.be/KXWurAmtf78?si=JLtbt3lzhO-yS63K LATEST TOPICS - MAGAZINE - FEATURES - COLLECTIONS - PODCASTS CHEMPICS JOBS (Q)

f 🞯 🕱 in

REKSISTENT PULLUTANTS

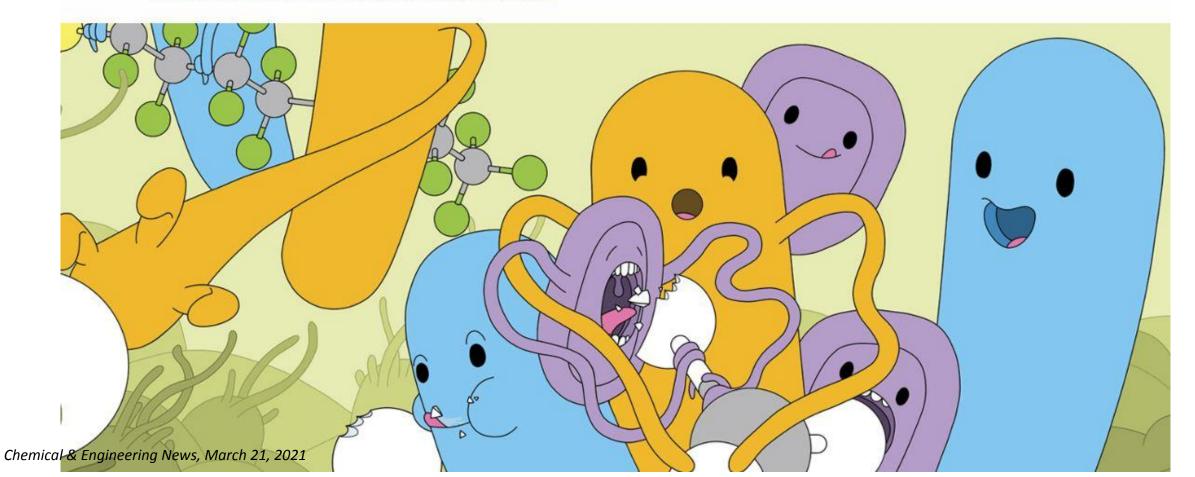
c&e

Can microbes save us from PFAS?

Researchers are investigating whether microbes can help break down fluorinated contaminants

by XiaoZhi Lim, special to C&EN

March 21, 2021 | A version of this story appeared in Volume 99, Issue 10



Microbes Collectively Organizing



• MICROBES COLLECTIVELY ORGANIZING

BIOFILM FORMATION

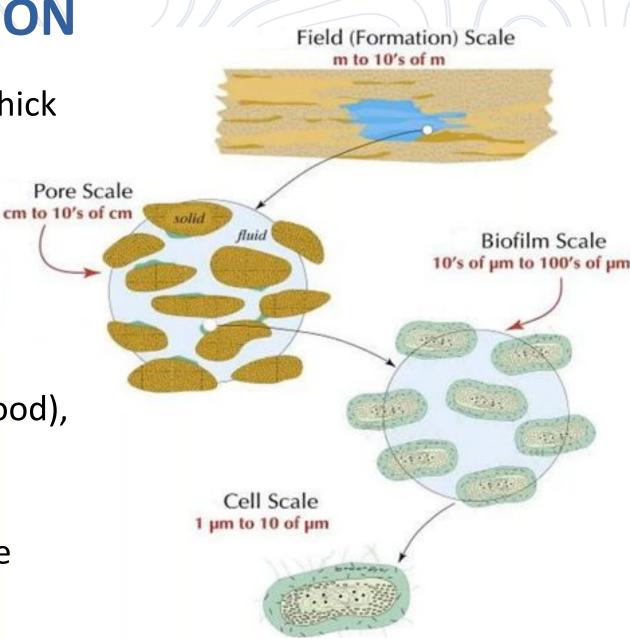


SCALE & COMPOSITION

- Biofilms ~97% water, ~10 to 100 μm thick
- EPS layer ~0.2 to 1.0 μm
- EPS 65–95%, microbes 5-35%

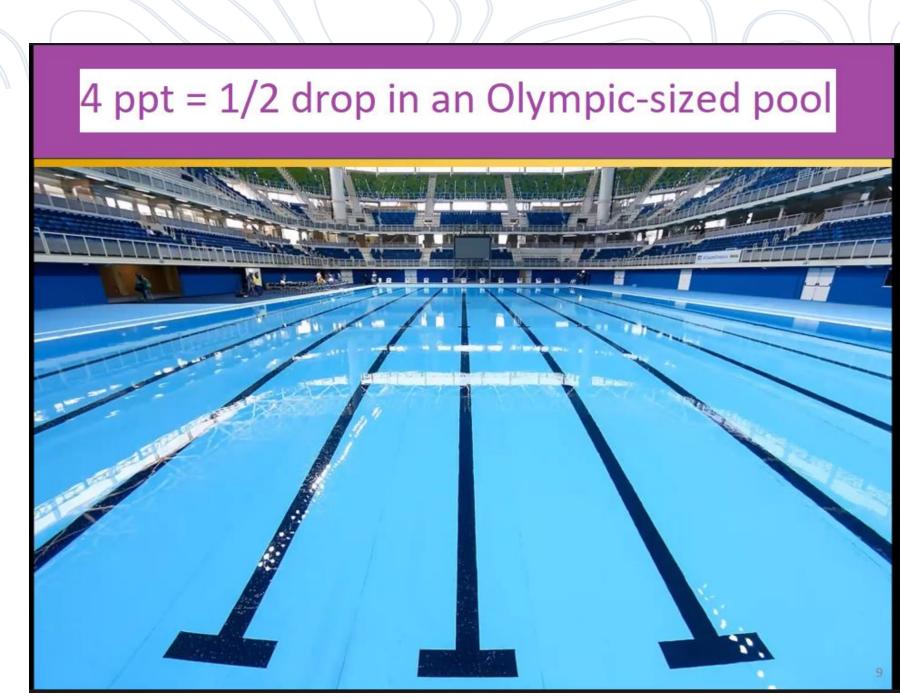
EPS composition determined by environment, growth circumstances, microbial strains (DNA-in-the-neighborhood), and nutrient supply

Lack of nutrients = biofilm cells disengage from surfaces



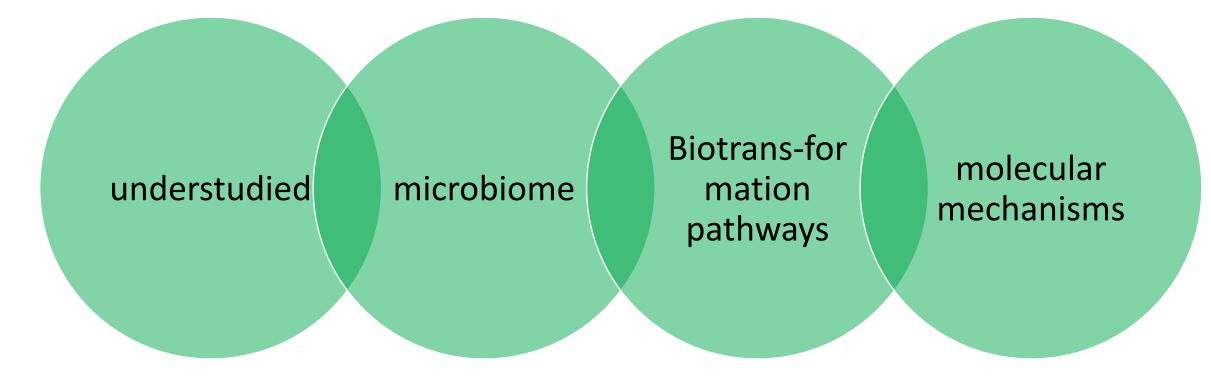


PFOA/PFOS standard is 1,000x more dilute than an equivalent TCE plume



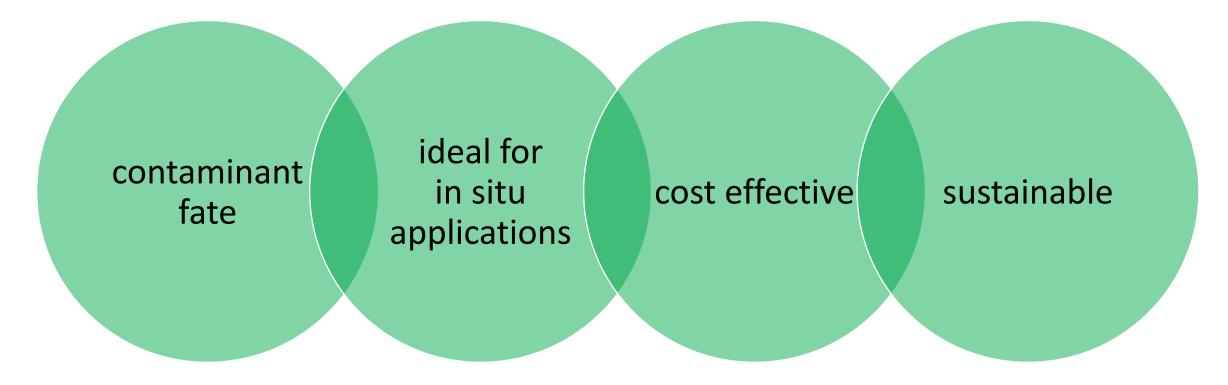
IN SITU PFAS BIODEGRADATION

UNKNOWNS



IN SITU PFAS BIODEGRADATION

BENEFITS



microbial biotechnology

Minireview

Nothing lasts forever: understanding microbial biodegradation of polyfluorinated compounds and perfluorinated alkyl substances



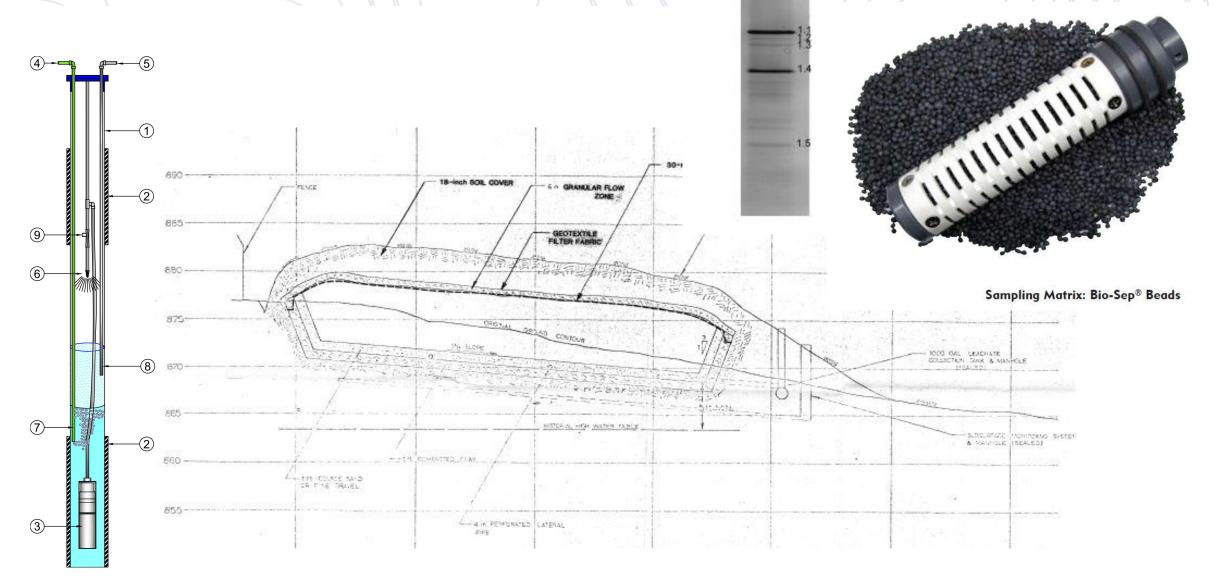
Open Access

MDPI

BIOFILM MEDIATED PFAS DESTRUCTION IN GROUNDWATER



MICROBE SOURCING

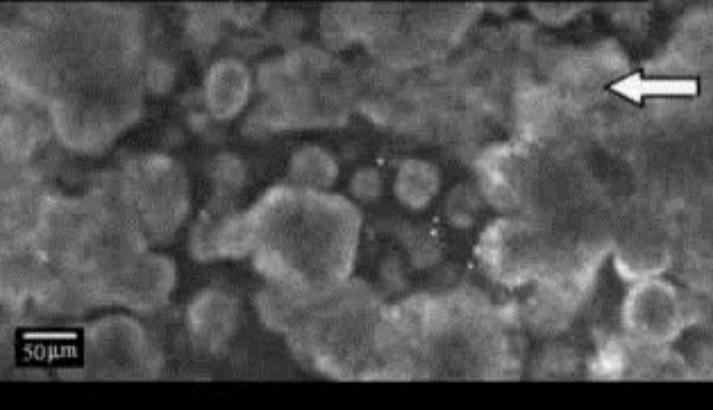


MW-118

Accelerated Remediation Technologies, Inc. - https://www.artinwell.com/about.asp

BIOFILM ENVIRONMENT

Channel flow occurs within pores & fractures & 10 – 1,000 X faster Liquid Flow Through Biofilm Channels Stoodley et al. 1994 Appl. Environ. Microbiol.60:2711-16 deBeer and Stoodley, 2000 in The Prokaryoles

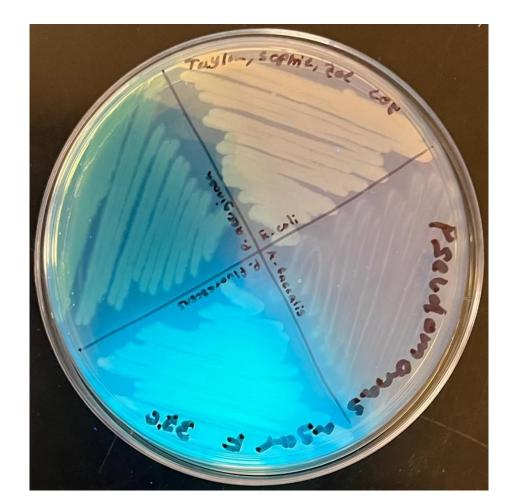


Biofilm of Pseudomonas aeruginosa, Pseudomonas fluorescens, and Klebsiella pneumoniae. P. Stoodley & Z. Lewandowski (1994) - Montana State University - Center for Biofilm Engineering (CBE)

ISOLATES V. CONSORTIUM

New and novel testing and destructive technologies

- Stable isotope probing
- Metagenomics
- Transcriptomics
- Metabolomics
- New genes
- Carbon tagging/ biodegradation



MICROBAC[®]

Genomic Sequencing Report

3/13/2024

Microbac Oak Ridge WO#

Client: Kent Armstrong

Sample Project:

Sample ID: DWH-Z

Sample Collected: 1/30/2024

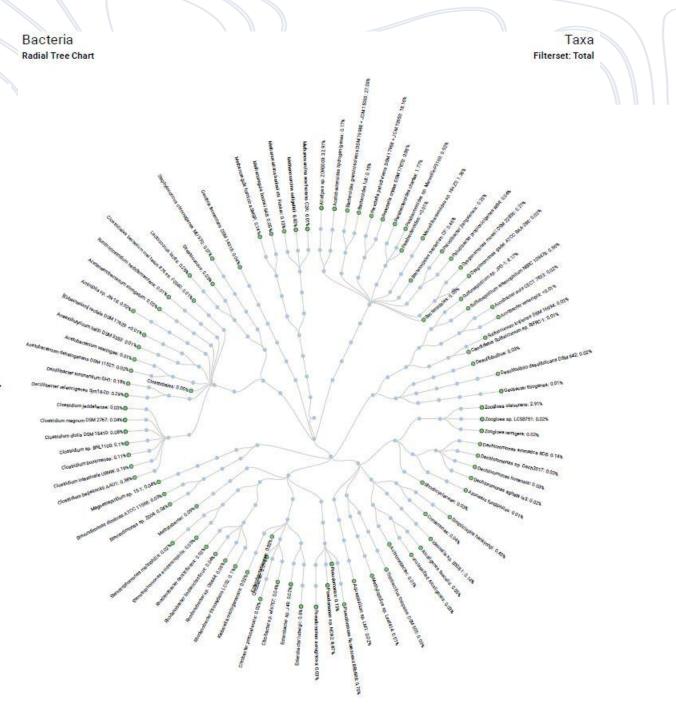
Sampe 16S Biomass: 2.9 X 10¹¹ gene copies/g

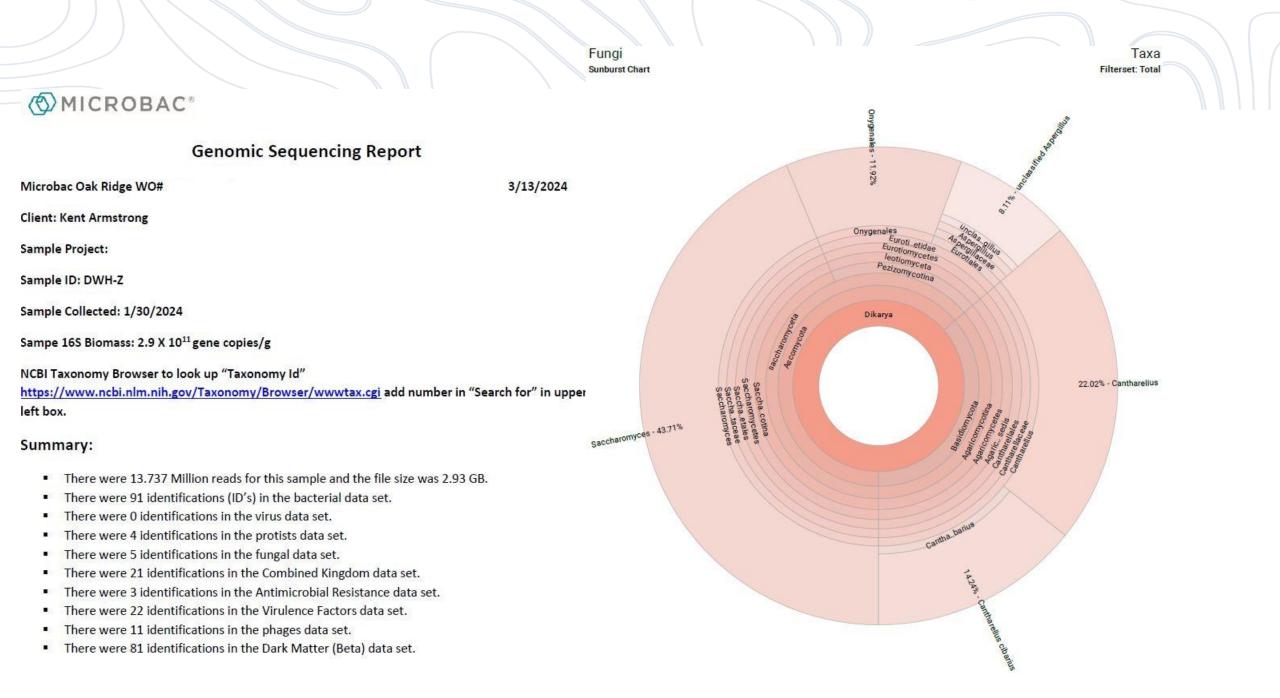
NCBI Taxonomy Browser to look up "Taxonomy Id"

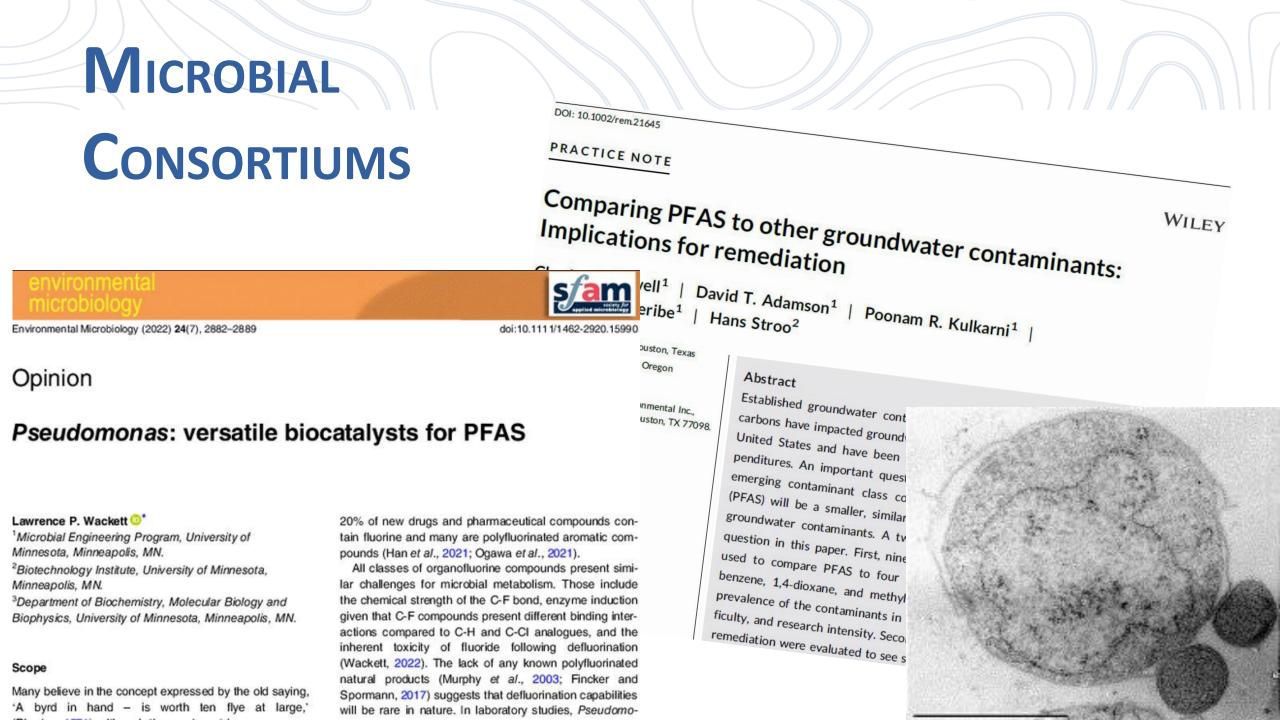
https://www.ncbi.nlm.nih.gov/Taxonomy/Browser/wwwtax.cgi add number in "Search for" in upper left box.

Summary:

- There were 13.737 Million reads for this sample and the file size was 2.93 GB.
- There were 91 identifications (ID's) in the bacterial data set.
- There were 0 identifications in the virus data set.
- There were 4 identifications in the protists data set.
- There were 5 identifications in the fungal data set.
- There were 21 identifications in the Combined Kingdom data set.
- There were 3 identifications in the Antimicrobial Resistance data set.
- There were 22 identifications in the Virulence Factors data set.
- There were 11 identifications in the phages data set.
- There were 81 identifications in the Dark Matter (Beta) data set.







FLUORIDE EFFECT

Applied

Role of the CrcB transporter of Pseudomonas putida in the RESEARCH ARTICLE multi-level stress response elicited by mineral fluoride Patricia Calero | Nicolás Gurdo | Pablo I. Nikel 💿

The Novo Nordisk Foundation Center for Biosustainability, Technical University of Denmark, Kongens Lyngby, Denmark

Pablo I. Nikel, The Novo Nordisk Foundation Center for Biosustainability, Technical University of Denmark, 2800 Kongens Lyngby, Denmark. Email: pabnik@biosustain.dtu.dk

H2020 Environment, Grant/Award Number: 814418; Novo Nordisk Fonden, Grant/Award Numbers: NNF180C0034818, NNF20CC0035580, NNF210C0067996; Cystic Fibrosis Trust, Strategic Research Centre Award-2019-SRC017; Danish Council for Independent Research Natural Sciences, Grant/Award Number: 8021-00039B

The presence of mineral fluoride (F⁻) in the and anthropogenic origin, and the halide ha tually all living organisms. While the evide species supports this notion, a systematic salts on the metabolism and physiology underexplored thus far. In this work, we mechanisms deployed by the model s KT2440 against NaF. By adopting syste functional genomics and metabolomics, at different regulatory levels under con Several genes involved in halide tolera Tn-Seq screening—among which crc was shown to play the predominant metabolomics, combined with the ass lar pH values and quantitative physio

nature communications

Perspective

https://doi.org/10.1038/s41467-024-49018-1

9

The link between ancient microbial fluoride resistance mechanisms and bioengineering organofluorine degradation or synthesis

Received: 10 January 2024

Randy B. Stockbridge O¹ & Lawrence P. Wackett O²

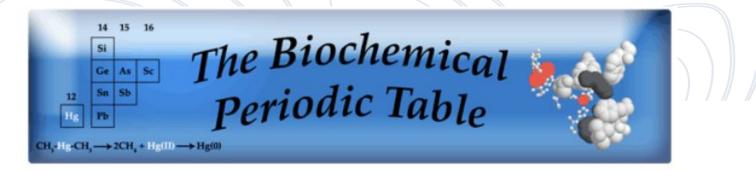
Accepted: 20 May 2024

Published online: 30 May 2024

Check for updates

Fluorinated organic chemicals, such as per- and polyfluorinated alkyl sub-

stances (PFAS) and fluorinated pesticides, are both broadly useful and unusually long-lived. To combat problems related to the accumulation of these compounds, microbial PFAS and organofluorine degradation and biosynthesis of less-fluorinated replacement chemicals are under intense study. Both efforts are undermined by the substantial toxicity of fluoride, an anion that powerfully inhibits metabolism. Microorganisms have contended with environmental mineral fluoride over evolutionary time, evolving a suite of detoxification mechanisms. In this perspective, we synthesize emerging ideas on microbial defluorination/fluorination and fluoride resistance mechanisms and identify best approaches for bioengineering new approaches for degrading and making organofluorine compounds.



He

Ne

Ar

Kr

Xe

Rn

Group 1 12 13 14 15

Pe	riod	

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<u>Li</u>

Na

K

Rb

Cs

Fr

Ra

		Key	E	Biolog	ically	releva	ant ele	ement	s colo	red							
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		N	Major	cations,	all life												
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	4		Major	biologica	al transit	ion meta	s					5	6	7	8	9	
	Be		Speci	alized us	es, some	e life						B	<u>C</u>	N	<u>0</u>	E	
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	<u>Ba</u>	Lu	Hf	<u>Ta</u>	W	<u>Re</u>	<u>Os</u>	Ir	<u>Pt</u>	<u>Au</u>	Hg	11	Pb	<u>Bi</u>	Po	At	

Lanthanoids	57	58	59	60	61	62	63	64	65	66	67	68	69	70
	<u>La</u>	<u>Ce</u>	<u>Pr</u>	<u>Nd</u>	<u>Pm</u>	<u>Sm</u>	<u>Eu</u>	<u>Gd</u>	<u>Tb</u>	<u>Dy</u>	<u>Ho</u>	<u>Er</u>	Tm	<u>Yb</u>
Actinoids	89	90	91	92	93	94	95	96	97	98	99	100	101	102
	Ac	<u>Th</u>	<u>Pa</u>	<u>U</u>	<u>Np</u>	<u>Pu</u>	<u>Am</u>	<u>Cm</u>	Bk	Cf	Es	Fm	Md	No

INTENSE FINANCIAL PRESSURE

DoD	Investigate	Cleanup
2019	\$256.5M	\$245.1M
2020	\$242.5M	\$28.8M
2021	\$174.1M	\$77.3M
2022	\$181.7M	\$78.5M

¹ Current removal cost equal to rate of production estimated \$20 to \$7,000 trillion/yr

² PFAS drinking water removal \$3.2 - \$5.7 billion/yr

2

DRINKING WATER TREATMENT

WaterWorld

DRINKING WATER

SMART WATER

PFAS remediation spending forecasted to triple by 2030

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ASSET MANAGEMENT

A new report form Bluefield Research anticipates that infrastructure investments, amid a changing regulatory env national spending on PFAS treatment systems.

ABOUT

WATER UTILITY MANAGEMENT

SMART WATER

WATER REUSE

WASTEWATER

SUMMIT

EHANDBO

RESIDENTIAL/COMMERCIA

May 17, 2022



PFAS SOURCE REDUCTION



SECTIONS | P

LOCAL

🖈 StarTribune

By 2032, almost anything you buy in Minnesota will come in recyclable, compostable or reusable packaging. Here's why.

Environmental groups and Twin Cities leaders say requiring packaging be recyclable is key to reducing the growing amount of waste.

By Christopher Magan Star Tribune | JUNE 4, 2024 - 9:29AM



ELIZABETH FLORES, STAR TRIBUNE

Earlier this year, the Food & Drug Administration tested for PFAS in a variety of foods. While the sample sizes were small and may not reflect typical contamination levels, here's what the FDA found.



Natural Resources Defense Council scorecard (Apparel)

https://www.nrdc.org/resources/going-out-f ashion-us-apparel-manufacturers-must-elimi nate-pfas-from-their-supply-chains

Green Science Policy Institute (Corporate PFAS evaluation)

https://pfascentral.org/pfas-free-products/

Center for Environmental Health (Foodware) https://ceh.org/wp-content/uploads/2021/0 8/foodware.jpg

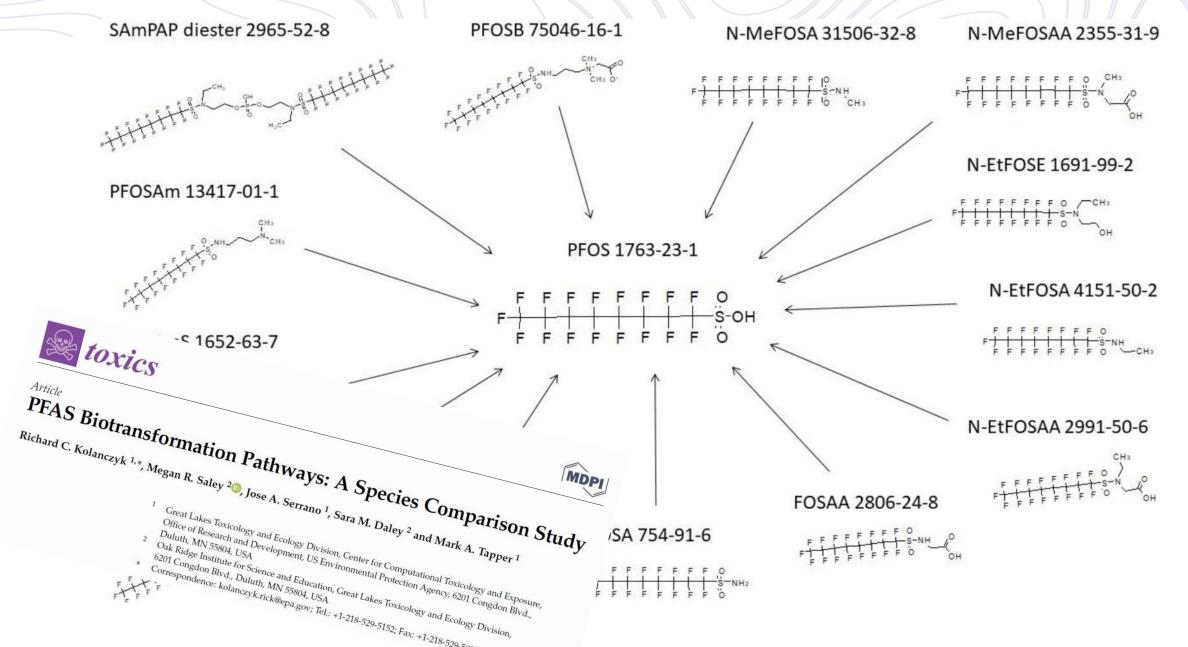
NOTHING LASTS FOREVER

QUESTIONS

Keith B. Rapp krapp@baywest.com (612) 382-3763



BIOTRANSFORMATION PATHWAYS

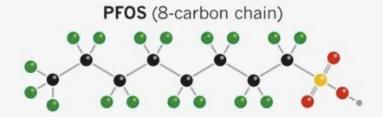


OLD PFAS

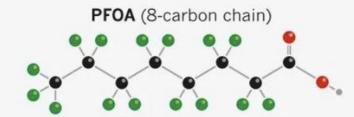
Carbon
Fluorine
Sulfur

Oxygen • Hydrogen • I

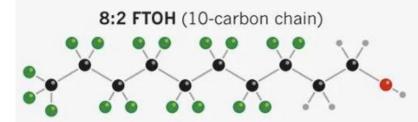
n 💿 Nitrogen



Production now heavily restricted.

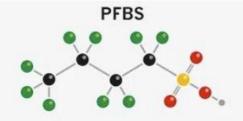


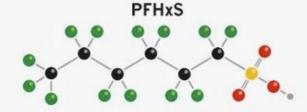
Expected to be similarly restricted this year.

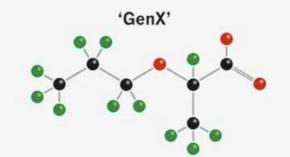


Hundreds of precursor compounds can degrade into PFOS or PFOA in the environment.

'New Gen PFAS'







Variations in chain length and branching produce dozens of variant structures.

A Stockholm Convention committee is reviewing whether to ban this substance.

US chemical firm Chemours is being sued over the presence of this chemical in North Carolina water supplies.

Scientific American, Feb. 19, 2019. The Fluorine Detectives.