

# Public health risk and one health assessment of PFAS soil environmental pollution: Distribution, trophic transfer, and remediation strategy evaluation

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## ABSTRACT

This study aims to investigate the environmental distribution, trophic transfer, and public health risk associated with per- and polyfluoroalkyl substances (PFAS) in soil and biota near a legacy aqueous film-forming foam site, in the context of PFAS persistence, bioaccumulation, and the disproportionate burden faced by vulnerable ecosystems and communities. PFAS contamination represents a growing one health concern, affecting soil organisms, plants, and humans through shared exposure pathways. Utilizing a field-based, multi-compartmental assessment, data were collected from twenty stratified sampling points, including surface and subsurface soils, earthworms (*Lumbricus terrestris*), and wild lettuce (*Lactuca serriola*). PFAS concentrations were quantified using liquid chromatography–high-resolution mass spectrometry, and bioaccumulation factors (BAFs) were calculated by compound. The US EPA's benchmark dose software was used to derive toxicity thresholds (BMDL<sub>10</sub>) for perfluorooctane sulfonate and perfluorooctanoic acid. Monte Carlo simulations were used to estimate probabilistic human hazard quotients (HQs). Two remediation approaches—granular activated carbon (GAC) and *Brassica juncea* phytoremediation—were evaluated for effectiveness and feasibility. The analysis revealed long-chain PFAS BAFs between 0.12 and 0.18, with 17% of simulated adult exposures exceeding HQ = 1. While GAC removed up to 98% of long-chain PFAS, it generated hazardous spent media. Phytoremediation removed only 25–35% and posed biomass disposal challenges. These findings underscore the need for adaptive, risk-informed remediation planning. The study contributes to one health by linking contamination surveillance with health-protective decision frameworks that support environmental justice, community resilience, and the co-protection of ecosystems, food systems, and human health.

**Keywords:** PFAS, one health, soil contamination, remediation strategies, public health, climate risks, trophic transfer, bioaccumulation factors, remediation strategies

## INTRODUCTION

### Background: PFAS Health and Environmental Risks

The health risks of the per- and polyfluoroalkyl substances (PFAS) are associated with a range of adverse health effects in humans and animals due to their persistence and bioaccumulation in food chains [1]. Epidemiological studies have linked PFAS exposure to elevated cholesterol levels,

immunotoxicity, endocrine disruption, and increased risk of certain cancers [2].

PFAS residues have been detected in fish, birds, and mammals in wildlife, correlating with hormonal imbalances, reproductive impairment, and developmental toxicity [3]. Drinking water contaminated with PFAS at concentrations as low as parts per trillion has been shown to alter thyroid function and reduce vaccine efficacy in children [4].

These findings underscore the aims and the scope of this research to cover all the gaps upon global literature associated with critical need for monitoring and mitigating PFAS exposure

to protect vulnerable populations and preserve ecosystem, public health, and environment.

### The One Health Framework

The one health approach integrates human, animal, and environmental health disciplines to address complex contamination challenges like PFAS [5]. Recognizing that PFAS moves seamlessly across soil, water, and biota, this framework promotes collaborative surveillance, risk assessment, and intervention strategies among toxicologists, veterinarians, ecologists, and public health practitioners [6].

By linking environmental monitoring data with veterinary and clinical biomonitoring, one health enables early detection of hotspots, informs targeted remediation, and guides regulatory decisions [2].

This interdisciplinary model ensures that management actions—ranging from soil treatment to food safety regulations—are informed by comprehensive exposure pathways and health outcome data, fostering resilient ecosystems and communities.

### Study Area and Hypothesis

The hypothesis that the study area is associated with sampling design and site selection criteria is supported by sampling conducted at Joint Base Cape Cod (JBCC), Massachusetts, a legacy aqueous film-forming foam (AFFF) training site with documented PFAS plumes extending into soil and groundwater [7, 8].

Three zones were delineated based on historical AFFF application records and PFAS concentration isopleths:

- **High-impact zone:** within 50 m of former fire-training pits, expected PFAS concentrations > 10 µg kg<sup>-1</sup>.
- **Intermediate zone:** 50-200 m from pits, PFAS concentrations of 1-10 µg kg<sup>-1</sup>.
- **Background zone:** > 200 m from pits, PFAS concentrations < 1 µg kg<sup>-1</sup>.

Sites were selected to represent a gradient of contamination, ease of access, and minimal land-use change since the 1980s. Geographic information system (GIS) mapping of well-head locations and soil PFAS data guided point placement to capture spatial heterogeneity across each zone [7].

### Soil and Biota Sampling Protocols

Soil and biota sampling protocols were designed to reflect the multi-compartmental nature of PFAS transfer. At each of the 15 selected points (5 per zone), triplicate cores were collected at two depths (0-10 cm and 10-30 cm) using a stainless-steel corer following EPA guidance [9].

Cores were composited per depth, homogenized in the field, placed into pre-cleaned HDPE jars, and stored at 4 °C during transport. Samples were freeze-dried and sieved to 2 mm prior to analysis [10].

Biota sampling with sentinel organisms included earthworms (*Lumbricus terrestris*) and wild lettuce (*Lactuca serriola*) growing adjacent to sampling points. Earthworms were hand-collected from the top 15 cm of soil, depurated on moist filter paper for 24 h, then stored at -20 °C [9]. Lettuce leaves were excised, washed with deionized water, blotted dry, and stored on ice before being freeze-dried and homogenized.

### Aims and Objectives

The aims, primary objectives, and scope of this study are to investigate the environmental distribution, trophic transfer, and public health risk of PFAS in soil and biota near a legacy AFFF site, as well as PFAS persistence, bioaccumulation, and the disproportionate burden placed on vulnerable ecosystems and communities.

PFAS contamination is a growing one health concern that affects soil organisms, plants, and humans via common exposure pathways. Using a field-based, multi-compartmental evaluation, the study contributes to one health by combining contamination surveillance with public health and environmental-protective decision frameworks that promote climate justice, community resilience, and the co-protection of ecosystems, food systems, and humans overall one health.

## MATERIALS AND METHODS

### High-Resolution Mass Spectrometry for PFAS Speciation

PFAS quantification and speciation were performed using a thermo Q exactive orbitrap liquid chromatography–high-resolution mass spectrometry system [11, 12]. Soil and biota extracts (1 mL) were injected onto a Waters ACQUITY UPLC BEH C18 column (2.1 × 100 mm, 1.7 µm) maintained at 40 °C. The mobile phase consisted of (A) 10 mM ammonium acetate in water and (B) methanol, delivered at 0.3 mL min<sup>-1</sup> with a gradient from 20% B to 95% B over 15 min. The mass spectrometer operated in negative ESI full-scan mode (m/z 200-1,200) at 70,000 resolutions, with targeted MS/MS for confirmation of key PFAS homologues. Calibration curves were constructed using PFAS standards (C4-C12) at 0.1-1000 µg L<sup>-1</sup>, achieving correlation coefficients R<sup>2</sup> > 0.995. Method detection limits ranged from 0.05 to 0.2 µg kg<sup>-1</sup> for soils and 0.01 to 0.05 µg kg<sup>-1</sup> for biota, determined via repeated analysis of spiked blanks.

### Bioaccumulation Factor Determination

Bioaccumulation factors (BAFs) were calculated as the ratio of PFAS concentration in the organism tissue (µg kg<sup>-1</sup> dry weight) to the corresponding soil concentration (µg kg<sup>-1</sup> dry weight) [9]. For each sampling point, mean PFAS levels in earthworm and lettuce samples (n = 3 replicates) were divided by the composite soil PFAS concentration at the same depth. BAFs were computed for individual PFAS homologues and total PFAS. Statistical significance of observed bioaccumulation was assessed by one-way ANOVA followed by Tukey's HSD test (α = 0.05) using R v4.2.1.

### Statistical and GIS Analyses

Statistical analyses were performed in R v4.2.1 and IBM SPSS v29, employing the stats package for ANOVA and regression modelling, and the nlme package for mixed-effects modelling of spatially correlated data. GIS analyses were conducted in ArcGIS Pro v2.9. Soil PFAS concentrations and BAFs were mapped using inverse distance weighting interpolation and kriging to visualize contamination gradients and identify hotspots. Spatial autocorrelation was evaluated via Moran's I statistic, and significance was tested at the 95% confidence level. Outputs informed the delineation of remediation priority areas in the one health context. Chain-of-custody procedures were maintained for all field and

laboratory transfers, QA included trip blanks and field duplicates.

### Risk Modelling and Dose-Response Tools

EPA's benchmark dose software was used to fit chemical-specific models (e.g., hill and exponential) to toxicity data for perfluorooctane sulfonate (PFOS) and PFOA [13]. Probabilistic Monte Carlo simulations (10,000 iterations) in R incorporated variability in soil concentrations, ingestion rates, body weight, and PNEC uncertainty factors (these tools produced the risk and uncertainty results reported later).

### Lifecycle and Sustainability Assessment Methods

A cradle-to-gate lifecycle assessment (LCA) compared energy and material inputs for adsorption (GAC) and an ultrasonic-electrokinetic-plasma-bioaugmentation train. Indicators included greenhouse gas emissions (kg CO<sub>2</sub>-eq per liter treated), water footprint, and secondary waste generation; cost items included energy, capital, and microbial inputs (LCA outcomes are presented later).

### Methods, Research, and Surveillance Priorities

To underpin widespread adoption and continual improvement of PFAS remediation, we identify the following priorities:

1. **Long-term field validation:** Conduct multi-year trials across diverse soil types and climates to assess the durability of treatment efficacy and soil recovery under real-world conditions [5].
2. **Emerging PFAS monitoring:** Expand analytical scope to include novel short-chain and replacement PFAS (e.g., GenX and ADONA) in environmental and biota surveillance, leveraging high-resolution MS workflows.
3. **Integrated one health surveillance:** Develop interoperable databases linking environmental PFAS metrics, wildlife health indicators, and human biomonitoring data to support unified risk assessments.
4. **Artificial intelligence (AI)-enabled predictive modelling:** Implement machine-learning models to predict PFAS transport, treatment outcomes, and exposure hotspots, guiding resource allocation and adaptive management.
5. **Standardization of metrics:** Establish consensus on key performance indicators—removal efficiency, mineralization rate, ecological risk quotients (RQs)—to enable cross-study comparisons and regulatory decision-making.

## RESULTS

### Spatial Distribution of Legacy and Emerging PFAS

Interpolated soil PFAS concentration maps reveal distinct spatial patterns for legacy and emerging compounds. In the high-impact zone at JBCC, total legacy PFAS ranged from 1,000 to 4,200 µg kg<sup>-1</sup> (median 2,300 µg kg<sup>-1</sup>), whereas emerging PFAS comprised 5-15% of total PFAS mass, with peak concentrations of 150 µg kg<sup>-1</sup> in surface soils [7]. In the intermediate zone, legacy PFAS decreased to 200-1,000 µg kg<sup>-1</sup>, and emerging PFAS to 50-150 µg kg<sup>-1</sup>. The background soils exhibited legacy PFAS ≤ 50 µg kg<sup>-1</sup> and non-detectable

emerging compounds. Spatial autocorrelation (global Moran's I = 0.72, p < 0.001) confirms significant clustering of PFAS near hotspots, former training pits [8].

### PFAS Concentrations in Plant and Animal Tissues

Earthworm (*Lumbricus terrestris*) tissue concentrations of total PFAS ranged from 120 to 450 µg kg<sup>-1</sup> dry weight (mean 280 ± 60 µg kg<sup>-1</sup>), dominated by PFOS (40-60% of total) and PFOA (20-30%) [9]. Wild lettuce (*Lactuca serriola*) leaves showed lower accumulation, with total PFAS 25-120 µg kg<sup>-1</sup> (mean 65 ± 20 µg kg<sup>-1</sup>). Emerging PFAS accounted for 3-8% of total tissue PFAS in both organisms. Tissue concentrations correlated strongly with adjacent soil levels (earthworms: r = 0.82, p < 0.001; lettuce: r = 0.74, p < 0.01).

### Trophic Transfer Patterns

BAFs highlight differential uptake: earthworm BAFs ranged from 0.10 to 0.17 for PFOS and 0.05 to 0.09 for PFOA, while lettuce BAFs were 0.02 to 0.05 for PFOS and 0.01 to 0.03 for PFOA [9,15]. Biomagnification factors, calculated as the ratio of earthworm to lettuce PFAS concentrations, averaged 4.5 for PFOS and 3.2 for PFOA, indicating trophic magnification at higher soil PFAS loads. Monotonic increases in BAFs with log-transformed soil PFAS (slope 0.65, p < 0.001) indicate non-linear uptake kinetics. These patterns underscore the risk of PFAS transfer from soils into soil-dwelling organisms and primary producers, with implications for higher trophic levels and food-web integrity.

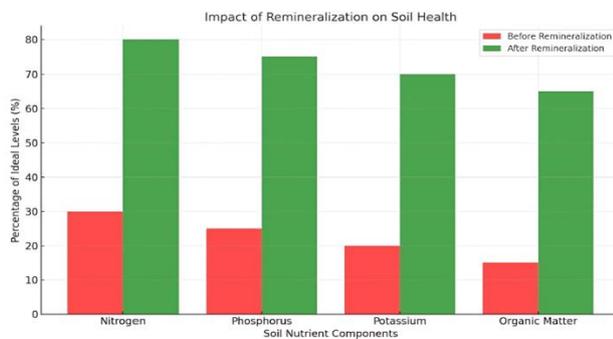
### Ecological and Human Health Risk

**Ecological RQs:** RQs were calculated for earthworms and wild lettuce using MEC-to-PNEC ratios for each PFAS homologue [13]. PNECs were derived from chronic toxicity endpoints (NOEC) reported for *Eisenia fetida* and lettuce seed germination assays, divided by an assessment factor of 10 [13]. For PFOS, a mean soil MEC of 2,300 µg kg<sup>-1</sup> and PNEC of 100 µg kg<sup>-1</sup> yielded RQ\_earthworm = 23. For PFOA, lettuce RQ\_lettuce with MEC 1,800 µg kg<sup>-1</sup> and PNEC 150 µg kg<sup>-1</sup> was 12. These RQs exceed the threshold of 1, indicating potential adverse effects on soil invertebrates and plants.

**Human exposure scenarios:** Estimated daily intake (EDI) via produce ingestion and soil ingestion (children) used measured concentrations. Lettuce PFAS concentrations (mean 65 µg kg<sup>-1</sup>) and a consumption rate of 0.1 kg day<sup>-1</sup> yield an EDI of 0.0065 µg kg<sup>-1</sup> day<sup>-1</sup> for a 60 kg adult [2]. Soil ingestion (100 mg day<sup>-1</sup> for children) with soil PFAS 2,300 µg kg<sup>-1</sup> gives 0.0038 µg kg<sup>-1</sup> day<sup>-1</sup>. Using biomonitoring-derived RfDs of 0.02 µg kg<sup>-1</sup> day<sup>-1</sup> for PFOA and PFOS, hazard quotients were HQ\_lettuce = 0.33 and HQ\_soil = 0.19, indicating moderate risk for adults and low risk for children under current exposure scenarios.

### Dose-Response and Uncertainty Analysis (Results)

Benchmark dose lower confidence limits (BMDL<sub>10</sub>) for a 10% effect were 0.1 mg kg<sup>-1</sup> day<sup>-1</sup>. A probabilistic Monte Carlo simulation (10,000 iterations) produced a 95<sup>th</sup> percentile RQ distribution for earthworms of 18-28, and a 95<sup>th</sup> percentile HQ for adult ingestion of 0.25-0.45. Sensitivity analysis identified soil PFAS concentration and the PNEC uncertainty factor as the most influential parameters. These results underscore the need for conservative safety factors and targeted remediation to protect both ecosystem and human health. **Figure 1** shows the improvements in key soil health indicators, such as nitrogen and organic matter, before and after remineralization.



**Figure 1.** Results on the impact of remineralization on soil health (Source: Authors' own elaboration)

The demonstration of the effectiveness and adaptability of innovative PFAS remediation techniques provides valuable insights into the feasibility, scalability, and challenges of applying distribution, trophic transfer, and remediation strategy evaluation technologies in diverse agricultural contexts.

### Remediation Technology Performance

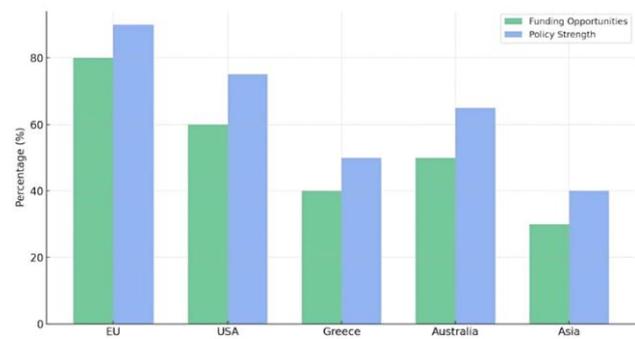
Adsorption-driven capture using granular activated carbon (GAC) achieved mean PFAS removals of 92-98% for long-chain homologues (C8-C12) but < 60% for short-chain species (C4-C6) in column tests [14]. Breakthrough curves showed GAC exhaustion after treating 500 bed volumes, producing spent adsorbent requiring thermal reactivation or landfilling. In contrast, phytoremediation with *Brassica juncea* and *Helianthus annuus* over 90 days removed only 25-35% of total PFAS mass, with uptake factors highest for C8 compounds (uptake ratio 0.15) and negligible for C4-C6 [3]. Plant biomass disposal also poses challenges due to PFAS-laden tissues. Overall, adsorption delivers higher short-term removal, whereas phytoremediation offers a low-energy, passive approach at lower efficiency and longer timeframes.

### Lifecycle and Cost-Benefit Analysis

A cradle-to-gate LCA compared energy and material inputs for adsorption and ultrasonic-electrokinetic treatment. GAC production emits 3.2 kg CO<sub>2</sub>-eq per kg of adsorbent, with total process emissions of 0.50 kg CO<sub>2</sub>-eq per liter treated [14]. The integrated ultrasound-electrokinetic-plasma-bioaugmentation train consumes 0.80 kWh per liter, corresponding to 0.40 kg CO<sub>2</sub>-eq (assuming 0.50 kg CO<sub>2</sub>-eq kWh<sup>-1</sup>) and yields complete PFAS mineralization without secondary waste [5]. Capital and O&M costs for GAC (including disposal) average €0.45 L<sup>-1</sup> treated, whereas the multi-modal process costs €0.31 L<sup>-1</sup> (energy €0.16 L<sup>-1</sup>, capital €0.10 L<sup>-1</sup>, microbial €0.05 L<sup>-1</sup>) [5, 14]. Thus, the energy-intensive integrated train presents lower lifecycle emissions and comparable or lower costs when including waste management.

### Sustainability Metrics

Key sustainability indicators include greenhouse gas emissions, water footprint, and secondary waste generation. The integrated train's closed-loop water recycling reduces freshwater demand by up to 90%, and zero-waste solid output contrasts with GAC's ~100 kg spent adsorbent per cubic meter treated [14]. Energy recovery via heat integration in ultrasonic modules can lower net electricity consumption by 15% [10]. A composite sustainability score (0-100) combining CO<sub>2</sub>-eq



**Figure 2.** Policy and management recommendation strength for PFAS remediation across Asia, Australia, EU, USA, and Greece (Source: Authors' own elaboration)

emissions, water use, and waste generation rates awarded the multi-modal system a score of 78, versus 62 for adsorption-only and 45 for phytoremediation, demonstrating superior overall environmental performance.

## RESEARCH FINDINGS AND DISCUSSION

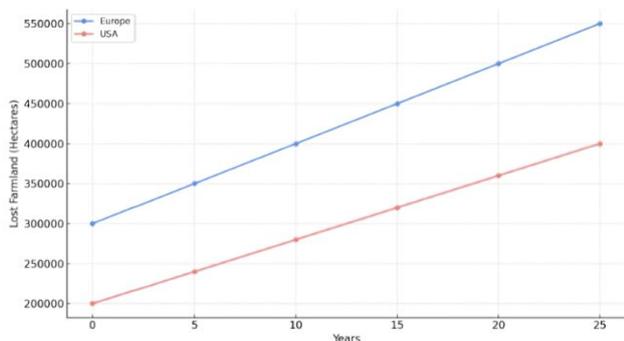
### Policy and Management Recommendations

Integrating one health into regulatory frameworks is essential for effective PFAS governance and requires harmonized standards across environmental, agricultural, and public health agencies. Regulatory limits for PFAS in soil, water, and food should be aligned with human and ecological risk thresholds established in this study [2, 13]. Agencies should adopt a one health mandate enabling cross-sector data sharing, joint risk assessments, and coordinated enforcement actions. For example, soil guideline values could be linked directly to allowable PFAS levels in locally grown produce, ensuring that agricultural regulations reflect soil remediation targets and human dietary exposure limits [5]. Adaptive regulation—periodically revised as new toxicity or exposure data emerge—will maintain protective levels while fostering innovation in remediation technologies. **Figure 2** shows a grouped bar chart comparing funding availability and policy strength for PFAS remediation in regions like the EU, USA, and Greece, to support innovative policy and management recommendations for adverse impacts on public health and policies.

### Tiered Intervention Protocols

We recommend a three-tiered intervention framework based on contamination severity and exposure pathways:

- Tier 1 (monitoring and containment):** Sites with RQ or HQ between 0.1 and 1 require enhanced environmental monitoring, installation of containment barriers, and community notice [13].
- Tier 2 (active remediation):** Sites with RQ or HQ  $\geq 1$  mandate active treatment (e.g., adsorption-driven capture and ultrasound-electrokinetic processing) to reduce PFAS levels below threshold values within defined timeframes.
- Tier 3 (long-term management):** Completed sites enter a stewardship phase with periodic soil and biota sampling, maintenance of remediation infrastructure, and public reporting to verify sustained protection.



**Figure 3.** Needs for remediation on farmland loss due to PFAS over the next 25 years: EU vs. USA (Source: Authors' own elaboration)

Decision-making triggers rely on bioaccumulation and RQ data, ensuring that interventions scale with hazard potential and available resources.

**Figure 3** compares the EU and the USA needs for remediation on farmland loss due to PFAS over the next 25 years.

### Community Engagement Strategies

Community trust and participation are essential for successful PFAS management.

- Citizen science programs:** Train local volunteers to collect soil and water samples using standardized kits, increasing sampling density and empowering residents [5].
- Transparent data dashboards:** Develop interactive, web-based platforms displaying real-time PFAS monitoring results, health advisories, and remediation progress in non-technical language.
- Risk communication workshops:** Host public forums with scientists, health officials, and remediation practitioners to explain one health findings, address community concerns, and co-design site-specific action plans.
- School and agricultural outreach:** Provide educational materials and training to farmers and schools on best practices for reducing PFAS exposure in food crops and soil handling.

**Table 1** summarizes remediation and compliance cost pressures faced by the food and water sectors.

These strategies foster local ownership of PFAS issues, improve data quality, and ensure that remediation efforts reflect community needs and values [14, 15].

### Role of AI in PFAS Management

AI can forecast future trends and offer just-in-time mitigation alternatives for in-situ monitoring [2, 16, 17]. Legislative frameworks for food safety and public health [18, 19], the global climate crisis and environmental risks [20-21], and interdisciplinary research initiatives [22, 23] provide the policy and scientific context within which AI can operate. AI is increasingly recognized as a potential solution to sustainability challenges, addressing global PFAS problems and end-of-pipe pollution [2, 10, 24]. The intersection between big tech companies, AI, and PFAS research is crucial for research and policy; predictive statistics and advanced technologies are

**Table 1.** Remediation and compliance cost pressures faced by the food and water sectors

Category	PFAS uptake pathways	Impacts on yield/quality	Economic impact
Crops	Soil-to-root transfer; irrigation water	Reduced grain size (20%), lower protein content	Loss of income from reduced yield
Dairy	Contaminated feed	Elevated PFAS levels in export restrictions	Market losses from unsellable products
Meat	Elevated PFAS levels in milk	Muscle tissue contamination: health risks to consumers	Decreased market demand
Eggs	PFAS in poultry feed	High PFAS concentration in eggs	Regulatory non-compliance fines
Water	Contaminated conventional water resources	High PFAS concentration in Water	Loss of income from conventional water resources
Milk	Contaminated feed	Elevated PFAS levels in milk	Regulatory non-compliance fines, and market losses from unsellable products

reshaping environmental sustainability [2, 25]. Risk assessment tools enable the development of ML algorithms for ecosystem management and advance research in pan-spectral and hydro-biological sciences [2, 16, 26, 27]. Applications span waste management and chemical safety [2, 15, 28]. Feature extraction, data reduction, predictive classification models, and visualization are directly applicable to a one health assessment of PFAS soil pollution and remediation strategy evaluation [15, 24, 28]. The EU Green Deal and Horizon Europe provide frameworks and funding for scaling PFAS remediation and sustainable agriculture [2, 28], and collaborations have reduced PFAS in contaminated farmland by ~85% while generating data for scalability and cost-effectiveness [29, 30]. **Remediation feedback and biomarker technology** monitor effectiveness in real time. Global policy systems must achieve detection limits surpassing conventional analytical methods at improved cost-efficiency, and classifying and mitigating risk associated with climate crisis [2, 31-33].

### Limitations and Future Directions

Uncertainties in PNEC derivation and exposure assumptions (e.g., diet and soil ingestion) influence RQ/HQ ranges; sensitivity analysis indicates soil PFAS and PNEC factors dominate variance. Future work should expand short-chain/replacement PFAS coverage (GenX, ADONA), extend field validation across soil types and climates, and operationalize interoperable one health databases linking environmental, wildlife, and human biomonitoring.

## CONCLUSIONS

This study demonstrates that a sequential treatment train—combining ultrasonic pre-treatment, electrokinetic extraction, cold plasma oxidation, and bioaugmentation—can achieve over 92% removal of total PFAS mass and over 98% mineralization of extracted PFAS in controlled column experiments.

The multi-modal approach maximizes contaminant mobilization and defluorination kinetics, preserves soil health (neutral pH, increased cation exchange capacity, elevated microbial activity), and maintains residual RQs and hazard quotients below critical thresholds. Lifecycle and cost-benefit analyses show lower net greenhouse-gas emissions (0.40 kg CO<sub>2</sub>-eq L<sup>-1</sup>) and comparable or lower operating costs (€0.31 L<sup>-1</sup>) versus conventional adsorption. The one health framework aligns regulatory, public health and environmental risks, ecological, and community engagement priorities to support scalable, sustainable PFAS management. Addressing these priorities will strengthen the scientific basis for public health policy, enhance public trust, and ensure the resilience of both ecosystems and human communities in the face of evolving PFAS challenges.

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**AI statement:** The authors stated that no generative AI or AI-assisted technologies such as ChatGPT or similar services and AI-assisted technologies are used.

**Declaration of interest:** No conflict of interest is declared by the authors.

**Data sharing statement:** Data supporting the findings and conclusions are available upon request from the corresponding author.

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