Journal of Hazardous Materials 446 (2023) 130629

Contents lists available at ScienceDirect

Journal of Hazardous Materials

journal homepage: www.elsevier.com/locate/jhazmat

Assessing aerobic biotransformation of 8:2 fluorotelomer alcohol in aqueous film-forming foam (AFFF)-impacted soils: Pathways and microbial community dynamics

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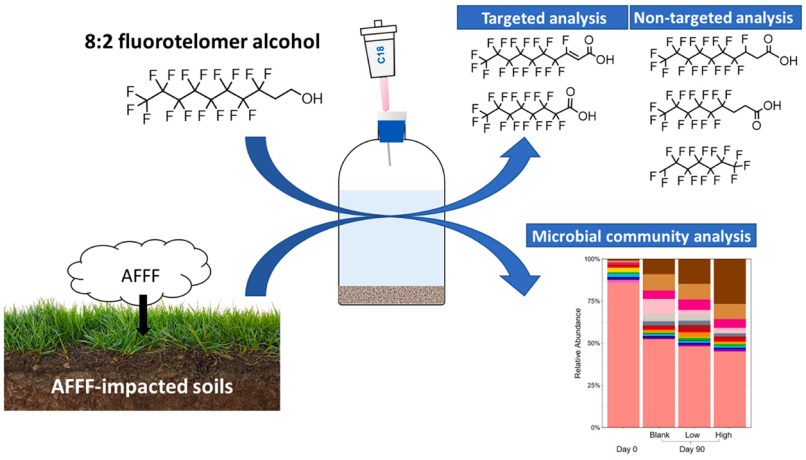
HIGHLIGHTS

- Aerobic 8:2 FTOH biotransformation was studied in AFFF-impacted soil microcosms.
- 8:2 FTOH can be transformed by soil microorganisms with a half-life of about 3 days.
- Perfluoroalkane-like compounds were identified as novel 8:2 FTOH transformation products.
- Species in genus *Sphingomonas* might be potential 8:2 FTOH degraders.

GRAPHICAL ABSTRACT



ARTICLE INFO

Keywords:
Aerobic PFAS biotransformation
8:2 FTOH
Microbial community analysis
Fluorotelomer alcohols
AFFF

ABSTRACT

Production of 8:2 fluorotelomer alcohol (8:2 FTOH) for industrial and consumer products, including aqueous film-forming foams (AFFFs) used for firefighting, has resulted in its widespread occurrence in the environment. However, the fate of 8:2 FTOH at AFFF-impacted sites remains largely unknown. Using AFFF-impacted soils from two United States Air Force Bases, microcosm experiments evaluated the aerobic biotransformation of 8:2 FTOH (extent and byproduct formation) and the dose-response on microbial communities due to 8:2 FTOH exposure. Despite different microbial communities, rapid transformation of 8:2 FTOH was observed during a 90-day incubation in the two soils, and 7:2 secondary fluorotelomer alcohol (7:2 sFTOH) and perfluorooctanoic acid (PFOA) were detected as major transformation products. Novel transformation products, including perfluoroalkane-like compounds (1H-perfluoroheptane, 1H-perfluorohexane, and perfluoroheptanal) were

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https://doi.org/10.1016/j.jhazmat.2022.130629
Received 30 September 2022; Received in revised form 12 December 2022; Accepted 16 December 2022
Available online 19 December 2022

identified by liquid chromatography-high resolution mass spectrometry (LC-HRMS) and used to develop aerobic 8:2 FTOH biotransformation pathways. Microbial community analysis suggests that species from genus *Sphingomonas* are potential 8:2 FTOH degraders based on increased abundance in both soils after exposure, and the genus *Afipia* may be more tolerant to and/or involved in the transformation of 8:2 FTOH at elevated concentrations. These findings demonstrate the potential role of biological processes on PFAS fate at AFFF-impacted sites through fluorotelomer biotransformation.

1. Introduction

Fluorotelomer alcohols [(n:2) FTOHs, $F(CF_2)_nCH_2CH_2OH$] are ubiquitous in the environment and are associated with a variety of adverse health and environmental impacts. FTOHs are a group of polyfluorinated compounds used in a wide range of industrial and consumer products, including waterproofing agents, polishes, lubricants, paints, paper, textiles, non-stick cookware, and firefighting agents [1,2]. As a result, FTOH contamination is present in many environmental matrices, including the atmosphere [3], indoor air [4], wastewater treatment plant (WWTP) effluent and sludge [5,6], and sludge-applied soils [7]. Adverse health effects resulting from exposure to FTOHs and associated transformation products, such as perfluoroalkyl carboxylic acids (PFCAs), have been documented in animal models [8,9] and human cells [10].

Among FTOHs of varying chain lengths, 8:2 FTOH [$F(CF_2)_8CH_2CH_2OH$] has received special attention because it is the dominant homologue in the synthesis of modern fluorotelomer-based products [11,12]. Also, 8:2 FTOH has been detected as an active ingredient (average concentrations ranging from 8 to 26.5 mg/L) in some aqueous film-forming foam (AFFF) formulations, which have been widely used to extinguish hydrocarbon-fuel fires [1,13]. Furthermore, intermediates in the biotransformation of FTOHs to PFCAs, such as saturated and unsaturated fluorotelomer carboxylic acids (FTCAs), are more toxic than the PFCAs to algae [14] and aquatic animals [15,16]. Given the potential for the release of 8:2 FTOH-based products into the environment and the toxicity of 8:2 FTOH and its biotransformation products, it is critical to understand 8:2 FTOH biotransformation pathways and mechanisms for assessing its environmental fate.

Microbial transformation of 8:2 FTOH has been investigated in different environmental matrices, including pristine soils, WWTP activated sludge and digester sludge, as well as landfill leachate sediment under aerobic [17-22] and anaerobic conditions [23,24]. These studies in various environmental matrices documented different 8:2 FTOH biotransformation products. For example, 7:2 ketone [$F(CF_2)_7C(O)CH_3$] was found in an 8:2 FTOH transformation experiment using a *Pseudomonas* culture and native microbial communities in pristine soil, while 7:3 U amide [$F(CF_2)_7CH{=}CHCONH_2$] was only documented in activated sludge [25,20,21]. These findings indicate that distinct microbial populations may lead to different 8:2 FTOH biotransformation pathways.

Due to the extensive and repeated application of AFFF at military, civilian aviation, and firefighting facilities, it is particularly relevant to understand 8:2 FTOH biotransformation in AFFF-impacted soils. However, no previous studies have investigated aerobic biotransformation of 8:2 FTOH in AFFF-impacted soil and the associated microbial communities. It is necessary to fill these research gaps in order to improve our understanding of the fate of 8:2 FTOH in the environment and facilitate the development of innovative microbial-based remediation strategies.

Therefore, this study aimed to (a) investigate 8:2 FTOH biotransformation (byproducts and pathways) under aerobic conditions in two AFFF-impacted soils, and (b) evaluate the composition shift of two soil microbial communities during 8:2 FTOH biotransformation. To achieve this, 90-day microcosm experiments using two geographically distinct soils were carried out. Targeted and non-targeted mass spectrometry were employed to identify the possible formation of previously documented 8:2 FTOH biotransformation products and unknown products, respectively. Both novel and previously identified transformation products were used to propose 8:2 FTOH transformation pathways under aerobic conditions. Illumina high-throughput sequencing was utilized to assess the response of soil microbial communities during 8:2 FTOH biotransformation.

2. Materials and methods

2.1. AFFF-impacted soils

Two soils were used in this study: one was collected from an AFFF spray test area with a hand auger down to 1.8 m at Robins Air Force Base (AFB) (Houston County, GA); and the other was collected by shovel proximate to a crash site and fire station at the former Loring AFB (Aroostook County, ME). Detailed information on physical and chemical properties of these two soils (moisture content, organic matter content, pH, cation exchange capacity (CEC), and particle size) are provided in the Supplementary Information (SI)-Section S1. Both locations were impacted by PFAS from historical application(s) of AFFF. Prior to microcosm preparation, the soils were homogenized, ground with a mortar and pestle, passed through a 2 mm sieve (ASTM E11 # 10 size), and stored at 4 °C.

2.2. Robins biotransformation experiment

The Robins biotransformation experiment was conducted in Wheaton glass serum bottles (160-mL) containing 100 mL of 30 mM bicarbonate-buffered, sterile mineral salts medium and capped with aluminum crimp-sealed butyl rubber septa (Chemglass; Vineland, NJ). Mineral medium was prepared according to the recipe by Löffler et al. [26] with modifications to allow for aerobic microbial processes. Specifically, resazurin and L-cysteine were omitted due to the potential to serve as carbon sources, while L-cysteine and sodium sulfide were eliminated since highly reduced medium was not required for microbial growth under aerobic conditions. Microcosms were prepared by adding approximately ten grams (dry weight) of damp Robins AFB soil into each bottle as the microbial inocula. Each bottle of live treatment was then dosed with 85 μL of a 200 mg/L 8:2 FTOH stock solution prepared in diethylene glycol butyl ether (DGBE) to yield an initial concentration of approximately 170 μg/L (see Table 1 for experiment set-up). DGBE is the primary organic solvent in AFFF formulations and was used as the carbon source and electron donor in the microcosms [27].

A set of abiotic controls was prepared by amending the medium with 1 g/L sodium azide (determined through a preliminary experiment detailed in SI-Section S2) to inhibit microbial activity, while the other aspects of the preparation procedures were identical to the live treatment (Table 1). The abiotic control was used to assess the potential abiotic transformation of 8:2 FTOH and other PFAS in the Robins AFB soil and to evaluate other potential losses. A 18 G× 1″ needle was connected to a pre-conditioned C18 SPE cartridge (Maxi-Clean™, Alltech, Deerfield, IL) and inserted into the headspace of each live treatment and abiotic control. The cartridge ensured aeration in the microcosms, while capturing 8:2 FTOH and other volatile transformation products during incubation. Additionally, live controls containing sterile mineral salts medium, Robins AFB soil, and 85 μL of DGBE were prepared to evaluate the background levels of PFAS in the soil and to monitor their potential biotransformation products (Table 1). In live control bottles where 8:2 FTOH was not added, a 0.22 μm sterile

polyethersulfone (PES) syringe filter was placed on top of the needle instead of the C18 cartridge to maintain aeration. Live treatments were run in triplicate and all controls were run in duplicate. All the bottles were shaken at 150 rpm in a 25 °C incubator (Model I 24, New Brunswick, NJ) for 90 days.

Samples from each phase (aqueous phase, solid phase, and headspace) were collected using methods described in Section 2.4 on day 0, 3, 7, 14, 28, 56, and 90 for targeted PFAS analysis. Day 0 and day 90 samples were also pooled from each phase for non-targeted PFAS analysis (described in Section 2.5). Additionally, slurry samples from the live treatments and live controls were collected according to methods in Section 2.6 at day 7 and day 90 for DNA extraction and microbial community analysis. Day 7 was used rather than day 0 because the native microbial community from Robins AFB soil (day 0) contained very low microbial biomass (< 1 ng/g) and could not provide enough DNA for quantification and subsequent sequencing (detailed information on DNA yields of Robins AFB soil is provided in SI-Section S3).

2.3. Loring biotransformation experiment

Since the DNA yield from Robins AFB soil was less than typical natural soils (at μg/g to mg/g level), additional evaluations of the soil microbial community dynamics during 8:2 FTOH biotransformation were investigated using Loring AFB soil, which contains a more abundant microbial quantity (detailed in SI-Section S2). The microcosms in the Loring biotransformation experiment were prepared similarly to the microcosms in the Robins biotransformation experiment as described in Section 2.2. To impose extreme pressure on the soil microbial community, a high-dose treatment was set up in the Loring biotransformation experiment in addition to the low-dose treatment which was same as the live treatment in the Robins biotransformation experiment, and live control (See Table 1). In the high-dose treatment, 1,700 μg/L of 8:2 FTOH was amended every two weeks. During the 90-day incubation, additional DGBE was supplemented as a carbon source in the live controls and low-dose treatments as needed based on total organic carbon (TOC) measurements every two weeks. Samples from the aqueous phase, solid phase, headspace, as well as slurry samples were collected on day 0 and 90 for targeted and non-targeted PFAS analysis and microbial community analysis using the method described in Section 2.5 and Section 2.6, respectively.

2.4. Sampling and PFAS extraction

At each sampling point, the headspace of the live treatments and abiotic controls was sampled by flushing with approximately 1.5 L/min sterile air through the C18 cartridge for 5 min to capture 8:2 FTOH and other volatile compounds. The C18 cartridges were subsequently removed and eluted with 5 mL methanol. A new pre-conditioned cartridge was then installed after aqueous and solid phase samples were collected. Approximately 2 mL of well-mixed slurry was withdrawn by a 3-mL disposable polypropylene syringe (BD; Franklin Lakes, NJ) from each bottle into a 2-mL microcentrifuge tube, and then centrifuged at 21,100g for 10 min.

Five hundred microliters of supernatant were immediately diluted in 9.5 mL methanol and filtered through a 0.2 μm nylon filter (Corning, Inc., Corning, NY) for quantification of PFAS in the aqueous phase. Three hundred microliters of supernatant were diluted in 8 mL ultrapure water for total organic carbon (TOC) analysis to estimate the consumption of DGBE on a Shimadzu TOC-L CPH analyzer (Shimadzu, Kyoto, Japan). The soil pellet that remained in the centrifuge tube was resuspended with 1.5 mL methanol, vortexed for 20 min, ultra-sonicated at 60 °C in a water bath for 20 min, followed by centrifugation at 21,100g for 10 min. One milliliter of the soil extract subsequently was diluted with 9 mL methanol and filtered through a 0.2 μm nylon filter for solid phase PFAS quantification. All headspace eluents, methanol-diluted aqueous phase samples and methanol-extracted solid phase samples were stored at −20 °C until PFAS analysis.

2.5. Targeted analysis by LC-MS/MS and non-targeted analysis by LC-HRMS

Parent 8:2 FTOH and target biotransformation products collected from the C18 cartridge eluent, methanol-diluted aqueous phase samples, and methanol-extracted solid phase samples were detected and quantified by a Waters ACQUITY ultra high-performance liquid chromatograph coupled with a Waters Xevo triple quadrupole mass spectrometer (LC-MS/MS) (Waters Corporation, Milford, MA). Analytical standards, methods and instrumental parameters are provided in SI-Sections S4–5, Table S1 and S2. The detection limits of target biotransformation products are shown in Table S3.

To identify potential novel 8:2 FTOH biotransformation products generated in both microcosm experiments, non-targeted PFAS analysis was performed using a Thermo Q Exactive HF-X Orbitrap liquid chromatography-high resolution mass spectrometry (LC-HRMS) system. Prior to the analysis, the C18 cartridge eluent, methanol-diluted aqueous phase samples, and methanol-extracted solid phase samples collected at the beginning and end of the experiment (day 0 and day 90) were pooled for each treatment and control. All pooled samples were injected three times, which facilitated filtering out the features that were not reproducible across replicate injections. Detailed information on instrumental settings and procedures of data processing are presented in SI-Section S6.

Table 1
Experimental setup examining aerobic biotransformation of 8:2 FTOH and soil microbial community dynamics during 8:2 FTOH biotransformation.

Experiment	Treatment	Components			Analyses performed	
		Soil type	Spiked 8:2 FTOH (μg/L) at day 0	Amended NaN_3		
Robins biotransformation experiment	Live treatment	Robins	170	N/A	PFAS analysis[a]	Microbial community analysis[b]
	Abiotic control	Robins	170	Yes[c]		
	Live control	Robins	N/A	N/A		
Loring biotransformation experiment	High-dose treatment	Loring	1,700[d]	N/A	PFAS analysis[e]	Microbial community analysis[f]
	Low-dose treatment	Loring	170	N/A		
	Live control	Loring	N/A	N/A		

[a] Samples of day 0, 3, 7, 14, 28, 56, 90 were analyzed using LC-MS/MS for targeted PFAS quantification; samples of day 0, 28, and 90 were also analyzed using LC-HRMS for non-targeted PFAS analysis.
[b] Biomass samples from day 7 and 90 were analyzed using 16S rRNA gene amplicon sequencing.
[c] 1 g/L NaN_3.
[d] Amended with additional 1,700 μg/L 8:2 FTOH at day 14, 28, 42, 67, and 82
[e] Samples of day 0 and 90 were analyzed for both targeted and non-targeted PFAS analysis.
[f] Biomass samples from day 0 and 90 were analyzed using 16S rRNA gene amplicon sequencing.

2.6. DNA extraction and microbial community analysis

Biomass samples were collected by mixing the medium and soil in the microcosm thoroughly, followed by transferring to a 2-mL centrifuge tube for centrifugation at 21,100g for 10 min. The supernatant was then removed, and the remaining pellets were stored at −80 °C until DNA extraction. DNeasy PowerSoil Kit (Qiagen, Hilden, Germany) was used for DNA extraction according to the manufacturer's protocols. The DNA samples extracted from replicate cultures were pooled and stored at −20 °C prior to sequencing. Amplification and sequencing of microcosm DNA samples were performed at the Alkek Center for Metagenomics and Microbiome Research at the Baylor College of Medicine. Briefly, the V4 region of the 16S rRNA gene was amplified by PCR using barcoded primer sets (515 F/806 R) and sequenced on the MiSeq platform (Illumina, San Diego, CA) using a 2 × 250 bp paired-end protocol [28,29]. The produced read pairs were demultiplexed, filtered and merged using parameters optimized form the 16Sv4 amplicon type [30,31]. Resulting reads were denoised using the Deblur algorithm following the default workflow and the length limit of 252 bp. The generated sequences were mapped against the latest SILVA Database [32]. ATIMA (Agile Toolkit for Incisive Microbial Analyses) was used to analyze and visualize trends in taxa abundance, alpha diversity, and beta diversity. Additionally, STAMP (Statistical Analysis of Metagenomics Profiles) software (version 2.1.3), was used to evaluate the significant differences in relative abundance at different taxonomic levels [33]. The statistical significance between samples was tested using the two-sided Fisher's exact test with the Benjamini-Hochberg False Discovery Rate procedure.

3. Results and discussion

In this study, two sets of microcosm experiments were conducted to study the aerobic biotransformation products of 8:2 FTOH after 90 days of incubation in Robins and Loring AFB soils. The microbial community composition changes in these two soils after 90 days were also evaluated. Aerobic 8:2 FTOH biotransformation pathways were proposed based on the biotransformation products identified in this study and the pathways proposed in previous studies on 8:2 FTOH biotransformation [25,23,34,20,21,24]. In addition, the 8:2 FTOH biotransformation kinetics in Robins AFB soil was determined. To reflect a worst-case scenario, the microbial community shift in Loring AFB soil during biotransformation of 8:2 FTOH with a high concentration was also investigated.

3.1. Biotransformation of 8:2 FTOH in Robins AFB soil

3.1.1. Parent compound biotransformation and kinetics

Throughout the 90-day Robins biotransformation experiment, 85.0–127.2 mol% of initially spiked 8:2 FTOH was measured in the abiotic controls without significant changes ($p > 0.05$) (Fig. 1a). The absence of microbial activity in the abiotic controls was confirmed through organic carbon measurements that did not change significantly ($C/C_0 = 106.5 \pm 6.0\%$, $p > 0.05$) (Fig. S1). In contrast, decreasing organic carbon in the live treatments and live controls by approximate 90% within 2–3 weeks after each addition of DGBE was indicative of microbial activity in these bottles (Fig. S1). These results confirm the integrity of the experimental systems and the abiotic stability of 8:2 FTOH under the experimental conditions, consistent with prior studies [17,19,20].

In the live treatment, 8:2 FTOH was transformed rapidly, with 7.0 mol% of 8:2 FTOH remaining in the systems after 90 days (Fig. 1a). The 8:2 FTOH biotransformation in Robins AFB soil was fitted with first-order exponential and first-order double exponential decay models (see Tables S4 and 2, respectively). First-order exponential models are frequently used to describe biodegradation kinetics and has been used to depict the biotransformation of 8:2 FTOH under nitrate-reducing conditions [35] and for the biotransformation of other PFAS [36]. However, it was found in this study that a first-order double exponential decay model better fit the data (R^2 of 0.995 vs 0.969); the associated fitted curve is presented in Fig. S2. It was hypothesized that biotransformation of 8:2 FTOH occurs following a two-step mechanism. This assertion is based on previously reported studies examining anaerobic biodegradation of 8:2 FTOH in activated sludge [23] and degradation of other organic compounds in natural environments [37,38]. As shown in Eq. (1), the molar percentage of 8:2 FTOH remaining in the system (M) at time t is described mathematically as the sum of two first-order kinetic expressions, where M_1 and M_2 are constants representing molar percentages of the initial mass, associated with two differing transformation

Table 2
Fitting parameters of the first-order double exponential decay model.

Parameter	Value	Standard Error	Statistics	
M_1	92.634	5.042	R^2	0.995
M_2	8.370	4.146	Adjusted R^2	0.991
k_1	0.228	0.028		
k_2	0.002	0.009		

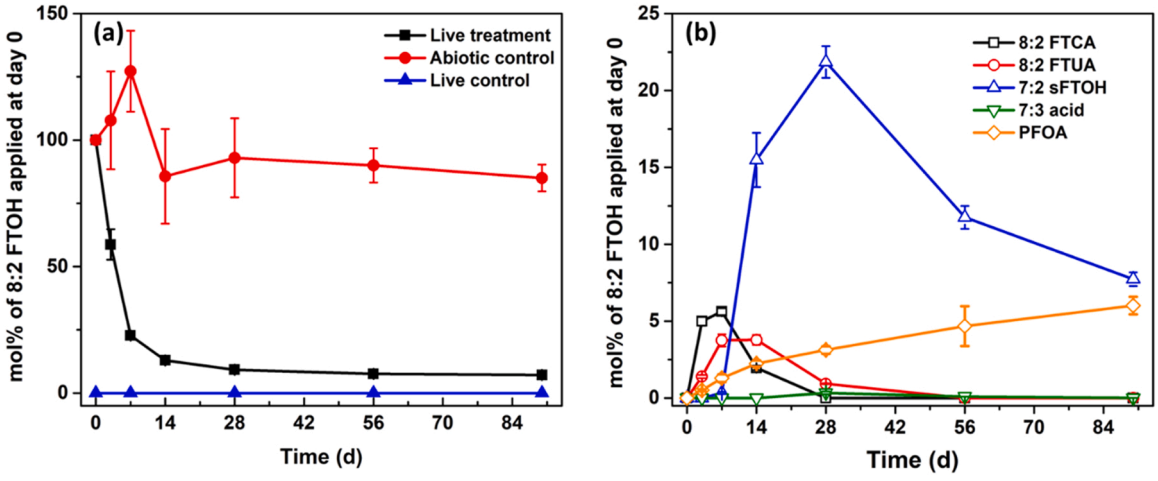

Fig. 1. Profiles of 8:2 FTOH and its biotransformation products from the Robins biotransformation experiment during 90-day incubation: (a) residual molar percentage of 8:2 FTOH in the live treatment, abiotic control, and live control; error bars represent one standard deviation of triplicate live treatments and duplicate abiotic/live controls; (b) molar yields of quantified biotransformation products in the live treatment; error bars represent one standard deviation of triplicate live treatments.

rate constants of k_1 and k_2, respectively.

$$M(t) = M_1 e^{-k_1 t} + M_2 e^{-k_2 t} \quad (1)$$

According to the model fits, 92.6 mol% of the initially spiked 8:2 FTOH was rapidly biotransformed with a half-life of about 3 days (first step), while transformation of the remaining 8.4 mol% 8:2 FTOH was much slower, with a half-life of more than 400 days (second step). The rapid biotransformation of the larger 8:2 FTOH fraction in the first step may be attributed to easier access to the compound by microorganisms in the aqueous phase. The lower biotransformation rate associated with the smaller fraction in the second step may be due to the limited bioavailability of 8:2 FTOH that resulted from kinetic limitations associated with desorption of 8:2 FTOH from the soil matrix or microbial biomass [21,39]. The overall half-life of 8:2 FTOH in the present study (~3 days) was similar to the findings for 8:2 FTOH biotransformation in aerobic soils (~5 days) [21], and activated sludge (< 5 days) [20,23], but considerably shorter than the half-life reported in aerobic landfill leachate-sediment (> 1 year) [17], water-saturated Appling soil (~210 days) [40], and anaerobic digestion sludge (~145 days) [24]. The rapid 8:2 FTOH biotransformation observed herein is likely attributed to the prior existence of effective enzymes in the Robins AFB soil microbial community, which were either innate or evolved during long term exposure to PFAS in the field.

3.1.2. Quantified biotransformation products by LC-MS/MS

Four polyfluorinated compounds, 8:2 FTCA [$F(CF_2)_8CH_2COOH$], 8:2 FTUA [$F(CF_2)_7CF{=}CHCOOH$], 7:2 sFTOH [$F(CF_2)_7CHOHCH_3$], and 7:3 acid [$F(CF_2)_7CH_2CH_2COOH$], were detected in the live treatment over the 90-day incubation period, while none of these compounds were detected in the control bottles. These data provide further evidence that aerobic biotransformation of 8:2 FTOH occurred in the live treatment.

Among the four polyfluorinated transformation products, 8:2 FTCA and 8:2 FTUA have been widely documented as major polyfluorinated metabolites [41,17,25,23,19,20,22,24]. During the first 14 days, the yields of 8:2 FTCA and 8:2 FTUA increased concurrently with the consumption of 8:2 FTOH and reached a peak of 5.6% and 3.8% at day 7 and 14, respectively (Fig. 1b). The lower yields of 8:2 FTCA and 8:2 FTUA observed in this study compared to those in previous 8:2 FTOH biotransformation studies [41,25,20,22] were likely due to their rapid conversion to downstream transformation products.

Although 7:2 sFTOH was the most abundant polyfluorinated biotransformation product after 14 days, it was not detected until day 7 (Fig. 1b). As shown in Fig. 1b, the molar yield of 7:2 sFTOH reached a peak at 21.9% of initial 8:2 FTOH at day 28 and then gradually decreased to 7.7% at the end of the experiment. A similar trend of 7:2 sFTOH appearance and disappearance was observed during the aerobic biotransformation of 8:2 FTOH in forest and agricultural soils, except that the peak concentration was reached faster (2–7 days) [21]. Similarly, 7:2 sFTOH was found to be the most prevalent product from 8:2 FTOH aerobic biotransformation in a mixed bacterial culture [20]. Furthermore, 7:2 sFTOH was one of the major 8:2 FTOH metabolites produced by native soil microbes [19] and by pure *Pseudomonas* strains under aerobic conditions [25].

Although previously reported as one of the major biotransformation products in aerobic soils [21] and landfill leachate-sediment [17], 7:3 acid was the least abundant polyfluorinated metabolite in the present study. The molar yield of 7:3 acid at day 28 was only 0.3% of the initial 8:2 FTOH and declined thereafter (Fig. 1b). One possible explanation for the low yield of 7:3 acid in this study is its rapid β-oxidation and further transformation to PFOA [23,20,22]. However, some studies have presented differing observations of 7:3 acid stability. For example, β-oxidation of 7:3 acid to PFOA was not observed in forest and agricultural soils [21]. It was also found that 7:3 acid could accumulate over periods of 90 days to 7 months in soils, digested sludge, and landfill leachate sediment [17,21,24].

In addition to polyfluorinated transformation products, PFOA has been widely reported as a major stable 8:2 FTOH transformation product [41,17,25,19-22,42]. Consistent with prior studies, PFOA was detected as an 8:2 FTOH biotransformation product in live treatments. The molar yield of PFOA was calculated by subtracting the background concentration in the live control samples from the live treatment samples (Fig. 1b), as biotransformation of both 8:2 FTOH and other unknown PFOA precursors could have contributed to the formation of PFOA. During the 90-day incubation, PFOA was detected at day 3 and increased continuously to 6.0% of the initial 8:2 FTOH concentration without reaching a steady-state concentration. Perfluoropentanoic acid (PFPeA), perfluorohexanoic acid (PFHxA), perfluoroheptanoic acid (PFHpA), perfluorononanoic acid (PFNA), and perfluorodecanoic acid (PFDA) were also detected throughout the experiment period in the live treatment, and abiotic and live control groups. The masses of PFPeA, PFHpA, PFNA, and PFDA remained relatively steady at low levels (<1 nmole) and did not change significantly ($p > 0.05$) over time (Fig. S3). Although the mass of PFHxA was measured at slightly over 2 nmole from the initial time point, no significant ($p > 0.05$) increase of PFHxA mass was observed (Fig. S3). These results are not consistent with prior studies that found PFCAs, including perfluorobutanoic acid (PFBA) [23,42], PFPeA [23,42], PFHxA [17,25,23,19-22,42,24], PFHpA [17,23,19,42], and PFNA [42], were formed during 8:2 FTOH biotransformation.

A possible explanation for the observed differences in formation of biotransformation products, including poly- and perfluorinated compounds, between this and earlier studies could be related to differences in microbial communities associated with different environmental matrices. Note that not all PFCAs (C4-C9) that have been identified in earlier studies were identified as biotransformation products in this study. It is possible that those PFCAs were produced as 8:2 FTOH biotransformation products in trace amounts but were masked by background levels of PFAS contaminants in the AFFF-impacted soils.

3.1.3. Mass balance

The mass balance was expressed as the % ratio of molar mass recovered at certain timepoints to the total molar mass of initially spiked 8:2 FTOH. The mass balance by day 90 reached 58.5 mol% (Fig. S5, Table S6), which is consistent with previous studies (~40–65%) [41,17,25,23]. There are several possible causes for the incomplete mass balance. First, irreversible sorption to microcosm solids could reduce the extraction efficiencies of 8:2 FTOH and its biotransformation products. Previous studies revealed that up to 35% of 8:2 FTOH was irreversibly bound to soils [21]. Also, some 8:2 FTOH transformation products (e.g., 8:2 FTUA) can covalently bond with biological macromolecules, hindering their extraction and quantification [41,25,43]. Second, mass loss could result from mineralization of β- and γ-carbon of 8:2 FTOH [20,22], masked PFCA generation by background contamination levels, and the potential loss of volatile biotransformation products during sampling. Third, additional unknown products formed during the biotransformation of 8:2 FTOH could account for some mass loss. Therefore, non-targeted PFAS analysis was performed to identify those unknown products, as discussed in Section 3.3.1.

3.2. Biotransformation of 8:2 FTOH in Loring AFB soil

Native microorganisms in the Loring AFB soil also showed effective transformation of 8:2 FTOH, including the treatments with a higher starting concentration of 8:2 FTOH (1,700 μg/L). In the Loring biotransformation experiment, 5.8–10.1 mol% of spiked 8:2 FTOH remained in the system after 90 days, which is comparable to the result in the Robins AFB soil (7.2 mol% of 8:2 FTOH remaining after 90 days).

The polyfluorinated compounds quantified at the end of the Loring biotransformation experiment are shown in Table S5. Similar to the Robins biotransformation experiment, in the low-dose treatment in the Loring biotransformation experiment, the concentrations of 8:2 FTCA and 8:2 FTUA at day 90 were lower than the limits of detection; the two

most abundant transformation products were 7:2 sFTOH and 7:3 acid with yields of 33.0 mol% and 3.0 mol%, respectively. In the high-dose treatment at day 90, 7:2 sFTOH was also the biotransformation product of measurable abundance (6.9 mol%), while the concentration of 7:3 acid was below the detection limit. In contrast to the low-dose treatment, 8:2 FTCA and 8:2 FTUA were still detected at day 90 with yields of 0.2 mol% and 1.0 mol%, respectively, in the high-dose treatment. While C4-C10 PFCAs were detected in both low-dose and high-dose treatments, the masses of these PFCAs were significantly higher ($p < 0.05$) in samples from the high-dose treatment at day 90 compared to those from the live control. However, significantly higher mass ($p < 0.05$) of only PFHpA and PFOA were detected in the low-dose treatment (Fig. S4). The total mass recovery by day 90 was 44.7% and 30.3 % in the Loring low-dose treatment and high-dose treatment, respectively, which are lower than that of the 8:2 FTOH biotransformation experiment with Robins AFB soil (58.5%).

3.3. Novel 8:2 FTOH biotransformation products and proposed pathways

3.3.1. Potential novel transformation products identified by non-targeted analysis

As shown in Table S7, ten candidate 8:2 FTOH transformation products were identified and assigned confidence levels 1–4, with 4 indicating the least likely and 1 being the most likely [44]. The change in peak area counts over time for the candidate products are plotted in Figs. S6-S8.

Three of the ten candidates were classified as level 4 (unequivocal molecular formula) due to the lack of MS^2 spectrum data. However, according to their exact mass ($m/z = 438.9822$, $m/z = 356.9785$, and $m/z = 391.0009$) and customized suspect mass list based on prior literature [45], 7:3 U amide, 6:2 FTUA [$F(CF_2)_5CF{=}CHCOOH$], and 6:3 acid [$F(CF_2)_6CH_2CH_2COOH$]) were proposed as the potential transformation products, respectively. Previously, 7:3 U amide was determined as an 8:2 FTOH biotransformation product in activated sludge, although the identification was not confirmed due to the lack of an authentic standard [20]. Neither 6:2 FTUA nor 6:3 acid has been previously reported as 8:2 FTOH transformation products. Longer chain fluorotelomer carboxylic acids have been documented as precursors of shorter chain fluorotelomer carboxylic acids. For example, 5:3 acid [$F(CF_2)_5CH_2CH_2COOH$] could produce 4:3 acid [$F(CF_2)_4CH_2CH_2COOH$] and 3:3 acid [$F(CF_2)_3CH_2CH_2COOH$] with 4:2 FTUA [$F(CF_2)_3CF{=}CHCOOH$] as an intermediate through the one-carbon removal pathway [34]. Similarly, it is possible that 6:2 FTUA and 6:3 acid could be generated from the biotransformation of 7:3 acid.

Five of the ten candidates were assigned as level 3 (tentative candidates). The structures of level 3 compounds were based on an exact mass, MS^2 spectrum data, and matching to MetFrag simulations [46]. Based on the results of suspect screening, compounds of $m/z = 438.9822$ and $m/z = 376.9691$ were proposed to be 7:3 U acid [$F(CF_2)_7CH{=}CHCOOH$] and 6:2 FTCA [$F(CF_2)_6CH_2COOH$], respectively. Although not quantified due to lack of analytical standard in this study, 7:3 U acid has been identified as an 8:2 FTOH biotransformation product in prior studies and reported as a precursor to 7:3 acid and other polyfluorinated transformation products [17,25,23,20,24]. The formation of 6:2 FTCA in the present study could be attributed to the one-carbon removal pathway as an intermediate from 7:3 acid to 6:3 acid. The compound with $m/z = 458.9885$ was identified as 3-fluoro-7:3 acid [$F(CF_2)_7CHFCH_2COOH$]. As 3-fluoro-5:3 acid [$F(CF_2)_5CHFCH_2COOH$] was proposed as a 6:2 FTUA biotransformation product, a similar reaction could occur to 8:2 FTUA, leading to the formation of 3-fluoro-7:3 acid by a reductase [24]. Compounds with $m/z = 346.9745$ and $m/z = 318.9797$ were identified as perfluoroalkane-like compounds, perfluoroheptanal [$F(CF_2)_6CH{=}O$] and 1H-perfluorohexane [$F(CF_2)_6H$], respectively. Both were identified as potential 8:2 FTOH transformation products for the first time during aerobic 8:2 FTOH biotransformation.

Additionally, one of the ten compounds, 3-OH-7:3 acid [$F(CF_2)_7CHOHCH_2COOH$] with $m/z = 456.9930$ was assigned as level 2 (probable structure) based on MS^2 fragmentation. It is hypothesized that 3-OH-7:3 acid is a 8:2 FTOH biotransformation product in soils, generating from 7:3 U acid [21]. No reference standards were available for the nine transformation product candidates with confidence levels 2–4 discussed above. Only one of the ten compounds ($m/z = 368.9764$) was classified as level 1, 1H-perfluoroheptane [$F(CF_2)_7H$]). Reference standards were available for 1H-perfluoroheptane and used to confirm the identification (Fig. S9).

All ten candidate products were detected in samples from the high-dose treatment in the Loring biotransformation experiment, while only three of the ten were detected in samples from the low-dose treatment in the Loring biotransformation experiment and four from the Robins biotransformation experiment (Table S7). The distinct transformation products could be associated with different metabolic pathways or due to lower yields in the low-dose experiments, making it difficult to detect in the non-targeted analysis or distinguish these compounds from the background levels.

3.3.2. Aerobic biotransformation pathways

The proposed 8:2 FTOH biotransformation pathway under aerobic conditions (Fig. 2) is based on biotransformation products detected by LC-MS/MS targeted analysis and by non-targeted HRMS in this study, and on pathways proposed in earlier publications [25,23,34,20,21,24]. First, 8:2 FTOH could be oxidized to 8:2 fluorotelomer aldehyde (8:2 FTAL) [$F(CF_2)_8CH_2CHO$] by alcohol dehydrogenase(s), and further oxidized to 8:2 FTCA by aldehyde dehydrogenase(s). Although 8:2 FTAL was commonly recognized as an 8:2 FTOH biotransformation product in aerobic soil and activated sludge [20,21], it was not identified in this study, consistent with studies using aerobic sludge and digester sludge under anaerobic conditions [23,24]. It is believed that 8:2 FTAL is unstable and could be rapidly oxidized to 8:2 FTCA, which was detected in this study. Subject to hydrogen fluoride (HF) elimination, 8:2 FTCA could form 8:2 FTUA, which was also detected. Fluoride ion was measured in this study, although no distinguishable increase of fluoride was observed in live treatments from the background levels (discussion on fluoride quantification is detailed in SI-Sections S7–8). As speculated in a previous study, 8:2 FTUA could be a key branching point for subsequent biotransformation pathways [20].

There are three potential pathways following 8:2 FTUA formation, including conversion to (I) 3-fluoro-7:3 acid, (II) 7:2 sFTOH, and (III) 7:3 U acid. In branch (I), 8:2 FTUA may undergo a reduction reaction to form 3-fluoro-7:3 acid, which was identified by non-targeted analysis in this study. Branch (II) involves conversion of 8:2 FTUA to 7:2 sFTOH, which was quantified in this study. However, the intermediates between 8:2 FTUA and 7:2 sFTOH have been a controversial point discussed in prior studies [25,23,20,21]. For example, 7:2 sFTOH was first identified in 8:2 FTOH biotransformation using aerobic activated sludge where 8:1 olefin [$(CF_3(CF_2)_6CF{=}CH_2$] and 7:2 olefin [$(CF_3(CF_2)_6CH{=}CH_2$] were proposed as its precursors [20]. A study investigating anaerobic 8:2 FTOH biotransformation using activated sludge surmised that 7:2 olefin could be a precursor to 7:2 sFTOH, although 7:2 olefin may come from other intermediates (7:3 acid and 7:3 U acid) [23]. As 7:2 ketone rather than olefins was experimentally detected in previous studies using aerobic soil or pure *Pseudomonas* culture [21,25], 7:2 ketone was assumed as the precursor to 7:2 sFTOH here. Then, 7:2 sFTOH was further catalyzed to form PFCAs (C4-C8), possibly by monooxygenases via several oxidation reactions, although the mechanisms have not yet been identified [25,23,21].

Branch (III) leads to 7:3 U acid via reductive defluorination by a possible dehalogenase [24]. The reaction pathway after the formation of 7:3 U acid is split into four routes. Route (III-a) converts 7:3 U acid to 7:3 acid by unknown reductase(s) [24]. Route (III-b) and (III-c) produce 7:3 U amide and 3-OH-7:3 acid, respectively [20,21]. Route (III-d) enters a single carbon removal process to generate 6:3 acid, 6:2 FTCA, 6:2 FTUA as well as short-chain PFCAs (C4-C7) by a one-carbon removal

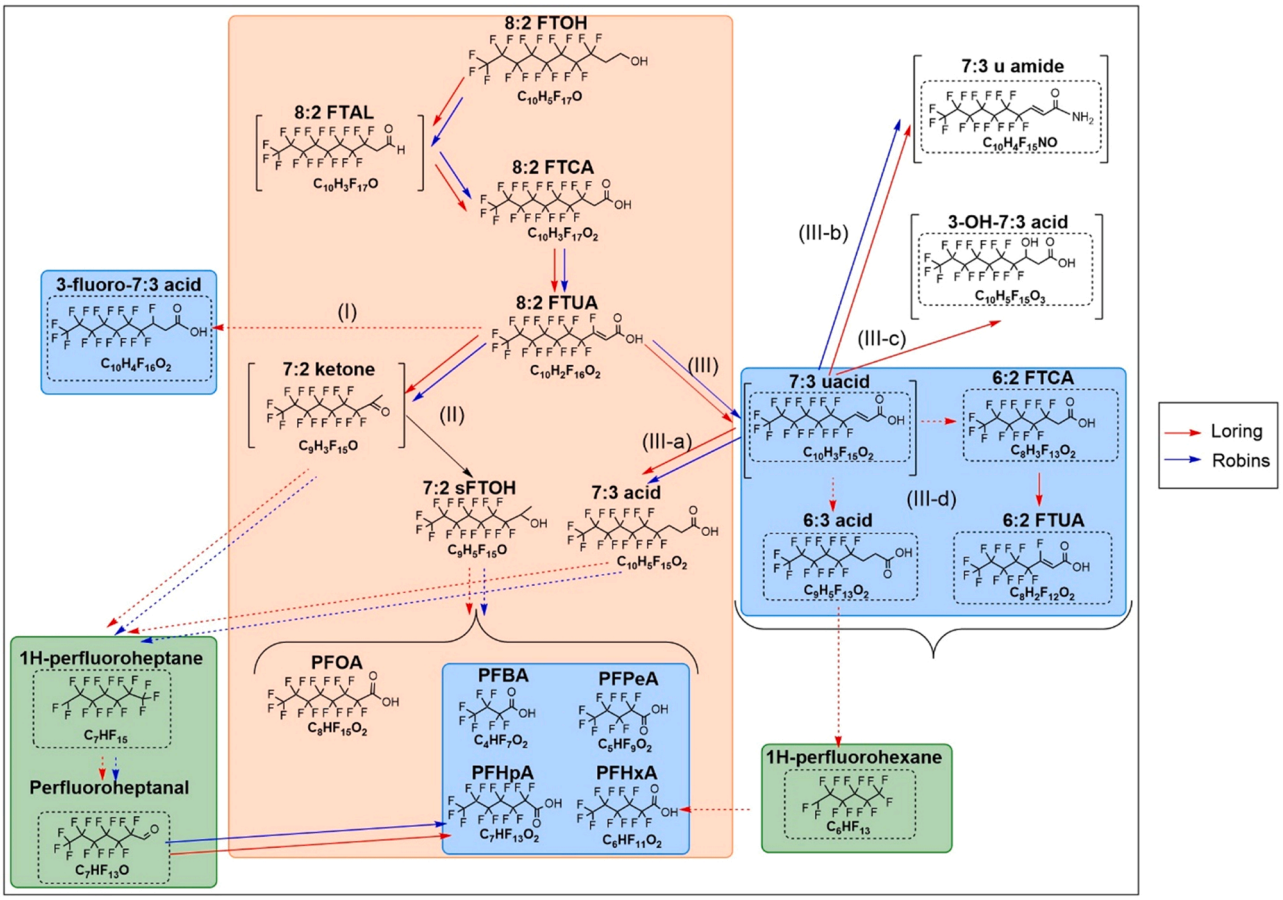

Fig. 2. Proposed aerobic biotransformation pathways of 8:2 FTOH based on this study and previous studies [25,23,34,20,21,24]. Red and blue arrows represent the pathways harbored by microbial communities in Loring and Robins AFB soils, respectively. Compounds in the orange box were directly quantified by LC-MS/MS in this study, except for those with brackets. Compounds in brackets were not detected by LC-MS/MS in this study but have been documented as 8:2 FTOH biotransformation products. Compounds in dashed boxes are tentative biotransformation products identified in non-targeted analysis by LC-HRMS in this study. Compounds/pathway with dashed arrows in blue boxes are proposed based on prior literature. Compounds/pathway in green boxes with dashed arrows are newly identified in this study.

process, which is proposed based on the pathways found in a study of 5:3 acid biotransformation [34]. Since 7:2 sFTOH and PFOA were the two major biotransformation products, and 7:3 acid and/or C4-C7 PFCAs were minor products (Fig. 1b, Table S5), the biotransformation pathway involving 7:2 sFTOH could be a major pathway that soil microbial communities in this study were able to utilize.

The newly identified perfluoroalkane-like compounds might be involved in a pathway after production of 7:2 ketone or 7:3 acid, where perfluoroheptanal is the intermediate between 1H-perfluoroheptane and PFHpA. Through a similar mechanism, 1H-perfluorohexane might be produced after 6:3 acid to generate PFHxA. No evidence has been documented that perfluoroalkane-like compounds are precursors to PFCAs. Additional experiments are needed to investigate the formation mechanisms of these novel transformation products.

3.4. Microbial community dynamics during aerobic biotransformation of 8:2 FTOH

3.4.1. Microbial community richness and diversity

The mapped reads of 16S rRNA gene amplicons obtained from the Robins and Loring biotransformation experiments varied in the range of 4,753 to 12,647 (Table S8). To compare the diversity and richness of the microbial communities between different samples, the number of reads was rarefied to an identical sequencing depth (4,753) and clustered into operational taxonomic units (OTUs) ranging from 51 to 312 at a 97% similarity level. Chao 1 and Shannon indices were calculated as microbial richness and diversity estimators, respectively (Table S8). The original Loring AFB soil had a higher Chao 1 index and numbers of OTUs, as well as higher Shannon indices than those of Robins AFB soil. This indicates that the original Loring AFB soil had both higher microbial richness and diversity than the Robins AFB soil. Principal coordinate analysis (PCoA) was also applied to visualize the microbial community composition alterteration across different treatments. Microcosms constructed with Loring AFB soil were clustered together, while all microcosms using Robins AFB soil were clustered together indicating that the microbial community composition was distinct between Loring AFB soil and Robins AFB soil (Fig. S11). Although both sites were highly contaminated with PFAS, the lower microbial richness and diversity in Robins AFB soil are possibly associated with its physical/chemical characteristics; for example, a lower organic carbon content ($< 0.5\%$) compared to Loring AFB soil (3.5%). In addition, based on the Shannon and Chao 1 indices (Table S8), different 8:2 FTOH doses in the Loring biotransformation experiment led to differences in microbial richness and diversity. Compared with the soil in live controls, soil used in the low-dose treatment had higher microbial richness and diversity, while soil used in the high-dose treatment had lower richness and diversity. This result suggests the possible toxicity of high concentrations of 8:2 FTOH and its biotransformation products and the microbial community shift towards specific microbial taxa during the biotransformation of higher 8:2 FTOH concentrations.

3.4.2. Microbial community composition shifts in the Robins biotransformation experiment

As shown in Fig. 3a, a total of fifteen phyla were identified in the Robins AFB soil microbial community. Proteobacteria occupied a high proportion (78.90%) in the initial Robins AFB soil after a 7-day incubation with a carbon source added. Other major phyla in the initial Robins AFB soil included Myxococcota (8.33%), Firmicutes (7.45%), Bacteroidota (2.88%), and Bdellovibrionota (1.41%). The phylum Proteobacteria dominated over the 90-day period in both the live treatment (69.09%) and live control (74.10%). The relative abundance of phyla Firmicutes, Acidobacteriota, Myxococcota, and Desulfobacterota were higher in the live treatment (16.20%, 2.74%, 1.03%, and 1.18%) than those in the live control (10.30%, 0.61%, 0.48%, and 0.42%).

Further analysis at the genus level revealed that obvious shifts towards an uncultured genus belonging to the family Symbiobacteraceae (phylum Firmicutes, 14.75% in the live treatment versus 4.02% in the live control) and the genus *Flavisolibacter* (phylum Bacteroidota, 7.41% in the live treatment versus 0.08% in the live control). The relative abundance of five genera including *Variovorax*, *Azospirillum*, *Ralstonia*, *Sphingomonas*, and *Ancylobacter* belonging to phylum Proteobacteria (2.42%−8.08%) in the live treatment, were much higher than those in the live control (0.17%−2.12%) after the 90-day incubation (Fig. 3b). Species in genera *Variovorax*, *Ralstonia*, *Sphingomonas*, and *Ancylobacter* have been previously found to be responsible for degradation of organic chlorinated compounds [47-50], indicating they may also be involved in 8:2 FTOH biotransformation.

3.4.3. Microbial community composition shifts in the Loring biotransformation experiment

Twenty-seven classified phyla in the domain Bacteria and three in domain Archaea were identified across all treatments over the 90-day Loring biotransformation experiment, though not all phyla were present in all groups. The top fifteen abundant taxa at the phylum level are depicted in Fig. 4a. In the original Loring AFB soil (day 0), the dominant bacterial phyla included Proteobacteria (35.01%), Bacteroidota (17.04%), Acidobacteriota (14.71%), Chloroflexi (11.66%), and Gemmatimonadota (3.79%). Generally, minor impacts of low doses of 8:2 FTOH on the microbial community composition in Loring AFB soil were observed after 90 days when compared to the live control. For example, the relative abundance of Proteobacteria and Bacteroidota, were 46.71% and 14.39%, respectively, in the low-dose treatment, which was comparable with the proportions of these phyla in the live control after incubation (44.94% and 14.22%). Conversely, in the high-dose group, the relative abundance of Proteobacteria was elevated to 56.07%. Proteobacteria, the most prominent phylum in the present study, was also identified by others as the most prominent phylum in surface soil with heavy PFAS pollution [51,52]. This increase in Proteobacteria abundance is also consistent with observations in river sediments dosed with 6:2 FTOH [53], and in wetland slurry spiked with 6:2 fluorotelmer sulfonate (FTS) [54]. Additionally, the proportions of Euryarchaeota, an archaeal phylum, as low as 0.19% in the original soil though, were enriched in the presence of 8:2 FTOH. The relative abundance of Euryarchaeota increased to 0.74% in the low-dose treatment and 1.24% in the high-dose treatment. These observations suggest the potential tolerance of Proteobacteria and Euryarchaeota to specific PFAS,

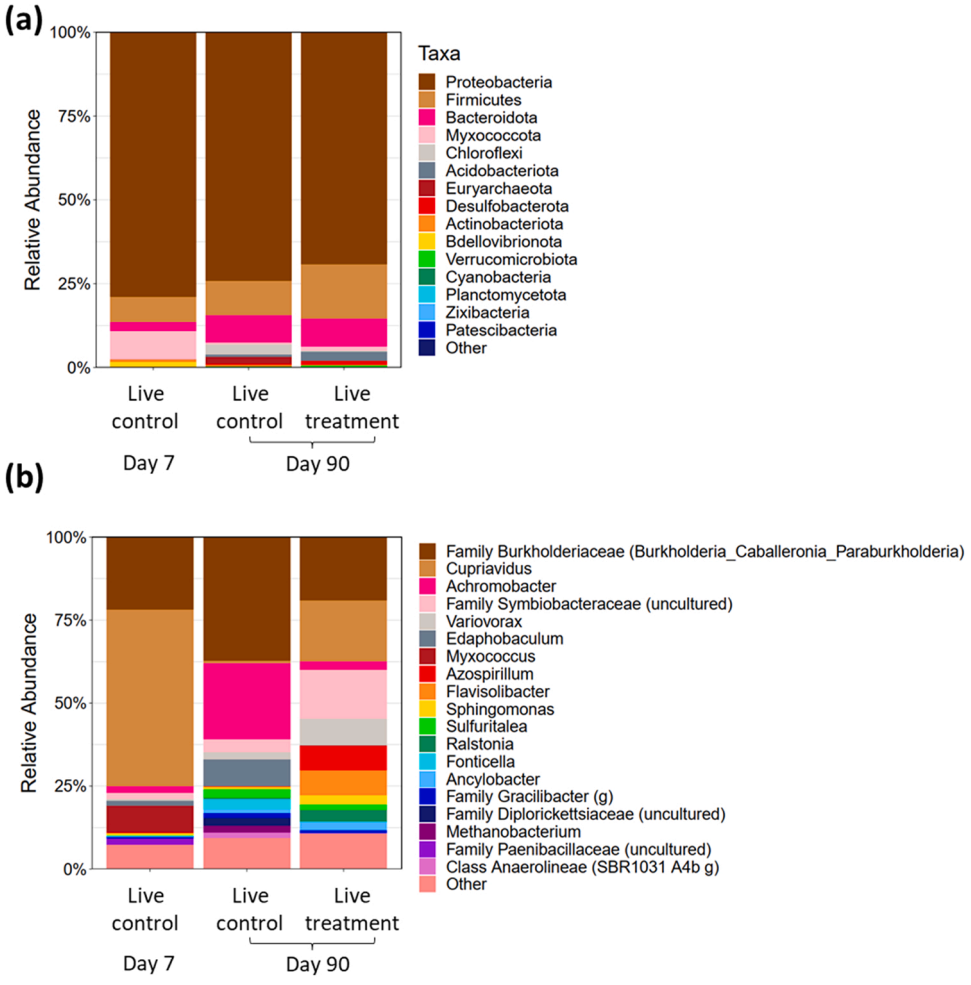

Fig. 3. Relative abundance of microbial community at the (a) phylum level and (b) genus level in the Robins biotransformation experiment.

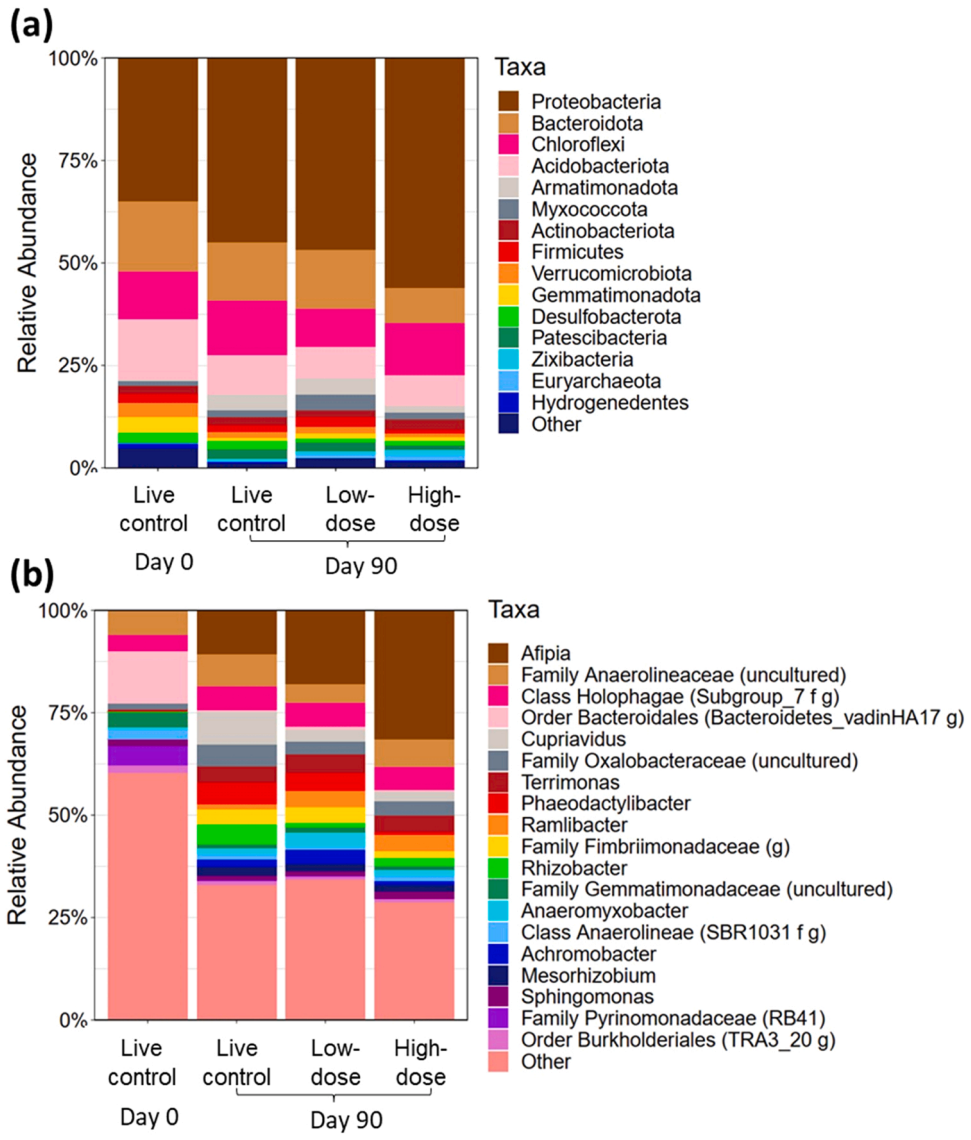

Fig. 4. Relative abundance of microbial community at the (a) phylum level and (b) genus level in the Loring biotransformation experiment.

including 8:2 FTOH.

To identify subtler shifts in the Loring AFB soil microbial community, analysis of microbial genera was performed. A sum of 246 genera was shared among day 90 samples of all three treatments. The predominant 19 genera are shown in Fig. 4b. The abundance of *Afipia* and *Sphingomonas*, both affiliated with phylum Alphaproteobacteria were positively correlated with the concentration of 8:2 FTOH. After the 90-day incubation, the relative abundance of *Afipia* increased from non-detectable levels to 10.58% in the live control. The relative abundance of *Afipia* was further enriched to 17.95% and 31.37% in the low-dose treatment and high-dose treatment, respectively. *Afipia* strains exist widely in freshwater, river sediments, Antarctic habitats, and soil [55,56]. Previous studies showed that isolated *Afipia* strains had the ability to degrade 1,4-dioxane [56] and methanesulfonate [57]. Interestingly, some *Afipia* spp. are capable of degrading different haloacetic acids as α-halocarboxylic acid dehalogenase genes are present in their genomes [58]. Similarly, the relative abundance of *Sphingomonas* was imperceptibly higher in the low-dose treatment (1.18%) at day 90 compared to that in the live control (1.14%), while it was higher in the high-dose treatment (1.81%). *Sphingomonas* spp. are also widespread in soil and aquatic systems [59,60], and are known to be metabolically versatile in degrading organic pollutants, including polycyclic aromatic hydrocarbons [49] and highly chlorinated pesticides using dehalogenases [61]. The relative abundance of *Methanobacterium* (Euryarchaeota) followed a similar trajectory with elevated 8:2 FTOH concentrations as *Afipia* and *Sphingomonas* and increased to 1.24% in the high-dosed treatment after incubation, although they accounted for less than 0.20% in the original soil. Members of *Methanobacterium* were also reported to have the ability to tolerate and degrade halogenated compounds [62,63]. The existence of *Afipia*, *Sphingomonas*, and *Methanobacterium* in AFFF-impacted soil and their increasing abundance after 8:2 FTOH exposure, particularly under the high-dose condition, indicated that species from these genera could be potential 8:2 FTOH degraders and tolerant towards 8:2 FTOH and its biotransformation products.

Despite the difference in microbial community composition, the occurrence of 8:2 FTOH biotransformation in both soils in the present study and in other environmental matrices from prior studies [23,20,21] suggests that microorganisms capable of transforming 8:2 FTOH are not rare in the environment. The similarities in major 8:2 FTOH transformation products detected in Loring AFB soil and Robins AFB soil incubations, indicate that these two soils may share key 8:2 FTOH degraders.

4. Conclusions

This study demonstrated that 8:2 FTOH can be transformed rapidly by indigenous microbial communities from two different AFFF-impacted surface soils under aerobic conditions. More than 90 mol% of 8:2 FTOH was removed, and similar major transformation products were detected during the 90-day incubation period irrespective of the amount of 8:2 FTOH spiked initially and the microbial communities present in the soil. Previously documented 8:2 FTOH biotransformation products, including polyfluorinated substances, like 8:2 FTCA, 8:2 FTUA, 7:2 sFTOH, and 7:3 acid, as well as perfluorinated compounds like PFOA were identified and quantified. Although rarely documented, 7:3 U amide and 3-OH-7:3 acid, which tentatively derived from 7:3 U acid with a non-fluorinated moiety changed, were identified in this study. Polyfluorinated compounds with shorter carbon-chain length (< 10), for example, 6:3 acid, which were produced via a single carbon removal pathway were also identified by non-targeted analysis. Importantly, perfluoroalkane-like compounds, including 1H-perfluoroheptane, 1H-perfluorohexane, and perfluoroheptanal were identified as novel transformation products that have not been previously reported. Of the newly identified transformation products, only 1 H-perfluoroheptane and perfluoroheptanal, were detected in all live reactors containing 8:2 FTOH with both Robins and Loring soils. This result may be due to different transformation extent or rates that resulted from the different microbial populations, and the lower yields of some compounds produced from the lower concentration of spiked 8:2 FTOH.

This work documented that the microbial community composition shifted over the 90-day incubation period after exposure to two dosages of 8:2 FTOH (one-time addition of 170 μg/L and repeated additions of 1,700 μg/L). Exposure to the higher concentration of 8:2 FTOH facilitated not only the detection of novel transformation products, but also the identification of microbes that may be more tolerant of 8:2 FTOH and its biotransformation products by imposing selective pressure. Results suggest that microorganisms in phylum Proteobacteria, such as genera *Afipia* and *Sphingomonas* may be more tolerant of high 8:2 FTOH concentrations. Microorganisms in these two and other genera including *Variovorax*, *Ralstonia*, *Sphingomonas*, *Ancylobacter*, and *Methanobacterium* may be involved as potential degraders. We recognize that phylogenetic identification of taxa using 16S rRNA gene sequencing does not necessarily reflect microbial biotransformation activity. Future studies are needed to assess the viability and function of these potential degraders using additional analyses, e.g., metatranscriptomics and quantitative PCR. Our findings provide insights on 8:2 FTOH biotransformation potential and a starting point for screening microbes engaged in fluorotelomer degradation in the future.

Overall, this study detailed microbial transformation pathways and microbial mechanisms that contribute to fluorotelomer transformations in AFFF-impacted soils. This information may also serve as a reference for predicting the fate of fluorotelomers in the environment that can be used in the development of improved future conceptual site models. Additionally, results from this study provide insights into microbial community changes in response to fluorotelomer contamination that could help in isolating microorganism(s) that can transform fluorotelomers and other PFAS.

CRediT authorship contribution statement

Sheng Dong: Investigation, Methodology, Visualization, Writing – original draft. **Peng-Fei Yan**: Investigation, Methodology, Visualization, Writing – review & editing. **Chen Liu**: Resources, Writing – review & editing. **Katherine E. Manz**: Resources, Writing – review & editing. **Melissa P. Mezzari**: Resources, Writing – review & editing. **Linda M Abriola**: Funding acquisition, Writing – review & editing. **Kurt D. Pennell**: Funding acquisition, Writing – review & editing. **Natalie L. Cápiro**: Conceptualization, Funding acquisition, Supervision, Writing – review & editing.

Environmental implications

Fluorotelomer alcohols (FTOHs) and their biotransformation products, particularly perfluoroalkyl carboxylic acids, are persistent in the environment and human body over time and cause many adverse health effects. This study demonstrated that 8:2 FTOH, a precursor to perfluorooctanoic acid (PFOA), can be biotransformed aerobically in aqueous film forming foam (AFFF)-impacted soils. Such information would be useful for predicting the stability of fluorotelomers in AFFF-impacted sites. It also provides information on biotransformation pathways and potential degraders of fluorotelomers, which is valuable for assessing fate of FTOHs in the environment and devising microbial-based remediation strategies.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

Acknowledgements

This study is supported by the Strategic Environmental Research and Development Program (SERDP), under contract W912HQ-18-C-0014 for project number ER18–1149, "Development and Laboratory Validation of Mathematical Modeling Tools for Prediction of PFAS Transformation, Transport, and Retention in AFFF Source Areas". The authors thank the Air Force Civil Engineer Center and Geosyntec Consultants for assistance in the collection of site materials at Robins AFB. Sheng Dong and Peng-Fei Yan also acknowledge the financial support from the China Scholarship Council, and Auburn University Graduate School and Office of International Programs. The high resolution mass spectrometer used to identify reaction byproducts was partially funded by National Science Foundation (NSF) Major Research Instrumentation (MRI) award CBET-1919870.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.jhazmat.2022.130629.

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