



# Environment and Natural Resources Trust Fund

2022 Request for Proposal

## General Information

**Proposal ID:** 2022-188

**Proposal Title:** PFAS Fungal-woodchip Filtering System

## Project Manager Information

**Name:** Jiwei Zhang

**Organization:** U of MN - College of Food, Agricultural and Natural Resource Sciences

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## Project Basic Information

**Project Summary:** Develop and implement a fungal filtering system that combines the benefits of both waste wood chips and soil fungi to sequester and degrade PFAS leachate from contaminated waste sites.

**Funds Requested:** \$189,000

**Proposed Project Completion:** June 30 2024

**LCCMR Funding Category:** Small Projects (H)

**Secondary Category:** Methods to Protect, Restore, and Enhance Land, Water, and Habitat (F)

## Project Location

**What is the best scale for describing where your work will take place?**

Statewide

**What is the best scale to describe the area impacted by your work?**

Statewide

**When will the work impact occur?**

During the Project and In the Future

## Narrative

**Describe the opportunity or problem your proposal seeks to address. Include any relevant background information.**

Per- and Polyfluoroalkyl Substances (PFAS) are a large group of emerging environmental contaminants that are globally ubiquitous. These “forever chemicals” are highly recalcitrant and possess the capability to persist in the environment and accumulate in living organisms. PFAS exposure is increasingly being linked to a variety of adverse health outcomes. Regarding this, strategies to remediate PFAS polluted environments are urgently needed.

Not only the water systems but also soils have been found to be large reservoirs of PFAS chemicals, even in locations distant from any known source. The ubiquitous presence of PFAS in soils around the world is concerning and will likely have long-term ramifications on attempts to remediate contaminated waters. In Minnesota, MPCA (Minnesota Pollution Control Agency) and MDH (Minnesota Department of Health) have discovered PFAS pollution throughout the state since the early 2000s. Among these sites, landfills represent a large repository for PFAS, with 98 out of 101 MPCA tested landfill sites found PFAS and alarmingly 59 sites had detectable levels exceeding health guidelines. A cost-effective means to intercept leachate from landfills and to remove PFAS pollutants, before they can escape to groundwater, will be critical for conserving Minnesota’s environment.

**What is your proposed solution to the problem or opportunity discussed above? i.e. What are you seeking funding to do? You will be asked to expand on this in Activities and Milestones.**

Using woodchips which have been proven to filter other pollutants such as nitrogen and metals from water combined with wood decay fungi which have demonstrated good results in removing contaminants.

**What are the specific project outcomes as they relate to the public purpose of protection, conservation, preservation, and enhancement of the state’s natural resources?**

Development of a cost-effective method to intercept PFAS before it enters groundwater and degrades it in-situ using wood-decay fungi to begin controlling PFAS pollution from the array of contaminated sites throughout Minnesota. Establishing a platform for the further study of PFAS bioremediation to inform long-term efforts to remove PFAS from soil and water providing insights into fungal metabolic capabilities that can be applied to improve not just PFAS remediation but a variety of other applications

## Activities and Milestones

### Activity 1: Screening for an Improved PFAS degrader

**Activity Budget:** \$37,200

#### Activity Description:

The goal of objective one is to identify fungal species able to grow using PFAS as a nutrient source. This will involve inoculating up to two dozen wood-decay fungi onto to growth media plates containing Perfluorooctanoic acid (PFOA) a representative PFAS as the sole carbon source meaning if a fungus is to grow it must be able to utilize PFOA. However, PFAS are known to be difficult to use as a food source so a second screening will be conducted using plates containing a small concentration of glucose in addition to PFOA as a carbon source if initial screening is not fruitful. Other's work has demonstrated that supplementation with additional nutrient sources can improve microbial degradation of PFAS as these substances can be too tough to swallow alone so to speak and the additional nutrients act to stimulate the production of fungal enzymes that can degrade recalcitrant substrates. This step is expected to take 2 to 3 months depending on fungal growth rates and if the second screening is required. The Zhang lab possesses the supplies and expertise to work with fungi so screening fungi for nutrient utilization is a routine task.

#### Activity Milestones:

Description	Completion Date
Setup Initial Cultures for Screening	July 31 2022
Complete Initial Screening	August 31 2022
Complete Second Screening	September 30 2022
Select Candidate	October 31 2022

### Activity 2: Laboratory Scale testing of Fungal-filtering system

**Activity Budget:** \$74,400

#### Activity Description:

Using the identified organism(s) from the initial screening a benchtop scale experiment using a soil jar test and default parameters (~2inch wood chips, pH 5, 50% moisture, defined nitrogen levels) will be conducted. Soil and wood chips will be inoculated with candidate fungi, and C13 labelled PFOA will be added to simulate PFAS pollution. These components will be sealed inside a sterile container creating a closed and controllable system in which to monitor the experiment. The C13 labelled PFOA will facilitate LC/MS measurement of PFOA and its degradation products as well as allowing for the carbon originating from the PFOA to be tracked through the fungal-woodchip system providing a means to confirm PFOA degradation by the fungi by detecting the incorporation of labelled PFOA carbon into fungal biomass. Parameters will be adjusted to optimize fungal growth and enzyme production to promote PFAS degradation. LC/MS will be performed by the University of Minnesota's Center for Mass Spectrometry and Proteomics to determine PFAS degradation. Multiple iterations of this experiment will be conducted to determine the optimal conditions for fungal PFAS degradation information that is currently lacking in the literature facilitating further studies into fungal PFAS metabolism.

#### Activity Milestones:

Description	Completion Date
Soil Jar Test Assembly	November 30 2022
Parameter optimization	May 31 2023
LC/MS Analysis of Samples	June 30 2023

### Activity 3: Field Application of Fungal-filtering System

**Activity Budget:** \$77,400

**Activity Description:**

Working with collaborators from Wenck Environmental Consulting a suitable field site near the Twin Cities will be identified. Wenck will support the Zhang lab by providing access to the site and equipment necessary for the installation of the pilot fungal-filter. Wenck has agreed to provide a modified jobox (secure steel chest) previously used by Wenck to test a different filter system. The jobox is a sealable container modified with a grate in the side allowing water and small particulates to run through the filter material installed inside. Concurrently with lab scale testing the fungal-filter system (woodchips and fungi identified in screening) will be installed in a contaminated landfill at a depth of one meter using the jobox to contain the experiment. Landfill leachate will filter through the fungi-woodchip application and PFAS concentrations will be monitored over the course of the experiment. PFAS and degradation products will be determined with LC/MS before and after pyrolysis to first determine PFAS absorption and degradation, and then to confirm removal via pyrolysis. Pyrolysis will be performed using the equipment and expertise of Dr. Ruan who has agreed to allow us the use of the well-established pyrolysis lab to produce biochar from the recycled filter system.

**Activity Milestones:**

Description	Completion Date
In Consultation with Wenck Select Field Site	August 31 2022
Install Field Application	October 31 2022
Project Cleanup- Removal of Field Application	May 31 2024
Pyrolysis and LC/MS analysis	May 31 2024
Final Data Compilation and Project Write-up	June 30 2024

## Project Partners and Collaborators

Name	Organization	Role	Receiving Funds
Roger Ruan	University of Minnesota	Co-PI on the project, working on using "pyrolysis" to heat treat the sequestered PFAS for the complete removal.	Yes

## Long-Term Implementation and Funding

**Describe how the results will be implemented and how any ongoing effort will be funded. If not already addressed as part of the project, how will findings, results, and products developed be implemented after project completion? If additional work is needed, how will this be funded?**

The data gathered will be used to inform further research into the genes and metabolic pathways involved in PFAS degradation and to further improve the field application.

## Project Manager and Organization Qualifications

**Project Manager Name:** Jiwei Zhang

**Job Title:** Assistant Professor

**Provide description of the project manager's qualifications to manage the proposed project.**

The project managers Dr. Jiwei Zhang (PI) and Dr. Roger Ruan (Co-PI) have enormous combined project management experiences, including LCCMR projects. Each manager's roles have been clearly defined, deliberated to avoid 'scope creep' that can overburden/underfund projects. In the proposal, we have stated feasibility statements and have outlined the pitfalls. We have also integrated tiered strategies with alternative means to achieve goals. In terms of project management, we plan to deliberately enable a science-forward and open group dynamic, with uniform data handling (see DMP), hands-on involvement of students, postdocs, and PIs in collaboration, and sharing results transparently via lectures and data sharing.

Managers' research experiences enable them to manage the proposed project. Dr. Zhang is currently leading a fungal lab at the University of Minnesota. Dr. Zhang and his team, including postdocs and students, are interested in dissecting the fungal pathways involved in organic polymers' degradation to sustain and conserve the local and national environments. These fungal pathways are believed to be fundamentally crucial for the biodegradation of pollutant polymers such as the "forever" chemical PFAS. Co-PI Dr. Ruan's team also has tremendous experience in hazardous pollutants' remediation.

Relevant to this proposal, Zhang lab has constructed a wealth of combined toolkits that can be adopted for fungal screening, culturing, polymer decomposition, and metabolites detection to ensure the proposed project's success. Also, Zhang lab is well equipped with cutting-edge systems biology approaches, enabling the successful dissection of the fundamental biodegradation pathways of PFAS in fungi. Ruan's lab is well equipped for using pyrolysis to remove PFAS and generate biochar as a valuable byproduct ultimately.

This project will generate breakthrough findings to the fungal remediation of PFAS. We know this from experience, and our proposal is poised for high-quality, mission-relevant science.

**Organization:** U of MN - College of Food, Agricultural and Natural Resource Sciences

**Organization Description:**

The College of Food, Agricultural, and Natural Resource Sciences (CFANS) is one of seventeen colleges and professional schools at the University of Minnesota. The Department of Bioproducts and Biosystems Engineering of CFANS, where Dr. Zhang serves as a faculty, is organized for discovering and teaching solutions for the sustainable use of renewable resources and the enhancement of the environment. These missions well align with this proposal, and the facilities, equipment, and other resources owned by both the College and Department will ensure the success of this project.



## Budget Summary

Category / Name	Subcategory or Type	Description	Purpose	Gen. Ineligible	% Benefits	# FTE	Classified Staff?	\$ Amount
<b>Personnel</b>								
Assistant Professor		PI			36.5%	0.5		\$30,526
Graduate Student Assistant		Assist PI in carrying out project			19.9%	2		\$124,543
Professor/Director		Co-PI			36.5%	0.04		\$9,995
							<b>Sub Total</b>	<b>\$165,064</b>
<b>Contracts and Services</b>								
Lab services	Professional or Technical Service Contract	CMSP will be performing LC/MS on our behalf				1		\$2,936
							<b>Sub Total</b>	<b>\$2,936</b>
<b>Equipment, Tools, and Supplies</b>								
	Tools and Supplies	Laboratory Supplies year 1	Materials to setup and perform laboratory procedures: screening, laboratory scale testing, and analysis					\$15,000
	Tools and Supplies	Laboratory supplies and equipment year 2	Tools to install field application and sample collection supplies to facilitate monitoring of field application					\$3,000
							<b>Sub Total</b>	<b>\$18,000</b>
<b>Capital Expenditures</b>								
							<b>Sub Total</b>	-
<b>Acquisitions and Stewardship</b>								
							<b>Sub Total</b>	-

<b>Travel In Minnesota</b>								
	Miles/ Meals/ Lodging	840 miles/yr at .56, Two people approx. 15 trips( up to two trips for installation and 13 trips for monitoring at monthly followed by quarterly intervals)intervals	Travel to and from field site for installation and monitoring (year 2)					\$3,000
							<b>Sub Total</b>	<b>\$3,000</b>
<b>Travel Outside Minnesota</b>								
							<b>Sub Total</b>	-
<b>Printing and Publication</b>								
							<b>Sub Total</b>	-
<b>Other Expenses</b>								
							<b>Sub Total</b>	-
							<b>Grand Total</b>	<b>\$189,000</b>

Classified Staff or Generally Ineligible Expenses

Category/Name	Subcategory or Type	Description	Justification Ineligible Expense or Classified Staff Request
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Non ENRTF Funds

Category	Specific Source	Use	Status	Amount
<b>State</b>				
			<b>State Sub Total</b>	-
<b>Non-State</b>				
			<b>Non State Sub Total</b>	-
			<b>Funds Total</b>	-

## Attachments

### Required Attachments

#### *Visual Component*

File: [145692a2-310.pdf](#)

#### *Alternate Text for Visual Component*

The use of a combination of woodchips and wood-decay fungi to control PFAS and metals leachate originating from contaminated waste sites. Rain or irrigation carries pollutants which can then contaminate areas outside the waste site eventually entering groundwater. This leachate is filtered through the fungal biofilter installed around the periphery of the site intercepting the pollutants allowing for the absorption, degradation, and removal of PFAS....

### Optional Attachments

#### *Support Letter or Other*

Title	File
Institutional Approval to submit	<a href="#">966a5844-045.pdf</a>
LCCMR Proposal	<a href="#">27b16835-b8a.pdf</a>
CMSP Letter of Support	<a href="#">b9d916f7-fdb.pdf</a>
Wenck Letter of Support	<a href="#">d0d00070-f03.pdf</a>

## Administrative Use

**Does your project include restoration or acquisition of land rights?**

No

**Does your project have potential for royalties, copyrights, patents, or sale of products and assets?**

Yes

**Do you understand and acknowledge IP and revenue-return and sharing requirements in 116P.10?**

Yes

**Do you wish to request reinvestment of any revenues into your project instead of returning revenue to the ENRTF?**

No

**Does your project include original, hypothesis-driven research?**

Yes

**Does the organization have a fiscal agent for this project?**

No



Figure 1: Using a fungal Filter to reduce PFAS leachate